UTS UNIVERSITY OF TECHNOLOGY SYDNEY

The Development of a Clinical Practice Guideline for the Management of Increased Intestinal Permeability

by Bradley Graeme Leech

Thesis submitted in fulfilment of the requirements for the degree of

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under the supervision of Professor David Sibbritt, Doctor Amie Steel and Doctor Erica McIntyre

University of Technology Sydney
Faculty of Faculty of Health, Australian Research Centre in Complementary and Integrative Medicine

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CERTIFICATE OF ORIGINAL AUTHORSHIP

I, Bradley Graeme Leech, declare that this thesis, is submitted in fulfilment of the requirements for the award of Doctor of Philosophy (Public Health), in the Faculty of Health at the University of Technology Sydney.

This thesis is wholly my own work unless otherwise referenced or acknowledged. In addition, I certify that all information sources and literature used are indicated in the thesis.

This document has not been submitted for qualifications at any other academic institution.

This research is supported by the Australian Government Research Training Program.

Signature:

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FORMAT OF THIS THESIS

This thesis is structured in the format of Thesis by Compilation. It presents a single body of work comprised of traditional thesis chapters and published articles or documents involved in the development of a clinical practice guideline. Where articles have been published, they have been embedded within the relevant chapters of this thesis. A description of the published publication with relevant notes are mentioned at the beginning of the relevant chapter. A copy of the published work with the journal formatting is included in the Appendices. A list of these articles and details on authorship contributions are provided below.

PUBLISHED WORK INCLUDED IN THE THESIS

Of the three manuscripts incorporated into this thesis, all have been published. A further three reports have been developed as part of the clinical practice guideline. The manuscripts and reports are as follows:

1. **Chapter 2** Leech, B, McIntyre, E, Steel, A, Sibbritt, D “Risk factors associated with intestinal permeability in an adult population: A systematic review", The International Journal of Clinical Practice, 2019, Vol 73, 10.
2. **Chapter 3** Leech, B, McIntyre, E, Steel, A, Sibbritt, D “Clinical practice guideline for the management of increased intestinal permeability: Guideline Development Process”, University of Technology Sydney.
3. **Chapter 4** Leech, B, McIntyre, E, Steel, A, Sibbritt, D “Health-seeking behaviour, views and preferences of adults with suspected increased

intestinal permeability: A cross-sectional survey of Australian adults", Integrative Medicine Research, 2022, Vol 11, 1.

4. **Chapter 5** Leech, B, McIntyre, E, Steel, A, Sibbritt, D "The Subjective Well-being and Health-Related Quality of Life of Australian Adults with Increased Intestinal Permeability and Associations with Treatment Interventions", The Journal of Alternative and Complementary Medicine, 2021, Vol 27, 12.
5. **Chapter 6** Leech, B, McIntyre, E, Steel, A, Sibbritt, D "Clinical practice guideline for the management of increased intestinal permeability: Technical Report", University of Technology Sydney.
6. **Chapter 8** Leech, B, McIntyre, E, Steel, A, Sibbritt, D "Clinical practice guideline for the management of increased intestinal permeability: IP Guideline", University of Technology Sydney.

STATEMENT OF AUTHOR CONTRIBUTIONS TO JOINTLY AUTHORED WORKS CONTAINED IN THIS THESIS

This thesis contains three articles as part of Chapters 2, 4 and 5 and three reports in Chapters 3, 6 and 8. The articles have been written in joint authorship and published in peer-reviewed journals, while the reports represent the collective body of work for a clinical practice guideline. As the lead researcher and candidate for the award, I have been the primary author of each article, providing majority of the contribution for the development of research topics and research questions, performing analysis, conceptualising the data collection, drafting manuscripts for publication, submitting the articles for publication, and responding to peer-review feedback. Professor David Sibbritt, Doctor Amie Steel and Doctor Erica McIntyre provided support in each of these areas.

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ABSTRACT

Background: Increased intestinal permeability (IP) may play an important role in health and disease. Clinicians treating people with IP use a combination of therapeutic interventions, with many interventions having little evidence. This thesis aims to improve the management of IP by clinicians in primary care settings in Australia by developing an evidence-based clinical practice guideline that considers the views and preferences of people with IP in the developed recommendations.

Methods: A cross-sectional survey design was employed to explore the views and preferences of Australian adults with suspected IP (Phase One n=589). A clinical practice guideline was developed based on the National Health and Medical Research Council (NHMRC) guidelines for guidelines (Phase Two). The level of evidence for each recommendation was determined based on the NHMRC grades for recommendations and the NHMRC Evaluation of Evidence process. Eight stakeholders participated in a cross-sectional survey to explore each recommendation's understanding, agreement, importance, and appropriateness.

Results: Phase One found that most Australian adults with suspected IP (56.2%) are self-diagnosing their condition, with many of these individuals (56.7%) preferring to be assessed using an accurate method by a general practitioner or naturopath. Regarding the treatment of IP, participants reported using dietary products (87.9%), dietary supplements (72.9%) and lifestyle therapies (54.6%) for managing IP. The out-of-pocket cost associated with managing IP suggests a financial burden; participants that struggle financially spend significantly more

(mean=$2963) on dietary supplements compared to participants who find it easy to live on their available income ($1918) (p=0.015). Participants had worse subjective well-being (SWB) compared to the Australian population ($p < 0.001$). Self-reported improvement in IP was a significant predictor of SWB and health-related quality of life (HRQoL) (β=10.70, $p < 0.001$). Furthermore, the number of days IP affects daily living correlated with SWB and HRQoL ($p < 0.001$). Phase Two produced a total of 38 recommendations consisting of 27 evidence-based recommendations, seven practice points and four consensus-based recommendations. These recommendations provide clinicians with beneficial dietary choices and dietary supplements while suggesting interventions that are ineffective and should be avoided. Furthermore, according to key stakeholders, most of the developed recommendations were accepted and acknowledged as important and appropriate for clinicians to follow.

Conclusion: The research findings presented in this thesis may optimise patient care, improve health outcomes, and reduce variation in care by clinicians in primary care settings in Australia. The recommendations align with consumer and stakeholder views and values, enabling clinicians to follow confidently. This thesis provides a comprehensive insight into the needs of this under-investigated population group while laying the foundations for multiple research opportunities, especially in exploring disease burden and IP. Ultimately, these results can be used to inform the design of clinical trials to explore the IP treatment strategies used by clinicians and consumers which has limited supporting evidence.

1. INTRODUCTION

1.1 INCREASED INTESTINAL PERMEABILITY

Impairment of the intestinal barrier function may be an early event in the pathogenesis of numerous systemic and intestinal health conditions.[1] As such, increased intestinal permeability (IP), which involves the loss of integrity between the epithelial cells within the small intestines, plays a vital role in health and disease.[2] The full extent of the impact caused by the loss of intestinal integrity continues to evolve with the advancement of the scientific understanding of IP. In the previous two decades, extensive evidence centred on animal and *in vitro* models has emerged to describe the sequence of events involved in IP and has illustrated the negative effect small intestinal permeability can have on systemic health and disease.[3] Within private practice, clinicians consider IP to contribute to several health conditions.[4] During treating IP, a whole system approach is often used, with clinicians frequently using dietary modifications, lifestyle alterations and dietary supplementation as their preferred treatment methods.[5] However, many of these treatment methods do not have appropriate evidence to support their use.[5] Therefore, providing clinicians with guidance on the treatment strategies with the most significant clinical impact and evidence aims to enhance patient care and improve health outcomes.

1.2 AIMS AND SCOPE OF THESIS

1.2.1 RESEARCH AIM

This thesis aims to identify evidence-based treatment recommendations that align with patients' views and preferences to improve clinicians management of IP in clinical practice.

1.2.2 RESEARCH QUESTIONS

1. What are the known risk factors associated with altered IP in adults?
2. Which views and preferences of patients with suspected IP should be considered during the management of IP?
3. What are the health-seeking behaviours of adults with suspected IP?
4. Do people with suspected IP experience a worsened subjective wellbeing compared to the Australian population?
5. Can health-seeking behaviours of people with suspected IP influence health-related quality of life and subjective wellbeing?
6. What are the evidence-based management options available for IP?
7. How can evidence-based management options also consider patients views and preferences towards treatment?

1.2.3 RESEARCH OBJECTIVES

1. Identify known risk factors associated with altered IP in adults.
2. Explore the views, preferences, and health-seeking behaviours of adults with suspected IP.
3. Examine the subjective wellbeing and health-related quality of life of adults with suspected IP.

4. Determine the management options available for IP.
5. Develop an evidence-based clinical practice guideline for the management of IP which considers patients' views and preferences.

1.2.4 SIGNIFICANCE AND SCOPE

Altered IP is estimated to have a prevalence of 10-87%[1] in diseases with a known association with IP, compared to about 5% in healthy individuals.[6,7] This original research project is the first to collectively consider the published literature and consumers' views and preferences for developing a clinical practice guideline in managing IP. Understanding the risk factors associated with IP in clinical practice may indicate the treatment interventions most appropriate for managing IP. Despite the involvement of IP in many health conditions, there is no guidance for the management in clinical practice. With over 80% of naturopaths and nutritionists identifying digestive health as a major practice interest area, the absence of any guidelines has resulted in clinicians having inconsistencies in managing IP.[5,8] Furthermore, given the community's interest in digestive health, other clinicians such as general practitioners, gastroenterologists and dieticians may require further guidance in managing IP.[4] Therefore, developing a clinical practice guideline for the management of IP may provide a foundation for addressing these gaps in knowledge. This guideline would need to consider the patient's preferred treatment methods and whether any of these treatment methods may affect the quality of life. Creating a transparent, evidence-based clinical practice guideline for managing IP may help inform clinical practice, optimise patient care, improve health outcomes, and reduce variation in care for clinicians while informing policymakers and researchers. A clinical practice

guideline aims to guide clinicians in their recommendations and inform their treatment strategies. Future research can also build on the gaps identified in the recommendations and use the results to inform intervention studies.

1.3 THESIS STRUCTURE

This body of work is structured in the format of Thesis by Compilation. It presents a single body of work comprised of traditional thesis chapters, published articles and documents involved in developing a clinical practice guideline.

Chapter 1 provides background knowledge to understand the research project and the following chapters. Key details on the function of the gastrointestinal system are provided with a comprehensive explanation of IP. The different assessment methods are discussed to provide context and understanding for the thesis.

Chapter 2 reviews the literature on the known risk factors associated with the development and exacerbation of IP. The identification of pathology markers, demographics, anthropometric measurements, disease status and dietary intake are explored. The results of this chapter have been published in the *International Journal of Clinical Practice*.

Chapter 3 describes the methodology of this research project, providing details on the study design, data collection and analysis. This chapter is comprised of one traditional methods section and presents the *Guideline Development Process* as part of the clinical practice guideline.

Chapter 4 presents the first section of the results from the cross-sectional survey of Australian adults with suspected IP. This chapter explores the health-seeking

behaviours of Australian adults with suspected IP considering the type of healthcare this population would prefer to receive, especially regarding the diagnosis of IP. Additionally, this chapter explores the length of time between first suspecting IP and receiving a diagnosis of IP. Finally, the out-of-pocket expenditure associated with the assessment and management of IP are described. The results of this chapter have been published in *Integrative Medicine Research*.

Chapter 5 further expands on Chapter 4, by describing the preferred treatment methods people with IP want to use and whether any applied treatment methods affect quality of life. This chapter draws links between the treatment methods patients use and the effect they may have on quality of life. The results of this chapter have been published in *The Journal of Alternative and Complementary Medicine (*now referred to as *The Journal of Integrative and Complementary Medicine).*

Chapter 6 provides detailed results of the IP Guideline with a comprehensive description of the evidence, risk of bias and drafted recommendations. This chapter presents the *Technical Report* as part of the clinical practice guideline.

Chapter 7 summarises stakeholders' views and preferences surrounding the implementation of the IP Guideline into clinical practice. A detailed description of their agreement of each recommendation and suggested changes to each recommendation is provided.

Chapter 8 summarises the clinical need for each clinical question and provide the evidence and justification for each recommendation. The final recommendations based on stakeholder feedback are included in this chapter.

Collectively, this chapter contains the *Clinical practice guideline for the management of increased intestinal permeability: IP Guideline*.

Chapter 9 provides a discussion on the findings of this thesis. Results are contextualised with previous research around patient care and IP management. This chapter identifies the clinical relevance of the results to clinicians in clinical practice, describes the limitation of the results and suggests a research agenda based on the findings of this thesis.

Chapter 10 summarises the thesis, provides a conclusion on the findings and relates the results back to the aim and objective of this thesis.

1.4 BACKGROUND

1.4.1 THE GASTROINTESTINAL TRACT

The gastrointestinal tract is estimated to have a surface area of 400m^2 and is the most extensive interface between the internal and external environment in the human body.[9] The small intestine contains four cell types: enterocytes, enteroendocrine cells, goblet cells and Paneth cells. The paracellular pathway forms the interconnection between enterocytes and is composed of tight junctions, desmosomes and adherens junctions.[10] The tight junctions consist of membrane proteins, mainly occluding and claudins, that act as a selective paracellular barrier.[11] The gastrointestinal tract, consisting of a physical and biochemical barrier, requires a stable interaction of all elements, including the mucus layers, microbiome, and intestinal cells, to maintain intestinal mucosal homeostasis.[12]

1.4.2 INTESTINAL MUCUS LAYERS

The gastrointestinal tract contains a mucus layer that acts as the first line of immune defence. Goblet cells produce glycoprotein, which forms the mucin in the mucus layer.[13] The mucin within the mucus layer provides an environment and energy source for the microbiome.[14] The thickness of the mucus layer can reduce in some disease states, health conditions and ageing, thereby exposing the microbiome to the intestinal epithelium.[15] A reduced mucus layer is thought to contribute to a cascade of events involved in the development of IP.[12]

1.4.3 INTESTINAL MICROBIOME

The microbiome may play a fundamental role in modulating the presence and severity of IP.[3] The complexity of the microbiome involves a bidirectional relationship between the colonocyte and intestinal microbiome, which may influence the balance of bacteria in the intestinal tract.[16] The microbiome is the collective genome of the microbiota, while the microbiota is the sum population of microbes found in the gastrointestinal system.[17] The gastrointestinal tract is estimated to contain 200g of bacteria belonging to more than 500 different species.[18,19] Furthermore, it has been reported that there are as many bacteria cells in the gastrointestinal system as human cells in the whole body.[18] The microbiome in the gastrointestinal tract is estimated to contain 150-fold more genetic material than all the genes in the human body.[20] A decrease in overall gene richness in the gastrointestinal system is associated with many health conditions such as obesity and systemic inflammation.[21]

The microbiome may influence metabolic, hormonal, neurological, and immune biochemical pathways, resulting in therapeutic and protective outcomes for particular health conditions.[22] This ecosystem found in the gastrointestinal system can change and modulate the immune system, playing an essential part in health and disease.[23] Any change to the intestinal microbiome composition relative to healthy individuals is collectively referred to as dysbiosis.[24] The three main categories for intestinal dysbiosis are (1) loss of beneficial microbial organisms; (2) expansion of potentially harmful microbial organisms; and (3) reduced biodiversity.[24] Any change to the microbiome's diversity has been suggested to influence the pathogenesis of inflammatory diseases.[25] Dysbiosis plays a fundamental role in the development and exacerbation of many health conditions, with the microbiome also capable of influencing IP.[3]

1.4.4 GASTROINTESTINAL HEALTH AND DISEASE

Intestinal integrity may influence many aspects of the gastrointestinal system. A disruption to the homeostasis of the gastrointestinal tract, especially the intestinal mucosa, may contribute to alterations in the mucus layer leading to intestinal inflammation, dysbiosis and IP.[12,26] A dysbiotic microbiome can alter short-chain fatty acid production,[27] stimulate IP,[28] and influence inflammatory expression.[29,30] The health and function of the gastrointestinal tract are suggested to be a contributing element in the aetiology of a growing number of health conditions.[31] Some health conditions linked with gastrointestinal health are obesity,[32] Crohn's disease,[33] type-1 diabetes,[34] mental health,[35] and multiple sclerosis.[36]

1.5 INCREASED INTESTINAL PERMEABILITY

1.5.1 DEFINING INCREASED INTESTINAL PERMEABILITY

The exact definition of IP remains poorly understood. The evidence suggests two subtypes of IP: *acute IP* and *low-grade chronic IP*. Acute IP is more common within a hospital setting and is primarily triggered by pathogenic bacteria, resulting in sepsis.[37] Acute IP is more prevalent in conditions such as pancreatitis and burn injuries.[38] Low-grade chronic IP appears to be more prominent in primary care settings, with naturopaths, nutritionists, herbalists and integrative medicine practitioners frequently treating this subtype.[4] There is no defined nomenclature for IP with many possible terms commonly used including intestinal permeability, increased intestinal permeability, leaky gut, leaky gut syndrome, hyperpermeability, intestinal integrity, increased gut permeability, small intestinal permeability and endotoxemia. Throughout this thesis, IP will refer to the *low-grade chronic IP* where there is a loss of integrity between the small intestines' epithelial cells, leading to IP.[2]

1.5.2 INTESTINAL PERMEABILITY IS NORMAL

The permeability of the small intestine is a natural and homeostatic mechanism required for human survival and homeostasis equilibrium. For instance, IP is considered a normal defence mechanism to wash out unwanted microorganisms colonising the small intestine.[39] During conditions like gastroenteritis, an increase in IP facilitates the removal of pathogenic bacteria by attracting fluid into the gastrointestinal tract.[39] Another time when IP is considered normal is during pregnancy. IP naturally occurs during pregnancy and is suggested to contribute

to neonate mucosal immune system development.[40] In addition to the involvement of IP during pregnancy, it has been suggested that altered intestinal integrity and changes in the microbiota in early life may impact metabolic health later in life.[40] These examples of IP naturally occurring throughout life provide the understanding that IP plays an important role in homeostasis.

1.5.3 EXOGENOUS FACTORS AFFECTING INTESTINAL PERMEABILITY

Exogenous and genetic elements may contribute to the aetiology of IP. These elements may influence IP by stimulating the release of *zonulin*, the only physiological mediator known to regulate IP.[2] Paracellular IP is controlled by intercellular junctions that regulate the space between the intestinal epithelial cells. One of the most prominent junctions are the tight junctions that regulate over 50 proteins, with zonulin being the most measured marker for tight junction dysfunction.[41] Zonulin has a direct action in the intestine, where it is responsible for the cascade of events resulting in the disassembling of the tight junctions.[2] The exogenous factors that affect IP are numerous in quantity and variety. These factors may include, but are not limited to, dietary components such as alcohol[42] and fructose consumption[43] and exposure to the gliadin protein found in gluten.[44] Other physical and psychological exogenous factors include acute psychological stress by releasing corticotropin-releasing hormone activating mast cells within the small intestine,[45] physical stress[46] and strenuous exercise.[47] Some medications are known to adversely impact IP, with most evidence identifying the negative consequences of nonsteroidal anti-inflammatory drugs.[48] These collective groups of exogenous environmental factors provide an example of the diversity of factors that may influence IP.

1.5.4 GENETIC AND EPIGENETIC FACTORS AFFECTING INTESTINAL PERMEABILITY

The influence of genetics in developing IP is a growing area of research. For example, healthy family members of people with Crohn's disease may have a higher IP prevalence than unrelated household members they live with.[49] The difference in IP prevalence may be due to genetic and epigenetic factors. Claudin proteins may contribute to IP's pathogenesis, with gene expression and gene mutation playing a role.[49,50] Growing evidence suggests epigenetics, which is the influence the environment has on gene expression, may also be a factor involved in IP.[51] An example of epigenetics involvement in IP is seen with an over-expression of the occludin proteins in tight junctions.[11] Two factors that may influence an epigenetic change in tight junction proteins that lead to IP are alcohol and oestrogen.[52,53] While probiotics such as *Lactobacillus plantarum* MB452 may change the expression of the tight junction, thereby reducing permeability.[54]

1.5.5 INTESTINAL PERMEABILITY IN HEALTH AND DISEASE

The prevalence of IP is difficult to quantify and estimate. However, studies involving a healthy control group generally observe IP in about 5% of participants.[6,7,55] A diverse range of conditions seen in primary care practice, including autoimmune conditions, liver-related conditions, metabolic conditions, digestive conditions and neurological conditions, are associated with IP with an estimated prevalence of 10 to 87% (Table 1.1).[1] It has also been associated with disease severity and particular clinical symptoms.[1]

Research has speculated that IP may play a role in the pathogenesis of chronic disease. From the health conditions explored, altered intestinal integrity may precede the clinical onset of Crohn's disease,[56,57] type 1 diabetes,[58] coeliac disease,[59] and gestational diabetes[60] with IP involved in the pathogenesis of chronic liver disease,[61] IgA nephropathy[62] and intrahepatic cholestasis of pregnancy.[63]

Table 1.1 The prevalence of increased intestinal permeability

Conditions	Increased Intestinal Permeability
Autoimmune Conditions	
Dermatitis herpetiformis	87%
Ulcerative colitis	10-43%
Crohn's disease	36%
Systemic sclerosis	34%
Type 1 diabetes	30%
Primary biliary cirrhosis	25%
Liver Related Conditions	
Chronic liver disease with type 2 diabetes	65%
Liver cirrhosis	35%
Chronic liver disease	15-35%
Non-alcoholic fatty liver disease	31%
Diabetic Conditions	
Chronic liver disease with type 2 diabetes	65%
Gestational diabetes	37%
Type 1 diabetes	30%
Neurological Conditions	
Autism	36%
Gastrointestinal Conditions	
Irritable bowel syndrome	35%

Source: Table from Leech et al., 2019[1]

1.5.6 CLINICAL PRESENTATION OF INTESTINAL PERMEABILITY

In the current literature, there is limited evidence on the clinical presentation of IP.[64,65] Clinicians report that people with IP present with food sensitivities, intestinal dysbiosis, abdominal pain, bloating, parasitic infection, brain fog, flatulence, inflammation, stress and obesity in addition to having IP.[4] These signs and symptoms are associated with numerous other conditions and are not specific to IP.[66] The clinical presentation with the greatest area of evidence appears to be disease association rather than clinical signs and symptoms. Clinicians that frequently treat patients with IP report the conditions most associated with IP as gastrointestinal, autoimmune, skin, neurological, respiratory and liver-related conditions.[1] The reported association between disease and IP appears to reflect the published literature with substantial research confirming an association between IP and autoimmune conditions, liver-related conditions, metabolic conditions, digestive conditions and neurological conditions.[1]

1.5.7 MEASURING INTESTINAL PERMEABILITY

The measurement and assessment of IP remain a highly controversial and debated area of research as there is no recognised gold standard.[67] There are five common techniques available for the assessment of intestinal integrity:

1) Measurement of an introduced medium: dual sugar test;
2) Measurement of a released biomarker: serum and stool zonulin;
3) Measurement of a consequence of IP: serum LPS;
4) Measurement of contributing factors: mucus barrier; and
5) Biopsy of intestinal cells: claudin.

In a research setting, there are four tests frequently used all of which have a diverse degree of strength, weakness and applicability (Table 1.2).

Table 1.2 Frequently used methods to measure intestinal permeability

Test	Method used	Strength	Weakness	Application
Dual sugar test	Urine collection for 6 hours	• Validated across multiple population groups • Controls for confounding factors	• Collection time validity • Lengthy collection process • Diet may impact results	• Identification of well-established intestinal permeability • Identification of disease severity
Zonulin, plasma	Blood collection	• Easy collection process • Suggested to reflect intestinal permeability	• Limited validation studies • Influenced by weight • Zonulin released from many tissues	• Used in research when confounding factors can be controlled • Should not be used in clinic
Zonulin, stool	Stool collection	• Identification of tight junction permeability • Limited confounding variables	• Limited validation studies • Unable to identify intestinal permeability when not stimulated	• Early-stage intestinal permeability • Intestinal permeability actively being stimulated • Identification of disease severity
Lipopolysaccharides	Blood collection	• Easy collection process • Suggested to reflect intestinal permeability	• Not available in clinical practice • Diet may impact results	• Used in late-stage disease • Prolonged intestinal permeability

1.5.7.1 DUAL SUGAR

The primary pathology test currently used in clinical practice and research to assess IP appears to be the dual sugar test.[4,68] The dual sugar test has been validated across multiple population groups and is considered an accurate and reliable measure of intestinal integrity.[69-72] The dual sugar test corresponds with biopsy-confirmed abnormalities in the small intestine and abnormal mucosa in people with organic and functional gastrointestinal conditions.[70,73,74] The repeatability of the dual sugar test has demonstrated a good to excellent linear relationship for the dual sugar tests laboratory assay.[69] The dual sugar test best identifies well-established IP, compared to early-stage IP.[75] This test has been shown to distinguish disease severity in patients with irritable bowel syndrome.[76] Furthermore, the dual sugar test correlates with disease severity in patients with liver steatosis, with a greater degree of permeability seen in patients with moderate or severe steatosis.[75]

There is a significant difference in the lactulose:mannitol ratio between healthy control and coeliac disease or Crohn's disease patients.[77] The dual suagr test controls for many confounding factors such as gastric emptying, renal function and intestinal transit time compared to using a disaccharide test alone.[78] The dual sugar ratio has been demonstrated to have higher specificity and sensitivity in identifying disease compared to the disaccharide.[79] Although there are conflicting ideas around the optimal duration of urine collection, the current consensus suggests that <5 hours of collection, not including the first-morning void, is the most accurate collection time.[80] The dual sugar test involves the oral consumption of two sugars, lactulose and mannitol, in roughly 100-300ml of water

after an overnight fast. The principle behind the lactulose and mannitol test is the different molecule sizes of the two sugars. When the intestinal integrity is healthy, the monosaccharide mannitol is readily absorbed, while lactulose being a disaccharide, remains within the intestine and is poorly absorbed. However, during a loss of intestinal integrity, the ratio of lactulose to mannitol is increased, as lactulose can permeate the intestinal mucosa and become present in the urine.[80]

1.5.7.2 STOOL ZONULIN

Stool zonulin as a marker for IP has been shown to be a marker of disease severity in several health conditions. Stool zonulin measures the amount of zonulin protein found in the faecal matter. The ability to measure stool zonulin in clinical practice is only a recent advancement, with stool zonulin first used in clinical studies around 15 years ago. Although a limited number of studies are available, there is some evidence of a relationship between stool zonulin in gastrointestinal and autoimmune health conditions. Firstly, IBS patients with a greater degree of disease severity have also been found to have higher levels of stool zonulin compared to IBS patients with less disease severity.[76] When considering autoimmune conditions, stool zonulin has been found to have a moderate positive correlation with Th17 cells, an immune cell implicated in the pathogenesis of many inflammatory and autoimmune diseases.[81] Additionally, stool zonulin has a high positive correlation with the severity of psoriatic arthritis.[81] The severity of stool zonulin appears synergistic with many factors, including medication and the microbiome influencing the level of zonulin. For instance, proton pump inhibitors in people with diagnosed intestinal dysbiosis appear to

have a greater impact on zonulin levels than in those not using proton pump inhibitors.[82] The relationship between stool zonulin and disease severity provides a piece of important case information in addition to the identification of IP.

The correlation between stool zonulin and the dual sugar test is poorly established, with many studies finding conflicting results.[75,83,84] The current understanding in the literature is that the two tests identify different stages of IP. As zonulin is an acute-phase protein, elevation occurs when actively stimulated due to the relatively short half-life.[85,86] Thus, stool zonulin can identify early-stage IP or when a stimulus is present in the environment.[86] Late-stage disease with established permeability is best identified using the dual sugar test.[75] Understanding the most appropriate method and time to assess IP is essential for clinicians.

1.5.7.3 SERUM ZONULIN

Early understanding of serum zonulin suggested that this serological integrity marker was an accurate indicator for IP.[87] However, the validity of serum zonulin as a marker of IP has recently been questioned.[88] The inaccuracy of serum zonulin may stem from where this protein is released throughout the body. The release of zonulin comes from the liver, enterocytes, adipose tissue, heart, kidney, brain, skin and immune cells.[89,90] With multiple sites capable of releasing zonulin, determining the origin remains a continued area of investigation. Some research suggests that zonulin released from the adipose tissue can influence serum levels of zonulin, thereby contributing to a false negative result for IP.[88]

The potential of a false negative only appears to impact overweight and obese people.[88]

Another factor to consider in using serum zonulin as a marker of intestinal integrity is the consequence of zonulin being a protein. An individual's nucleotide sequence ultimately determines the synthesis of zonulin. Someone with a homozygous Hp1-1 polymorphism cannot produce zonulin. However, this polymorphism remains low in the population (8-15%), regardless of their disease state.[91,92] The consequence of zonulin being a protein continues, as the commercial assay available to clinicians in Australia has recently been found not to detect actual zonulin.[91,92] The protein(s) in which the commercial zonulin assay detects are unknown yet are within the zonulin family peptides. [91-93] These zonulin family peptides have been demonstrated to reflect IP in some health conditions, as indicated in the association with the dual sugar test.[94,95] The same protein(s) collectively referred to as serum zonulin have been used extensively in the literature, with many disease states and biochemical reactions associated with elevated levels.[1]

With these limitations of serum zonulin, there are factors to consider when interpreting serum zonulin as a marker for IP. As adipose tissue can release zonulin, the use of serum zonulin in clinical practice, especially in overweight patients, should be used with a high degree of caution. However, the accuracy of serum zonulin as an IP indicator in research studies depends on the analysis. Specifically, if studies control for participant's body mass index in the analysis, serum zonulin may be considered a suggestive marker of IP.[88]

1.5.7.4 LIPOPOLYSACCHARIDE

Lipopolysaccharides (LPS), also referred to as endotoxins, are derived from the cell wall of gram-negative bacteria.[96] LPS and dead bacteria are major contributors to endotoxins circulating through the portal vein and systemic circulation.[97] Under normal conditions, a healthy intestinal lining only absorbs a small amount of LPS into portal circulation, where Kupffer cells in the liver can process the endotoxins.[96] High-density lipoproteins (HDL) can also clear LPS by binding and neutralising the LPS in circulation.[98] However, during prolonged LPS exposure or increased absorption of endotoxins from the small intestine, the normal processes cannot mitigate the LPS leading to cytokine-mediated systemic inflammation and oxidative stress.[99] A major factor determining the end consequence of LPS absorption dramatically depends on the microbiome. The microbiome can produce multiple types of LPS such as hepta-, hexa-, penta-, and tetra-acylated LPS.[100] Although this area of research is in its infancy, hexa-LPS appears to be responsible for a more significant inflammatory response.[100]

There remains uncertainty surrounding whether LPS is a cause or consequence of IP. For instance, some research suggests serum LPS may be an indicator for prolonged IP or when there is a high degree of permeability.[101,102] Other research identifies LPS as a catalyst for IP due to the pro-inflammatory action.[103] Although LPS may be associated with several health conditions and disease severity, there remains a large body of conflicting evidence surrounding LPS and disease association.[76,104]

There is a lack of research on the association between IP and LPS. The evidence suggests LPS does not reflect mild IP, however, it indicates severe IP.[37,105] This lack of association in mild IP and more so in severe IP may be due to a few confounding variables affecting LPS as a marker of IP. Two primary confounding variables to consider in the interpretation of serum LPS are the amount of dietary fat and the half-life of LPS.[106] Firstly, when LPS is present in the lumen of the intestine, saturated fatty acids and chylomicrons facilitate the absorption of LPS into circulation.[107] Therefore, a high-fat diet may provide a false-positive result. The other confounding variable is the relatively short half-life of LPS.[108] As a result of the short half-life, serum LPS may be considered a marker for IP in late-stage disease or when there is prolonged severe IP, as LPS is continuously high in these situations.[37]

1.5.8 INTESTINAL PERMEABILITY IN CLINICAL PRACTICE

While biomedicine places little emphasis on digestive health in managing chronic disease, modulating gastrointestinal health is a fundamental principle of naturopathic care.[109] The vast majority of complementary medicine practitioners in Australia report that digestive disorders are a special interest in their clinical practice.[8] Previous research has explored the management of IP from the clinician's perspective, where they acknowledge the involvement of IP in many health conditions seen in clinical practice.[4] In Australia, approximately two-thirds of naturopaths and nutritionists would often or always treat IP in their clinical practice but only 20% of practitioners reported testing for IP in their patients.[4] The pathology tests available in clinical practice are invasive, require patients to pay out-of-pocket, and involve a long time to perform. Many naturopaths and

nutritionists report the avoidance of validated pathology tests due to the financial cost to the patient and utilise case history to identify patients with IP.[4]

1.5.9 TREATMENT OF INCREASED INTESTINAL PERMEABILITY

Currently, there are no guidelines for clinicians to follow regarding the management of IP in clinical practice. As many clinicians treat patients with digestive disorders, the absence of any guidelines has resulted in some inconsistencies by clinicians.[5,8] The clinicians that frequently treat patients with IP have reported using diverse treatment strategies.[5] These treatment interventions include dietary modifications, lifestyle alterations and dietary supplements.[5] Providing clinicians with guidance on the treatment strategies with the greatest clinical impact and evidence aims to enhance patient care.

Normalising IP through treatment interventions or reducing known risk factors for the development and exacerbation of IP has been suggested to improve disease severity.[75,76] Currently, there are no pharmacological agents developed for the treatment of IP however, early evidence suggests a drug known as larazotide acetate may temporarily reduce zonulin-induced permeability by acting as a tight junction regulator.[110] Although larazotide acetate is a pharmaceutical advancement for the treatment of IP, this drug is still in phase III clinical trials and has the limitation of only lasting 2-3 hours.[111] Beyond this recent progress in the pharmaceutical management of IP, environmental and genetic factors are important elements identified through research to influence the risk of IP.

1.5.10 CLINICAL PRACTICE GUIDELINES IN CLINICAL PRACTICE

Clinical practice guidelines are defined as "statements that include recommendations intended to optimise patient care that are informed by a systematic review of evidence and an assessment of the benefits and harms of alternative care options".[112] These guidelines are designed to support clinicians in their decision making for the management of specific health conditions in clinical practice. Clinical practice guidelines are considered one of the best ways to present evidence-based recommendations to clinicians while reducing inappropriate care and supporting new knowledge to the clinician.[113,114] However, clinical practice guidelines are not intended to supersede professional judgement, with clinicians always advised to act with the patients' best interest and needs first.[115] Many clinical practice guidelines have been developed for a diverse range of gastrointestinal conditions, including irritable bowel syndrome,[116] Barrett's oesophagus,[117] small intestinal bacterial overgrowth,[118] ulcerative colitis,[119] Crohn's disease,[120] dyspepsia,[121] constipation,[122] infectious diarrhoea,[123] diarrhoea[124] and acute abdominal pain[125] Integrative medicine practitioners and general practitioners frequently use these guidelines to inform disease management.[126] Conversely, naturopaths and nutritionists rely less on clinical practice guidelines, with their use mainly informing care rather than treatment interventions.[127] This may be due to multiple factors such as the lack of guidelines that involve these clinicians as key stakeholders, the shortage of naturopathic specific treatment interventions or the perception of conflict between individualised care and clinical practice guidelines.[127] Therefore, a clinical practice guideline for the management of IP ought to consider the values of key stakeholders in the guideline development.

1.6 CHAPTER SUMMARY

This introduction has provided an overview of IP and the implication dysfunction of the intestinal barrier may have on health and disease. Key details of the gastrointestinal system and its relationship with IP have been described. Furthermore, a fundamental discussion on the assessment methods was also explained, providing important context for the following chapters. The pathogenesis and aetiology of IP have been briefly discussed in this chapter however, a comprehensive understanding of the potential risk factors associated with IP is lacking in the existing published literature. Understanding any potential risk factors associated with IP may direct potential interventions that are indicated for people with IP.

2. LITERATURE REVIEW

The concept of IP and its implication for health and disease has been briefly discussed in the preceding chapter. This research project seeks to develop evidence-based treatment recommendations to improve clinicians' management of IP in clinical practice. However, to establish context for this research project, a literature review was undertaken to explore the most significant risk factors associated with IP. Understanding the risk factors associated with IP in clinical practice may indicate the treatment interventions most appropriate for managing IP, including the context in which those treatments may be employed. This chapter presents the methods used to search the literature and the literature review results.

2.1 PUBLICATION OF REVIEW

This chapter contains a systematic review, which has been published (see Appendix 1.1) with details as follows:

> Leech, B, McIntyre, E, Steel, A, Sibbritt, D (2019) "Risk factors associated with intestinal permeability in an adult population: A systematic review", The International Journal of Clinical Practice, Vol 73, 10.

2.2 RISK FACTORS ASSOCIATED WITH INTESTINAL PERMEABILITY IN AN ADULT POPULATION: A SYSTEMATIC REVIEW

2.2.1 INTRODUCTION & BACKGROUND

Increased intestinal permeability (IP) involves the loss of integrity between the cells of the small intestine.[2] The prevalence of altered IP is estimated to be 10-87%[1] in diseases with a known association compared to about 5% in healthy subjects.[6,7] Furthermore, approximately 1 in 3 individuals are suggested to experience IP when diagnosed with a disease associated with IP.[1] Although the concept of IP was first mentioned in the literature during the 1960s[128] and further explored in relation to disease during the 1970s[129] it was not until the 2000s that the mechanism of action for IP development was discovered, providing further clarification into the role IP plays in health and disease.[130] While IP may be considered an emerging health condition that clinicians should be aware of, the consequence of impaired barrier function remains underexamined.[131]

The loss of intestinal integrity occurs when the transmembrane proteins connecting the cells of the small intestine disassemble in response to a cascade of events involving the protein zonulin.[2] As a result of altered IP, particular aspects of disease such as clinical symptoms, severity and activity have been found to be exacerbated in the presence of IP.[132,133] In addition, preliminary evidence suggests that IP may be involved in the pathogenesis of type 1 diabetes,[134,135] Crohn's disease,[136] coeliac disease[59] and diarrhoea-predominant irritable bowel syndrome (IBS-D).[137,138] Altered IP has also been associated with

many autoimmune conditions, liver diseases, gastrointestinal conditions and metabolic conditions.[1] Although the pathogenesis is not clearly defined, inflammation appears to be involved both as a driving factor for and consequence of altered IP.[1] Furthermore, the aetiology of IP is poorly understood, with early research indicating that two aspects, namely pathogenic bacteria and gliadin from gluten, are responsible for triggering IP.[139] Although, recent research suggests that the pathogenesis of IP is multifactorial and different for each individual.[140]

There are two tests primarily used for the clinical diagnosis of IP, namely the dual sugar test and serum zonulin; with many others used in a research setting. However, there remains controversy surrounding the gold standard of IP testing and the consistency between measurement methods.[141] The dual sugar test involves the oral consumption of two sugars after an overnight fast followed by the collection of urine for a given period of time. The fundamental principle behind the dual sugar test is the different molecule size of monosaccharide and disaccharide. When the integrity of the intestine is healthy the monosaccharide (mannitol) is easily absorbed whereas the disaccharide (lactulose) is poorly absorbed and remains in the intestine. During altered IP the disaccharide is readily absorbed resulting in an increased ratio between lactulose and mannitol in the urine.[80] Whereas zonulin, the protein responsible for the disassembling of the tight junctions, can be measured in either the serum or stool.[2] Zonulin is considered to be the only measurable biomarker that reflects an impairment of the intestinal barrier.[2,58] However, zonulin has been reported to be released from many tissues including adipose tissue and proposed to be a biomarker of metabolic syndrome, obesity, inflammation and poor health more so than IP.[88]

Nevertheless, zonulin is recognised as an accurate measurement of IP.[58] Another method of measuring IP is the level of lipopolysaccharide (LPS) found within the blood. LPS is suggested to be an exacerbator and marker of IP and is mostly increased at the later stage of disease or in advanced IP.[142,143] Collectively, these markers of IP provide healthcare practitioners with a method to measure and assess IP in clinical practice.

Correctly identifying patients at risk of IP may allow for timely testing to determine the potential severity of IP and facilitate access to appropriate treatment interventions if required. Although the full extent of untreated IP remains underexamined, there is a considerable amount of research linking the health and integrity of the intestine to chronic disease.[140] The purpose of this review is to summarise the known risk factors for IP and identify the most significant of these risk factors.

2.2.2 METHODS

The reporting of this systematic review followed the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA)[144] statement and the Meta-analysis Of Observational Studies in Epidemiology (MOOSE) checklist.[145] The protocol was registered with the International Prospective Register of Systematic Reviews (PROSPERO) (#CRD42018109384).

2.2.2.1 SEARCH STRATEGY

The databases PubMed, EMBASE, CINAHL, and Scopus were searched for articles published up until September 2018 by the lead researcher (BL). The single-arm search terms used were: 'intestinal permeability' OR 'intestinal integrity' OR 'intestinal barrier dysfunction' OR 'gastrointestinal permeability' OR 'gut permeability' OR 'zonulin' OR 'dual sugar' OR 'lactulose AND mannitol' OR 'lactulose AND rhamnose' OR 'cellobiose AND mannitol'. A hand search of the reference list from the included articles was also carried out.

2.2.2.2 ELIGIBILITY CRITERIA

Included articles were original observational studies reporting on risk factors associated with IP in an adult population. These risk factors are in relation to low-grade chronic IP rather than acutely induced IP caused by sepsis in critically ill patients. Articles were excluded if subjects were under the age of 18, were critically ill (i.e. in intensive care or palliative care), involved an experimental design or used a method of diagnosing IP other than zonulin (serum, plasma, stool), dual sugar urinary test (lactulose/mannitol, lactulose/rhamnose, cellobiose/mannitol) and serum LPS. These methods were selected to ensure clinical relevance of the review. There was no exclusion based on language, geographical location or publication date.

2.2.2.3 STUDY SELECTION AND DATA EXTRACTION

All identified citations were imported to Endnote (Version X9) and duplicates removed. The citations were independently screened for eligibility by the lead

author (BL). A sample (20%) of the eligibility citations were reviewed by a second author (EM). When uncertainty of eligibility criteria arose the corresponding author of the article in question was contacted for clarification.

2.2.2.4 CRITICAL APPRAISAL ANALYSIS AND RISK OF BIAS ASSESSMENT

The quality of the included articles was assessed (by BL) and reviewed (by EM) using the Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) statement.[146] In addition, the included articles were assessed for risk of bias using a previously established tool for prevalence studies. The assessment tool is composed of 10 items covering four main domains of bias including external validity, internal validity, measurement bias, and bias relating to analysis.[147]

2.2.2.5 STATISTICAL ANALYSIS

A thematic synthesis of the association between risk factors of IP and altered IP was carried out. Three categories of association namely odds ratio (OR), beta coefficient and correlation coefficient were collectively assessed for associated risk factors with IP. Only statistically significant risk factors were extracted from the included articles, along with the confidence interval (CI). Furthermore, only ORs and beta coefficients that adjusted for confounders were extracted. Unadjusted correlation coefficients were extracted; however, precedence was given to adjusted correlation coefficients when available. Interpretation of both Spearman's (ρ) and Pearson's (*r*) correlation coefficient were as followed: little

(0.00 to 0.29), weak (0.30 to 0.49), moderate (0.50 to 0.69), high (0.70 to 0.89), and very high (0.90 to 1.00) correlation[148]. Variables with a little correlation coefficient were omitted from the results to minimise misinterpretation where results remain uncertain. However, these variables were still reported in study characteristics and considered in the discussion as the articles met the inclusion criteria. When associations were determined by a *coefficient of determination* (R^2), this value was converted to a correlation coefficient by taking the square root of the R^2 value.

2.2.3 RESULTS

A total of 22,118 articles were identified through the key database searches, of which 10,914 duplicates were removed. After title and abstract screening 149 potentially relevant full-text articles were reviewed, of which 42 articles were considered eligible. Hand searching the reference list of the 42 eligible articles identified an additional five articles. A total of 47 articles were included in this systematic review (Figure 2.1). From the sample of eligible studies reviewed (by EM), a strong agreement (Kappa score 0.90) was achieved.

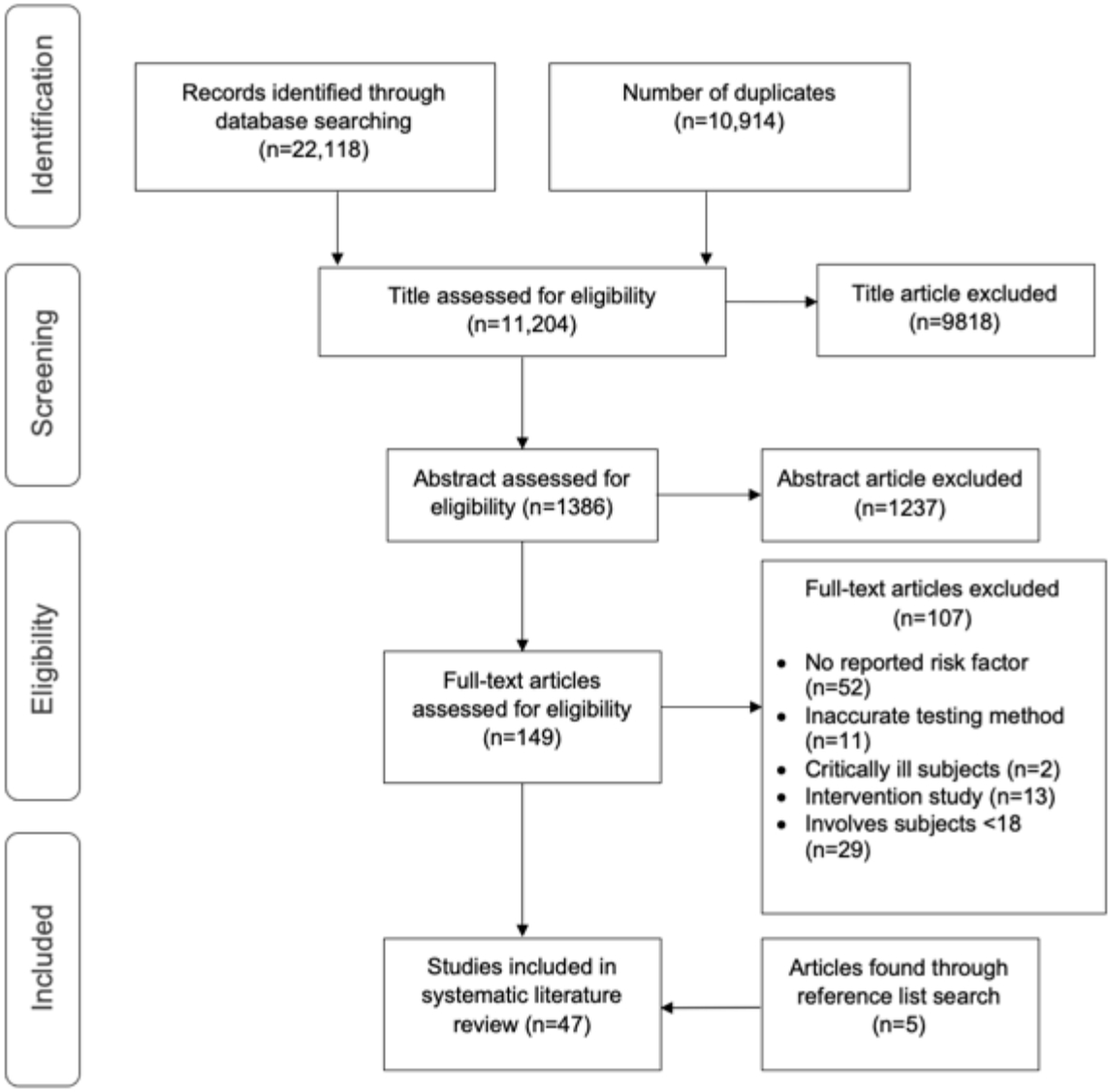

Figure 2.1 PRISMA flow diagram of study selection. Starting with 22 118 identified citations, 47 articles were included in the final systematic review

The sample size of each study varied from 21 to 1,015[106,149] (mean=155) with the majority of the studies carried out in Europe (n=34) followed by Asia (n=5), America (n=5), Africa (n=2), and Australia (n=1). The laboratory markers of IP used in each study were zonulin (n=24), dual sugar (n=13), LPS (n=10) and stool zonulin (n=3). A total of 30 different study populations were measured for IP with findings suggesting 101 statistically significant risk factors associated with IP. Risk factors were identified in study populations with glucose metabolism

disorders (n=57), body mass index (BMI) >29 (n=42), pregnancy (n=39), liver conditions (n=34), general population (n=29), polycystic ovarian syndrome (PCOS) (n=14), digestive conditions (n=13), kidney disease (n=12), obstructive sleep apnoea (n=12), respiratory conditions (n=6), pain conditions (n=4), alcohol use disorder (n=2), Parkinson's disease (n=2), ankylosing spondylitis (n=1), and systemic sclerosis (n=1). These risk factors were grouped into five major domains; *medical history and disease, dietary factors, anthropometric measurements, biomarkers,* and *demographic factors*.

2.2.3.1 CRITICAL APPRAISAL AND RISK OF BIAS ASSESSMENT

STROBE evaluation identified that the majority of the included articles provided an inadequate indication of study design, methods of addressing bias, study size calculation or consider the use of a flowchart (Table 2.1). Three articles were recognised as low-quality.[150-152] During risk of bias assessment, no articles were identified as high risk of bias; although, 27 of the 47 articles were classified as having a moderate risk of bias. This moderate risk of bias was primarily due to the articles demonstrating large gaps in the external validity criteria. Internal validity assessment showed a low risk of bias with a large degree of consistency between articles. Results from the risk of bias assessment are presented in Table 2.2.

STROBE critical appraisal tool.	Title, abstract & Introduction								Methods					Results																Discussion & other information					
Study	Title & Abstract		Background	Objectives	Study design	Setting	Participants		Variables	Data sources		Bias	Study size	Quantitative variables	Statistical methods					Participants			Descriptive data			Outcome data	Main results			Other analyses	Key results	Limitations	Interpretation	Generalizability	Funding
	1a	1b	2	3	4	5	6a	6b	7	8	9	10	11	12a	12b	12c	12d	12e	13a	13b	13c	14a	14b	14c	15	16a	16b	16c	17	18	19	20	21	22
Amar et al., 2008[106]	x	x	x	x	x	x	x	/	x	x	-	-	x	x	x	x	x	x	x	x	-	x	x	/	x	x	x	/	x	x	x	x	x	x
Barcelo et al., 2016[153]	x	x	x	x	x	x	x	x	x	x	x	-	x	x	x	-	x	x	x	x	-	x	x	/	x	x	x	/	x	x	x	x	x	x
Cangemi et al., 2016[154]	x	x	x	x	x	x	x	x	x	x	-	-	x	x	x	x	/	x	-	-	-	x	x	-	x	x	x	/	x	x	x	x	x	x
Cariello et al., 2010[155]	x	x	x	x	x	x	x	x	x	x	-	-	x	x	x	x	x	x	x	x	-	x	x	/	x	x	x	/	x	x	-	x	x	x
Carnevale et al., 2017[156]	x	x	x	x	x	x	x	/	x	x	-	x	x	x	x	x	x	x	x	-	-	x	x	/	x	x	x	/	x	x	x	x	x	x
Caserta et al., 2003[157]	-	x	x	x	x	x	x	x	x	x	-	x	-	x	x	x	x	x	x	x	-	x	x	/	x	x	x	/	x	x	-	x	x	-
Caviglia et al., 2018[158]	x	x	x	x	x	x	-	-	x	x	-	-	-	x	x	-	-	x	x	x	-	x	x	/	x	x	x	/	x	x	x	x	x	x
Ciccia et al., 2017[159]	-	x	x	x	x	x	x	/	x	x	-	-	x	x	x	x	x	x	x	x	-	x	x	/	x	x	x	/	x	x	-	x	-	x
Di Leo et al., 2003[160]	x	x	x	x	x	x	x	/	x	x	x	-	x	x	x	x	-	x	x	x	-	x	x	x	x	x	x	x	x	x	-	x	x	x
Donnadieu-Rigole et al., 2018[161]	-	x	x	x	x	x	x	-	x	x	-	-	x	x	x	-	-	x	x	x	x	x	x	x	x	x	x	/	x	x	-	x	x	x
Du Plessis et al., 2013[162]	-	x	x	x	x	x	x	x	x	x	-	-	x	x	x	x	x	x	-	-	-	x	x	/	x	x	x	/	x	x	x	x	x	x
Duerksen et al., 2010[149]	-	x	x	x	x	x	x	x	x	x	x	-	x	x	-	-	-	-	x	x	-	x	x	/	x	x	x	/	x	x	x	x	x	x
Ficek et al., 2017[163]	-	x	x	x	x	-	x	x	x	x	-	-	x	x	x	x	x	x	x	x	x	x	x	/	x	x	x	/	x	x	x	x	x	x
Goebel et al., 2008[164]	-	x	x	x	x	x	x	x	x	x	-	-	x	x	x	x	x	x	x	x	-	x	x	/	x	x	x	/	x	x	x	x	x	x
Hendy et al., 2017[165]	-	x	x	x	x	x	x	x	x	x	-	-	x	x	x	x	x	x	x	x	-	x	x	/	x	x	x	/	x	x	-	x	x	x
Hilsden et al., 1999[166]	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	-	x	x	x	x	-	x	x	x	x	x	x	/	x	x	-	x	x	x

STROBE critical appraisal tool.	Title, abstract & Introduction											Methods										Results									Discussion & other information					
Study	Title & Abstract		Background	Objectives	Study design	Setting	Participants		Variables	Data sources	Bias	Study size	Quantitative variables	Statistical methods					Participants			Descriptive data			Outcome data	Main results			Other analyses	Key results	Limitations	Interpretation	Generalizability	Funding
Jayashree et al., 2014[167]	-	x	x	x	x	x	x	x	x	x	-	-	x	x	x	x	x	x	x	x	-	x	x	/	x	x	x	/	x	x	-	x	x	x
Johnston et al., 2000[168]	x	x	x	x	x	x	x	x	x	x	x	-	x	x	-	x	x	x	x	-	-	-	x	-	x	x	x	/	x	x	-	x	x	x
Karthikeyan et al., 2018[169]	-	x	x	x	x	x	x	/	x	x	-	-	x	x	x	x	-	x	x	x	-	x	x	/	x	x	x	/	x	x	-	x	x	x
Kim et al., 2018[170]	-	x	x	x	x	x	x	/	x	x	-	-	x	x	x	-	-	x	x	x	-	x	x	/	x	x	x	x	x	x	x	x	x	x
Kvehaugen et al., 2017[171]	x	x	x	x	x	x	x	/	x	x	x	-	x	x	x	x	x	x	x	x	x	x	x	/	x	x	x	x	x	x	x	x	x	x
Lassenius et al., 2011[172]	-	x	x	x	x	x	x	x	x	x	x	-	x	x	x	x	x	x	x	x	-	x	x	/	x	x	x	/	x	x	-	x	x	x
Lindheim et al., 2017[173]	x	x	x	x	x	x	x	x	x	x	-	-	x	x	-	x	x	x	x	x	-	x	x	/	x	x	x	/	x	x	x	x	x	x
Lukaszyk et al., 2018[152]	-	x	x	x	x	-	x	/	x	x	-	-	x	x	-	-	x	x	-	-	-	x	x	/	x	x	x	/	x	x	x	x	x	x
Malickova et al., 2017[174]	-	x	x	x	x	x	x	x	x	x	-	-	x	x	x	x	x	x	x	x	-	x	x	/	x	x	x	/	x	x	x	x	x	x
Malyszko et al., 2014[175]	-	x	x	x	x	x	x	/	x	x	-	-	x	x	x	x	-	x	x	-	-	x	x	/	x	x	x	/	x	x	-	x	x	-
Mokkala et al., 2017[176]	x	x	x	x	x	x	x	/	x	x	x	x	x	x	x	x	x	x	x	x	-	-	x	/	x	x	x	x	x	x	x	x	x	x
Mokkala et al., 2016[177]	x	x	x	x	x	x	x	/	x	x	x	x	x	x	x	x	x	x	x	x	-	-	x	/	x	x	x	x	x	x	x	x	x	x
Mokkala et al., 2017[60]	x	x	x	x	x	x	x	/	x	x	x	x	x	x	-	-	x	x	x	x	-	x	x	/	x	x	x	/	x	x	x	x	x	x
Moreno-Navarrete et al., 2012[178]	-	x	x	x	x	x	x	/	x	x	x	-	x	x	x	x	x	x	x	x	-	x	x	/	x	x	x	/	x	x	x	x	x	x
Morkl et al., 2018[179]	-	x	x	x	x	x	x	/	x	x	-	-	x	x	x	x	x	x	x	x	-	x	x	/	x	x	x	/	x	x	x	x	x	x

STROBE critical appraisal tool.	Title, abstract & Introduction				Methods														Results															Discussion & other information				
Study	Title & Abstract		Background	Objectives	Study design	Setting	Participants		Variables	Data sources	Bias	Study size	Quantitative variables	Statistical methods					Participants			Descriptive data			Outcome data	Main results			Other analyses	Key results	Limitations	Interpretation	Generalizability	Funding
Mujagic et al., 2014[180]	-	x	x	x	x	x	x	/	x	x	x	-	x	x	x	x	x	x	x	x	-	x	x	/	x	x	x	/	x	x	x	x	x	x
Nymark et al., 2009[181]	x	x	x	x	x	x	x	x	x	x	-	-	x	x	x	x	x	x	x	x	-	x	x	-	x	x	x	x	x	x	-	x	x	x
Ohlsson et al., 2017[88]	-	x	x	x	x	x	x	/	x	x	x	x	x	x	x	x	x	x	x	x	-	x	x	/	x	x	x	/	x	x	x	x	x	x
Qi et al., 2017[182]	-	x	x	x	x	x	x	/	x	x	-	-	x	x	x	-	-	x	x	-	-	x	x	/	x	x	x	/	x	x	x	x	x	x
Raparelli et al., 2017[102]	x	x	x	x	x	x	x	/	x	x	-	-	x	x	x	x	x	x	x	x	-	x	x	/	x	x	x	/	x	x	x	x	-	x
Riordan et al., 1997[151]	-	x	x	x	x	-	x	/	x	x	-	-	x	x	-	-	-	x	x	x	-	-	x	/	x	x	x	/	x	x	-	x	x	x
Rutten et al., 2014[183]	-	x	x	x	x	x	x	x	x	x	x	x	x	x	-	-	-	x	x	x	x	x	x	/	x	x	x	/	x	x	x	x	x	x
Schwiertz et al., 2018[184]	x	x	x	x	x	x	-	x	x	x	x	-	x	x	x	x	-	x	x	x	-	x	x	/	x	x	x	/	x	x	x	x	x	x
Swanson et al., 2015[185]	-	x	x	x	x	x	x	x	x	x	x	x	x	x	-	-	x	x	x	x	x	x	x	/	x	x	x	/	x	x	-	x	x	x
Teixeira et al., 2012[186]	-	x	x	x	x	x	x	/	x	x	-	x	x	x	x	x	-	x	x	x	-	x	x	/	x	x	x	/	x	x	-	x	x	x
Troseid et al., 2013[187]	x	x	x	x	x	x	x	x	x	x	-	-	x	x	-	x	x	x	x	-	-	x	x	/	x	x	x	-	x	x	x	x	x	x
Volynets et al., 2012[143]	-	x	x	x	x	x	x	/	x	x	-	-	x	x	x	-	x	x	x	x	-	x	x	/	x	x	x	/	x	x	-	x	x	x
Wyatt et al., 1993[150]	x	-	-	x	x	x	x	x	x	x	-	-	x	x	-	-	-	x	x	x	x	x	-	x	x	x	x	x	x	-	-	x	x	-
Zak-Golab et al., 2013[188]	-	x	x	x	x	x	x	/	x	x	-	-	x	x	x	-	x	x	x	x	x	x	x	/	x	x	x	/	x	x	x	x	x	x
Zhang et al., 2015[133]	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	-	x	x	/	x	x	x	/	x	x	x	x	x	x

STROBE critical appraisal tool.	Title, abstract & Introduction			Methods								Results							Discussion & other information					
Study	Title & Abstract	Background	Objectives	Study design	Setting	Participants	Variables	Data sources	Bias	Study size	Quantitative variables	Statistical methods	Participants	Descriptive data	Outcome data	Main results	Other analyses	Key results	Limitations	Interpretation	Generalizability	Funding
Zhang et al., 2014[189]	-	x	x	x	x	x	x	/	x	x	-	-	x	x	x	-	x	x	x	x	-	x	x	/	x	x	x	x	x	x

Table 2.1 Results from critical appraisal according to STROBE guidelines for included articles
'x', found within study; '-', not found within study; '/', not applicable.

Table 2.2 Risk of bias assessment of the included articles

Risk of bias assessment	External validity				Internal validity						Score
	Representativeness to national population	True representation of the target population	Random sampling methods	Likelihood of nonresponse bias minimal	Data directly collected from participants	Acceptable case definition	Validated study tool used	Consistent data collections methods	Appropriate parameters of numerator & denominator	Summary of overall risk of study bias	
Amar et al., 2008[106]	-	-	-	x	-	-	-	-	-	-	1
Barcelo et al., 2016[153]	x	x	x	x	-	-	-	-	-	-	4
Cangemi et al., 2016[154]	x	-	x	x	-	-	-	-	-	-	3
Cariello et al., 2010[155]	x	x	x	x	-	-	-	-	-	-	4
Carnevale et al., 2017[156]	x	x	x	x	-	-	-	-	-	-	4
Caserta et al., 2003[157]	x	x	x	x	-	-	-	-	-	-	4

Risk of bias assessment	External validity				Internal validity						Score
	Representativeness to national population	True representation of the target population	Random sampling methods	Likelihood of nonresponse bias minimal	Data directly collected from participants	Acceptable case definition	Validated study tool used	Consistent data collections methods	Appropriate parameters of numerator & denominator	Summary of overall risk of study bias	
Caviglia et al., 2018[158]	x	x	x	x	-	-	-	x	-	x	6
Ciccia et al., 2017[159]	x	x	x	x	-	-	-	-	-	-	4
Di Leo et al., 2003[160]	x	-	x	x	-	-	-	-	-	-	3
Donnadieu-Rigole et al., 2018[161]	x	-	x	x	-	-	-	-	-	-	3
Du Plessis et al., 2013[162]	x	x	x	x	-	-	-	-	-	-	4
Duerksen et al., 2010[149]	x	-	x	x	-	-	-	-	-	x	4
Ficek et al., 2017[163]	x	x	x	x	-	-	-	-	-	-	4
Goebel et al., 2008[164]	x	-	x	x	-	-	-	-	-	x	4
Hendy et al., 2017[165]	x	-	x	x	-	-	-	-	-	-	3
Hilsden et al., 1999[166]	x	-	x	x	-	-	-	-	-	-	3
Jayashree et al., 2014[167]	-	-	-	x	-	-	-	-	-	-	1
Johnston et al., 2000[168]	-	-	-	x	-	-	-	-	-	x	2
Karthikeyan et al., 2018[169]	x	x	x	x	-	-	-	-	-	-	4
Kim et al., 2018[170]	x	-	x	x	-	-	-	-	-	-	3
Kvehaugen et al., 2017[171]	x	-	x	x	-	-	-	-	-	-	3

Risk of bias assessment	External validity				Internal validity						Score
	Representativeness to national population	True representation of the target population	Random sampling methods	Likelihood of nonresponse bias minimal	Data directly collected from participants	Acceptable case definition	Validated study tool used	Consistent data collections methods	Appropriate parameters of numerator & denominator	Summary of overall risk of study bias	
Lassenius et al., 2011[172]	x	-	x	x	-	-	-	-	-	x	4
Lindheim et al., 2017[173]	x	x	x	x	-	-	-	-	-	-	4
Lukaszyk et al., 2018[152]	x	x	x	x	-	-	-	-	-	x	5
Malickova et al., 2017[174]	x	-	x	x	-	-	-	-	-	-	3
Malyszko et al., 2014[175]	x	x	x	x	-	-	-	-	-	x	5
Mokkala et al., 2017[176]	x	-	x	x	-	-	-	-	-	-	3
Mokkala et al., 2016[177]	x	-	x	x	-	-	-	-	-	-	3
Mokkala et al., 2017[60]	x	x	x	x	-	-	-	-	-	-	4
Moreno-Navarrete et al., 2012[178]	x	-	-	x	-	-	-	-	-	-	2
Morkl et al., 2018[179]	x	x	x	x	-	-	-	-	-	-	4
Mujagic et al., 2014[180]	-	-	x	x	-	-	-	-	-	-	2
Nymark et al., 2009[181]	-	-	x	x	-	-	-	-	-	-	2
Ohlsson et al., 2017[88]	-	-	-	x	-	-	-	-	-	-	1
Qi et al., 2017[182]	x	x	x	x	-	-	-	-	-	x	5
Raparelli et al., 2017[102]	x	x	x	x	-	-	-	-	-	-	4

Risk of bias assessment	External validity				Internal validity						Score
	Representativeness to national population	True representation of the target population	Random sampling methods	Likelihood of nonresponse bias minimal	Data directly collected from participants	Acceptable case definition	Validated study tool used	Consistent data collections methods	Appropriate parameters of numerator & denominator	Summary of overall risk of study bias	
Riordan et al., 1997[151]	x	x	x	x	-	x	-	-	-	x	6
Rutten et al., 2014[183]	x	x	x	x	-	-	-	-	x	x	6
Schwiertz et al., 2018[184]	x	-	-	x	-	-	-	-	-	x	3
Swanson et al., 2015[185]	x	x	x	x	-	-	-	-	-	-	4
Teixeira et al., 2012[186]	x	x	x	x	-	-	-	-	-	-	4
Troseid et al., 2013[187]	x	x	x	x	-	-	-	-	-	x	5
Volynets et al., 2012[143]	x	x	x	x	-	-	-	-	-	-	4
Wyatt et al., 1993[150]	x	x	x	x	-	x	-	-	-	x	6
Zak-Golab et al., 2013[188]	x	x	x	x	-	-	-	-	-	-	4
Zhang et al., 2015[133]	x	-	x	x	-	-	-	-	-	-	3
Zhang et al., 2014[189]	x	-	x	x	-	-	-	-	-	-	3

'x', risk of bias; '-', low risk of bias.

2.2.3.2 MEDICAL HISTORY AND DISEASE RISK FACTORS

Twenty studies reported a statistically significant association between IP and 19 reported medical history attributes. The diagnosis of particular health conditions such as diabetes,[60,172,189] liver disease,[143,155,170] and gastrointestinal conditions[150,160,166,180] were reported to be associated with altered IP. First, the likelihood of altered IP in type 2 diabetes ranges from OR=1.080 (95% CI: 1.005, 1.161; p=0.037) to OR=2.888 (95% CI: 1.553, 5.370; p<0.001) with the severity of IP associated with the odds of type 2 diabetes[189] (Table 2.3). Gestational diabetes was also reported to have a similar association with altered IP (OR=1.08; 95% CI: 1.02, 1.15; p=0.009).[60] Furthermore, the age of type 1 diabetes onset was reported to correlate with IP (β= -0.14; p<0.001)[172] (Table 2.4). The degree of liver damage in non-alcoholic fatty liver disease (NAFLD) was reported to have a moderate positive correlation with IP (r=0.69; p=0.01)[143] while the diagnosis of moderate-to-severe fatty liver was associated with altered IP (OR = 1.77; 95% CI: 1.13, 2.76; p=0.015).[170] Altered IP was reported to be associated with underlying organic digestive diseases (OR=1.56; 95% CI: 1.32, 1.85; p<0.0001);[160] although, altered IP was also reported to be an independent risk factors for diarrhoea-predominant irritable bowel syndrome (IBS-D) (β=0.63; 95% CI: 0.09, 1.16; p=0.022)[180] and has a moderate positive correlation with Crohn's disease relapse (r=0.48; p=0.008) (Table 2.5).[166] The association between disease duration of Parkinson's disease, inflammatory bowel disease and systemic sclerosis and IP were reported to have a weak to high correlation in three studies (r=0.73; p<0.011) with altered IP reported in the early stages of disease manifestation.[157,158,184]

Table 2.3 Risk factors associated with intestinal permeability according to adjusted odds ratio

Author	Study population	Sample size	Age (range)	Country	Test	Cut-off	Risk factor	Odds ratio	95% CI	*p* value
Medical history and disease										
Zhang et al., 2014[189]	Type 2 diabetes	102	47±12	China	Zonulin	6±4 ng/mL	Type 2 diabetes	1.140[g] 1.117[h] 1.080[i]	1.076, 1.208 1.051, 1.187 1.005, 1.161	*p*<0.001 *p*<0.001 *p*=0.037
Zhang et al., 2014[189]	Various glucose tolerance	130	47±12	China	Zonulin	4.3-7.6 ng/mL	Type 2 diabetes	1.966[i]	1.068, 3.618	*p*<0.001
Zhang et al., 2014[189]	Various glucose tolerance	120	47±12	China	Zonulin	7.6-27.2 ng/mL	Type 2 diabetes	2.888[i]	1.553, 5.370	*p*<0.001
Cariello et al., 2010[155]	Chronic liver disease	83	54.1 (28-78)	Italy	L/M	0.030%	Type 2 diabetes	2.7[a]	0.8, 3.0	*p*<0.01
Mokkala et al., 2017[60]	Pregnancy	88	30.1±4.9	Finland	Zonulin	45±10 ng/mL	Gestational diabetes	1.08[f]	1.02, 1.15	*p*=0.009
Kim et al., 2018[170]	Moderate-to-severe fatty liver	34	44.7±9.2 (30-60)	South Korea	Zonulin	6-21 ng/mL	Moderate-to-severe fatty liver	1.83[b] 1.77[c]	1.18, 2.84 1.13, 2.76	*p*=0.007 *p*=0.015

Author	Study population	Sample size	Age (range)	Country	Test	Cut-off	Risk factor	Odds ratio	95% CI	*p* value
Cariello et al., 2010[155]	Chronic liver disease	83	54.1 (28-78)	Italy	L/M	0.030%	Portal hypertension degree 1 Portal hypertension degree 2-3	2.0[a] 3.1[a]	0.7, 2.6 1.1, 4.2	*p*<0.01 *p*<0.01
Ohlsson et al., 2017[88]	General population	363	43 (28-53)	Sweden	Zonulin	34±14 ng/mL	Diastolic blood pressure 66-72 mm Hg	2.82[d]	1.43, 5.58	*p*=0.003
Di Leo et al., 2003[160]	Chronic diarrhoea	261	37±15 (18-83)	Italy	L/M	0.030%	Underlying organic disease	1.56[j]	1.32, 1.85	*p*<0.0001
Dietary factors										
Cariello et al., 2010	Chronic liver disease	83	54.1 (28-78)	Italy	L/M	0.030%	Alcohol use	2.1[a]	0.6, 2.8	*p*<0.01
Kim et al., 2018[170]	Moderate-to-severe fatty liver	34	44.7±9.2 (30-60)	South Korea	Zonulin	6-21 ng/mL	<14 standard drinks per week >15 standard drinks per week/>5 standard drinks in one setting	1.91[c] 1.56[c]	1.01, 3.95 1.02, 2.67	*p*=0.05 *p*=0.05
Anthropometric measurements										
Ohlsson et al., 2017[88]	General population	363	43 (28-53)	Sweden	Zonulin	54-64 ng/mL	Overweight, BMI >25	2.36[e]	1.07, 5.21[b]	*p*=0.033[b]
Ohlsson et al., 2017[88]	General population	363	43 (28-53)	Sweden	Zonulin	>64 ng/mL	Overweight, BMI >25 Obesity, BMI >30	4.10[e] 4.90[e]	1.87, 8.97[b]	*p*<0.001[b] *p*=0.047[b]

Author	Study population	Sample size	Age (range)	Country	Test	Cut-off	Risk factor	Odds ratio	95% CI	*p* value
Ohlsson et al., 2017[88]	General population	363	43 (28-53)	Sweden	Zonulin	34±14 ng/mL	Waist circumference >97cm	7.03[d]	1.49, 31.65[b] 1.97, 25.11	*p*=0.003
Biomarkers										
Ohlsson et al., 2017[88]	General population	363	43 (28-53)	Sweden	Zonulin	34±14 ng/mL	Plasma glucose levels (mmol/L) >5.7	2.09[d]	1.05, 4.18	*p*=0.036
Demographic factors										
Cariello et al., 2010[155]	Chronic liver disease	83	54.1 (28-78)	Italy	L/M	0.030%	Age > 50 years	1.9[a]	1.1, 2.3	*p*<0.001

BMI, body mass index; CI, confidence interval; L/M, lactulose/mannitol.
[a] Adjusted for age, alcohol use, associated diabetes, and degree of portal hypertension; [b] Adjusted for HbA1c, LDL, HDL, and BMI; [c] Adjusted for HbA1c, LDL, HDL, BMI, age, smoking status, alcohol consumption, physical activity and the use of diabetes, and dyslipidemia medication; [d] Adjusted for weight, BMI, waist and hip circumference, systolic and diastolic blood pressure, and fasting glucose levels; [e] Adjusted for systolic and diastolic blood pressure, and fasting glucose levels; [f] Adjusted for BMI, previous gestational diabetes, original intervention group; [g] Adjusted for age and gender; [h] Adjusted for age, gender, BMI and waist to hip ratio; [i] Adjusted for age, gender, BMI, waist to hip ratio, LDL, HDL, triglycerides and total cholesterol; [j] Adjusted for age.

Table 2.4 Risk factors associated with intestinal permeability according to beta correlation coefficient

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	*β*	95% CI	*p* value
Medical history and disease									
Mujagic et al., 2014[180]	IBS	91	44.4±1.6 (18-75)	Netherlands	L/R	IBS-D	0.63[f]	0.09, 1.16	*p*=0.022
Wyatt et al., 1993[150]	Crohn's disease	72	37 (>18)	Austria	L/M	Crohn's disease relapse	3.54[n]	-	*p*<0.0001

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	*β*	95% CI	*p* value
Lassenius et al., 2011[172]	Type 1 diabetes	911	46 (36-56)	Finland	LPS	Age of type 1 diabetes onset	-0.14[q]	-	*p<0.001*
Lassenius et al., 2011[172]	Type 1 diabetes	911	46 (36-56)	Finland	LPS	Diastolic blood pressure	0.10[q]	-	*p=0.004*
Dietary factors									
Amar et al., 2008[106]	General population	201	53.9±6.1 (45-64)	France	LPS	Total energy intake	132.2[n] 121.8[o]	62.7 (SE) 57.7 (SE)	*p=0.04* *p=0.04*
Mokkala et al., 2016[177]	Pregnancy (BMI 30)	100	29.4±4.9 (18-45)	Finland	Zonulin	Dietary protein	-0.139[d]	-0.247, 0.031	*p*=0.01
Zak-Golab et al., 2013[188]	Various BMI	80	48 (32-63)	Poland	Zonulin	Fat percentage in diet	0.23[i]	±0.11	*p*<0.05
Anthropometric factors									
Zak-Golab et al., 2013[188]	Various BMI	80	48 (32-63)	Poland	Zonulin	BMI	0.26[h]	±0.10	*p*<0.05
Donnadieu-Rigole et al., 2018[161]	Alcohol use disorder	41	48.2±8.7 (>18)	France	Zonulin	BMI	1.507[m]	0.34 (SEM)	*p*<0.01
Biomarkers									
Kvehaugen et al., 2017[171]	Obesity (BMI 35-55)	140	43.1 (>18)	Norway	Zonulin	CRP	3.28[c]	1.10, 5.46	*p*<0.01
Mokkala et al., 2017[176]	Pregnancy (BMI 30)	100	29.4±4.9 (18-45)	Finland	Zonulin	hsCRP	0.013[b]	0.003, 0.023	*p*=0.015

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	β	95% CI	*p* value
Barcelo et al., 2016[153]	Obstructive sleep apnoea	38	50±12 (18-74)	Spain	Zonulin	hsCRP	0.075[r]	0.008, 0.158	*p*=0.046
Moreno-Navarrete et al., 2012[178]	Various glucose tolerance	123	52±11.7	Spain	Zonulin	Circulating IL-6	0.23[l]	-	*p*=0.04
Ficek et al., 2017[163]	Haemodialysis patients	150	62 (59-64)	Poland	LPS	Circulating IL-6	0.171[s]	-	*p*=0.04
Mokkala et al., 2017[176]	Pregnancy (BMI 30)	100	29.4±4.9 (18-45)	Finland	Zonulin	GlycA	0.004[b]	0.002, 0.006	*p*<.001
Mokkala et al., 2017[176]	Pregnancy (BMI 30)	100	29.4±4.9 (18-45)	Finland	Zonulin	Insulin	0.015[b]	0.007, 0.022	*p*<0.001
Mokkala et al., 2017[176]	Pregnancy (BMI 30)	100	29.4±4.9 (18-45)	Finland	Zonulin	Insulin resistance (HOMA)	0.015[b]	0.007, 0.022	*p*<0.001
Zhang et al., 2014[189]	Type 2 diabetes	102	47±12	China	Zonulin	Insulin resistance (HOMA)	0.024[e]	0.009 (SE)	*p*=0.005
Mokkala et al., 2017[176]	Pregnancy (BMI 30)	100	29.4±4.9 (18-45)	Finland	Zonulin	Insulin sensitivity (QUICKI)	-0.002[b]	- 0.003, -0.001	*p*<0.001
Moreno-Navarrete et al., 2012[178]	Various glucose tolerance	123	52±11.7	Spain	Zonulin	Insulin sensitivity	-0.263[k]	-	*p*=0.004
Zak-Golab et al., 2013[188]	Various BMI	80	48 (32-63)	Poland	Zonulin	Glucose	0.38[j]	±0.12	*p*<0.05
Mokkala et al., 2017[176]	Pregnancy (BMI 30)	100	29.4±4.9 (18-45)	Finland	Zonulin	Triglycerides	0.009[b]	0.003, 0.015	*p*=0.003

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	*β*	95% CI	*p* value
Lassenius et al., 2011[172]	Type 1 diabetes	911	46 (36-56)	Finland	LPS	Triglycerides	0.69[q]	-	*p<0.001*
Mokkala et al., 2017[176]	Pregnancy (BMI 30)	100	29.4±4.9 (18-45)	Finland	Zonulin	Total cholesterol	0.004[b]	0.000, 0.007	*p*=0.032
Barcelo et al., 2016[153]	Obstructive sleep apnoea	38	50±12 (18-74)	Spain	Zonulin	Alanine transaminase	0.014[r]	0.001, 0.028	*p*=0.04
Barcelo et al., 2016[153]	Obstructive sleep apnoea	38	50±12 (18-74)	Spain	Zonulin	Aspartate transaminase	0.02[r]	0.002, 0.037	*p*=0.04
Cangemi et al., 2016[154]	Pneumonia	278	70±16	Italy	LPS	sP-selection	0.415[p]	-	*p<0.001*
Raparelli et al., 2017[102]	Liver cirrhosis	69	62.6±13.5	Italy	LPS	sCD40L	0.43[a]	-	*p*<0.0001
Mokkala et al., 2017[176]	Pregnancy (BMI 30)	100	29.4±4.9 (18-45)	Finland	LPS	Zonulin	0.002[b]	0.001, 0.003	*p*=0.002
Malyszko et al., 2014[175]	Kidney transplant recipients	72	45.5±12.2	Poland	Zonulin	Total serum protein	-0.51[g]	-	*p*=0.014
Malyszko et al., 2014[175]	Kidney transplant recipients	72	45.5±12.2	Poland	Zonulin	Thyroglobulin-binding protein	0.47[g]	-	*p*=0.03
Zak-Golab et al., 2013[188]	Various BMI	80	48 (32-63)	Poland	Zonulin	Microbiota bacteria count	0.33[h]	±0.13	*p*<0.05
Lassenius et al., 2011[172]	Type 1 diabetes	911	46 (36-56)	Finland	LPS	uMCP1/ creatinine ratio	0.10[q]	-	*p=0.003*

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	*β*	95% CI	*p* value
Demographic factors									
Zak-Golab et al., 2013[188]	Various BMI	80	48 (32-63)	Poland	Zonulin	Age	0.31[h]	±0.06	*p*<0.05

BMI, body mass index; CI, confidence interval; GlycA, glycoprotein acetylation; hsCRP, high sensitivity C-reactive protein; HOMA-IR, homeostatic model assessment; IBS-D, diarrhoea predominant irritable bowel syndrome; IBS, irritable bowel syndrome; IL-6, interleukin 6; L/M, lactulose/mannitol; LPS, lipopolysaccharides; L/R, lactulose/rhamnose; QUICKI, quantitative insulin-sensitivity check index; SE, standard error; sP-selectin, plasma soluble P-selectin; SEM, structural equation modelling; sCD40L, soluble cluster of differentiation 40 ligand; uMCP1, urinary monocyte chemoattractant protein-1.

[a] Multivariate analysis adjusted for sex, age, Child Pugh score and LPS; [b] Multiple linear regression analysis adjusted for log-transformed BMI and gestational weeks; [c] Linear regression adjusting for age, sex, and BMI; [d] Multiple linear regression analysis model including protein and polyunsaturated fatty acids; [e] Multiple stepwise linear regression analysis adjusted for age, BMI, waist to hip ratio, triglycerides, total cholesterol, HbA1c, HDL, LDL, IL-6, TNF-α, uric acid and zonulin; [f] Linear regression analysis model including demographical factors, medication, psychological symptoms and lifestyle; [g] Multiple regression analysis model including systolic and diastolic blood pressure, hemoglobin, erythrocyte count, fasting glucose, thyroglobulin-binding protein, total protein, and treatment with angiotensin-converting enzyme inhibitors; [h] Multiple regression analysis model including age, BMI, total bacterial, Bacteroides and Firmicutes counts; [i] Multiple regression analysis model including energy intake and macronutrients content; [j] Multiple regression analysis model including parameters of carbohydrate and lipid metabolism; [k] Multiple regression analysis model including age, BMI, log insulin sensitivity and log fasting triglycerides; [l] Multiple regression analysis model including age, BMI, log insulin sensitivity, log fasting triglycerides and IL-6; [m] Multiple regression analysis model including factors known to influence bacteria translocation and associated factors; [n] Multivariate analysis adjusted for age, physical activity, BMI, and residuals from linear regression of energy on protein, carbohydrates and alcohol in subjects with LPS >39 U/L; [o] Multivariate analysis adjusted for age, physical activity, BMI, and residuals from linear regression of energy on protein carbohydrates and alcohol in subjects with LPS >39 U/L excluding energy from alcohol; [p] Multivariable regression analysis adjusting for clinical characteristics; [q] Multivariate linear regression analysis adjusted for clinical variables; [r] Multiple regression analysis adjusted for age, sex, BMI, and metabolic syndrome components; [s] Multivariable regression analysis adjusting for zonulin, LPS, D-lactates.

Table 2.5 Risk factors associated with intestinal permeability according to correlation coefficient

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	Correlation	*p* value
Medical history and disease								
Caserta et al., 2003[157]	Systemic sclerosis	32	45.7±10.9	Italy	C/M	Disease duration	*r*=0.73	*p*<0.011
Caviglia et al., 2018[158]	Inflammatory bowel disease	118	49 (18-77)	Italy	Zonulin	Disease duration	ρ=-0.30	*p*=0.001

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	Correlation	*p* value
Schwiertz et al., 2018[184]	Parkinson's disease	36	65.5 (44-78)	Germany	Stool zonulin	Disease duration	*r*=-0.34	*p*=0.042
Hilsden et al., 1999[166]	Crohn's disease	61	36 (18-66)	Canada	L/M	Crohn's disease relapse	*r*=0.48	*p*=0.008
Mujagic et al., 2014[180]	IBS	91	44.4±1.6 (18-75)	Netherlands	L/R	Diarrhoea	ρ=0.17	*p*<0.05
Mujagic et al., 2014[180]	IBS	91	44.4±1.6 (18-75)	Netherlands	L/R	Indigestion syndrome	ρ=0.17	*p*<0.05
Goebel et al., 2008[164]	Fibromyalgia	40	48±11 (18-65)	Germany	L/M	Pain (NRS)	*r*=-0.3	*p*<0.05
Goebel et al., 2008[164]	Complex regional pain syndrome	17	43±13 (18-65)	Germany	L/M	Pain (NRS)	*r*=0.19	*p*<0.05
Du Plessis et al., 2013[162]	Liver cirrhosis	29	60±10 (44-63)	South Africa	LPS	Child-Pugh score	*r*=0.292	*p*=0.03
Malyszko et al., 2014[175]	Kidney transplant recipients	72	45.5±12.2	Poland	Zonulin	Systolic blood pressure	*r*=-0.33	*p*<0.05
Ohlsson et al., 2017[88]	General population	363	43 (28-53)	Sweden	Zonulin	Systolic blood pressure	ρ=0.120	*p*=0.024
Troseid et al., 2013[187]	Obesity (BMI 45)	49	42.9±9.2 (28-55)	Norway	LPS	Systolic blood pressure	*r*=0.40	*p*=0.009

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	Correlation	*p* value
Ohlsson et al., 2017[88]	General population	363	43 (28-53)	Sweden	Zonulin	Diastolic blood pressure	ρ=0.178	*p*=0.001
Rutten et al., 2014[183]	COPD	18	63.6±1.3	Netherlands	L/R	COPD	*r*=0.67	*p*<0.01
Riordan et al., 1997[151]	SIBO	34	64 (22-95)	Australia	L/M	SIBO	*r*=0.61	*p*<0.0005
Zhang et al., 2015[133]	PCOS	78	29±5	China	Zonulin	Number of menstrual cycles	ρ=-0.401[a]	*p*<0.001
Schwiertz et al., 2018[184]	Parkinson's disease	36	65.5 (44-78)	Germany	Zonulin	Levodopa dose	*r*=-0.39	*p*=0.019
Volynets et al., 2012[143]	NAFLD	20	41.9±2.3	Germany	LPS	Degree of liver damage	*r*=0.69	*p*=0.01
Dietary factors								
Morkl et al., 2018[179]	Women (BMI 13-46)	102	24.6±4.6	Austria	Zonulin	Total energy intake	ρ=0.230	*p*=0.036
Zak-Golab et al., 2013[188]	Various BMI	80	48 (32-63)	Poland	Zonulin	Total energy intake	ρ=0.27	*p*<0.05
Zak-Golab et al., 2013[188]	Various BMI	80	48 (32-63)	Poland	Zonulin	Protein intake	ρ=-0.23	*p*<0.05
Mokkala et al., 2016[177]	Pregnancy (BMI 30)	95	29.4±4.9 (18-45)	Finland	Zonulin	Protein intake	ρ=-0.291	*p*=0.004

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	Correlation	*p* value
Morkl et al., 2018[179]	Women (BMI 13-46)	102	24.6±4.6	Austria	Zonulin	Protein intake	ρ=0.208	*p*=0.036
Volynets et al., 2012[143]	NAFLD	20	41.9±2.3	Germany	LPS	Protein intake	ρ=0.59	*p*=0.001
Volynets et al., 2012[143]	NAFLD	20	41.9±2.3	Germany	LPS	Animal-derived protein intake	ρ=0.54	*p*=0.002
Morkl et al., 2018[179]	Women (BMI 13-46)	102	24.6±4.6	Austria	Zonulin	Carbohydrate intake	ρ=0.221	*p*=0.025
Mokkala et al., 2016[177]	Pregnancy (BMI 30)	95	29.4±4.9 (18-45)	Finland	Zonulin	PUFAs intake	ρ=-0.224	*p*=0.03
Mokkala et al., 2016[177]	Pregnancy (BMI 30)	95	29.4±4.9 (18-45)	Finland	Zonulin	PUFAs n-6 intake	ρ=-0.247	*p*=0.01
Mokkala et al., 2016[177]	Pregnancy (BMI 30)	95	29.4±4.9 (18-45)	Finland	Zonulin	Vitamin E intake	ρ=-0.228	*p*=0.02
Mokkala et al., 2016[177]	Pregnancy (BMI 30)	95	29.4±4.9 (18-45)	Finland	Zonulin	Magnesium intake	ρ=-0.291	*p*=0.004
Mokkala et al., 2016[177]	Pregnancy (BMI 30)	95	29.4±4.9 (18-45)	Finland	Zonulin	Niacin intake	ρ=-0.291	*p*=0.004
Mokkala et al., 2016[177]	Pregnancy (BMI 30)	95	29.4±4.9 (18-45)	Finland	Zonulin	Iron intake	ρ=-0.228	*p*=0.02
Mokkala et al., 2016[177]	Pregnancy (BMI 30)	95	29.4±4.9 (18-45)	Finland	Zonulin	Potassium intake	ρ=-0.343	*p*=0.001

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	Correlation	*p* value
Morkl et al., 2018[179]	Women (BMI 13-46)	102	24.6±4.6	Austria	Zonulin	Sodium intake	ρ=0.207	*p*=0.037
Morkl et al., 2018[179]	Women (BMI 13-46)	102	24.6±4.6	Austria	Zonulin	Vitamin B12 intake	ρ=0.198	*p*=0.046
Anthropometric measurements								
Barcelo et al., 2016[153]	Overweight (BMI 29)	38	49±12 (18-73)	Spain	Zonulin	Waist circumference	ρ=0.382	*p*=0.04
Barcelo et al., 2016[153]	Obstructive sleep apnoea	38	50±12 (18-74)	Spain	Zonulin	Waist circumference	ρ=0.442	*p*=0.004
Morkl et al., 2018[179]	Women (BMI 13-46)	102	24.6±4.6	Austria	Zonulin	Waist circumference	ρ=0.263	*p*=0.007
Ohlsson et al., 2017[88]	General population	363	43 (28-53)	Sweden	Zonulin	Waist circumference	ρ=0.271	*p*<0.001
Morkl et al., 2018[179]	Women (BMI 13-46)	102	24.6±4.6	Austria	Zonulin	Hip circumference	ρ=0.231	*p*=0.202
Ohlsson et al., 2017[88]	General population	363	43 (28-53)	Sweden	Zonulin	Hip circumference	ρ=0.173	*p*=0.001
Moreno-Navarrete et al., 2012[178]	Various glucose tolerance	123	52±11.7	Spain	Zonulin	Waist to hip ratio	*r*=0.2	*p*=0.025
Zhang et al., 2015[133]	PCOS	78	29±5	China	Zonulin	Waist-to-hip ratio	*r*=0.401	*p*=0.015

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	Correlation	*p* value
Zhang et al., 2014[189]	General population	95	34±11	China	Zonulin	Waist-to-hip ratio	*r*=0.241[b]	*p*=0.011
Zhang et al., 2014[189]	Various glucose tolerance	290	40±19	China	Zonulin	Waist-to-hip ratio	*r*=0.200[b]	*p*=0.022
Teixeira et al., 2012[186]	Various BMI	40	28.5±7.6/30.7±6.5	Brazil	L/M	Abdomen circumference	ρ=0.30	*p*=0.05
Hendy et al., 2017[165]	NAFLD	56	37.2±6.8 (29-46)	Egypt	Zonulin	BMI	*r*=0.378	*p*<0.05
Zak-Golab et al., 2013[188]	Various BMI	80	48 (32-63)	Poland	Zonulin	BMI	ρ=0.41	*p*<0.001
Moreno-Navarrete et al., 2012[178]	Various glucose tolerance	123	52±11.7	Spain	Zonulin	BMI	*r*=0.28	*p*=0.002
Moreno-Navarrete et al., 2012[178]	Glucose intolerance	41	55.9±10.3	Spain	Zonulin	BMI	*r*=0.42	*p*=0.007
Morkl et al., 2018[179]	Women (BMI 13-46)	102	24.6±4.6	Austria	Zonulin	BMI	ρ=0.235	*p*=0.017
Zhang et al., 2015[133]	PCOS	78	29±5	China	Zonulin	BMI	*r*=0.535	*p*<0.05
Zhang et al., 2014[189]	Various glucose tolerance	290	40±19	China	Zonulin	BMI	*r*=0.201	*p*=0.020

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	Correlation	*p* value
Ohlsson et al., 2017[88]	General population	363	43 (28-53)	Sweden	Zonulin	BMI	ρ=0.213	*p*<0.001
Troseid et al., 2013[187]	Obesity (BMI 45)	49	42.9±9.2 (28-55)	Norway	LPS	BMI	*r*=0.37	*p*=0.017
Zak-Golab et al., 2013[188]	Various BMI	80	48 (32-63)	Poland	Zonulin	Weight	ρ=0.34	*p*<0.01
Ohlsson et al., 2017[88]	General population	363	43 (28-53)	Sweden	Zonulin	Weight	ρ=0.193	*p*<0.001
Zak-Golab et al., 2013[188]	Various BMI	80	48 (32-63)	Poland	Zonulin	Fat mass	ρ=0.42	*p*<0.001
Zak-Golab et al., 2013[188]	Various BMI	80	48 (32-63)	Poland	Zonulin	Fat percentage	ρ=0.40	*p*<0.001
Morkl et al., 2018[179]	Women (BMI 13-46)	102	24.6±4.6	Austria	Zonulin	Fat percentage	ρ=0.205	*p*=0.039
Morkl et al., 2018[179]	Women (BMI 13-46)	102	24.6±4.6	Austria	Zonulin	Subcutaneous fat	ρ=0.244	*p*=0.013
Troseid et al., 2013[187]	Obesity (BMI 45)	49	42.9±9.2 (28-55)	Norway	LPS	Subcutaneous fat	*r*=0.33	*p*=0.038
Zhang et al., 2015[133]	PCOS	78	29±5	China	Zonulin	Visceral adiposity index	*r*=0.432	*p*=0.011
Troseid et al., 2013[187]	Obesity (BMI 45)	49	42.9±9.2 (28-55)	Norway	LPS	Intra-abdominal fat	*r*=0.61	*p*<0.001

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	Correlation	*p* value
Qi et al., 2018[182]	General population	37	23.1±3.9/76.7±5.2 (18->70)	America	Zonulin	Muscle strength	*r*=-0.332	*p*=0.048
Qi et al., 2018[182]	General population	37	23.1±3.9/76.7±5.2 (18->70)	America	Zonulin	Steps per day	*r*=-0.410	*p*=0.016
Biomarkers								
Glucose metabolism								
Barcelo et al., 2016[153]	Obstructive sleep apnoea	38	50±12 (18-74)	Spain	Zonulin	Fasting glucose	ρ=0.321	*p*=0.04
Barcelo et al., 2016[153]	Overweight (BMI 29)	38	49±12 (18-73)	Spain	Zonulin	Fasting glucose	ρ=0.343	*p*=0.035
Ohlsson et al., 2017[88]	General population	363	43 (28-53)	Sweden	Zonulin	Fasting glucose	ρ=0.138	*p*=0.009
Malyszko et al., 2014[175]	Kidney transplant recipients	72	45.5±12.2	Poland	Zonulin	Fasting glucose	*r*=-0.25	*p*<0.05
Zhang et al., 2014[189]	Various glucose tolerance	290	40±19	China	Zonulin	Fasting glucose	*r*=0.300[b]	*p*=0.001
Zhang et al., 2014[189]	Type 2 diabetes	102	47±12	China	Zonulin	Fasting glucose	*r*=0.299[b]	*p*=0.010
Zak-Golab et al., 2013[188]	Various BMI	80	48 (32-63)	Poland	Zonulin	Fasting glucose	ρ=0.18	*p*<0.05

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	Correlation	*p* value
Jayashree et al., 2014[167]	Type 2 diabetes	45	51±6 (30-60)	India	LPS	Fasting glucose	*r*=0.229	*p*=0.026
Zhang et al., 2015[133]	PCOS	78	29±5	China	Zonulin	Glucose tolerance at 0 min	*r*=0.351[b]	*p*=0.045
Zhang et al., 2015[133]	PCOS	78	29±5	China	Zonulin	Glucose tolerance at 120 min	*r*=0.347[b]	*p*=0.045
Zhang et al., 2014[189]	Various glucose tolerance	290	40±19	China	Zonulin	Glucose tolerance at 120 min	*r*=0.213[b]	*p*=0.016
Zhang et al., 2014[189]	Glucose intolerance	92	39±13	China	Zonulin	Glucose tolerance at 120 min	*r*=0.325[b]	*p*<0.05
Zhang et al., 2014[189]	Type 2 diabetes	102	47±12	China	Zonulin	Glucose tolerance at 120 min	*r*=0.342[b]	*p*<0.05
Jayashree et al., 2014[167]	Type 2 diabetes	45	51±6 (30-60)	India	LPS	Glucose tolerance at 120 min	*r*=0.341	p<0.001
Jayashree et al., 2014[167]	Type 2 diabetes	45	51±6 (30-60)	India	LPS	HbA1c	*r*=0.334	*p*<0.001
Troseid et al., 2013[187]	Obesity (BMI 45)	49	42.9±9.2 (28-55)	Norway	LPS	HbA1c	*r*=0.56	*p*=0.001
Moreno-Navarrete et al., 2012[178]	General population	82	48.3±11.7	Spain	Zonulin	HbA1c	*r*=0.24	*p*=0.03

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	Correlation	*p* value
Zhang et al., 2014[189]	Various glucose tolerance	290	40±19	China	Zonulin	HbA1c	r=0.302[b]	p=0.002
Zhang et al., 2014[189]	Type 2 diabetes	102	47±12	China	Zonulin	HbA1c	r=0.231[b]	p=0.048
Barcelo et al., 2016[153]	Obstructive sleep apnoea	38	50±12 (18-74)	Spain	Zonulin	Fasting insulin	ρ=0.351	p=0.03
Barcelo et al., 2016[153]	Overweight (BMI 29)	38	49±12 (18-73)	Spain	Zonulin	Fasting insulin	ρ=0.328	p=0.041
Hendy et al., 2017[165]	NAFLD	56	37.2±6.8 (29-46)	Egypt	Zonulin	Fasting insulin	r=0.305	p<0.05
Zhang et al., 2014[189]	Glucose intolerance	92	39±13	China	Zonulin	Fasting insulin	r=0.267[b]	p=0.004
Zhang et al., 2014[189]	Type 2 diabetes	102	47±12	China	Zonulin	Fasting insulin	r=0.325[b]	p=0.005
Mokkala et al., 2017[176]	Pregnancy (BMI 30)	75	29.4±4.9 (18-45)	Finland	Zonulin	Fasting insulin	ρ=0.616[c]	p<0.001
Mokkala et al., 2017[176]	Pregnancy (BMI 30)	75	29.4±4.9 (18-45)	Finland	LPS	Fasting insulin	ρ=0.264[d]	p=0.02
Zhang et al., 2015[133]	PCOS	78	29±5	China	Zonulin	Insulin sensitivity (OGTT at 0 min)	r=0.605	p<0.05
Zhang et al., 2015[133]	PCOS	78	29±5	China	Zonulin	Insulin sensitivity (OGTT at 120 min)	r=0.527	p=0.001

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	Correlation	*p* value
Zhang et al., 2015[133]	PCOS	78	29±5	China	Zonulin	Insulin sensitivity index	*r*=0.262[b]	*p*=0.019
Moreno-Navarrete et al., 2012[178]	Various glucose tolerance	123	52±11.7	Spain	Zonulin	Insulin sensitivity	*r*=-0.28	*p*=0.002
Moreno-Navarrete et al., 2012[178]	General population	82	48.3±11.7	Spain	Zonulin	Insulin sensitivity	*r*=-0.22	*p*=0.045
Moreno-Navarrete et al., 2012[178]	Glucose intolerance	41	55.9±10.3	Spain	Zonulin	Insulin sensitivity	*r*=-0.36	*p*=0.02
Mokkala et al., 2017[176]	Pregnancy (BMI 30)	75	29.4±4.9 (18-45)	Finland	LPS	Insulin sensitivity (QUICKI)	ρ=-0.245[d]	*p*=0.03
Mokkala et al., 2017[176]	Pregnancy (BMI 30)	75	29.4±4.9 (18-45)	Finland	Zonulin	Insulin sensitivity (QUICKI)	ρ=-0.600[c]	*p*<0.001
Zhang et al., 2014[189]	Glucose intolerance	92	39±13	China	Zonulin	Insulin sensitivity (QUICKI)	*r*=-0.311[b]	*p*=0.001
Zhang et al., 2014[189]	Various glucose tolerance	290	40±19	China	Zonulin	Insulin sensitivity (QUICKI)	*r*=-0.214[b]	*p*=0.016
Zhang et al., 2014[189]	Type 2 diabetes	102	47±12	China	Zonulin	Insulin sensitivity (QUICKI)	*r*=-0.295[b]	*p*=0.001

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	Correlation	*p* value
Teixeira et al., 2012[186]	Various BMI	40	28.5±7.6/30.7±6.5	Brazil	L/M	Insulin resistance (HOMA)	ρ=0.3	*p*=0.014
Hendy et al., 2017[165]	NAFLD	56	37.2±6.8 (29-46)	Egypt	Zonulin	Insulin resistance (HOMA)	*r*=0.413	*p*<0.01
Mokkala et al., 2017[176]	Pregnancy (BMI 30)	75	29.4±4.9 (18-45)	Finland	Zonulin	Insulin resistance (HOMA)	ρ=0.616[c]	*p*<0.001
Zhang et al., 2015[133]	PCOS	78	29±5	China	Zonulin	Insulin resistance (HOMA)	*r*=0.315[b]	*p*=0.044
Zhang et al., 2014[189]	Various glucose tolerance	290	40±19	China	Zonulin	Insulin resistance (HOMA)	*r*=0.281[b]	*p*=0.001
Zhang et al., 2014[189]	Glucose intolerance	92	39±13	China	Zonulin	Insulin resistance (HOMA)	*r*=0.274[b]	*p*=0.003
Zhang et al., 2014[189]	Type 2 diabetes	102	47±12	China	Zonulin	Insulin resistance (HOMA)	*r*=0.434[b]	*p*<0.05
Mokkala et al., 2017[176]	Pregnancy (BMI 30)	75	29.4±4.9 (18-45)	Finland	LPS	Insulin resistance (HOMA)	ρ=0.264[d]	*p*=0.02
Cholesterol and triglycerides								
Barcelo et al., 2016[153]	Obstructive sleep apnoea	38	50±12 (18-74)	Spain	Zonulin	Total cholesterol	ρ=0.397	*p*=0.011
Mokkala et al., 2017[176]	Pregnancy (BMI 30)	75	29.4±4.9 (18-45)	Finland	Zonulin	Total cholesterol	ρ=0.566[c]	*p*<0.001

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	Correlation	*p* value
Zhang et al., 2014[189]	Various glucose tolerance	290	40±19	China	Zonulin	Total cholesterol	r=0.333[b]	p<0.05
Zhang et al., 2014[189]	Type 2 diabetes	102	47±12	China	Zonulin	Total cholesterol	r=0.245[b]	p=0.018
Nymark et al., 2009[181]	Type one diabetes	477	36.5±11	Finland	LPS	Total cholesterol	r=0.34	p<0.001
Mokkala et al., 2017[176]	Pregnancy (BMI 30)	75	29.4±4.9 (18-45)	Finland	LPS	Total cholesterol	ρ=0.374[d]	p=0.001
Mokkala et al., 2017[176]	Pregnancy (BMI 30)	75	29.4±4.9 (18-45)	Finland	Zonulin	LDL cholesterol	ρ=0.458[c]	p=0.001
Zhang et al., 2014[189]	Type 2 diabetes	102	47±12	China	Zonulin	LDL cholesterol	r=0.362[b]	p=0.002
Mokkala et al., 2017[176]	Pregnancy (BMI 30)	75	29.4±4.9 (18-45)	Finland	LPS	LDL cholesterol	ρ=0.264[d]	p=0.01
Moreno-Navarrete et al., 2012[178]	General population	82	48.3±11.7	Spain	Zonulin	HDL cholesterol	r=-0.27	p=0.01
Moreno-Navarrete et al., 2012[178]	Various glucose tolerance	123	52±11.7	Spain	Zonulin	HDL cholesterol	r=-0.21	p=0.02
Zhang et al., 2014[189]	Various glucose tolerance	290	40±19	China	Zonulin	HDL cholesterol	r=-0.342[b]	p<0.05

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	Correlation	*p* value
Zhang et al., 2015[133]	PCOS	78	29±5	China	Zonulin	HDL cholesterol	*r*=-0.412	*p*=0.031
Zhang et al., 2014[189]	General population	95	34±11	China	Zonulin	HDL cholesterol	*r*=-0.390[b]	*p*<0.05
Hendy et al., 2017[165]	NAFLD	56	37.2±6.8 (29-46)	Egypt	Zonulin	HDL cholesterol	*r*=-0.397	*p*<0.01
Nymark et al., 2009[181]	Type one diabetes	477	36.5±11	Finland	LPS	HDL cholesterol	*r*=-0.24	*p*<0.001
Troseid et al., 2013[187]	Obesity (BMI 45)	49	42.9±9.2 (28-55)	Norway	LPS	HDL cholesterol	*r*=-0.43	*p*=0.006
Mokkala et al., 2017[176]	Pregnancy (BMI 30)	75	29.4±4.9 (18-45)	Finland	LPS	HDL cholesterol	ρ=0.240[d]	*p*=0.03
Jayashree et al., 2014[167]	Type 2 diabetes	45	51±6 (30-60)	India	LPS	HDL cholesterol	*r*=0.531	*p*<0.001
Teixeira et al., 2012[186]	Various BMI	40	28.5±7.6/30.7±6.5	Brazil	L/M	HDL cholesterol	ρ=-0.39	*p*=0.01
Hendy et al., 2017[165]	NAFLD	56	37.2±6.8 (29-46)	Egypt	Zonulin	Triglycerides	*r*=0.296	*p*<0.05
Mokkala et al., 2017[176]	Pregnancy (BMI 30)	75	29.4±4.9 (18-45)	Finland	Zonulin	Triglycerides	ρ=0.529[c]	*p*<0.001
Morkl et al., 2018[179]	Women (BMI 13-46)	102	24.6±4.6	Austria	Zonulin	Triglycerides	ρ=0.283	*p*=0.004

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	Correlation	*p* value
Moreno-Navarrete et al., 2012[178]	Various glucose tolerance	123	52±11.7	Spain	Zonulin	Triglycerides	*r*=0.21	*p*=0.02
Moreno-Navarrete et al., 2012[178]	General population	82	48.3±11.7	Spain	Zonulin	Triglycerides	*r*=0.22	*p*=0.045
Zhang et al., 2015[133]	PCOS	78	29±5	China	Zonulin	Triglycerides	*r*=0.422	*p*=0.031
Zhang et al., 2014[189]	Various glucose tolerance	290	40±19	China	Zonulin	Triglycerides	*r*=0.449[b]	*p*<0.05
Zhang et al., 2014[189]	General population	95	34±11	China	Zonulin	Triglycerides	*r*=0.329[b]	*p*=0.002
Zhang et al., 2014[189]	Glucose intolerance	92	39±13	China	Zonulin	Triglycerides	*r*=0.501[b]	*p*<0.05
Lassenius et al., 2011[172]	Type one diabetes	904	46 (36-56)	Finland	LPS	Triglycerides	*r*=0.73	*p*<0.001
Troseid et al., 2013[187]	Obesity (BMI 45)	49	42.9±9.2 (28-55)	Norway	LPS	Triglycerides	*r*=0.52	*p*=0.001
Nymark et al., 2009[181]	Type one diabetes	477	36.5±11	Finland	LPS	Triglycerides	*r*=0.61	*p*<0.001
Jayashree et al., 2014[167]	Type 2 diabetes	45	51±6 (30-60)	India	LPS	Triglycerides	*r*=0.353	*p*<0.001

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	Correlation	*p* value
Mokkala et al., 2017[176]	Pregnancy (BMI 30)	75	29.4±4.9 (18-45)	Finland	LPS	Triglycerides	ρ=0.245[d]	*p*<0.001
Nymark et al., 2009[181]	Type one diabetes	477	36.5±11	Finland	LPS	ApoB	*r*=0.34	*p*<0.001
Inflammatory markers								
Barcelo et al., 2016[153]	Obstructive sleep apnoea	38	50±12 (18-74)	Spain	Zonulin	hsCRP	ρ=0.372	*p*=0.02
Karthikeyan et al., 2018[169]	Liver cirrhosis	30	47.7±1.4 (18-60)	India	Zonulin	hsCRP	ρ=0.482	*p*=0.0063
Mokkala et al., 2017[176]	Pregnancy (BMI 30)	75	29.4±4.9 (18-45)	Finland	Zonulin	hsCRP	ρ=0.412[c]	*p*=0.004
Morkl et al., 2018[179]	Women (BMI 13-46)	102	24.6±4.6	Austria	Zonulin	CRP	ρ=0.293	P=0.003
Hendy et al., 2017[165]	NAFLD	56	37.2±6.8 (29-46)	Egypt	Zonulin	Circulating IL-6	*r*=0.288	*p*<0.05
Moreno-Navarrete et al., 2012[178]	Various glucose tolerance	123	52±11.7	Spain	Zonulin	Circulating IL-6	*r*=0.29	*p*=0.008
Moreno-Navarrete et al., 2012[178]	General population	82	48.3±11.7	Spain	Zonulin	Circulating IL-6	*r*=0.31	*p*=0.01

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	Correlation	*p* value
Morkl et al., 2018[179]	Women (BMI 13-46)	102	24.6±4.6	Austria	Zonulin	Circulating IL-6	ρ=0.317	*p*=0.001
Qi et al., 2018[182]	General population	37	23.1±3.9/ 76.7±5.2 (18->70)	America	Zonulin	Circulating IL-6	*r*=0.345	*p*=0.043
Ficek et al., 2017[163]	Haemodialysis patients	150	62 (59-64)	Poland	LPS	Circulating IL-6	ρ=0.241	*p*=0.003
Jayashree et al., 2014[167]	Type 2 diabetes	45	51±6 (30-60)	India	LPS	Circulating IL-6	*r*=0.542	*p*<0.001
Qi et al., 2018[182]	General population	37	23.1±3.9/ 76.7±5.2 (18->70)	America	Zonulin	TNF-α	*r*=0.357	*p*=0.032
Zhang et al., 2014[189]	Various glucose tolerance	290	40±19	China	Zonulin	TNF-α	*r*=0.296[b]	*p*=0.010
Zhang et al., 2014[189]	General population	95	34±11	China	Zonulin	TNF-α	*r*=0.623[b]	*p*<0.05
Zhang et al., 2014[189]	Glucose intolerance	92	39±13	China	Zonulin	TNF-α	*r*=0.647[b]	*p*<0.05
Zhang et al., 2014[189]	Type 2 diabetes	102	47±12	China	Zonulin	TNF-α	*r*=0.352[b]	*p*=0.001
Jayashree et al., 2014[167]	Type 2 diabetes	45	51±6 (30-60)	India	LPS	TNF-α	*r*=0.407	*p*<0.001
Zak-Golab et al., 2013[188]	Various BMI	80	48 (32-63)	Poland	Zonulin	sTNFR1	ρ=0.34	*p*<0.001

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	Correlation	*p* value
Mokkala et al., 2017[176]	Pregnancy (BMI 30)	75	29.4±4.9 (18-45)	Finland	LPS	GlycA	ρ=0.387[d]	*p*=0.001
Mokkala et al., 2017[176]	Pregnancy (BMI 30)	75	29.4±4.9 (18-45)	Finland	Zonulin	GlycA	ρ=0.616[c]	*p*<0.001
Malickova et al., 2017[174]	Inflammatory bowel disease	40	(18-65)	Czech Republic	Stool zonulin	Stool calprotectin	ρ=0.430	*p*=0.006
Markers of intestinal permeability								
Ciccia et al., 2017[159]	Ankylosing spondylitis	20	47 (23-58)	Italy	L/M	Serum zonulin	ρ=0.851	*p*=0.0177
Duerksen et al., 2010[149]	Coeliac disease (Marsh type 3)	6	53.5	Canada	Zonulin	L/M	ρ=0.891	*p*=0.05
Jayashree et al., 2014[167]	Type 2 diabetes	45	51±6 (30-60)	India	Zonulin	LPS	*r*=0.252	*p*<0.01
Carnevale et al., 2017[156]	Glucose intolerance	70	62.5±13.2	Italy	Zonulin	LPS	*r*=0.529	*p*=0.001
Raparelli et al., 2017[102]	Liver cirrhosis (Child-Pugh B+C)	34	62.5±13.4	Italy	Zonulin	LPS	ρ=0.48	*p*<0.05
Mokkala et al., 2017[176]	Pregnancy (BMI 30)	75	29.4±4.9	Finland	Zonulin	LPS	ρ=0.458[c]	*p*=0.001
Cangemi et al., 2016[154]	Pneumonia	278	70±16	Italy	Zonulin	LPS	ρ=0.545	*p*<0.001

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	Correlation	*p* value
Goebel et al., 2008[164]	Fibromyalgia	40	48±11 (18-65)	Germany	L/M	Gastroduodenal permeability	*r*=0.68	*p*<0.0001
Goebel et al., 2008[164]	Complex regional pain syndrome	17	43±13 (18-65)	Germany	L/M	Gastroduodenal permeability	*r*=0.88	*p*<0.0001
Liver pathology								
Barcelo et al., 2016[153]	Obstructive sleep apnoea	38	50±12 (18-74)	Spain	Zonulin	Alanine transaminase	ρ=0.484	*p*=0.002
Hendy et al., 2017[165]	NAFLD	56	37.2±6.8 (29-46)	Egypt	Zonulin	Alanine transaminase	*r*=0.312	*p*<0.05
Volynets et al., 2012[143]	NAFLD	20	41.9±2.3	Germany	LPS	Alanine transaminase	ρ=0.50	*p*=0.005
Barcelo et al., 2016[153]	Obstructive sleep apnoea	38	50±12 (18-74)	Spain	Zonulin	Aspartate transaminase	ρ=0.426	*p*=0.006
Barcelo et al., 2016[153]	Obstructive sleep apnoea	38	50±12 (18-74)	Spain	Zonulin	Gamma glutamyltransferase	ρ=0.444	*p*=0.004
Hendy et al., 2017[165]	NAFLD	56	37.2±6.8 (29-46)	Egypt	Zonulin	Liver histopathology	*r*=0.518	*p*<0.001
Intestinal microbiome markers								
Lindheim et al., 2017[173]	PCOS	24	27	Austria	Zonulin	Microbial diversity	*r*=-0.334	*p*=0.029

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	Correlation	*p* value
Lindheim et al., 2017[173]	PCOS	24	27	Austria	Stool zonulin	Microbial diversity	ρ=-0.366	*p*=0.016
Zak-Golab et al., 2013[188]	Various BMI	80	48 (32-63)	Poland	Zonulin	Total bacteria count	ρ=0.26	*p*<0.05
Mokkala et al., 2016[177]	Pregnancy (BMI 30)	95	29.4±4.9 (18-45)	Finland	Zonulin	*Faecalibacterium* (genus)	ρ=0.29	*p*=0.004
Mokkala et al., 2016[177]	Pregnancy (BMI 30)	95	29.4±4.9 (18-45)	Finland	Zonulin	*Blautia* (genus)	ρ=-0.25	*p*=0.018
Mokkala et al., 2016[177]	Pregnancy (BMI 30)	95	29.4±4.9 (18-45)	Finland	Zonulin	*F. prausnitzii* (species)	ρ=0.29	*p*=0.005
Mokkala et al., 2016[177]	Pregnancy (BMI 30)	95	29.4±4.9 (18-45)	Finland	Zonulin	*Blautia* (species)	ρ=-0.25	*p*=0.018
Other biomarkers								
Barcelo et al., 2016[153]	Obstructive sleep apnoea	38	50±12 (18-74)	Spain	Zonulin	Mean oxygen saturation	ρ=-0.378	*p*=0.019
Lukaszyk et al., 2018[152]	Chronic kidney disease	35	73.9±10.9	Poland	Zonulin	Hepcidin with hsCRP >10mg/dL	*r*=-0.37	*p*<0.05
Malyszko et al., 2014[175]	Kidney transplant recipients	72	45.5±12.2	Poland	Zonulin	Thyroglobulin-binding protein	*r*=0.24	*p*<0.05

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	Correlation	*p* value
Malyszko et al., 2014[175]	Kidney transplant recipients	72	45.5±12.2	Poland	Zonulin	Haematocrit	r=0.28	p<0.05
Malyszko et al., 2014[175]	Kidney transplant recipients	72	45.5±12.2	Poland	Zonulin	Haemoglobin	r=0.32	p<0.01
Malyszko et al., 2014[175]	Kidney transplant recipients	72	45.5±12.2	Poland	Zonulin	Total protein	r=-0.33	p<0.05
Malyszko et al., 2014[175]	Kidney transplant recipients	72	45.5±12.2	Poland	Zonulin	Erythrocyte count	r=0.26	p<0.05
Moreno-Navarrete et al., 2012[178]	Various glucose tolerance	123	52±11.7	Spain	Zonulin	Uric acid	r=0.2	p=0.025
Moreno-Navarrete et al., 2012[178]	General population	82	48.3±11.7	Spain	Zonulin	Uric acid	r=0.24	p=0.03
Raparelli et al., 2017[102]	Liver cirrhosis	69	62.6±13.5	Italy	LPS	Platelet activation	ρ=0.55	p<0.001
Rutten et al., 2014[183]	COPD	18	63.6±1.3	Netherlands	L/R	Plasma lactic acid	r=0.66	p=0.01

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	Correlation	*p* value
Swanson et al., 2015[185]	Alcohol use disorder	20	45.9±12.2	America	L/M	Plasma melatonin	*r*=-0.39	*p*=0.03
Volynets et al., 2012[143]	NAFLD	20	41.9±2.3	Germany	LPS	Plasminogen activator inhibitor – 1	ρ=0.46	*p*=0.01
Raparelli et al., 2017[102]	Liver cirrhosis	69	62.6±13.5	Italy	LPS	sP-Selectin	ρ=0.32	*p*=0.008
Cangemi et al., 2016[154]	Pneumonia	278	70±16 (>18)	Italy	LPS	sP-selectin	ρ=0.362	*p*<0.001
Cangemi et al., 2016[154]	Pneumonia	278	70±16 (>18)	Italy	LPS	sNOX2-dp	ρ=0.455	*p*<0.001
Demographic factors								
Johnston et al., 2000[168]	Coeliac disease	77	35.0 (25-64)	Ireland	L/M	Age	*r*=0.34	*p*=0.001
Moreno-Navarrete et al., 2012[178]	General population	82	48.3±11.7	Spain	Zonulin	Age	*r*=0.22	*p*=0.045
Zak-Golab et al., 2013[188]	Various BMI	80	48 (32-63)	Poland	Zonulin	Age	ρ=0.43	*p*<0.001

ApoB, apolipoprotein; C/M, cellobiose/mannitol; COPD, chronic obstructive pulmonary disease; GlycA, glycoprotein acetylation; HbA1c, glycated haemoglobin A1c; HDL, high-density lipoprotein; hsCRP, high sensitivity C-reactive protein; HOMA, homeostatic model assessment; IBS-D, diarrhoea predominant irritable bowel syndrome; IBS, irritable bowel syndrome; IL-6, interleukin 6; LDL, low-density lipoprotein; L/M, lactulose/mannitol; LPS, lipopolysaccharides; L/R, lactulose/rhamnose; n-6, omega-6 fatty acid; NAFLD, non-alcoholic fatty liver disease; NRS, numeric rating scale; OGTT, oral glucose tolerance test; ρ, Spearman's correlation coefficient; PCOS, polycystic ovarian syndrome; PUFA, polyunsaturated fatty acids; QUICKI, quantitative insulin-sensitivity check index; r, Pearson's correlation coefficient for the sample; sNOX2-dp, soluble NOX2-deriver peptide; sP-selectin, plasma soluble P-selectin; SIBO, small intestine bacteria overgrowth; sTNFR1, soluble tumour necrosis factor receptor-1; TNF-α, tumor necrosis factor alpha.

[a] Adjusted for BMI; [b] Adjusted for BMI and age; [c] Multiple linear regression adjusted for log-transformed BMI and gestational weeks in subjects with hs-CRP> 3mg/L; [d] Only including subjects with hsCRP >3mg/L.

The strength of association between IP and disease severity varied depending on the study population and the nature of disease severity classification. Pain, as measured by numeric rating scale, was reported to have a weak positive correlation with IP.[164] Whereas conflicting evidence was reported for blood pressure and the strength of association with altered IP. The association between portal hypertension and altered IP was only reported for second and third-degree portal hypertension (OR=3.1; 95% CI: 1.1, 4.2; p<0.01).[155] A moderate positive correlation was reported between IP and systolic blood pressure in obesity (r=0.40; p=0.009).[187] Whereas diastolic blood pressure (66-72 mmHg) was reported to be an independent risk factor for altered IP (OR=2.82; 95% CI: 1.43, 5.58; p=0.003) in the general population.[88]

2.2.3.3 DIETARY RISK FACTORS

Five studies reported nine statistically significant dietary factors that were associated with IP.[106,143,170,177,188] Intake of >2616 kcal/day was reported as an independent risk factor for altered IP (β=121.8; p=0.04) as measured by LPS[106] (Table 2.4). Total fat percentage in the diet was also reported as an independent risk factor for altered IP (β=0.23; 95% CI: ±0.11; p<0.05).[188] One study reported protein intake as an independent risk factor for altered IP (β=-0.139; 95% CI: -0.247, -0.031; p=0.01).[177] While one other study[143] reported a moderate positive correlation between total protein intake and IP (ρ=0.59; p=0.001) with sub-analysis on protein source reporting that animal-derived protein intake had a moderate positive correlation with altered IP (ρ=0.54; p=0.002)[143] (Table 2.5). One study reported alcohol consumption to be a predictive risk factor for altered IP, with <14 standard drinks per week (OR=1.91; 95% CI: 1.01, 3.95; p=0.05)

and above >15 standard drinks per week (OR=1.56; 95% CI: 1.02, 2.67; p=0.05) associated with altered IP.[170]

2.2.3.4 ANTHROPOMETRIC RISK FACTORS

Ten studies reported a statistically significant association between 12 anthropometric measurements and IP.[88,133,153,161,165,178,182,186-188] The correlation between BMI and IP ranged from a weak to moderate positive correlation, of which most were reported to have a weak positive correlation[88,165,178,179,188,189] (Table 2.5). Two studies report BMI as an independent risk factor for altered IP as measured by zonulin levels (β=0.26; ±0.10; p<0.05, β=1.507; 0.34 SEM; p<0.01)[161,188] (Table 2.4). Furthermore, it was reported in the general population that a BMI of >25.0 and BMI of >30.0 were associated with altered IP OR=4.10 (95% CI: 1.87, 8.97; p<0.001) and OR=4.90 (95% CI: 1.49, 31.65; p=0.047) respectively as measured by zonulin (>64 ng/mL)[88] (Table 2.3). Two studies reported the strength of association between IP and both waist circumference and waist to hip ratio.[133,153] Although only a weak positive correlation was reported between waist circumference and IP[153] one study reported an association between altered IP and waist circumference >97cm (OR=7.03; 95% CI: 1.97, 25.11; p=0.003).[88]

2.2.3.5 BIOMARKER RISK FACTORS

Twenty-four studies reported on 29 statistically significant biomarkers and association with altered IP.[88,102,133,143,149,152-154,156,159,163-165,167,169,171-179,181-183,185-189] Two studies reported that fasting glucose had a weak positive correlation with

IP[153,189] (Table 2.5). Moreover, an additional study reported that a plasma glucose level >5.7 nmol/L is associated with a greater odds of having altered IP (OR=2.09; 95% CI: 2.09, 4.18; p=0.036) in the general population[88] (Table 2.3). In addition, fasting glucose was reported to be an independent risk factor associated with altered IP as measured by zonulin (β=0.38; ±0.12; p<0.05)[188] (Table 2.4). In contrast, a 120-minute glucose tolerance test was reported in three studies to have a weak positive correlation with IP.[133,167,189] Three studies reported a weak positive correlation between glycated haemoglobin A1c (HbA1c) and IP.[167,187,189]

From the four studies that reported a statistically significant association between fasting insulin and IP a weak to moderate positive correlation was reported (ρ=0.616; p<0.001)[153,165,176,189] (Table 2.5). Furthermore, one study reported fasting insulin to be associated with IP as measured by zonulin levels (β=0.015; 95% CI: 0.007, 0.022; p<0.001).[176] Four slightly different methods were used to measure insulin sensitivity with the strength of association varying from a weak to moderate correlation between insulin sensitivity and IP (r=0.605; p<0.05).[133,176,178,189] Moreover, insulin sensitivity was reported to be associated with IP as measured by zonulin (β=-0.263; p=0.004, β=-0.002; 95% CI: -0.003, -0.001; p<0.001)[176,178] (Table 2.4). Five studies found a similar strength of association between markers of insulin resistance and altered IP; with a weak to moderate positive correlation reported (ρ=0.616; p<0.001).[133,165,176,186,189]

Serum lipids and lipoproteins were measured in ten studies with a varying degree of strength of association with IP.[133,153,165,167,172,176,181,186,187,189] Total cholesterol was reported to have a statistically significant association with IP in four studies,

these studies report a weak to moderate positive correlation between total cholesterol and IP (ρ=0.566; p<0.001)[153,176,181,189] (Table 2.5). Furthermore, total cholesterol was reported to correlate with IP as measured by zonulin (β=0.004; 95% CI: 0.000, 0.007; p=0.032)[176] (Table 2.4); whereas, low-density lipoprotein (LDL) cholesterol was reported to have a weak positive correlation with IP.[176,189] Five studies reported high-density lipoprotein (HDL) cholesterol to have a weak negative correlation with zonulin and the dual sugar test.[133,165,167,186,187,189] Seven studies reported triglycerides to have a weak to high positive correlation with IP (r=0.73; p<0.001).[133,167,172,176,181,187,189] Triglycerides were further reported as an independent risk factor for IP as measured by zonulin (β=0.009; 95% CI: 0.003, 0.015; p=0.003).[176]

Numerous inflammatory markers were measured in a total of 11 studies and were reported to have an association with IP.[153,167,169,171,175,176,178,179,182,188,189] High sensitivity C-Reactive protein (hsCRP) were reported to have a weak positive correlation IP[153,169,176] (Table 2.5) with two studies also reporting that hsCRP correlates with IP as measured by zonulin (β=0.013; 95% CI: 0.003, 0.023; p=0.015, β=0.075; 95% CI: 0.008, 0.158; p=0.046)[153,176] (Table 2.4). In contrast, another study reported CRP to be an independent risk factor for IP (β=3.28; 95% CI: 1.28, 5.46; p<0.01).[171] Circulating interleukin-6 (IL-6) was reported to have a weak to moderate positive correlation with IP according to four studies (r=0.542; p<0.001).[167,178,179,182] Furthermore, two studies reported circulating IL-6 to independently correlate with levels of zonulin (β=0.23; p=0.04)[178] and LPS (β=0.171; p=0.04);[163] whereas, tumor necrosis factor alpha (TNF-α)—the other

major inflammatory marker measured in the included studies—was reported to have a weak to moderate positive correlation with IP (r=0.647; p<0.05).[167,182,189]

Three studies report the liver enzyme alanine transaminase (ALT) to have a statistically significant weak to moderate positive correlation with IP (ρ=0.50; p=0.005)[143,153,165] (Table 2.5), with one study reporting ALT to correlate with IP as measured by zonulin (β=0.014; 95% CI: 0.001, 0.028; p=0.04)[153] (Table 2.4). Two other liver enzymes aspartate transaminase (AST) and gamma glutamyltransferase (GGT) were reported to have a weak positive correlation with IP, with only AST reported as an independent risk factor for IP (β=0.02; 95% CI: 0.002, 0.037; p=0.04).[153] One study reported that microbial diversity had a weak negative correlation with serum zonulin and stool zonulin.[173]

The strongest association among biomarkers and IP were markers of IP themselves. Two studies used both zonulin and the dual sugar test and reported a high positive correlation between the two tests (ρ=0.891; p=0.05)[149,159] (Table 2.5). However, mixed evidence was reported for the strength of association between zonulin and LPS with a weak to moderate positive correlation reported (ρ=0.545; p<0.001);[102,154,156,176] although, one study reported LPS to independently correlate with zonulin levels (β=0.002; 95% CI: 0.001, 0.003; p=0.002)[176] (Table 2.4). The dual sugar test was also reported to have a moderate to high positive correlation with gastroduodenal permeability according to one study (r=0.68; p<0.0001, r=0.88; p<0.0001).[164]

2.2.3.6 DEMOGRAPHIC RISK FACTORS

Although most studies evaluated basic demographic characteristics only age was reported to have a statistically significant association with IP in three studies.[155,168,188] Study populations that were diagnosed with a health condition reported a weak positive correlation between age and IP according to both zonulin and the dual sugar test[155,168,188] (Table 2.5). Age was reported an independent risk factor for altered IP[188] with the increase of IP more likely over the age of 50 (OR=1.9; 95% CI: 1.1, 2.3; p<0.001)[155] (Table 2.3).

2.2.4 DISCUSSION

This is the first systematic review to explore the potential risk factors associated with IP in an adult population. This review identified over 100 potential risk factors associated with IP that had a varying degree of strength of association. The majority of the identified risk factors were only found to have a weak association with IP; however, there were similarities with many of the risk factors measured and reported to be associated with IP in numerous instances. This similarity further strengthens the identified risk factors as valuable clinical features healthcare professionals may consider as part of their differential diagnosis. Many of the risk factors identified have previously been reported as major risk factors for morbidity and mortality in chronic diseases worldwide.[190,191] Therefore, IP may be considered a feature of chronic disease rather than merely a digestive health issue.

2.2.4.1 STRONGEST RISK FACTORS FOR ALTERED INTESTINAL PERMEABILITY

Elevated levels of proinflammatory markers, dyslipidaemia, hyperglycaemia, anthropometric measurements resembling obesity, advanced disease severity with comorbidity and the consumption of a Western-style diet were identified as the strongest risk factors for altered IP (Figure 2.2). An unexpected finding of our review was the paucity of digestive health symptoms reported to be associated with IP alongside the magnitude of risk factors that resemble a metabolic-like condition. Although digestive health symptoms such as bloating, abdominal cramps and pain, heartburn, reflux, nausea and flatulent were measured in a few of the included studies, none were reported to be significantly correlated with the risk of IP.[88,164,180] The digestive health issues that were reported to be associated with IP were diseases situated primarily within the gastrointestinal system such as inflammatory bowel disease,[150,158] diarrhoea predominant irritable bowel syndrome (IBS-D),[180] intestinal dysbiosis,[173,177,188] symptoms like diarrhoea (especially from an organic disease)[160] and indigestion syndrome.[180] However, these digestive health symptoms were not found to be associated with IP in the general population.[88] Although digestive health symptoms appear to lack association with IP, this should not undermine the association between gastrointestinal conditions and IP, especially provided the high correlation between the improvement of altered IP and a reduction in postinfectious IBS disease severity purported in the literature.[192] Conversely, many of the risk factors that resemble a metabolic-like condition were found to be associated with IP in the general population.[88,178,189] However, risk factors such as waist-to-hip ratio, waist circumference, and elevated triglycerides, were less associated with

IP in the general population when compared to a disease state. It appears that the identified risk factors have a stronger association with altered IP within a disease state rather than in the general population.

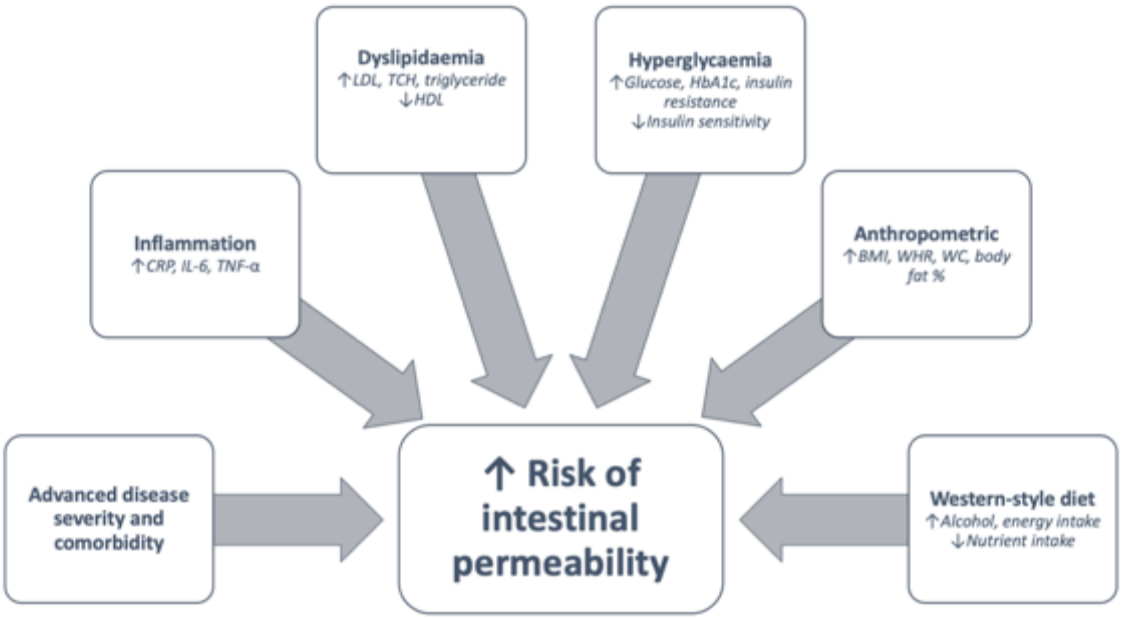

Figure 2.2 The strongest identified risk factors identified to be associated with intestinal permeability

Many hypotheses exist detailing the mechanism of action linking the health and integrity of the digestive system to inflammation, obesity, poor glycaemic control and dyslipidaemia.[167,170,177] One of the most prominent theories suggests IP is both a cause and consequence of LPS absorption. The translocation of LPS as the result of IP may contribute to the cascade of events that is responsible for the metabolic-like risk factors.[193,194] First, LPS have been demonstrated to trigger inflammation that may alter glucose metabolism resulting in poor glycaemic control and insulin resistance.[193] The occurrence of dyslipidaemia may contribute to the loss of intestinal integrity as HDL is in part responsible for neutralising LPS,

whereby low levels of HDL may result in inflammation and LPS exacerbating IP.[195] The link between metabolic factors and altered IP is further strengthened as improvement in some of the identified risk factors such as BMI,[75] HbA1c,[187] and inflammation[196] have been shown to be associated with the improvement of IP.

2.2.4.2 CHRONIC DISEASE AND MULTIPLE RISK FACTORS

Comorbidity of chronic diseases such as diabetes, liver disease, metabolic syndrome, kidney disease, and obesity were identified to increase the risk of IP.[155,170,172,181] Moreover, the severity and activity of chronic health conditions including liver disease,[102,143,155,162,170] metabolic syndrome,[172] PCOS,[133] coeliac disease[149] and BMI[161] were reported to correlate with the degree of IP. The notion that IP correlates with disease severity is further supported as the severity of particular risk factors also increase alongside the degree of IP.[88,172,176,177,179,189] The involvement of IP in chronic disease, especially with advanced disease severity, highlights the potential importance of intestinal integrity in health and disease. This review also suggests a synergistic effect is possible when more than one risk factor is experienced. In particular, BMI, age, alcohol consumption and inflammation were all identified as having some degree of synergistic effect;[170,176,178,182,188] although, inflammation appears to be the driving factor in many of the risk factors.[188]

2.2.4.3 DIETARY AND LIFESTYLE HABITS AND INTESTINAL PERMEABILITY

It appears that a high energy, nutrient-depleted diet with either inadequate protein intake or excess animal-derived protein in combination with alcohol consumption is a potential risk factor for IP. This dietary pattern closely resembles that of the Western diet, which has been suggested to increase the risk of chronic disease[197] and metabolic disease such as obesity.[191] Dietary intervention studies are limited; however, one study suggests an increase in dietary protein is associated with elevated zonulin and inflammation.[198] Furthermore, it has been demonstrated that an increase in dietary fibre reduces zonulin.[199] While alcohol withdrawal is associated with a reduction of IP with a greater result seen in patients with a high BMI.[161] Based on these findings, dietary and lifestyle habits may present a key clinical feature that healthcare professionals may utilise in identifying patients at risk of altered IP.

2.2.4.4 SIMILARITIES AND DIFFERENCES BETWEEN MARKERS OF INTESTINAL PERMEABILITY

Both zonulin and the dual sugar test were reported to highly correlated with each other. However, only two risk factors namely HDL cholesterol and insulin resistance were shown to be associated with both zonulin and the dual sugar. This finding may be due to the limited number of studies using the dual sugar test included in this review. Previous research has suggested that zonulin is a biomarker of metabolic syndrome, obesity, inflammation and poor health rather than an indicator of IP;[88] although, zonulin is associated with many of the risk factors that resemble a metabolic-like condition after adjusting for metabolic

syndrome, obesity and inflammation, implicating zonulin as a true marker of IP. However, the mixed evidence surrounding the association between serum and stool zonulin could be explained by zonulin being described as an acute phase biomarker of coeliac disease[200] and IBD.[201] This feature of zonulin being an acute phase biomarker may also explain the lack of consistency between the dual sugar test and zonulin. For example, IBS-D is a condition known to be related to IP and has recently been shown to be associated with the dual sugar test but not zonulin.[104] Another potential factor influencing the results is the accuracy of the commercial zonulin assay; with recent research advocating caution in using the commercial zonulin assay as a means of evaluating intestinal integrity.[91] Limited studies have used both the zonulin and dual sugar test; however, our review found that these two tests have the highest association with each other compared to all the risk factors identified. Whether zonulin is a more sensitive marker of IP for particular risk factors compared to the dual sugar test is yet to be investigated. Moreover, the ideal test for specific disease diagnosis and the stage and activity of the disease requires further investigation. Healthcare professionals may find clinical benefit from using both the serum zonulin and dual sugar test for an accurate diagnosis of IP when patients present with the risk factors for IP.

2.2.4.5 LIMITATIONS

This systematic review has some limitations worth mentioning. As a result of limited research examining risk factors associated with IP, this review consisted of a heterogeneous range of health conditions, preventing cumulative statistical meta-analysis. The target population for our review were adults 18 years and over; however, many large cohort studies involved adolescents. A number of

articles were included when age range was unknown yet evaluated to be adults 18 years and over. Future similar reviews are suggested to incorporate adolescents into the target population. Numerous studies were excluded as they did not use measurable and comparable analysis of IP risk factors. In addition, risk factors that were not statically significant were not included potentially increasing selection bias. Other potential risk factors may have been missed due to the nature of the risk factor only being included in experimental research designs.

2.2.4.6 CLINICAL SIGNIFICANCE

The clinical relevance of the identified risk factors warrants the attention of healthcare practitioners in their differential diagnosis. IP has previously been recognised by healthcare practitioners to be associated with gastrointestinal conditions more so than any other group of diseases, including metabolic conditions.[4] In our review digestive health symptoms were not identified as a major risk factor for IP. In contrast, many conditions such as food sensitivities[132] and histamine intolerance,[202] were found to be clinically relevant in the identification of patients at risk of IP. Lastly, until there is a comprehensive understanding of the clinical diagnosis of IP healthcare professionals are advised to consider multiple methods of IP testing, and to account for the identified risk factors to ensure the most accurate diagnosis of intestinal integrity.

2.2.4.7 FURTHER RESEARCH

Further research needs to examine whether the identified risk factors are solely linked with the diagnosis of IP or whether the disease state influences the association. Further evidence is necessary to distinguish which marker of IP is most appropriate and accurate for measuring IP in different conditions and at different stages of disease manifestation. Longitudinal studies measuring the identified risk factors may provide increased understanding of the cause or consequence of IP. Lastly, the validation of serum zonulin, stool zonulin and the dual sugar test as markers for altered IP are necessary to be undertaken for the advancement of IP research.

2.2.4.8 CONCLUSION

Dyslipidaemia, poor glycaemic control, inflammation, anthropometric measurements that resemble obesity, and Western-style dietary habits have the strongest association with altered IP—which amplify when combined. In addition, comorbidity of chronic diseases and advanced disease severity are also strong risk factors of altered IP. These risk factors warrant the attention of clinicians and other healthcare providers to aid in the identification of potential patients at risk of altered IP.

2.3 CHAPTER SUMMARY

This was the first systematic review to explore the known risk factors associated with IP. Several risk factors associated with IP were identified, including biomarkers, anthropometric measurements, demographics, dietary intake and chronic diseases. These risk factors warrant the attention of clinicians and other healthcare providers to aid the identification of potential patients at risk of altered IP. These identified risk factors further direct the type of interventions and therapeutic action that may be required to have a beneficial impact in people with IP. Although some risk factors may have been missed due to the inclusion criteria and searched databases, this review provides a comprehensive summary of the possible risk factors associated with IP. The results from this chapter provide direction for the clinical questions the IP Guideline may consider exploring. With many known risk factors, there remains uncertainty surrounding the evidence-based treatment interventions for managing IP in clinical practice.

3. METHODOLOGY

This research project employs the most appropriate methodology to answer the objectives outlined in Chapter 1. To achieve the research aim, this project was conducted over two phases: Phase One, a cross-sectional survey of Australian adults with suspected IP; and Phase Two, the developing a clinical practice guideline for the management of IP. The findings from Phase One directly inform Phase Two by facilitating the developed recommendations that consider the views and values of people with IP. Phase Two also included a cross-sectional survey of key stakeholders involved in the management of IP. This chapter describes the rationale and methods used to address the research objectives.

3.1 RESEARCH METHODOLOGY AND DESIGN FRAMEWORK

The Leaky Gut Survey aimed to explore and describe the health-seeking behaviours of Australian adults with suspected IP. Secondly, after the IP Guideline had been drafted, stakeholder feedback was required to assess the agreement with each recommendation. Therefore, a quantitative methodology approach that employed an observational, cross-sectional, self-administered online questionnaire was adopted for Phase One and Phase Two of this research project. Phase One and Phase Two intended to understand the participants' views and perspectives thus, an observational rather than experimental framework was most suitable for this research project.[203] Utilising an observational framework allowed for greater external validity, allowing the results to be generalised across Australia.[204]

3.2 PHASE ONE: THE LEAKY GUT SURVEY

3.2.1 PHASE ONE (LEAKY GUT SURVEY) OVERVIEW

Phase One, also referred to as the *Leaky Gut Survey*, involved a cross-sectional survey of Australian adults with suspected IP. This was the first survey to investigate this population group nationally or internationally. Collectively, Phase One aimed to address Research Objectives 2, 3 and 5. Specifically, the findings found from the Leaky Gut Survey described the views, preferences, and health-seeking behaviours of adults with suspected IP (Objective 2) (see Chapter 4), quantify the subjective wellbeing and health-related quality of life of adults with suspected IP (Objective 3) (see Chapter 5). In addition, Phase One provided input for the development of a clinical practice guideline for the management of IP, which considered patients' views and preferences (Objective 5) (see Chapter 6 and 8).

3.2.2 LEAKY GUT SURVEY DESIGN FRAMEWORK

A survey is a single snapshot in time which is a cost-effective method that can gather the necessary data across a large geographical area in a short time.[205] The use of an online questionnaire may reduce the risk of non-completion rates and user fatigue by employing logic formatting where the questionnaire only showed relevant questions to the participants.[205] A cross-sectional observational survey design has been used in health services research investigating treatment methods for the management of IP.[5] The disadvantage of a cross-sectional study design, is that no causation can be anticipated, only associations.[204] However,

this limitation does not impact the current research objectives due to the exploratory aim of the research project. Therefore, a cross-sectional observational survey design was an appropriate method of data collection to address the research objectives.[206]

3.2.3 SETTING AND SAMPLING

The target population of Phase One are Australian adults with IP. To accurately represent the target population, the inclusion criteria were individuals that either suspected or know they have altered IP, were over the age of 18 years, were living in Australia and had internet access. The target population represents an under-examined group, with the study designed to sample people with suspected IP or confirmed IP. Including participants who self-diagnosed their IP best reflects the target population as IP is suggested to be under-diagnosed in clinical practice.[4] A snowball sampling method was used to best sample the target population. This method involved the development of a purpose-built webpage that could be shared via social media platforms. The link to the questionnaire was initially shared on known social media sites, including Leaky Gut and Microbiome Support Group Australia (600 members), Leaky Gut Research (5,681 followers), Leaky Gut Syndrome/ Food Allergies/Candida (8,719 members) and Gut Healing: Exploring diets/food sensitivities/leaky gut/root causes/etc (6,552 members). This wide survey distribution allowed the greatest possibility of capturing eligible participants. Furthermore, the survey was open for two months, between September 2019 and November 2019.

3.2.4 DATA COLLECTION

Data collection was undertaken by a cross-sectional, self-administered, online questionnaire through *SurveyGizmo. SurveyGizmo is* a commercial platform for building online surveys. After data collection, complete and incomplete data were transferred to a spreadsheet where it was cleaned and checked for duplicates before analysis. The full questionnaire has been included in Appendix 3.1. The questionnaire was composed of 51 to 62 questions, with the number of questions being dependent on the participant's answers to piping questions. The questionnaire utilised items that were previously used in published literature and were modified to suit Australians with suspected IP.[4,5] The types of questions utilised in the survey were Likert scales, multi-choice questions and open-ended questions. The use of Five-point Likert scales was chosen to accurately gauge participants' views and perspectives throughout the survey.[207] The survey included seven main domains: *demographic characteristics, diagnosis of IP, treatment methods for altered IP, financial expenditure related to IP, self-reported outcome of IP, subjective wellbeing and Health-related quality of life.*

3.2.4.1 DEMOGRAPHIC CHARACTERISTICS

The participants were asked about their gender, age, height, and weight. Body mass index (BMI) was calculated from height and weight measurements. BMI was then categorised as underweight, healthy weight, overweight, and obese.[21] The participants were further asked about their country of birth, the state or territory where they live and whether this was in an urban, rural, or remote location.

3.2.4.2 DIAGNOSIS OF INCREASED INTESTINAL PERMEABILITY

To better understand the way people with suspected IP are diagnosed, participants were asked questions relating to how they were assessed and how they would like to be assessed. Firstly, participants were asked to report the year they believed their IP started, the year their IP was diagnosed, at what point their IP was assessed, the method used to confirm their IP, the number of times their IP was assessed, and the qualifications of the practitioner involved in the assessment of their IP. These questions were used to better understand the accuracy of self-diagnosis of IP. Secondly, 5-point Likert scales were used to explore participants' views and perspectives surrounding IP. To understand the importance participants' place on clinicians' ability to measure them for IP in clinical practice, a 5-point Likert scale ranging from "not important" to "very important" was used. Other areas where the 5-point Likert scales were used included participants' preference for IP testing method characteristics and the likelihood of treatment adherence after a positive result.

3.2.4.3 TREATMENT OF INCREASED INTESTINAL PERMEABILITY

A series of questions were asked about dietary products, lifestyle therapies, dietary supplements and medications that may influence IP. The selection of prepopulated answers was based on pre-existing literature.[5] A six-point scale ('never', 'less than once a month', '1-3 times a month', 'once a week', '2-6 times a week', 'every day') was used to evaluate the frequency that dietary products, lifestyle therapies, dietary supplements and medications were used. To explore participants' preferred treatment method, a 5-point Likert scales ranging from "no preference" to "very strongly prefer" was used in relation to the four treatment

categories: dietary products, lifestyle therapies, dietary supplements and medications.

3.2.4.4 FINANCIAL EXPENDITURE AND MANAGEABILITY

Financial expenditures relating to the management of IP were also explored. Participants were asked to report the out-of-pocket expenditure for the treatment of IP and practitioner consultation fees in the previous 12 months. One question on the amount spent on the assessment of IP was also asked. To determine participant's income manageability, they were asked to select how well they manage their household income: 'difficult all the time', 'difficult some of the time', 'not too bad' or 'easy'. All amounts were reported in Australian dollars (AUD).

3.2.4.5 SELF-REPORTED OUTCOME OF INTESTINAL PERMEABILITY

To gain a deeper understanding for the potential severity of participants IP, two questions were asked. Firstly, participants were asked how many days a week their IP affects their daily living (0 days - 7 days). Participants were then asked whether they believed their IP had become 'better', 'worse' or 'no change' over the previous 12 months.

3.2.4.6 SUBJECTIVE WELLBEING

Subjective wellbeing also referred to as life satisfaction, is comprised of cognitive and affective components that can suggest an individual's appraisal of their satisfaction with their life.[208,209] A widely used assessment method is the Personal Wellbeing Index - Adult (PWI-A) scale - an instrument validated in Australian

population samples.[210] This scale is made up of seven domains evaluating satisfaction including; *standard of living*, *personal health*, *achieving in life*, *personal relationships*, *personal safety*, *community-connectedness* and *future security*.[210] Each domain is reported on a 0-10 scale, with 0 indicating no satisfaction at all and 10 being completely satisfied.

3.2.4.7 QUALITY OF LIFE

The 20-Item Short Form Health Survey is a measure of health-related quality of life which measures the impact of health status on quality of life.[211] This includes mental, physical, emotional, and social functioning.[212] Therefore, the 20-Item Short Form Health Survey was used in this study. A total of six health domains are assessed in this validated patient reported outcome measure. These domains include physical functioning (6 questions), role functioning (2 questions), social functioning (1 question), mental health (5 questions), current health perceptions (5 questions), and bodily pain (1 question).

3.2.5 STATISTICAL ANALYSIS

Data obtained from the survey was exported to STATA® 16 for statistical analyses. Missing data was excluded from analysis. Responses to questionnaire items were reported as means, standard deviations, 95% confidence intervals (CIs) or frequencies and percentages, where appropriate. Displaying the variables in this format is the most appropriate method for an exploratory survey. Chi-square analysis was used to examine the association between two categorical variables with Student's t-tests used for continuous variables across

a binary variable. Chi-square analysis only reports the existence of relationships, but cannot provide explanation for the strength or direction and is unable to account for confounding variables.[213] Spearman's rank-order correlation coefficient analysis was used to measure the correlation between the number of days IP affects daily living, subjective wellbeing and health-related quality of life. Ordinal variables including those based on Likert scales were analysed with non-parametric tests, including the Wilcoxon signed ranks test and Mann-Whitney U test, where appropriate. Analysis of variance (ANOVA) was used to measure the difference between a continuous variable across a categorical variable. Variables found to be associated with subjective wellbeing, health-related quality of life or the number of days IP affects daily living - with a bivariate p-value < 0.25[214] - were entered into the respective multivariate logistic or linear regression models, to adjust for potential confounders. Independent predictors were identified by a stepwise backward elimination process.

3.2.6 ETHICAL CONSIDERATIONS LEAKY GUT SURVEY

This thesis involved two ethics applications for Phase One and Phase Two. The Leaky Gut Survey received ethical approval from Human Research Ethics Committees (HREC) of the University of Technology Sydney (#ETH19-4012) (see Appendix 3.2). No funding was received for this project. Partaking in the survey had a minimal risk to participants, as the discomfort anticipated from their involvement does not exceed any ordinarily daily task. Discomfort may have occurred as the time required for participants to complete the survey was 30 minutes in length. To reduce discomfort, the survey was available online to allow participants enough time to participate at a time most convenient to them. Furthermore, using logic, the survey only showed relevant questions to the

participants to reduce fatigue. No pressure was placed on the applicants to participate in this survey. Completing the survey was completely voluntary and there was no consequence for choosing not to participate, as described in the participation information sheet, located at the beginning of the survey (see Appendix 3.3). Consent was obtained before the participants were able to commence the survey (see Appendix 3.4). No personal information that may identify the participants was gathered throughout the survey. However, at the completion of the survey, participants were asked if they wished to be sent a copy of the results/research findings. They had the option of entering their email address in a separate link, which would subscribe them to the email list. This sign-up list was separate from the main survey and was not linked to their results to ensure the results of the survey were anonymous. Participants were not identified within the publication of the research nor in conference presentations. Data was securely stored in Office 365 documents on CloudStor on a password protected computer while the project was in progress. At the end of the project, data was archived by creating an Archival Data Record through the University of Technology Sydney.

3.3 PHASE TWO: IP GUIDELINE

3.3.1 PHASE TWO (IP GUIDELINE) OVERVIEW

Phase Two, also referred to as the *IP Guideline*, involved developing a clinical practice guideline for managing IP. This guideline provides clinicians with evidence-based recommendations for managing IP in clinical practice. Phase Two was based on the NHMRC *Guidelines for Guidelines* Handbook to meet the 2016 *NHMRC Standards for Guidelines.*[215] This structured approach is

considered a leader in guideline development process and was used as a guide rather than a checklist for guideline submission to the NHMRC.[113] Phase Two has been developed to answer Research Objectives 4 and 5. Specifically, a systematic approach was followed to identify the treatment options available for patients with IP (Objective 4) with the development of an evidence-based clinical practice guideline for the management of IP which considers patients' views and preferences to follow (Objective 5) (see Chapter 6 and 8). A complete description of the methods used in Phase Two are outlined in the report: *Clinical practice guideline for the management of increased intestinal permeability: Guideline Development Process* found below in section 3.4. As part of the IP Guideline development process, a cross-sectional survey of stakeholders was undertaken to evaluate the developed recommendations with the results provided in Chapter 7. The methods involved in this survey are described in section 3.5.

3.3.2 CLINICAL PRACTICE GUIDELINE DESIGN FRAMEWORK

A clinical practice guideline is designed to support clinicians in their decision-making for the diagnosis and management of specific areas of healthcare. Clinical practice guidelines are considered one of the best ways to present evidence-based recommendations to clinicians while reducing inappropriate care and supporting new knowledge to the clinician.[113,114] However, clinical practice guidelines are not intended to supersede professional judgement, with clinicians always advised to act in the patients' best interest.[115] A potential disadvantage of clinical practice guidelines is that if they are formulated without a structured approach, inconsistent recommendations can be produced.[114] However, this limitation can be addressed by following an evidence-based and structured

approach to guideline development. The NHMRC Guideline is considered one of the world leaders in developing and supporting the development of clinical practice guidelines.[113] Other clinical practice guidelines have used the NHMRC *Guidelines for Guidelines* Handbook in recent years.[216] Therefore, a clinical practice guideline that follows the NHMRC *Guidelines for Guidelines* Handbook to meet the 2016 *NHMRC Standards for Guidelines* was the most appropriate method to address the research objectives.[215]

3.3.3 ETHICAL CONSIDERATIONS IP GUIDELINE

The HREC approved the IP Guideline of the University of Technology Sydney (#ETH20-5291) (see Appendix 3.5). Funding was acquired through the Australian Research Centre in Complementary and Integrative Medicine. The Working Group identified participants as suitable and appropriate participants based on their clinical experience and research interest. Participating in the survey was completely voluntary and there was no consequence for choosing not to participate as described in the participation information sheet, located at the beginning of the survey (see Appendix 3.6). Before the start of the survey, a declaration of consent was obtained (see Appendix 3.7). Participant's contact details were obtained via publicly available websites. Participants are recognised within the published IP Guideline. Participants were provided with a printed and PDF copy of the final IP Guideline. Furthermore, participants were reimbursed with a $100 Visa card upon completing their Terms of Reference. The reimbursement of $100 is to cover the 2 hours involved in participating in this study.

3.4 CLINICAL PRACTICE GUIDELINE FOR THE MANAGEMENT OF INCREASED INTESTINAL PERMEABILITY: GUIDELINE DEVELOPMENT PROCESS

The following section contains the *Guideline Development Process*, a document that forms part of the IP Guideline. The *Guideline Development Process* has been formatted based on the NHMRC *Guidelines for Guidelines* Handbook to meet the 2016 *NHMRC Standards for Guidelines.*[215] Details are as follows:

> Leech, B, McIntyre, E, Steel, A, Sibbritt, D (2022) "Clinical practice guideline for the management of increased intestinal permeability: Guideline Development Process", University of Technology Sydney.

3.4.1 INTRODUCTION

The need to develop a *clinical practice guideline for the management of increased intestinal permeability* (IP Guideline) was identified after health services research revealed gaps in both the published literature and clinical practice.[4,5] Upon searching grey literature, published literature and the Guidelines International Network Library, this clinical practice guideline was identified as the first guideline for increased intestinal permeability (IP) as no guideline surrounding any part of the management of IP has been developed in Australia or internationally.

3.4.2 AIM AND OBJECTIVE

The IP Guideline aims to improve the management of altered IP by clinicians in private practice of Australia. The IP Guideline provides clinicians with evidence-based recommendations for the management of IP.

The objectives are:

1. to identify any dietary choices available for the management of altered IP in Australian adults;
2. to identify any probiotic, prebiotic and synbiotic supplementation available for the management of altered IP in Australian adults;
3. to identify any amino acid supplementation available for the management of altered IP in Australian adults,;
4. to identify any plant-based medicine supplementation available for the management of altered IP in Australian adults;
5. to identify any essential fatty acid supplementation available for the management of altered IP in Australian adults;
6. to identify any mineral supplementation available for the management of altered IP in Australian adults;
7. to identify any vitamin supplementation available for the management of altered IP in Australian adults; and
8. to identify any colostrum supplementation available for the management of altered IP in Australian adults.

3.4.3 RESPONSIBLE ORGANISATION

The development of the IP Guideline was coordinated by the University of Technology Sydney (UTS), Faculty of Health, Australian Research Centre in

Complementary and Integrative Medicine (ARCCIM) as part of the PhD candidature for Bradley Leech. The Society of Intestinal Permeability Research (SIP Research) was involved in the dissemination and implementation.

3.4.4 SOURCE OF FUNDING

The development of the IP Guideline was funded by ARCCIM, providing a total of $4470 in support of guideline development, publication, and dissemination. The Australian Government Research Training Program Scholarship provided Bradley Leech with a scholarship. The scholarship funding did not influence the development or content of the guidelines.

3.4.5 STEPS IN PREPARING CLINICAL PRACTICE GUIDELINES

The IP Guideline followed the NHMRC *Guidelines for Guidelines* Handbook to meet the 2016 *NHMRC Standards for Guidelines*.[215] The level of evidence for each recommendation was determined based on the NHMRC grades for recommendations and the NHMRC Evaluation of Evidence process.[217] The reporting of the IP Guideline followed the RIGHT statement.[218] Six steps were undertaken to develop the recommendations:

1) Develop a structured clinical question;
2) Perform a systematic review;
3) Summarise the relevant data;
4) Risk of bias assessment;
5) Assess the body of evidence and formulate recommendations; and
6) Write the content narrative.

3.4.5.1 STEP 1: DEVELOP A STRUCTURED CLINICAL QUESTION

The first draft of the clinical questions was comprised from scoping the literature. To help inform the clinical questions, the views, preferences and experiences of both consumers and clinicians were drawn upon from published literature.[4,5,219,220] The Working Group was responsible for prioritising the questions based on the purpose, scope and clinical importance, taking into account the views, preferences and experiences of both consumers and clinicians. When appropriate, the clinical questions were structured to the PICO (patient/population/problem, interventions, comparison/control, outcome) framework. When the PICO framework was not applicable, a structured question was formulated. The complete list of questions is as follows:

3.4.6 CLINICAL QUESTION LIST

CQ.1). In Australian adults with increased intestinal permeability, what are the benefits of dietary choices for the treatment of increased IP?

CQ.2). In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for dietary choices?

CQ.3). In Australian adults with increased intestinal permeability, what are the benefits of oral probiotic, prebiotic or synbiotic supplementation for the treatment of increased intestinal permeability?

CQ.4). In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for oral probiotic, prebiotic or synbiotic supplementation use?

CQ.5). In Australian adults with increased intestinal permeability, what are the benefits of oral amino acid supplementation for the treatment of increased intestinal permeability?

CQ.6). In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for oral amino acid supplementation use?

CQ.7). In Australian adults with increased intestinal permeability, what are the benefits of oral plant-based medicine supplementation for the treatment of increased intestinal permeability?

CQ.8). In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for oral plant-based medicines use?

CQ.9). In Australian adults with increased intestinal permeability, what are the benefits of oral essential fatty acid supplementation for the treatment of increased intestinal permeability?

CQ.10). In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for oral essential fatty acid supplementation use?

CQ.11). In Australian adults with increased intestinal permeability, what are the benefits of oral mineral supplementation for the treatment of increased intestinal permeability?

CQ.12). In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for oral mineral supplementation use?

CQ.13). In Australian adults with increased intestinal permeability, what are the benefits of oral vitamin supplementation for the treatment of increased intestinal permeability?

CQ.14). In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for oral vitamin supplementation use?

CQ.15). In Australian adults with increased intestinal permeability, what are the benefits of oral colostrum supplementation for the treatment of increased intestinal permeability?

CQ.16). In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for oral colostrum use?

3.4.6.1 STEP 2: PERFORM A SYSTEMATIC REVIEW

3.4.7 SEARCH METHODS

A single literature search was carried out to identify the relevant articles for each clinical question. All related articles were identified and grouped according to clinical questions. Target searching was carried out when needed during the write-up of the guideline.

3.4.8 SEARCH STRATEGY

The databases PubMed, Embase, CINAHL, and Scopus were searched for articles published between January 2000 up until July 2020 by the Working Group. A single-arm search strategy was used: 'intestinal permeability' OR 'intestinal integrity' OR 'intestinal barrier dysfunction' OR 'intestinal epithelial barrier dysfunction' OR 'gastrointestinal permeability' OR 'gut permeability' OR 'gut barrier' OR 'zonulin' OR 'dual sugar' OR 'lactulose AND mannitol' OR 'lactulose AND rhamnose' OR 'cellobiose AND mannitol' OR 'Intestinal fatty acid binding protein'. The human filter was applied to the search. A hand search of the

reference list from the included articles and web search for any recently published articles was carried out.

3.4.9 ARTICLE CRITERIA FOR CLINICAL PRACTICE GUIDELINE

Included systematic reviews were original research articles exploring topics relevant to the clinical questions published between January 2000 and July 2020. The primary focus of the included systematic reviews were adults; however, systematic reviews were excluded if articles included participants under 18 years of age. However, including young adults has been suggested as a method to improve the search strategy for IP.[221] Therefore, at least 80% of the enrolled study population must be over 18 years of age. Included systematic reviews were also required to clearly illustrate a search strategy and use valid data extraction methods. Systematic reviews were excluded if the primary focus was on critically ill patients (i.e., in intensive care or palliative care) or includes patients with HIV, acute appendicitis, receiving chemotherapy, undergoing dialysis or abdominal surgery as the IP Guideline is focused on private practice in the community. Articles were also excluded if the primary focus was on genetic testing, polymorphism research or involve the treatment of exercise induced IP. When inaccurate testing method were used, these articles were excluded. Examples of inaccurate testing methods includes the dual sugar urinary test where the collection is over 6 hours or measured in the serum. Furthermore, in studies assessing the effectiveness of an intervention for IP management where clear evidence suggest that the patients do not have IP, these studies were excluded. Articles may include animal studies as supporting evidence for human research;

however, must not be the focus of the systematic review. There was no exclusion based on geographical location. Only articles published in English were included.

When a comprehensive systematic review was not available additional original research articles were included to fill the gaps. These articles were subject to the same inclusion and exclusion criteria as the systematic reviews and consist of both experimental and observational studies. Case studies and case series were excluded.

3.4.9.1 STEP 3: SUMMARISE THE RELEVANT DATA

For each article used to answer one of the clinical questions, the level of evidence was determined according to NHMRC (Table 3.1). The applicable articles for each clinical question were reported in a systematic review format located in the *Technical Report* (Section 6).

After the systematic review process, relevant articles were grouped according to the specific topic or clinical question. At least one article for each clinical question was required to have a level of evidence of I or II according the NHMRC. In the event no relevant article has sufficient evidence, these articles were excluded from the clinical practice guideline.

Table 3.1 Designations of levels of evidence according to type of research question

Level	Intervention	Diagnosis	Prognosis	Aetiology	Screening
I	A systematic review of level II studies	A systematic review of level II studies	A systematic review of level II studies	A systematic review of level II studies	A systematic review of level II studies
II	A randomised controlled trial	A study of test accuracy with: an independent, blinded comparison with a valid reference standard, among consecutive patients with a defined clinical presentation	A prospective cohort study	A prospective cohort study	A randomised controlled trial
III-1	A pseudo-randomised controlled trial (i.e. alternate allocation or some other method)	A study of test accuracy with: an independent, blinded comparison with a valid reference standard, among non-consecutive patients with a defined clinical presentation	All or none	All or none	A pseudo-randomised controlled trial (i.e. alternate allocation or some other method)
III-2	A comparative study with concurrent controls: Non-randomised, experimental trial Cohort study Case-control study	A comparison with reference standard that does not meet the criteria required for Level II and III-1 evidence	Analysis of prognostic factors amongst untreated control patients in a randomised controlled trial	A retrospective cohort study	A comparative study with concurrent controls: Non-randomised, experimental trial Cohort study Case-control study

	Interrupted time series with a control group A comparative study without concurrent controls: Historical control study				A comparative study without concurrent controls: Historical control study
III-3	Two or more single arm study Interrupted time series without a parallel control group	Diagnostic case-control study	A retrospective cohort study	A case-control study	Two or more single arm study
IV	Case series with either post-test or pre-test/post-test outcomes	Study of diagnostic yield (no reference standard)	Case series, or cohort study of patients at different stages of disease	A cross-sectional study	Case series

Source[222]

3.4.9.2 STEP 4: RISK OF BIAS ASSESSMENT

All included articles were assessed for risk of bias using the most appropriate tool identified by the Working Group. The risk of bias assessment for each included article is included in the *Technical Report* (Section 6).

3.4.10 SYSTEMATIC REVIEWS

Risk Of Bias In Systematic reviews (ROBIS) assesses the risk of bias in systematic reviews and covers research questions relevant to interventions,

diagnosis, prognosis and aetiology.[223] The ROBIS tool is composed of 3 phases: 1) assess relevance; 2) identify concerns with the review process; and 3) judge risk of bias. The overall risk of bias of a systematic review using the ROBIS tool provides a risk of bias rating of “High”, “Low” or “Unclear”. The corresponding rating of risk of bias was used in the evidence base section of the NHMRC body of evidence matrix to grade the potential recommendation.

3.4.11 RANDOMISED TRIALS

The Cochrane tool for assessing risk of bias in randomised trials (Cochrane RoB 2.0) is the most appropriate and widely used tool for assessing randomised trials for risk of bias.[224] The updated Cochrane RoB 2.0 tool is structured into five domains: 1) bias arising from the randomization process, 2) bias due to deviations from intended interventions, 3) bias due to missing outcome data, 4) bias in measurement of the outcome, and 5) bias in selection of the reported result. The risk of bias judgment for the previous domains and the overall risk of bias uses a rating of “high risk of bias”, “some concerns” and “low risk of bias”. The corresponding rating of risk of bias was used in the evidence base section of the HNMRC body of evidence matrix to grade the potential recommendation.

3.4.12 NON-RANDOMISED STUDIES OF INTERVENTIONS

The Risk Of Bias In Non-randomised Studies - of Interventions (ROBINS-I) was used to evaluate the risk of bias in intervention studies that are not randomised.[225] The ROBINS-I tool is structured into seven domains: 1) bias due to confounding; 2) bias in selection of participants into the study; 3) bias in classification of

interventions; 4) bias due to deviations from intended interventions; 5) bias due to missing data; 6) bias in measurement of outcomes; and 7) bias in selection of the report results. The risk of bias judgment for the previous domains and the overall risk of bias uses a rating of “low risk of bias”, “moderate risk of bias”, “serious risk of bias”, “critical risk of bias” and “no information”. The corresponding rating of risk of bias was used in the evidence base section of the HNMRC body of evidence matrix to grade the potential recommendation.

3.4.13 OBSERVATIONAL STUDIES

Risk of bias of observational studies was assessed by a previously established tool for prevalence studies.[147] The assessment tool is composed of 10 items covering four main domains of bias including external validity, internal validity, measurement bias and bias relating to analysis. Each risk of bias item will receive a binary response (score) of “low risk” (0) or “high risk” (1). Bias will be calculated by combining the score of all 10 items and classified as “high risk” (7-9), “moderate risk” (4-6) or “low risk” (0-3) according to the established checklist. The corresponding rating of risk of bias was used in the evidence base section of the NHMRC body of evidence matrix to grade the potential recommendation.

3.4.13.1 STEP 5: ASSESS THE BODY OF EVIDENCE AND FORMULATE RECOMMENDATIONS

Formulating and grading recommendations followed the NHMRC FORM (Australian method for formulating and grading recommendations) methodology.[113] The Working Group assessed the body of evidence by

completing the NHMRC Evidence Statement (see Appendix 3.8) which was used to evaluate the volume of evidence, the consistency, clinical impact, generalisability, applicability and an evidence statement (Table 3.2). To facilitate this process, the Working Group communicated via emails, teleconferences, and face-to-face meetings. After the completion of the NHMRC Evidence Statement, an evidence-based recommendation relating to the body of evidence was formed. The overall grade of the recommendation was reported according to the *NHMRC levels of evidence and grades for the recommendations for developers of guidelines* (Table 3.3). In situations where there was insufficient evidence, the Working Group developed consensus-based recommendations or practice points (Table 3.4). The recommendations became final once all members of the Working Group reached a consensus on the wording and content of each recommendation. The IP Guideline adapted the National Institute for Health and Care Excellence methodology when developing the wording of the recommendations (Table 3.5).[226]

Table 3.2 NHMRC body of evidence matrix

Component of Recommendation	**Recommendation Grade**			
	A - Excellent	**B - Good**	**C - Satisfactory**	**D - Poor**
Evidence base[1]	One or more level I studies with a low risk of bias or several level II studies with a low risk of bias	One or two level II studies with a low risk of bias or a systematic review/several level III studies with a low risk of bias	One or two level III studies with a low risk of bias, or level I or II studies with a moderate risk of bias	Level IV studies, or level I to III studies/systematic reviews with a high risk of bias

Consistency [2]	All studies consistent	Most studies consistent and inconsistency may be explained	Some inconsistency reflecting genuine uncertainty around clinical question	Evidence is inconsistent
Clinical impact	Very large	Substantial	Moderate	Slight or restricted
Generalisability	Population/s studied in body of evidence are the same as the target population for the guideline	Population/s studied in the body of evidence are similar to the target population for the guideline	Population/s studied in body of evidence differ to target population for guideline but it is clinically sensible to apply this evidence to target population[3]	Population/s studied in body of evidence different to target population and hard to judge whether it is sensible to generalise to target population
Applicability	Directly applicable to Australian healthcare context	Applicable to Australian healthcare context with few caveats	Probably applicable to Australian healthcare context with some caveats	Not applicable to Australian healthcare context

[1] Level of evidence determined from the NHMRC Evidence Hirerarchy level of evidence criteria

[2] If there is only one study, rank this component as 'not applicable'

[3] For example, results in adults that are clinically sensible to apply children OR psychosocial outcomes for one cancer that may be applicable to patients with another cancer.

Source[113]

Table 3.3 Definition of NHMRC grade of recommendations

Grade of recommendation	Description
A	Body of evidence can be trusted to guide practice
B	Body of evidence can be trusted to guide practice in most situations
C	Body of evidence provides some support for recommendation(s), but care should be taken in its application
D	Body of evidence is weak, and recommendation must be applied with caution

Source[113]

Table 3.4 NHMRC approved recommendation types and definitions

Type of recommendation	Definition
Evidence-based recommendation	A recommendation formulated after a systematic review of the evidence, indicating supporting references
Consensus-based recommendation	A recommendation formulated in the absence of quality evidence, after a systematic review of the evidence was conducted and failed to identify admissible evidence on the clinical question
Practice point	A recommendation on a subject that is outside the scope of the search strategy for the systematic review, based on expert opinion and formulated by a consensus process

Source[227]

Table 3.5 Guideline definitions for evidence-based statements and wording

Statement	Definition	Implication	Wording
Strong Recommendation	A strong recommendation means the benefits of the recommended approach clearly exceed the harms (or that the harms clearly exceed the benefits in the case of a strong negative recommendation), and that the quality of the supporting evidence is excellent (Grade A or B). In some clearly identified	Clinicians should follow a strong recommendation unless a clear and compelling rationale for an alternative approach is present.	Recommendations should contain the term “offer”, “advise”, “do NOT offer”, or “do NOT advise”.

	circumstances, strong recommendations may be made based on lesser evidence when high-quality evidence is impossible to obtain, and the anticipated benefits strongly outweigh the harms		
Recommendation	A recommendation means the benefits exceed the harms (or that the harms exceed the benefits in the case of a negative recommendation), but the quality of evidence is not as strong (Grade B or C). In some clearly identified circumstances, recommendations may be made based on lesser evidence when high-quality evidence is impossible to obtain, and the anticipated benefits outweigh the harms.	Clinicians should also generally follow a recommendation but should remain alert to new information and sensitive to patient preferences.	Recommendations should contain the term “consider” or “consider NOT”.
Option	An option means that either the quality of evidence that exists is suspect (Grade D) or that well-done studies (Grade A, B, or C) show little clear.	Clinicians should be flexible in their decision making regarding appropriate practice, although they may set bounds on alternatives; patient preference should have a substantial influencing role.	Recommendations should contain the term “may consider”.

No Recommendation	No recommendation means there is both a lack of pertinent evidence (Grade D) and an unclear balance between benefits and harms.	Clinicians should feel little constraint in their decision making and be alert to new published evidence that clarifies the balance of benefit versus harm; patient preference should have a substantial influencing role.	Clearly state no recommendation can be made.

Source[228]

3.4.13.2 STEP 5: WRITE THE CONTENT NARRATIVE

Each member of the Working Group reviewed and revised the content and structure of the guideline. An online meeting with all Working Group members was held to finalise the draft guideline before stakeholder consultation. During the meeting, each recommendation was reviewed and approved by consensus.

3.4.14 STAKEHOLDER ENGAGEMENT

In accordance with the NHMRC guideline, key stakeholders are required to be a part of the development of a clinical practice guideline to ensure their values and preferences have been considered. Essential stakeholders include patients with suspected or diagnosed IP (target population), clinicians (target users), lecturers and subject coordinators at educational institutions, members of professional associations and societies, and commercial companies dealing with pathology

and supplementation. The early and continuous involvement of stakeholders was incorporated throughout the guideline development according to strategies recommended by the NHMRC (Table 3.6). The values, preferences, health-seeking behaviours, and experience of stakeholders were acquired through appropriate methods to capture the necessary information. Below are the methods that were used to involve stakeholders.

3.4.15 TARGET USER GROUP INVOLVEMENT

Target users of the IP Guideline are clinicians within a private practice in Australia, identifying as diagnosing and treating patients with altered IP. Pre-existing research that investigated the experience of clinicians in the diagnosis and management of IP informed the scoping of the guideline development. Target users also form part of the Stakeholder Group; therefore, directly contribute to reviewing the IP Guideline. These target users are clinicians with a diverse degree of clinical experience in the diagnosis and treatment of IP.

3.4.16 CONSUMER INVOLVEMENT

The target population are Australian adults with suspected or diagnosed altered IP. The views and preferences of consumers/target population were continuously integrated into the development of the IP Guideline from the initial scoping and planning through to the implementation of the IP Guideline according to the NHMRC requirement A.4. The views and preferences of Australian adults with suspected IP were drawn upon by undertaking a cross-sectional survey of the target population, ensuring the IP Guideline is both relevant and appropriate. This

survey informed the Working Group as to the needs, priorities, gaps, and scope within clinical practice. Although the Stakeholder Group did not include a representative from Aboriginal and Torres Strait Islander Peoples, the views and preferences of the community were incorporated during The Leaky Gut Survey as participants included in this study identified as Aboriginal and Torres Strait Islander.

3.4.17 STAKEHOLDER GROUP INVOLVEMENT

The Stakeholder Group was comprised from clinicians, lecturers and subject coordinators at educational institutions, members of professional associations and societies, commercial companies dealing with pathology and supplementation. This group was formed after the initial scoping of the guideline. The specific clinicians were identified after investigating the health-seeking behaviours the target population. A survey was used during the guideline development to gain an understanding of the stakeholders' values and preferences towards the developed recommendations. Stakeholders were asked whether they foresee any barriers to the implementation of these recommendations.

Table 3.6 Methods used to include views and preferences of stakeholders in guideline development

	Stakeholder Group		
	Target user	*Target population*	*Other stakeholders**
Scoping the guideline	Survey	Survey	-
Planning the evidence review	Survey	Survey	-
Conducting the evidence review	Survey	Survey	-

Reviewing draft recommendations	Survey	-	Survey
Develop consumer or companion resources	Survey	Survey	Survey
Planning implementation of the guideline	Survey	Survey	Survey

*Other stakeholders include lecturers and subject coordinators from leading educational institutions, members of professional associations and societies, commercial companies dealing with pathology and supplementation.

3.4.18 GUIDELINE DEVELOPMENT GROUP

The IP Guideline was intended for use by clinicians in Australian private practice. The Guideline Development Group was comprised of two main groups: the Working Group and Stakeholder Group. The Working Group was involved in the development of the IP Guideline. Whereas the Stakeholder Group was included in providing their views and feedback on the developed recommendations. These groups were formulated from a multidisciplinary background of health professionals representing all potential clinicians which may see patients with IP, content experts and other major stakeholders. Bradley Leech chaired the Guideline Development Group.

3.4.18.1 WORKING GROUP

The multidiscipline Working Group initiated the IP Guideline after identifying gaps in both the published literature and clinical practice for the management of IP (Table 3.7). The role of the Working Group was to conduct, coordinate and collaborate on all aspects of the development and implementation, ensuring the completion of the IP Guideline.

Table 3.7 The Working Group members

Name	Discipline/Role	Affiliation
Bradley Leech	Project lead (Chair)	University of Technology Sydney
Prof David Sibbritt	Professor of Epidemiology	University of Technology Sydney
Dr Amie Steel	Senior Research Fellow	University of Technology Sydney
Dr Erica McIntyre	Postdoctoral Research Fellow	University of Technology Sydney

3.4.18.2 STAKEHOLDER GROUP

During the planning stages of the IP Guideline, the Working Group identified key stakeholders who should be involved in the development of the IP Guideline. The relevant stakeholders include content experts, health professionals, lecturers and subject coordinators from leading educational institutions, members of professional associations and societies, commercial companies dealing with pathology and supplementation (Table 3.8). Members for the Stakeholder Group were put forward by the Working Group as potential candidates. A consensus agreement between all members of the Working Group was reached for inviting each member of the Stakeholder Group to participate in the IP Guideline development. Stakeholder Group members were invited as representatives of their field or discipline, and not necessary content experts. Members of the Stakeholder Group were either nominated by the Working Group or their relevant professional organisations to represent the discipline. The Working Group worked closely with the Stakeholder Group to support the active feedback on the developed recommendations. The views, preferences and involvement of stakeholders were acquired through survey design (Table 3.6).

Table 3.8 The Stakeholder Group members

Name	Discipline/Role	Affiliation(s)
Dr Jason Hawrelak	Naturopath, gastrointestinal health research	University of Tasmania
Dr Nirala Jacobi	Naturopath, pathology company	The SIBO Doctor
Dr Michael Osiecki	Professional association, supplement company	Bio Concepts Pty Ltd, Complementary Medicine Australia
Dr Christine Houghton	Supplement company, nutritionist	Cell-Logic Pty Ltd, University of Queensland
Dr Ronald Goedeke	GP, Integrative medical practitioner	Appearance Medicine and Wellness Centre
Benedict Freudenmann	Clinical nutritionist	Learn to Nourish
Kirsty Wirth	Food company, health clinic	Kultured Wellness
Vanita Dahia	Pharmacist, pathology company	Alchemy of Health, NutriPATH

3.4.18.3 MANAGING CONFLICTS OF INTEREST

All members of the Stakeholder Group were required to declare any or actual conflicts of interest before the development of the IP Guideline. The Working Group modelled a disclosure of interest form in accordance with the NHMRC, which all members in the Stakeholder Groups were required to fill out (see Appendix 3.9). The register of disclosures of interest for each member and the required methods used to manage conflict of interest are detailed in Appendix 3.10. Any identified conflict of interest were managed by following the conflict-of-interest management plan (see Appendix 3.11) (Figure 3.1). Where a member declared any direct or indirect conflict of interest that could not be managed, the members response to the survey question(s) were excluded. All Disclosure of Interests form were reviewed by the chair when a conflict of interest was reported and identified, the Working Group discussed the appropriate management plan.

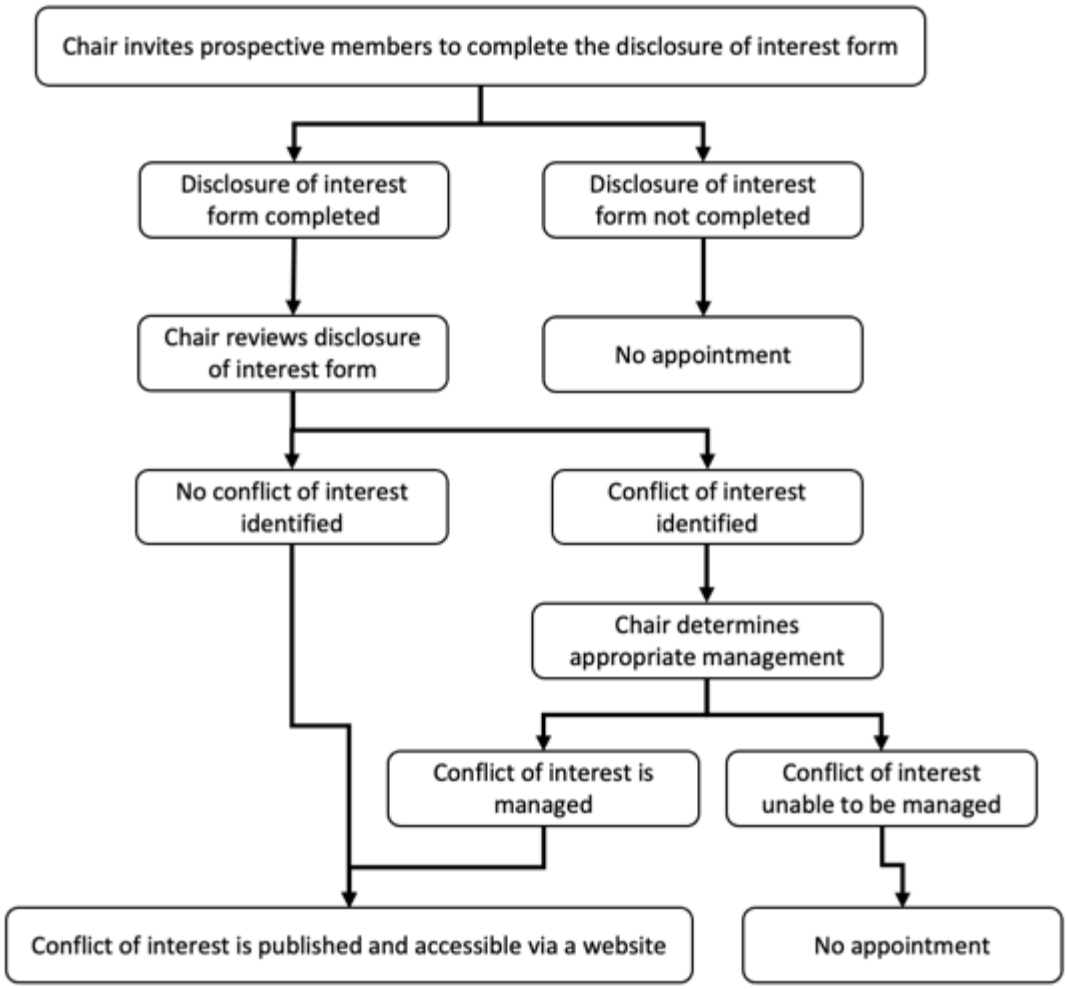

Figure 3.1 Disclosure of interest process flow chart

3.4.19 TERMS OF REFERENCE

All members were supplied with a *Terms of Reference* document (adopted from NHMRC 2019) that outlines the purpose of the group, expectations and responsibilities (see Appendix 3.12). Below are the responsibilities for each member of the Guideline Development Group.

3.4.19.1 RESPONSIBILITIES OF THE WORKING GROUP

The responsibilities of the Working Group are as follows:

- Develop the formation of the guideline development group,
- Overseeing the development of the IP Guideline,
- Agree on the scope and clinical questions based on consideration of the research-evidence,
- Collaborate in Working Group meetings,
- Manage all conflicts of interests in the Stakeholder Group,
- Identify, critically appraise and synthesise evidence for developing recommendations,
- Formulate recommendations based on evidence,
- Ensure effective consumer involvement
- Develop a dissemination and implementation plan,
- Evaluate the acceptability and feasibility of each recommendation,
- Assess the risks and benefits of treatment recommendations,
- Organise and manage the Stakeholder Group.

3.4.19.2 RESPONSIBILITIES OF THE STAKEHOLDER GROUP

The responsibilities of the Stakeholder Group are as follows:

- Participate in the Stakeholder Group survey,
- Report all relevant conflicts of interests,
- Evaluate the acceptability and feasibility of each recommendation,
- Provide insight on the extent published evidence reflects outcomes,
- Provide feedback on guideline wording to ensure recommendations are understandable.

3.4.20 DISSEMINATION AND IMPLEMENTATION

The IP Guideline was publicly available online via a purpose-built website for the guideline dissemination. A link to SIP Research (www.sipresearch.org) was shared on social media and sent directly to relevant stakeholders.

3.5 STAKEHOLDER SURVEY METHODOLOGY

3.5.1 AIM AND OBJECTIVES

This survey aims to assess whether the developed recommendations are clear and suitable for clinicians to follow in the management of IP by addressing the following objectives:

1. Evaluate the level understanding of each recommendation;
2. Assess the appropriateness of each recommendation for clinicians to follow in the management of IP; and
3. Evaluate the importance of each recommendation for clinicians to follow in the management of IP.

3.5.2 SETTING AND SAMPLING

The target population of this survey are stakeholders that have expertise, experience or involved in some area of IP management. To accurately represent the target population, a diverse range of stakeholders where included. This includes content experts, health professionals, lecturers and subject coordinators from leading educational institutions, members of professional associations and societies, commercial companies dealing with pathology and supplementation. In

accordance with the NHMRC Guidelines for Guidelines Handbook, a total of 8 stakeholders with a multidisciplinary approach were included in the Stakeholder Group.[215] A group of 8-12 members with a multidisciplinary background is ideal to reduce bias and increase efficiency in guideline development.[215] To capture the most appropriate stakeholders, the Working Group put forward potential members with a consensus agreement reached by the Working Group for the involvement of each member of the Stakeholder Group. Targeting the individuals involved in the survey allowed the greatest possibility to capture a diverse range of expertise, views and values towards the treatment of IP. The survey was open for 1 month between April 2022 and May 2022.

3.5.3 DATA COLLECTION

Data collection was undertaken by a cross-sectional, self-administered, online questionnaire through *Qualtrics software.* After data collection, complete and incomplete data was transferred to Microsoft Excel for analysis. The full questionnaire has been included in Appendix 3.13. The questionnaire was composed of 191 to 229 questions which was dependent on the participants answers to piping questions. A series of five to six questions were asked for each of the 38 different recommendations. The use of Five-point Likert scales were chosen to accurately gauge participants' level of understanding, agreement for the appropriateness and importance for the recommendations throughout the survey.[207] [207] The responses ranged from "strongly disagree" to "strongly agree". In the event the participant was not familiar with the content or wanted to abstain from providing feedback, the middle response of the Likert scale was "natural". One additional question was asked providing stakeholders with the binary option

("yes" or "no") to indicate if they would change anything to the recommendation. If changes were suggested, an additional open-ended space became available for participants to describe the required changes.

3.5.4 STATISTICAL ANALYSIS

Data obtained from the questionnaire was exported to Microsoft Excel for statistical analyses. Responses to questionnaire items were reported as frequencies and percentages, where appropriate.

3.6 CHAPTER SUMMARY

In order to develop evidence-based recommendations for the management of IP in clinical practice, a two-phase approach is required. By first exploring the views and perspectives of Australian adults with suspected IP via a cross-sectional survey and then applying the findings to guide recommendation development, ensures the IP Guideline is relevant and appropriate for the target population. Following the NHMRC guidelines for guideline development, a structured and evidence-based approach is used in the development of the IP Guideline. The methods described in this chapter provides the best possibility to address the objectives of this research project.

4. VIEWS AND PREFERENCES OF AUSTRALIAN ADULTS WITH SUSPECTED INCREASED INTESTINAL PERMEABILITY

The most significant risk factors associated with IP have been extensively explored in Chapter 2. This research project seeks to develop evidence-based treatment recommendations to improve clinicians' management of IP in clinical practice. However, to create appropriate recommendations for people with IP, an understanding of their health-seeking behaviour is required. Therefore, to provide essential knowledge for this research project, a cross-sectional survey of Australian adults with suspected IP was undertaken to explore their health-seeking behaviour towards the management of IP. This chapter presents the methods undertaken to survey the target population and the results of this study.

4.1 PUBLICATION OF RESULTS

The results presented within this chapter have been published (see Appendix 4.1) as follows:

> Leech, B, McIntyre, E, Steel, A, Sibbritt, D (2019) "Health-seeking behaviour, views and preferences of adults with suspected increased intestinal permeability: A cross-sectional survey of Australian adults", Integrative Medicine Research, 2022, Vol 11, 1.

4.2 HEALTH-SEEKING BEHAVIOUR, VIEWS AND PREFERENCES OF ADULTS WITH SUSPECTED INCREASED INTESTINAL PERMEABILITY: A CROSS-SECTIONAL SURVEY OF AUSTRALIAN ADULTS

4.2.1 INTRODUCTION AND BACKGROUND

The single layer of epithelial cells that separate the internal and external environment of the small intestine is renewed every four to five days, playing an essential role in maintaining intestinal homeostasis.[229] Increased intestinal permeability (IP) involves the disassembling of tight junction proteins between the cells of the small intestine, resulting in a loss of intestinal barrier integrity.[2] With an estimated prevalence of 10-87% in health conditions with a known association,[1] altered IP has been suggested to play an important role in health and disease in both public and private healthcare.[3]

The clinical relevance and consequence of altered IP remain a controversial topic within conventional medicine.[67] Yet, published literature continues to identify IP as a target for disease prevention and therapeutic intervention.[3] IP has been suggested to precede the onset of a number of chronic health conditions such as Crohn's disease,[230] liver disease,[231] type 1 diabetes,[58,59,134,232] coeliac disease,[59] rheumatoid arthritis,[233] gestational diabetes,[60] and diarrhoea-predominant irritable bowel syndrome.[137,138] Altered IP is also associated with autoimmune conditions, metabolic conditions, liver diseases, and gastrointestinal conditions.[1,221] Although IP is a reaction within the small intestine, many of the

measurable and clinically relevant risk factors are systemic, suggesting that IP is more than a digestive health issue and a possible feature of disease.[221]

Previous research has investigated the assessment and management of IP from the practitioner standpoint, where practitioners acknowledge the involvement of IP in many health conditions found in clinical practice.[4,5] Within clinical practice, the pathology tests available are invasive, require patients to pay out-of-pocket, and involve a substantial amount of time to perform.[4] Practitioners that frequently treat IP in clinical practice are reported to avoid using validated pathology tests due to the financial cost to the patient and prioritise case history to diagnose IP.[4] While the frequency of methods used by patients, including the accuracy of self-diagnosis remains unknown, the self-diagnosis of other chronic illnesses such as diabetes is considered to be somewhat accurate.[234] Furthermore, no research to date has considered patients views and preferences towards the assessment and management of IP, resulting in knowledge gaps for evidence-based practice. Incorporating patients views and preferences in the decision-making process is often overlooked however, a positive impact on the outcome of healthcare is observed when patients views and preferences are considered.[235] As such, this study aims to describe the health-seeking behaviour of Australian adults with suspected IP while also exploring the views and preferences surrounding the assessment and management of IP.

4.2.2 METHODS

4.2.2.1 STUDY DESIGN AND SETTING

A cross-sectional study design using an online self-reported survey was utilised with approval from the Human Research Ethics Committees (HREC) of the University of Technology Sydney (#ETH19-4012).

4.2.2.2 PARTICIPANTS AND RECRUITMENT

Participants were recruited via social media platforms and a purpose-built webpage, with snowball sampling methods also used. The survey was open for two months between September 2019 and November 2019. Eligibility to participate in this study required participants to either suspect or know they have altered IP, be aged 18 years or more, living in Australia and have internet access. Survey responders with incomplete demographic characteristics, accounting for <5% of total data were excluded from analysis. This study was designed to capture people that may have suspected IP or confirmed IP, to best reflect the type of patients that present to clinical practice for the treatment of IP.[4]

4.2.2.3 SURVEY INSTRUMENT

The developed online survey utilised the questionnaire items which were obtained from published literature and modified to suit Australians with suspected IP.[4,5] To improve the survey's reliability, standardised five-point Likert scales were used for scaling questions. The survey included three main domains: demographic characteristics, diagnosis of IP, and the financial expenditure related to IP. The questionnaire was first pilot tested using lay people to assess

the time required to complete the survey and language clarity, with corrections made accordingly.

4.2.3 DEMOGRAPHIC CHARACTERISTICS

The participants were asked about their gender, age, height, and weight. Body mass index (BMI) was calculated from height and weight measurements. BMI was then categorised to underweight, healthy weight, overweight, and obese.[236] The participants were also asked their country of birth, the state or territory where they live, and whether they live in an urban, rural or remote location.

4.2.4 DIAGNOSIS OF INCREASED INTESTINAL PERMEABILITY

Participants were asked a number of questions in relation to the assessment of IP including: the year they believed their IP started, the year their IP was diagnosed, the method used to confirm their IP, at what point their IP was assessed, the number of times their IP was assessed, and the qualifications of the practitioner involved in the assessment of their IP. In addition, participants preference for IP testing method characteristics, the preferred method and time point for IP assessment, and the qualifications of their preferred practitioner were all asked. To gauge the preference and importance towards being assessed for IP and the likelihood of treatment adherence if results returned a positive test of altered IP, 5-point Likert scales were used. The term ‘assessed’ and ‘assessment’ are used throughout this article to describe the action participants used for measuring, evaluating or identifying IP.

4.2.5 FINANCIAL EXPENDITURE

A number of items participants were asked to report: the out-of-pocket expenditure of treating IP, practitioner consultation fees, and cost of measuring IP. Participant's income manageability was determined by how well they manage their household income, categorised as 'difficult all the time', 'difficult some of the time', 'not too bad', or 'easy'. The amounts are reported in Australian dollars (AUD).

4.2.5.1 DATA COLLECTION

The survey was administered through the online platform *SurveyGizmo*. After data collection, data was exported to a statistical software program STATA® 16 for data checking and statistical analyses.

4.2.5.2 STATISTICAL ANALYSES

Responses to questionnaire items were reported as means, standard deviations, 95% confidence intervals (CIs) or frequencies and percentages. Chi-square analysis was used for tests of association between categorical variables and Student's t-tests were used for continuous variables across a binary variable. Ordinal variables such as those on Likert scales were assessed with non-parametric tests, including Mann-Whitney U test and the Wilcoxon signed ranks test, where appropriate. Analysis of variance (ANOVA) was used to measure the difference between a continuous variable across a categorical variable.

4.2.6 RESULTS

4.2.6.1 DEMOGRAPHIC CHARACTERISTICS

A total of 982 people responded to the survey, of which 393 responses did not meet the eligibility criteria or were classified as having too much incomplete data, leaving a total of 589 participants. Most participants were female (n=548, 93%), living within an urban area (n=416, 70.6%) in either New South Wales (n=175, 29.7%) or Queensland (n=161, 27.3%) (Table 4.1). The mean age of the participants was 45.0 (SD=12.1) with a mean BMI of 27.0 (SD=6.9). The income manageability of participants was described most commonly as 'easy or not too bad' (n=209, 46.5%) and 'difficult some of the time' (n=145, 32.3%). Half the surveyed population reported altered IP as their primary health concern (n=300, 50.9%) with a range of other autoimmune, inflammatory gastrointestinal, and metabolic conditions reported for the other half (Table 4.1).

Table 4.1 Demographic characteristics of study participants (n=589)

Demographics	**Distribution of responses**	
	n	**%**
Gender		
Female	548	93.0
Male	41	7.0
BMI classification		
Underweight	19	3.3
Healthy weight	268	46.1
Overweight	138	23.8
Obese	156	26.9
Country of birth		
Australia	476	81.0
Other	112	19.0
State or territory		
New South Wales	175	29.7
Queensland	161	27.3

Victoria	103	17.5
Western Australia	63	10.7
South Australia	36	6.1
Australian Capital Territory	23	3.9
Tasmania	18	3.1
Northern Territory	10	1.7
Area of residence		
Urban	416	70.6
Rural	161	27.3
Remote	12	2.0
Income manageability		
Easy or not too bad	209	46.5
Difficult some of the time	145	32.3
Difficult all the time	95	21.2
Primary health concern		
Increased intestinal permeability	300	50.9
Other autoimmune diseases	40	6.8
Hashimoto's thyroiditis	28	4.8
Gastrointestinal issues	24	4.1
Chronic fatigue syndrome	21	3.6
Rheumatoid arthritis	18	3.1
Obesity	15	2.6
Mental health	13	2.2
Hormonal issues	10	1.7
Fibromyalgia	9	1.5
Gastrointestinal *Candida albicans*	8	1.4
Psoriatic arthritis	7	1.2
Mould exposure	7	1.2
Irritable bowel syndrome	6	1.0
Ankylosing spondylitis	6	1.0
Asthma	6	1.0
Food intolerances	6	1.0
Cardiovascular disease	6	1.0
Mast cell activation syndrome	6	1.0
Other health conditions	53	9.0
	Mean	**SD (range)**
Age in years	45.0	12.1 (18-82)
Body Mass Index (BMI)	27.0	6.9 (15.4-64.5)

4.2.6.2 DIAGNOSIS OF INCREASED INTESTINAL PERMEABILITY

The most frequently used methods to diagnose IP were self-diagnoses (n=330, 56.2%) and case history, according to a healthcare practitioner (n=130, 22.2%) (Table 4.2). From the participants that were assessed for IP, 17.3% (n=102) were assessed before receiving treatment, 4.1% (n=24) during the treatment phase, and only 1.4% (n=8) after treatment was completed. Of the participants who were diagnosed with IP, 59.1% (n=140) reported being diagnosed within the last three years. However, on average, participants with suspected IP spent 11.1 (95% CI: 9.5, 12.8) years between first suspecting IP and receiving a diagnosis. No statistically significant difference was found between the length of time between when participants first suspected IP to the year they were diagnosed and whether they were diagnosed by a medical practitioner or another healthcare practitioner ($p=0.120$). The vast majority of participants were not assessed for IP (n=459, 77.9%) with only 17.7% (n=104) assessed once, and 4.4% (n=26) assessed more than twice. For the participants that were assessed two or more times, the second assessment of IP typically took place between 6 and 12 months (n=11, 42.3%). A significant association between the number of times IP was assessed and the person (practitioner or self) who diagnosed IP was found ($p<0.001$). Specifically, healthcare practitioners and medical practitioners more frequently assessed IP (n=74, 33.9%; n=39, 33.6%, respectively) compared to those who self-diagnosed (n=4, 1.9%).

Table 4.2 Health seeking behaviour for the assessment and management of increased intestinal permeability by Australian adults (n=589)

Health Seeking Behaviour	Distribution of Response	
	n	%
Method of assessment (n=587)		
Self-diagnosed	330	56.2
Case history according to a practitioner	130	22.2
IgG food sensitivity test	29	4.9
Hemaview - live blood analysis	23	3.9
Stool zonulin	22	3.8
Lactulose/mannitol urine test	17	2.9
I don't know	16	2.7
Iridology	12	2.0
Serum zonulin	4	0.7
Kinesiology	4	0.7
Stage that IP was measured (n=134)		
Before treatment	102	17.3
During the treatment phase	24	4.1
After treatment was completed	8	1.4
Number of times measured for IP (n=589)		
0	459	77.9
1	104	17.7
2 +	26	4.4
Time between initial and second assessment (n=26)		
Between 1 and 6 months	2	7.7
Between 6 and 12 months	11	42.3
Between 12 and 24 months	6	23.1
Over 2 years	7	26.9
Year IP was diagnosed (n=237)		
< 3 years	140	59.1
4-6 years	46	19.4
7-9 years	22	9.3
> 10 years	29	12.2
Year participant believe IP started (n=498)		
< 3 years	84	16.9
4-6 years	82	16.5
7-9 years	77	15.5
> 10 years	255	51.2

IP: increased intestinal permeability

4.2.6.3 PRACTITIONERS INVOLVED IN THE DIAGNOSIS OF INCREASED INTESTINAL PERMEABILITY

Most participants (n=374, 67.4%) first suspected they had IP, whereas 32.6% (n=181) had a practitioner first suggest IP as a possible diagnosis. Participants were most frequently diagnosed with IP by self-diagnosing (n=274, 47.9%), followed by a naturopath (n=207, 36.2%), integrative medicine practitioner (n=82, 14.3%), nutritionist (n=53, 9.3%), and general practitioner (n=50, 8.7%) (Table 4.3). Most participants preferred their IP to be assessed by a naturopath (n=363, 63.5%), followed by a general practitioner (n=310, 54.2%), integrative medicine practitioner (n=259, 45.3%), nutritionist (n=225, 39.3%), gastroenterologist (n=221, 38.6%) or a dietitian (n=162, 28.3%). From the participants that self-diagnosed, their preferred practitioner for the assessment of IP was a general practitioner (n=118, 56.7%) or a naturopath (n=118, 56.7%).

Table 4.3 Practitioners involved in the diagnosis of increased intestinal permeability (n=572)

Who diagnosed increased intestinal permeability	Diagnosis of increased intestinal permeability					
	Initial diagnosis		Total preferred practitioner		Preferred practitioner for self-diagnosed	
	n	%	n	%	n	%
Naturopath	207	36.2	363	63.5	118	56.7
Integrative medicine practitioner	82	14.3	259	45.3	75	36.1
Nutritionist	53	9.3	225	39.3	69	33.2
General practitioner	50	8.7	310	54.2	118	56.7
Herbalist	19	3.3	101	17.7	31	14.9
Kinesiologist	19	3.3	86	15.0	22	10.6
Dietitian	17	3.0	162	28.3	60	28.9
Chinese medicine practitioner	15	2.6	110	19.2	35	16.8
Homeopath	13	2.3	77	13.5	24	11.5

Acupuncturist	11	1.9	78	13.6	26	12.5
Chiropractor	11	1.9	58	10.1	18	8.7
Gastroenterologist	10	1.8	221	38.6	90	43.3
Ayurvedic practitioner	6	1.1	73	12.8	25	12.0
Osteopath	4	0.7	40	7.0	13	6.3
Nurse	3	0.5	53	9.3	19	9.1
Nurse practitioner	3	0.5	52	9.1	18	8.7
Pharmacist	1	0.2	54	9.4	14	6.7
Self-diagnosed	274	47.9	-	-	-	-

4.2.6.4 EXPENDITURES RELATED TO THE ASSESSMENT AND MANAGEMENT OF INTESTINAL PERMEABILITY

On average, participants reported spending $698.78 on consultation fees and $2175.96 on dietary supplements over the previous 12 months (Table 4.4). There was a statistically significant difference between income manageability and the average amount spent on dietary supplements. Specifically, participants who find it ‘difficult all the time’ to live on their available household income spend significantly more (mean=$2963.28) on dietary supplements over 12 months compared to participants who described their income manageability as ‘easy or not too bad’ ($1918.56; p=0.015). No significant differences were found between who diagnosed their IP and the average amount spent on dietary supplements in the previous 12 months (p=0.167). However, participants that were diagnosed by a medical practitioner spent on average $2309.16 on dietary supplements over the previous 12 months, whereas those who were self-diagnosed spent on average of $1793.40. Participants on average spent $286.76 on the assessment of IP with no significant difference found with either income manageability or the source of diagnosis.

Table 4.4 Expenditures related to the assessment and management of increased intestinal permeability and association with income manageability (n=447)

Expenditures	Income manageability								
	Mean expenses per person		Difficult all the time		Difficult some of the time		Easy or not too bad		p-value
	n	Mean	n	Mean	n	Mean	n	Mean	
Expenses for the management of IP in the previous 12 months									
Consultation fees	424	$698.78	91	$903.49	136	$745.99	197	$571.62	0.057
Dietary supplements	309	$2175.96	67	$2963.28	96	$2019.36	146	$1918.56	0.015
Expenses for the assessment of IP									
All assessment methods	74	$286.76	13	$238.46	24	$315.21	37	$285.27	0.847
Food sensitives - IgG	19	$515.53	4	$385.00	4	$738.75	11	$481.81	0.746
Stool zonulin	16	$329.38	4	$252.50	4	$500.00	8	$282.50	0.089
Hemaview - live blood analysis	15	$204.33	1	$120.00	8	$243.13	6	$166.66	0.620
Iridology	10	$167.50	3	$110.00	2	$100.00	5	$229.00	0.515
Lactulose/mannitol urine test	9	$115.00	-	-	4	$63.75	5	$156.00	0.030
Kinesiology	4	$77.50	1	$100.00	2	$105.00	1	$0.00	0.484
Serum zonulin	1	$70.00	-	-	-	-	1	$70.00	-
	n	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**p-value**
Importance of cost in the decision to be tested									
Very important	260	58.8	72	75.8	94	65.3	94	46.3	
Important	112	25.3	13	13.7	37	25.7	62	30.5	<0.001
Not important	70	15.8	10	10.5	13	9.0	47	23.2	
Amount willing to spend on IP assessment									
$0-$50	107	23.9	43	45.3	38	26.2	26	12.6	
$51-$150	218	48.8	41	43.2	67	46.2	110	53.1	<0.001
$151 or over	122	27.3	11	11.6	40	27.6	71	34.3	
Preference towards expense allocation to:									
Dietary treatments									
Very important	309	70.6	65	70.7	101	70.1	143	70.8	0.953

Important	101	23.1	20	21.7	33	22.9	48	23.8	
Not important	28	6.4	7	7.6	10	6.9	11	5.5	
Dietary supplements									
Very important	265	60.9	53	60.2	90	63.4	122	59.5	
Important	107	24.6	22	25.0	33	23.2	52	25.4	0.968
Not important	63	14.5	13	14.8	19	13.4	31	15.1	
The assessment of IP									
Very important	248	56.5	46	51.7	88	61.1	114	55.3	
Important	117	26.7	18	20.2	39	27.1	60	29.1	0.018
Not important	74	16.9	25	28.1	17	11.8	32	15.5	
Lifestyle treatments									
Very important	240	55.4	58	64.4	78	55.3	104	51.5	
Important	135	31.2	22	24.4	43	30.5	70	34.6	0.349
Not important	58	13.4	10	11.1	20	14.2	28	13.9	
Medications									
Very important	56	13.6	13	15.3	18	13.7	25	12.8	
Important	61	14.8	11	12.9	17	13.0	33	16.8	0.841
Not important	295	71.6	61	71.8	96	73.3	138	70.4	

IP: increased intestinal permeability

There is a statistically significant difference between who diagnosed their IP and the average amount spent on consultation fees in the previous 12 months ($p<0.001$). Specifically, those who were diagnosed by a medical practitioner, or another kind of healthcare practitioner spent significantly more (mean=$980.63 and $996.29 respectively) on consultation fees compared to participants who self-diagnosed IP ($226.45). No difference was found for the average amount spent on consultation fees between a medical practitioner or healthcare practitioners.

4.2.6.5 VIEWS AND PREFERENCES TOWARDS THE COSTS INVOLVED WITH INTESTINAL PERMEABILITY

Participants reported that the cost involved in testing IP is 'very important' in their decision to be tested (n=260, 58.8%), with many participants (n=218, 48.8%) indicating they are willing to spend between $51 and $150 on the testing procedure for IP (Table 4.4). However, the importance of cost in the decision to be tested decreased as income manageability increased ($p<0.001$). Furthermore, as income manageability increased, so did the amount participants were willing to spend on the testing procedure for IP ($p<0.001$).

Regardless of income manageability, participants reported a preference towards allocating finances to dietary treatment interventions (n=309, 70.6%) for the management of IP followed by dietary supplements (n=265, 60.9%) and lifestyle treatments (n=240, 55.4%) (Table 4.4). Although half the participants (n=248, 56.5%) reported the financial allocation for the assessment of IP to be 'very important', increased income manageability was associated with the preference

towards allocating finances to the assessment of IP (p=0.018). Irrespective of income manageability, participants reported medication use to be 'not important' for financial allocation (n=296, 71.6%).

4.2.6.6 VIEWS AND PREFERENCES TOWARDS THE ASSESSMENT AND MANAGEMENT OF INTESTINAL PERMEABILITY

The majority of participants (n=527, 89.6%) would prefer to be assessed for IP regardless of income manageability (p=0.054) with 75.0% (n=442) reporting the assessment of IP to be 'very important' (Table 4.5). Accuracy (n=554, 94.9%), accessibility (n=476, 81.4%), and affordability (n=408, 69.5%) were all commonly reported to be 'very important' characteristics for the assessment of IP; whereas non-invasive methods (n=470, 80.6%) and length of time involved to perform the assessment (n=352, 61.1%) were both commonly reported to be 'not important' characteristics for the assessment of IP. Participants further commonly reported the preference to be assessed for IP using blood pathology (n=459, 78.1%) followed by urine collection (n=354, 60.2%) and a stool test (n=325, 44.3%), with a case history assessment by a practitioner (n=242, 41.2%) to be the least preferred method of IP assessment. The time point that participants commonly prefer to be assessed for IP were; before receiving treatment for IP (n=354, 60.1%), for monitoring disease (n=231, 39.2%), when asked by the patient (n=213, 36.2%), for monitoring IP (n=204, 34.6%), after receiving treatment for IP (n=169, 28.7%), when advised by the practitioner (n=160, 27.2%), and during the treatment of IP (n=117, 19.9%).

The majority of participants (n=549, 93.2%) reported they would be 'very likely' to adhere to a treatment protocol if assessed and diagnosed with altered IP (Table 5). In terms of the preferred method of treating IP, participants 'strongly prefer' the use of dietary products (n=392, 82.2%), followed by lifestyle habits (n=357, 76.5%), and dietary supplements (n=324, 68.6%). On the contrary, 82.8% (n=351) of participants 'slightly prefer' the use of medications to treat IP, representing the least preferred method of IP treatment.

Table 4.5 Views and preferences towards the assessment and management of increased intestinal permeability by Australian adults (n=589)

Views and preferences	**Distribution of Responses**	
	n	**%**
Preference to be assessed for IP		
Prefer to be assessed	527	89.6
Prefer not to be assessed	61	10.4
Importance to be assessed for IP		
Very important	442	75.0
Important	78	13.2
Not important	69	11.7
Likelihood of adhering to treatment if assessed and positive for IP		
Very likely	549	93.2
Neutral	23	3.9
Very unlikely	17	2.9
Importance of various assessment characteristics for IP:		
Accuracy		
Very important	554	94.9
Important	25	4.3
Not important	5	0.9
Accessibility		
Very important	476	81.4
Important	78	13.3
Not important	31	5.3
Affordability		

Very important	408	69.5
Important	122	20.8
Not important	57	9.7
Time involved		
Very important	113	19.6
Important	111	19.3
Not important	352	61.1
Non-invasive method		
Very important	56	9.6
Important	57	9.8
Not important	470	80.6
Preference of assessment method		
Blood test	459	78.1
Urine collection	354	60.2
Stool test	325	55.3
Case history	242	41.2
Preference for assessment time point		
Before treatment	354	60.1
For monitoring disease	231	39.2
When asked by the patient	213	36.2
For monitoring IP	204	34.6
After treatment	169	28.7
When advised by the practitioner	160	27.2
During treatment	117	19.9
Preference for treatment method:		
Dietary products		
Strongly prefer	392	82.2
Prefer	50	10.5
Slightly prefer	35	7.3
Lifestyle habits		
Strongly prefer	357	76.5
Prefer	73	15.6
Slightly prefer	37	7.9
Dietary supplements		
Strongly prefer	324	68.6
Prefer	88	18.6
Slightly prefer	60	12.7
Medication		
Strongly prefer	46	10.9
Prefer	27	6.4

Slightly prefer	351	82.8
Important areas for practitioners to comprehend:		
Dietary treatments for IP		
Very important	395	95.2
Important	18	4.3
Not important	2	0.5
Lifestyle treatments for IP		
Very important	389	94.0
Important	23	5.6
Not important	2	0.5
Signs and symptoms of IP		
Very important	390	93.8
Important	25	6.0
Not important	1	0.2
Biomarkers associated with IP		
Very important	378	91.5
Important	28	6.8
Not important	7	1.7
Risk factors for IP		
Very important	373	90.8
Important	34	8.3
Not important	4	1.0
Methods to accurately assess IP		
Very important	376	90.4
Important	31	7.5
Not important	9	2.2
Conditions associated with IP		
Very important	376	90.2
Important	36	8.6
Not important	5	1.2
Dietary supplements for IP		
Very important	366	89.1
Important	33	8.0
Not important	12	2.9
Time point that IP should be assessed		
Very important	362	87.4
Important	45	10.9
Not important	7	1.7
Individuals that require to be assessed for IP		
Very important	354	85.1

Important	53	12.7
Not important	9	2.2
Medications for IP		
Very important	215	52.4
Important	85	20.7
Not important	110	26.8

IP: increased intestinal permeability

4.2.7 DISCUSSION

This is the first study to describe the health-seeking behaviours and explore the views and preferences of adults with suspected or diagnosed IP. The results of this study suggest inconsistencies between the healthcare provided to Australian adults with suspected IP and the healthcare this patient population would prefer to receive. Most notably, the majority of participants experienced a considerable length of time between first suspecting IP and receiving a diagnosis of IP. They also reported challenges involved in the accurate diagnosis of IP and the out-of-pocket expenditure associated with IP.

4.2.7.1 DIAGNOSIS OF INCREASED INTESTINAL PERMEABILITY

Our results indicate that those participants without a formal diagnosis of IP are self-diagnosing; however, have a desire to be assessed using an accurate method by a healthcare practitioner. This discrepancy in the assessment of IP may be contributed in part to the common practices of healthcare practitioners. Practitioners that frequently treat IP in clinical practice avoid measuring IP due to the financial cost to the patient and prioritise case history assessment for diagnosing IP.[4] However, the results of this study suggest that Australian adults with suspected IP are willing to allocate finances to an accurate and accessible

method of IP assessment before receiving treatment. The inconsistencies between the healthcare provided to Australian adults with suspected IP and their preferred healthcare suggest the preferences of the consumer may not always be considered.

As with other health-related conditions, IP is subject to under-diagnosis, over-diagnosis and misdiagnosis within clinical practice.[237,238] Of particular concern from our findings is the high rate of self-diagnosis of IP. This high self-diagnosis rate may result in a misdiagnosis, causing potential anxiety to the patient, unnecessary treatment burden when not required or result in other more serious health conditions being undiagnosed. The high self-diagnosis rate may also have an overall negative effect on practitioner-patient relationship with the potential utilisation of inaccurate or inappropriate treatments.[239] Our study also revealed that Australian adults with suspected IP would prefer a general practitioner or naturopath to assess them for IP. However, the lack of acknowledgement of IP by medical practitioners[67] may be a driving factor for the large number of Australian adults with suspected IP not receiving a formal diagnosis and a contributing factor as to why it takes 11 years for IP to be diagnosed. Whether the length of time for a formal diagnosis of altered IP is contributed to behaviours of the patient or the practitioner is unknown; however, the shortage of validated testing methods and no gold standard testing method are factors influencing healthcare practitioners not to measure IP and to treat regardless.[4]

A common practice for practitioners is the use of case history in the diagnosis of a number of health conditions, especially functional bowel disorders.[240,241] Even

with the extensive algorithms of patients case history, there is still a poor agreement between practitioners and the diagnostic criteria of functional bowel disorders.[242] A concern when applying an algorithmic model of diagnosis to IP is that there is no validated algorithm and the associated case history features of IP remain uncertain, especially as previously perceived symptoms of IP are not associated with diagnostic markers of IP.[4,221] The clinical similarities between gastrointestinal conditions[241] and the under examined clinical features of IP, limits the accuracy of case history as a diagnostic method for IP.

4.2.7.2 FINANCIAL EXPENDITURE OF INCREASED INTESTINAL PERMEABILITY

The out-of-pocket expenditure associated with the assessment and management of IP suggests a financial burden for Australian adults with suspected IP. Although a financial burden calculation is not possible with the data collected in this study, other Australian based studies provide further support for a potential financial burden. For instance, the mean out-of-pocket expenditure for the assessment and management of suspected IP is similar to the amount spent on chronic health conditions in Australia.[243-245] Furthermore, the out-of-pocket expenditure for consultation fees and dietary supplements over a 12 month period is greater than the mean annual expense for Australian adults with gastrointestinal disorders.[246] As Australia has one of the highest out-of-pocket expenditure for medication in the world,[243,247] healthcare practitioners should consider the out-of-pocket expenses related to IP management, especially people with a low income manageability.

The results of this study suggest a significant difference between the income manageability and the average amount spent on dietary supplements, with those who find it 'difficult all the time' to live on their available household income spending significantly more on dietary supplements compared to the 'easy or not too bad' income groups. Other studies suggest people with poor financial status are more likely to face a financial burden in relation to the out-of-pocket expenditure.[244,248] Whether a person's income manageability is a cause or consequence for the out-of-pocket expenditure on IP remains unknown; however, is a worthy area for further investigation.

4.2.7.3 VIEWS AND PREFERENCES OF INCREASED INTESTINAL PERMEABILITY

The results of this study suggest that Australian adults with suspected IP place little importance or value on medication use for the treatment of IP. The strong aversion towards medication use highlights a potential barrier for future pharmacological treatments under development.[111,249] Whether Australian adults with suspected IP will use such medication remains an area for future research. However, what this study does suggest is dietary products (dietary interventions) are the preferred method for the treatment of IP. Dietary interventions are also the most frequently used type of treatment for IP by practitioners in Australia,[5] highlighting agreement between the preferred treatment method for IP and the care given by healthcare practitioners. Utilising the results of this study, patients' views and preferences can help inform the development of a clinical practice guideline for the assessment and management of IP.

4.2.8 LIMITATIONS

There are a number of potential limitations of our study that need to be considered when interpreting our findings. Our sample has a greater percentage of females than the Australian general population, hence caution is required if generalising findings to the Australian population. Although this study aimed to explore Australian adults with suspected IP, whether or not participants have diagnosed IP is unknown. Therefore, these results are more relevant to those who suspect they have IP rather than those with a confirmed diagnosis. Self-reported data collection has the potential for recall bias. However, as this was the first survey to describe the health-seeking behaviours of Australian adults with suspected IP, this study does provide new and important information, thus advancing the research agenda on this topic.

4.2.9 CONCLUSION

The investigation of Australian adults with suspected IP has highlighted major inconsistencies between the healthcare provided and the healthcare this patient population would prefer to receive, especially regarding the diagnosis of IP. Most notably, the majority of participants experienced a considerable length of time between first suspecting IP and receiving a diagnosis of IP. The out-of-pocket expenditure associated with the assessment and management of IP suggests a financial burden for people with suspected IP. The results of this study provide novel patient-centred considerations that can be used to inform a clinical practice guideline for the assessment and management of IP as an important public health initiative.

4.3 CHAPTER SUMMARY

This cross-sectional survey was the first study to explore any aspect of patient-centred care in individuals with suspected IP. The results presented in this chapter provide a preliminary overview of the health-seeking behaviours of people with suspected IP. In response to the research question outlined in this thesis, most participants with suspected IP reported self-diagnosing their condition, with the majority of these participants preferring to be assessed using an accurate method by a general practitioner or naturopath. Furthermore, there is a considerable length of time between first suspecting IP and receiving a diagnosis of IP with Australian adults with suspected IP spending over 11 years between first suspecting IP and receiving a formal diagnosis. Our results also suggest the out-of-pocket expenditure associated with the management of IP were on average $699 for consultation fees, $2176 for dietary supplements, and $287 for the assessment of IP. Collectively, these findings provide valuable insight that can be used when developing treatment recommendations for the management of IP. Further investigation is needed to understand the treatment methods used by this population group.

5. QUALITY OF LIFE AND TREATMENT METHODS OF AUSTRALIAN ADULTS WITH SUSPECTED INCREASED INTESTINAL PERMEABILITY

The health-seeking behaviour towards the management of IP has been explored in the previous chapter. This project seeks to develop evidence-based treatment recommendations for the management of IP in clinical practice. However, to create recommendations that apply to people with IP, understanding their preferred treatment methods is required. Furthermore, identifying any treatment methods known to have a relationship with patient quality of life may provide direction for appropriate recommendations as part of the IP Guideline. To provide essential understanding for this research project, further analysis was undertaken to explore the management methods used and the association with subjective wellbeing and health-related quality of life. This chapter presents the methods undertaken to survey the target population and the results of this study.

5.1 PUBLICATION OF RESULTS

The results presented within this chapter have been published (see Appendix 5.1) as follows:

> Leech, B, McIntyre, E, Steel, A, Sibbritt, D (2019) “The Subjective Well-being and Health-Related Quality of Life of Australian Adults with Increased Intestinal Permeability and Associations with Treatment Interventions”, The Journal of Alternative and Complementary Medicine, 2021, Vol 27, 12.

5.2 THE SUBJECTIVE WELLBEING AND HEALTH-RELATED QUALITY OF LIFE OF AUSTRALIAN ADULTS WITH INCREASED INTESTINAL PERMEABILITY AND ASSOCIATIONS WITH TREATMENT INTERVENTIONS

5.2.1 INTRODUCTION AND BACKGROUND

The health of the gastrointestinal system has become a target of interest for disease prevention.[250] One specific gastrointestinal target area is the integrity of the intestinal barrier of the small intestine. During increased intestinal permeability (IP), the tight junction proteins between the cells of the small intestine disassemble in response to the protein zonulin.[2] The single layer of epithelium cells in the small intestine contributes to the biochemical and physical barrier to the array of foreign pathogens, allergens and other toxins.[251] The prevalence of altered IP has been suggested to be 10%-87% in health conditions with a known association.[1] During a loss of intestinal integrity, a cascade of reactions contributes to systemic symptoms and disease progression with the mitigation of zonulin suggested to inhibit or reduce disease onset.[221] Although no defined symptoms of IP have been identified,[221] a range of risk factors are known to be associated with altered IP.[221] The clinical risk factors associated with IP provide a potential platform for treatment interventions and areas for further investigation.

The management of altered IP may involve the use or avoidance of dietary products (e.g. increasing dietary fibre, avoidance of gluten and alcohol), lifestyle therapies (e.g. stress management, vagus nerve stimulation), dietary

supplements (e.g. vitamin A, probiotics, *Curcuma longa*, fish oil) and medication evaluation (e.g. avoidance of nonsteroidal anti-inflammatory drugs and antibiotics or the use of larazotide acetate).[233,252-254] These methods are proposed to have multiple direct and indirect modulatory actions that regulate intestinal integrity.[252,253] Many of the treatments used by practitioners for the management of IP have previously been shown to align with preclinical research.[253] Although these treatment methods are frequently used in clinical practice, there still remains limited evidence for the effective management of altered IP. A broad health services research-based study may help identify the potential areas for further clinical trials.[220]

The clinical relevance and consequence of altered IP in clinical practice have recently been questioned[131] despite identified associations between IP and a wide range of health conditions.[1] Questions regarding the clinical relevance and consequence of altered IP may stem from a low level of awareness and understanding regarding the potential effect of altered IP on individuals, especially their quality of life (QoL) and subjective wellbeing (SWB). QoL is an important contributor to overall disease burden alongside financial burden, mortality and morbidity.[255,256] Health-related quality of life (HRQoL) is a multi-dimensional concept that measures the impact of health status on QoL and includes mental, physical, emotional, and social functioning.[212] In addition to HRQoL, a person's SWB—also referred to as life satisfaction—can be a determinant in quantifying the clinical relevance and consequence of ill health. SWB is a multidimensional construct comprised of cognitive and affective components that reflect an individual's appraisal of their satisfaction with their

life.[208,209] Understanding the SWB of individuals with particular health conditions may help identify populations with greater mortality risk[257] and guide the development of targeted supportive interventions.

The impact of altered IP on individuals' HRQoL and SWB, and the treatments used in the management of IP, remains under-examined. As such, this study has two primary aims: to describe the SWB and HRQoL of Australian adults with suspected IP and explore the treatment methods used by this population group.

5.2.2 MATERIALS AND METHODS

5.2.2.1 STUDY DESIGN AND SETTING

A cross-sectional study design using an online self-reported survey was deployed. Approval for the study was obtained from the Human Research Ethics Committees (HREC) of the University of Technology Sydney (ETH19-4012). The health-seeking behaviour, views and preferences of this study cohort have previously been published.[220]

5.2.2.2 PARTICIPANTS AND RECRUITMENT

Participants were recruited via social media platforms and a purpose-built webpage, with snowball sampling methods used. The authors shared the survey on their social media and known Facebook groups such as Leaky Gut and Microbiome Support Group Australia. The survey was open for two months between September 2019 and November 2019. Eligibility questions asked participants whether they believe they have IP (self-diagnosed) or have been

diagnosed with IP. To participate in the study, participants were also required to be 18 years of age or older, living in Australia and have Internet access. The target population although broad, represents an under examined population group; as such, this study was designed to capture people with suspected IP or confirmed IP. As IP is suggested to be under diagnosed, including participants who self-diagnose IP best reflect the target population and the patients that present to clinical practice for the treatment of IP.[4] Survey responders with incomplete demographic characteristics, accounting for <5% of total data were excluded from analysis.

5.2.2.3 SURVEY AND DATA COLLECTION

The online survey administered through the online platform *SurveyGizmo* utilised questionnaire items previously developed to investigate IP in Australia.[4,253] The survey was pilot tested by four lay individuals to assess language clarity, with the required corrections made. The survey included four main domains: demographic characteristics, treatment methods for altered IP, SWB and HRQoL.

5.2.3 DEMOGRAPHIC CHARACTERISTICS

The participants were asked about their gender, age, height and weight from which body mass index (BMI) was calculated and categorised to underweight, healthy weight, overweight, and obese.[236] The participants were further asked about their country of birth, the state or territory where they reside and whether this was in an urban, rural or remote location. Participant's income manageability

was determined by how well they manage their household income, categorised as ‘difficult all the time’, ‘difficult some of the time’, ‘not too bad’ or ‘easy’.

5.2.4 SELF-REPORTED OUTCOME OF INCREASED INTESTINAL PERMEABILITY

Two questions were asked to explore the potential severity of IP. Firstly, participants were asked whether they believed their IP has become ‘better’, ‘worse’ or ‘no change’ over the previous 12 months. Participants were then asked how many days a week does their IP affects their daily living with the option of 0-7 days.

5.2.5 TREATMENT OF INCREASED INTESTINAL PERMEABILITY

A selection of survey items involving dietary products, lifestyle therapies, dietary supplements and medications that may either improve or exacerbate IP along with open-ended questions were used to document how frequently these methods were used. The frequency of use for dietary products, lifestyle therapies, dietary supplements and medications were measured using a six-point scale (‘never’, ‘less than once a month’, ‘1-3 times a month’, ‘once a week’, ‘2-6 times a week’, ‘every day’). These treatment methods were further explored in relation to the person who prescribed the treatment, mainly the qualification of the practitioner or whether the treatment was self-prescribed.

5.2.6 SUBJECTIVE WELLBEING

Participants' SWB was measured using the Personal Wellbeing Index - Adult (PWI-A) scale - an instrument validated in Australian population samples.[210] The PWI scale consisted of seven domains of satisfaction: *standard of living*, *personal health*, *achieving in life*, *personal relationships*, *personal safety*, *community-connectedness* and *future security*.[210] The PWI scoring system of each domain is reported on a 0-10 scale, with 0 representing no satisfaction at all and 10 being completely satisfied.

5.2.7 QUALITY OF LIFE

The 20-Item Short Form Health Survey (SF-20) was used to measure participants HRQoL.[211] The SF-20 assesses six health domains: physical functioning (6 questions), role functioning (2 questions), social functioning (1 question), mental health (5 questions), current health perceptions (5 questions), and bodily pain (1 question).

5.2.7.1 STATISTICAL ANALYSES

Data were exported to STATA® 16 for statistical analyses. Variables were reported as means, standard deviations, 95% confidence intervals (CIs) or frequencies and percentages, where appropriate. Chi-square analysis was used to examine the association between two categorical variables with Student's t-tests used for continuous variables across a binary variable. Analysis of variance (ANOVA) was used to measure the difference between a continuous variable across a categorical variable. Spearman's correlation analysis was used to

measure the correlation between the number of days IP affects daily living, SWB and HRQoL. Logistic and linear regression models were used when considering multiple factors. Variables associated with SWB, HRQoL, or the number of days IP affects daily living—with a bivariate p-value < 0.25[214]—were entered into the respective multivariate logistic or linear regression models, to adjust for potential confounders. A stepwise backward elimination process was then used to identify the most important independent predictors.

For analysis, participants use of dietary products, lifestyle therapies, dietary supplements and medications were grouped as frequently ('once a week', '2-6 times a week' and 'every day') and infrequently ('less than once a month', '1-3 times a month' and 'never'). Although participants were able to select either 'exacerbation', 'improvement' or 'no change' for the self-reported outcome of IP in the previous 12 months only data from exacerbation and improvement were used during analysis. Practitioners were categorised as 'medical practitioners' (integrative medicine practitioners, general practitioners, and gastroenterologists) and 'healthcare practitioners' (all practitioners).

Analysis and interpretation of the data collected from the PWI-A scale were undertaken according to previously published work.[258] Participants who answered consistently 0/10 or 10/10 across all PWI domains were excluded due to risk of response bias.[258] For analysis, the raw scores were transformed to a 0-100 scale. The combined mean score from the seven domains represents the participants overall SWB. A two-sample t-test was used to compare the normative mean of the surveyed sample and the Australian population.[259]

The analysis and interpretation of the SF-20 were undertaken according to previously published work.[211] For analysis, the SF-20 item scores were transformed to a scale of 0 to 100, with 0 representing the worst perceived health-related outcome. Item scores for each domain were combined and averaged to produce the final domain score (0-100). Higher scores reflect better perceived health-related outcomes, except for bodily pain where a higher score indicates more bodily pain.

5.2.8 RESULTS

5.2.8.1 DEMOGRAPHIC CHARACTERISTICS

There were 982 responses to the survey, of which 393 responses were excluded as the initial eligibility questions were not answered and thereby classified as not meeting the eligibility criteria; this left a total of 589 participants. Most participants were female (93%) with a mean age of 45.0 years (SD=12.1; range 18-82) and a mean body mass index (BMI) of 27.0 (SD=6.9). Participants' BMI were classified as healthy weight (46.1%), obese (26.9%), overweight (23.8%), and underweight (3.3%). Most participants were born in Australia (81.0%) and resided in New South Wales (29.7%), Queensland (27.3%), Victoria (17.5%), or Western Australia (10.7%); in an urban (70.6%), rural (27.3%) or remote area (2.0%). Most participants described their income manageability as 'easy or not too bad' (46.5%), followed by 'difficult some of the time' (32.3%) or 'difficult all the time' (21.2%). The major health concerns reported by participants were IP (n=300, 50.9%), autoimmune conditions (n=40, 6.8%), Hashimoto's thyroiditis (n=28,

4.8%), gastrointestinal issues (n=24, 4.1%), chronic fatigue syndrome (n=21, 3.6%) and rheumatoid arthritis (n=18, 3.1%).

5.2.8.2 PRACTITIONERS CONSULTED WITH, AND TREATMENTS USED, FOR MANAGING INCREASED INTESTINAL PERMEABILITY

Participants most frequently reported using dietary products (87.9%), dietary supplements (72.9%) and lifestyle therapies (54.6%) for the management of IP. Medications were infrequently used by participants for the treatment of IP (8.5%). Self-prescribing of treatment methods for the management of IP was most frequently reported (59.6%), followed by prescription from a naturopath (43.1%), integrative medicine practitioner (19.3%), general practitioner (16.8%), and nutritionist (12.4%) (Table 5.1). Dietary products (53.0%) and lifestyle therapies (33.8%) were both frequently self-prescribed. Whereas, dietary supplements and medications were most frequently prescribed by a naturopath (37.3%) and general practitioner (4.4%), respectively.

Table 5.1 Frequency of treatment methods used for increased intestinal permeability (n=483)

Who prescribed treatment	Treatment methods used for increased intestinal permeability[a]									
	Total		Dietary products		Lifestyle therapies		Dietary supplements		Medications	
	n	%	n	%	n	%	n	%	n	%
Self-prescribed	288	59.6	256	53.0	163	33.8	156	32.3	11	2.3
Naturopath	208	43.1	175	36.2	104	21.5	180	37.3	0	0.0
Integrative medicine practitioner	93	19.3	77	15.9	47	9.7	84	17.4	11	2.3
General practitioner	81	16.8	60	12.4	30	6.2	34	7.0	21	4.4
Nutritionist	60	12.4	57	11.8	28	5.8	47	9.7	0	0.0
Dietitian	37	7.7	34	7.0	12	2.5	15	3.1	0	0.0
Chinese medicine practitioner	28	5.8	17	3.5	12	2.5	24	5.0	0	0.0
Chiropractor	28	5.8	16	3.3	22	4.6	15	3.1	0	0.0
Acupuncturist	24	5.0	12	2.5	11	2.3	18	3.7	0	0.0
Herbalist	24	5.0	15	3.1	14	2.9	22	4.6	0	0.0
Gastroenterologist	20	4.1	13	2.7	5	1.0	8	1.7	3	0.6
Kinesiologist	20	4.1	14	2.9	12	2.5	12	2.5	0	0.0
Ayurvedic practitioner	12	2.5	10	2.1	5	1.0	9	1.9	0	0.0
Homeopath	12	2.5	8	1.7	7	1.5	9	1.9	0	0.0
Osteopath	7	1.5	6	1.2	5	1.0	5	1.0	0	0.0
Pharmacist	5	1.0	1	0.2	1	0.2	5	1.0	1	0.2
Nurse	4	0.8	4	0.8	2	0.4	2	0.4	2	0.4
Nurse practitioner	2	0.4	2	0.4	2	0.4	1	0.2	1	0.2

[a] Participants were able to select multiple treatment methods.

5.2.8.3 SELF-REPORTED OUTCOME OF INCREASED INTESTINAL PERMEABILITY

In the previous 12 months, more participants reported that their IP had improved (55.8%). Half of the participants (50.0%) reported IP affected their daily living seven days a week. Furthermore, participants who described an improvement in

their IP during the previous 12 months reported IP affected their daily life 4.0 days a week (95% CI: 3.6, 4.4); whereas, participants that described exacerbation of their IP in the previous 12 months reported IP affected their daily life 6.0 days a week (95% CI: 5.7, 6.3; p<0.001).

A self-reported improvement in IP was associated with participants who were treated by a practitioner compared to those who were not treated by a practitioner (76.1% vs. 23.9%; p<0.001). Participants who reported their IP had worsened in the previous 12 months had a significantly higher mean BMI compared to those who reported an improvement in their IP in the last 12 months (28.4 vs. 25.5; p<0.001). Multivariate logistic regression analysis found the use of nonsteroidal anti-inflammatory drugs (NSAIDs) (β= 1.08; 95% CI: 0.17, 1.98; p=0.021), lifestyle therapies (β= 1.08; 95% CI: 0.46, 1.70; p=0.001) and *Saccharomyces boulardii* (β= 1.56; 95% CI: 0.46, 2.67; p=0.006) were predictors of a greater number of days each week that IP was reported to affect daily living. Whereas, reporting an improvement of their IP in the previous 12 months (β= -1.78; 95% CI: -2.39, -1.17; p<0.001), and infrequently (β= -0.90; 95% CI: -1.64, -0.16; p=0.017) or frequently (β= -0.82; 95% CI: -1.49, -0.16; p=0.016) practicing yoga were found to be predictors for a fewer number of days affecting daily living each week.

5.2.8.4 TREATMENT RELATED CHARACTERISTICS OF INCREASED INTESTINAL PERMEABILITY IMPROVEMENT

Participants who reported an improvement in their IP in the previous 12 months were more likely to be treated by a healthcare practitioner (OR=2.04, p=0.015),

use dietary supplements (OR=2.66, p=0.003), participate in vigorous exercise (OR=2.99, p<0.001) and employ vagus nerve stimulation (OR=3.10, p=0.010) (Table 5.2). Furthermore, participants that reported an improvement in their IP during the previous 12 months were also less likely to consume gluten (OR=0.35, p<0.001) or use NSAIDs (OR=0.35, p=0.022).

Table 5.2 Treatment related characteristics and the improvement of increased intestinal permeability in the previous 12 months (n=287)

Characteristics	**Odds Ratio (95% CI)**	**p value**
Treating person		
Self	1.00	
Healthcare practitioner	2.04 (1.15 to 3.61)	0.015
Gluten		
Never	1.00	
Frequently	0.35 (0.20 to 0.61)	<0.001
Vigorous exercise		
Never	1.00	
Frequently	2.99 (1.61 to 5.53)	<0.001
Vagus nerve stimulation		
Never	1.00	
Frequently	3.10 (1.31 to 7.31)	0.010
Nonsteroidal anti-inflammatory drugs		
Never	1.00	
Infrequently	0.48 (0.26 to 0.86)	0.014
Frequently	0.35 (0.15 to 0.86)	0.022
Using dietary supplements		
No	1.00	
Yes	2.66 (1.40 to 5.05)	0.003

5.2.8.5 ASSOCIATIONS BETWEEN COMMON DIETARY PRODUCTS, LIFESTYLE THERAPIES, MEDICATIONS AND THE SELF-REPORTED OUTCOME OF INCREASED INTESTINAL PERMEABILITY

Participants who reported frequently consuming organic foods ($p<0.001$), fermented foods ($p=0.004$), bone broth ($p=0.001$), collagen ($p<0.001$), or apple cider vinegar ($p=0.026$) described an improvement in their IP in the previous 12 months compared to those who infrequently consumed these dietary products (Table 5.3). Furthermore, participants who reported infrequently consuming dairy products ($p=0.012$), refined sugar ($p<0.001$), or gluten-containing products ($p<0.001$) described an improvement in their IP in the previous 12 months compared to participants who reported frequently consuming these dietary products. Participants who reported frequently practising breathing exercises ($p<0.001$), stress management ($p<0.001$), meditation ($p=0.037$), vigorous exercise ($p<0.001$), yoga ($p=0.001$), or vagus nerve stimulation ($p<0.001$) more commonly described an improvement in their IP in the previous 12 months compared to participants who infrequently reported practising these lifestyle therapies. Participants who infrequently used NSAIDs ($p=0.001$) more commonly described an improvement in their IP in the previous 12 months compared to participants who frequently used NSAIDs.

Table 5.3 Associations between common dietary products, lifestyle therapies, medications and the self-reported outcome of intestinal permeability in the previous 12 months (n=483)

				Self-reported outcome of increased intestinal permeability in the previous 12 months		
		Total		Exacerbation	Improvement	
Dietary products		**n**	**%**	**%**	**%**	**p value**
Red meat	Frequently	357	74.2	44.6	55.4	0.909
	Infrequently	124	25.8	43.9	56.1	
Organic foods	Frequently	331	69.4	37.6	62.4	<0.001
	Infrequently	146	30.6	58.8	41.2	
Dairy	Frequently	279	57.9	50.6	49.4	0.012
	Infrequently	203	42.1	36.4	63.6	
Refined sugar	Frequently	239	49.8	59.6	40.4	<0.001
	Infrequently	241	50.2	30.8	69.2	
Fermented foods	Frequently	220	46.2	36.1	63.9	0.004
	Infrequently	256	53.8	52.3	47.7	
Gluten	Frequently	213	44.4	64.1	35.9	<0.001
	Infrequently	267	55.6	30.3	69.7	
Apple cider vinegar	Frequently	179	37.3	36.1	63.9	0.026
	Infrequently	301	62.7	49.0	51.0	
Bone broth	Frequently	173	36.1	32.5	67.5	0.001
	Infrequently	306	63.9	51.3	48.7	
Collagen	Frequently	166	35.0	31.1	68.9	<0.001
	Infrequently	308	65.0	51.3	48.7	
Alcohol	Frequently	148	30.9	47.4	52.6	0.472
	Infrequently	331	69.1	43.1	56.9	
Lifestyle therapies						
Breathing exercises	Frequently	212	45.5	34.4	65.6	<0.001
	Infrequently	254	54.5	55.3	44.7	
Stress management	Frequently	210	45.2	34.0	66.0	<0.001
	Infrequently	255	54.8	57.1	42.9	
Meditation	Frequently	191	40.8	38.7	61.3	0.037
	Infrequently	277	59.2	50.6	49.4	
Vigorous exercise	Frequently	146	30.9	28.4	71.6	<0.001
	Infrequently	327	69.1	51.4	48.6	
Yoga	Frequently	133	28.5	31.3	68.8	0.001

	Infrequently	333	71.5	50.7	49.3	
Vagus nerve stimulation	Frequently	61	13.3	20.5	79.6	<0.001
	Infrequently	399	86.7	48.8	51.2	
Medications						
Nonsteroidal anti-inflammatory drugs	Frequently	63	13.4	69.2	30.8	0.001
	Infrequently	407	86.6	40.2	59.8	
Prednisone	Frequently	16	3.5	41.7	58.3	0.827
	Infrequently	447	96.5	44.9	55.1	
Methotrexate	Frequently	11	2.4	57.1	42.9	0.704
	Infrequently	448	97.6	43.9	56.1	
Antibiotics	Frequently	6	1.3	60.0	40.0	0.657
	Infrequently	460	98.7	43.6	56.4	

5.2.8.6 FREQUENCY OF DIETARY SUPPLEMENTS USE FOR THE TREATMENT OF INCREASED INTESTINAL PERMEABILITY

The most frequently used dietary supplements for the management of IP were probiotics (36.1%), herbal mixtures (26.6%), prebiotics (21.7%), zinc (21.7%), glutamine (19.4%), magnesium (19.1%), and vitamin D (15.6%) (Table 5.4). Dietary supplements were most frequently used by participants who described an improvement in their IP during the previous 12 months compared to those who described exacerbation of their IP (63.3-86.8% vs 13.2-36.7%). Participants frequently reported using dietary supplements as prescribed by a practitioner rather than self-prescribed (66.7-87.8% vs 12.2-33.3%) (Table 5.4). There was a statistically significant association between the use of dietary supplements and the self-reported outcome of IP. Specifically, participants who used zinc (p=0.05), glutamine (p=0.02), magnesium (p=0.006), vitamin C (p=0.03), or vitamin B complex (p=0.001) described an improvement in their IP during the previous 12 months.

Table 5.4 Associations between dietary supplements and self-reported outcome of increased intestinal permeability in the previous 12 months and percentage for person prescribing each treatment (n=346)

Dietary supplements	Self-reported outcome of intestinal permeability in the previous 12 months					Person who prescribed treatment	
	Total		Exacerbation	Improvement		Self-prescribed	Practitioner prescribed
	n	%	%	%	P value	%	%
Probiotic	125	36.1	33.3	66.7	0.483	27.0	73.0
Herbal mixtures	92	26.6	28.4	71.6	0.111	26.4	73.6
Prebiotic	75	21.7	27.6	72.4	0.113	23.0	77.0
Zinc	75	21.7	25.4	74.6	0.05	20.0	80.0
Glutamine	67	19.4	22.5	77.6	0.02	25.8	74.2
Magnesium	66	19.1	19.2	80.9	0.006	29.2	70.8
Vitamin D	54	15.6	35.9	64.1	0.956	30.2	69.8
Vitamin C	50	14.5	18.8	81.3	0.03	20.4	79.6
Vitamin B complex	49	14.2	13.2	86.8	0.001	12.2	87.8
Omega 3	48	13.9	33.3	66.7	0.689	33.3	66.7
Curcuma longa	42	12.1	23.3	76.7	0.114	30.9	69.1
Slippery elm	41	11.9	28.6	71.4	0.366	17.1	82.9
Aloe vera	39	11.3	24.1	75.9	0.146	23.7	76.3
Digestive enzyme	37	10.7	36.7	63.3	0.963	13.5	86.5
Multivitamin	37	10.7	20.7	79.3	0.062	24.3	75.7
Amino acid mix	31	9.0	32.0	68.0	0.637	29.0	71.0
Saccharomyces boulardii	21	6.1	18.8	81.3	0.131	14.3	85.7
Vitamin A	19	5.5	33.3	66.7	0.806	26.3	73.7

5.2.8.7 SUBJECTIVE WELLBEING AND HEALTH-RELATED QUALITY OF LIFE

There was a statistically significant difference in overall SWB and each domain of SWB between Australian adults with suspected IP and the Australian population ($p<0.001$). Specifically, Australian adults with suspected IP had lower (i.e. worse) average scores for all domains compared to the Australian population. A t-test showed that participants who described exacerbation of their IP had a worse ($M=54.7$, $SD=20.3$) SWB than those reporting an improvement ($M=66.1$, $SD=19.6$) in their IP ($p<0.001$). Spearman's correlation analysis revealed that the number of days IP affects daily life had a negative correlation with SWB and HRQoL ($p<0.001$). Results for correlation analyses are summarised in Table 5.5.

Table 5.5 Spearman's correlation between quality of life and subjective wellbeing with the number of days increased intestinal permeability affects daily life each week (0-7 days).

	n	Mean	SD	Correlation coefficient	p value
Subjective wellbeing					
Personal wellbeing index	422	60.3	20.3	-0.402	<0.001
Standard of living	422	65.0	25.5	-0.313	<0.001
Health	422	43.4	24.6	-0.453	<0.001
Achieving in life	422	56.1	25.6	-0.377	<0.001
Personal relationship	422	64.2	26.3	-0.261	<0.001
Personal safety	422	75.3	24.3	-0.193	<0.001
Community connectedness	422	59.3	27.2	-0.277	<0.001
Future security	422	58.8	27.9	-0.273	<0.001
Quality of life					
Physical functioning	423	61.9	33.8	-0.275	<0.001
Role functioning	423	57.3	42.5	-0.335	<0.001
Social functioning	423	60.5	32.3	-0.388	<0.001
Mental health	423	55.0	21.6	-0.294	<0.001
Health perception	423	37.2	28.5	-0.474	<0.001
Bodily pain	423	50.4	25.1	0.316	<0.001

Note: Score ranges from 0-100. A high score indicates better health except for pain, where a high score indicates more pain.

5.2.8.8 SUBJECTIVE WELLBEING AND COMMON DIETARY PRODUCTS, LIFESTYLE THERAPIES AND MEDICATIONS

Pairwise comparison found a statistically significant difference between the overall SWB of participants, and the frequency of common dietary products, lifestyle therapies and medications use. Participants who used any form of dietary products (M=61.0, SD=20.4) for the treatment of IP were found to have better SWB compared to those that never used dietary products (M=54.6, SD=18.7) (p=0.023). Furthermore, participants who never consumed gluten-containing foods (M=65.2, SD=21.5) were found to have better SWB compared to participants that frequently consumed gluten (M=59.1, SD=19.8) (p=0.037). Whereas, participants who frequently consumed alcohol (M=64.9, SD=18.7)

were found to have better SWB compared to those who never consumed alcohol (M=54.0, SD=22.4) (p<0.001). Furthermore, participants who frequently practiced breathing exercises (M=63.0, SD=19.4; M=56.2, SD=21.5) (p=0.014), stress management (M=62.4, SD=19.0; M=56.1, SD=23.7) (p=0.036), vigorous exercise (M=66.2, SD=18.4; M=55.3, SD=20.9) (p<0.001), or yoga (M=68.0, SD=17.0; M=56.0, SD=20.9) (p<0.001) were found to have better SWB compared to participants that never participated in these lifestyle therapies. Lastly, participants that never used NSAIDs (M=62.5, SD=21.5) were found to have better SWB compared to those that frequently used them (M=54.3, SD=19.5) (p=0.026).

5.2.8.9 MULTIPLE REGRESSION PREDICTING SWB AND HRQOL

Seven regression models predicting overall SWB, and each HRQoL domain were undertaken. The results of these regression models found the outcome of IP in the previous 12 months, BMI, the treating person and the use of dietary products and lifestyle therapies were all statistically significant predictors of overall SWB and each HRQoL domain (Table 5.6). Specifically, improvement of IP (β= 10.70, p<0.001) and using dietary products (β= 12.12, p=0.008) were predictors of better SWB whereas being obese (β= -5.70, p=0.035), treated by a medical practitioner (β= -6.35, p=0.016) and using lifestyle therapies (β= -6.30, p=0.010) were predictors of worse SWB. Regarding HRQoL, all domains except physical functioning saw improvement in IP as a statistically significant predictor for higher HRQoL (Table 5.6).

Table 5.6 Multiple regression predicting subjective wellbeing and health-related quality of life

		Health-related quality of life					
	Subjective wellbeing (n=301) β (95% CI) p value	**Physical functioning (n=417) β (95% CI) p value**	**Role functioning (n=306) β (95% CI) p value**	**Social functioning (n=306) β (95% CI) p value**	**Mental health (n=304) β (95% CI) p value**	**Health perception (n=302) β (95% CI) p value**	**Bodily pain (n=304) β (95% CI) p value**
Improvement of IP in previous 12 months	10.70 (6.01, 15.39) <0.001		21.06 (11.60, 30.51) <0.001	18.83 (11.72, 25.94) <0.001	10.57 (5.56, 15.58) <0.001	21.88 (15.76, 27.99) <0.001	-11.74 (-17.53, -5.94) <0.001
Using dietary products	12.12 (3.21, 21.03) 0.008				15.79 (6.24, 25.33) 0.001		
Using lifestyle therapies	-6.30 (-11.05, -1.54) 0.010		-14.97 (-24.59, -5.35) 0.002	-9.30 (-16.53, -2.07) 0.012	-7.30 (-12.36, -2.23) 0.005	-7.45 (-13.58, -1.33) 0.017	5.86 (0.07, 11.64) 0.047
BMI							
Normal weight	1.00	1.00			1.00	1.00	1.00
Obese	-5.70 (-10.99, -0.41) 0.035	-15.51 (-22.59, -8.43) <0.001			-5.91 (-11.58, -0.24) 0.041	-12.89 (-19.88, -5.91) <0.001	12.69 (6.05, 19.33) <0.001
Treating person							
Self	1.00	1.00				1.00	
Medical practitioner	-6.35 (-11.52, -1.18), 0.016	-13.06 (-20.46, -5.66) 0.001				-9.76 (-16.57, -2.95) 0.005	

5.2.9 DISCUSSION

This study is the first to explore the HRQoL and SWB of Australian adults with suspected IP. Our results suggest that altered IP may pose a greater health burden than previously thought, providing the first indication that Australian adults with altered IP are susceptible to poor SWB and HRQoL. Furthermore, several participant characteristics were found to be associated with the improvement or exacerbation of IP (Figure 5.1).

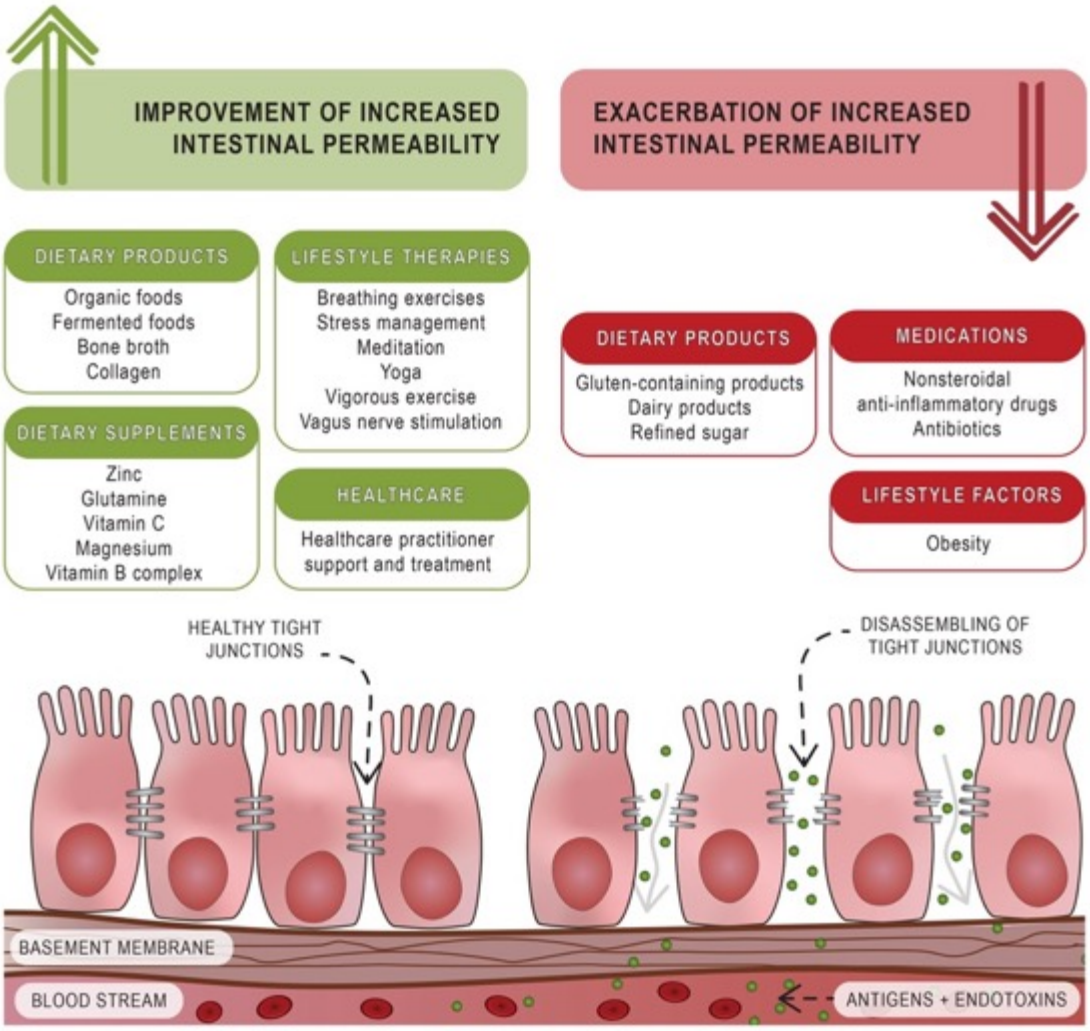

Figure 5.1 Participant's characteristics found to be associated with the improvement and exacerbation of increased intestinal permeability.

5.2.9.1 INCREASED INTESTINAL PERMEABILITY AND SWB AND HRQOL

Our results suggest that Australian adults with suspected IP have a lower SWB compared to the Australian population. Furthermore, improvement in IP is suggested to be a significant predictor of SWB and HRQoL. These results provide the first indication that a relationship between both SWB and HRQoL and altered IP exists in a diverse range of health conditions. In support of this relationship, Australian adults with gastrointestinal disorders (many of which are associated with altered IP),[1] have been found to have a lower HRQoL compared to Australian adults without gastrointestinal disorders.[246] Furthermore, a lower QoL has been reported in diarrhoea-predominant irritable bowel syndrome patients with IP compared to those with a normal intestinal integrity.[260] The association between altered IP and both SWB and HRQoL contributes to a much needed clinical understanding of altered IP, especially as the consequence and clinical relevance of altered IP in clinical practice have recently been questioned.[131] Furthermore, the correlation found between both SWB and HRQoL and the number of days IP affects daily living suggests the previously reported symptoms[4] and biomarkers[221] of altered IP are not the only clinical consequence of altered IP with both SWB and HRQoL now suggested to be involved.

5.2.9.2 HEALTHCARE AND INCREASED INTESTINAL PERMEABILITY

The care provided by a healthcare practitioners compared to self-care differs not only in the treatments used but the reported outcomes. First, this study identified a high prevalence of self-prescription of treatment interventions, primarily dietary

products, and lifestyle therapies, for the management of IP. Dietary supplements and medication were most frequently prescribed by a healthcare practitioner. These findings coincide with existing research that suggests complementary and integrative medicine practitioners frequently use dietary supplements while also using a multimodal and personalised approach for the management of altered IP.[253] Working alongside a healthcare practitioner has also been suggested to provide greater health outcomes compared with no clinic-based support.[261-263] This may explain why in this study Australian adults that report an improvement in their IP are two times more likely to be treated by a healthcare practitioner. Second, our study found only 24% of self-treated participants reported an improvement in their IP compared to 76% of practitioner treated participants. These findings suggest the care provided by healthcare practitioners to Australian adults with suspected IP may have beneficial effects on the outcomes of altered IP. Furthermore, healthcare practitioners, especially those with limited experience in the management of altered IP may draw upon the findings of this study to gain a deeper understanding as to the treatment methods used by Australian adults with IP.

5.2.9.3 FEATURES ASSOCIATED WITH INCREASED INTESTINAL PERMEABILITY IMPROVEMENT

Participants who reported an improvement in their IP were 35% less likely to consume gluten or use NSAIDs. Our results also found that participants who indicated they avoided consuming gluten-containing foods and never used NSAIDs were associated with a better SWB. These results concur with clinical studies that show the consumption of gluten-containing products and the use of

NSAIDs induce IP.[264,265] Practitioners that treat IP also advocate for their patients with IP to avoid gluten and NSAIDs.[253]

The finding that vitamin C and vigorous exercise is associated with improvement of IP conflicts with existing research. First, preliminary research suggests that 500 mg of vitamin C (ascorbic acid) may induce a rearrangement of the actin cytoskeleton and thereby an exacerbation of IP.[264,266] Potentially, the association between vitamin C intake and improvement of IP may be the result of the frequent use in dietary supplements, especially as participants who reported an improvement in their IP were 2.7 times more likely to use dietary supplements. Research has demonstrated a causative link between vigorous exercise and altered IP.[47] As a result of redistribution of blood flow and splanchnic hypoperfusion during vigorous exercise damage to mucosal and epithelial cells may occur, thereby pathing the path for exacerbation of IP.[47] The improvement associated with vigorous exercise in our study may be the result of improved health; for example, as health and wellbeing improve so does the ability to participate in exercise.[267] Further large-scale trials and epidemiological research is needed to confirm both of these hypotheses.

5.2.9.4 RESEARCH AGENDA

Our study provides useful information where further research can draw upon the findings to inform clinical trials and clinical practice guidelines. The identified characteristics found to be associated with the improvement and exacerbation of IP warrant further investigation (Figure 5.1). Many of these associated features are yet to be investigated for their effect on IP, with clinical research focusing

primarily on dietary supplements and dietary products for the treatment of IP. Yet, there has been limited investigation exploring the effectiveness of lifestyle therapies in the management of IP.[253] Nevertheless, many of these lifestyle therapies are reported to have beneficial health outcomes in health conditions with a known association with altered IP.[268,269] These results provide a foundation for future clinical trials where a study exclusively conducted in primary care ensuring a homogenous study population and standardised diagnostic criteria may confirm the results of this study.

The findings from this study may also help to inform the development of a clinical practice guideline for the management of altered IP. By understanding the treatment methods used the development of recommendations can incorporate the views and preferences of Australian adults with suspected IP to enable relevant and appropriate recommendations for this patient group.

5.2.10 LIMITATIONS

Although this study involved participants with self-reported suspected IP, whether there was a confirmed diagnosis of IP is unknown. However, previous research has shown that people with self-reported IBS have similar health care utilization and QoL as those with diagnosed IBS.[270] Many of the health conditions participants report experiencing are known to be more prevalent in females and are suggested to be associated with IP, which may explain why 93% of participants were female.[1] Therefore, these results are considered relevant to females who suspect they have IP rather than Australian adults with a confirmed diagnosis of altered IP. The self-reported outcome of IP has the potential for recall

bias and may not reflect improvement or exacerbation of IP. Therefore, to confirm the relationship between both SWB and HRQoL and altered IP, a clinical study that measures IP and evaluates both SWB and HRQoL is required. However, this study provides important and novel information, advancing the research agenda on the clinical consequence of altered IP and suggest potential treatment strategies worth investigating.

5.2.11 CONCLUSION

The integrity of the small intestine may pose a greater health burden than previously thought, with susceptibility to poor SWB and HRQoL reported in Australian adults with altered IP. Our results strengthen the clinical relevance and consequence of altered IP, providing the first indication that a relationship between both SWB and HRQoL and altered IP exists. Clinical trials may use these findings to further explore the potential use of the treatment interventions used by Australian adults with suspected IP.

5.3 CHAPTER SUMMARY

The results presented in this chapter provide a preliminary overview of the preferred treatment methods people with IP want to use and the impact they may have on quality of life. In response to the research question outlined in this thesis, most participants with suspected IP reported using dietary products, dietary supplements and lifestyle therapies for managing IP. Furthermore, the use of some products and participating in certain activities were identified by participants as contributing to a change in IP status. The results also suggest IP may pose a greater health burden than previously thought, with poor SWB and HRQoL reported in Australian adults with self-reported IP. Collectively, these findings provide the core foundations needed to develop a clinical practice guideline for the management of IP that incorporates the views and preferences of Australian adults with suspected IP.

6. CLINICAL PRACTICE GUIDELINE FOR THE MANAGEMENT OF INCREASED INTESTINAL PERMEABILITY: TECHNICAL REPORT

In the preceding two chapters, the health-seeking behaviour towards the management of IP and the association with subjective wellbeing and health-related quality of life were explored. As this thesis seeks to develop evidence-based treatment recommendations for the management of IP in clinical practice, the results from Chapter 4 and 5 inform the work presented in the following chapter to provide novel patient-centred considerations that can be used to inform a clinical practice guideline for the management of IP as an important public health initiative. This chapter provides a comprehensive summary of the evidence used in the IP Guideline. The chapter also presents the complete risk of bias assessment and data extraction for each study used in the IP Guideline.

6.1 IP GUIDELINE: TECHNICAL REPORT

The following section contains the *Technical Report*, a document that forms part of the IP Guideline. The *Technical Report* has been formatted based on the NHMRC *Guidelines for Guidelines* Handbook to meet the 2016 *NHMRC Standards for Guidelines.*[215] Details are as followed:

Leech, B, McIntyre, E, Steel, A, Sibbritt, D (2022) “Clinical practice guideline for the management of increased intestinal permeability: Technical Report”, University of Technology Sydney.

6.1.1 INTRODUCTION

The need to develop a *clinical practice guideline for the management of increased intestinal permeability* (IP Guideline) was identified after health services research revealed gaps in both the published literature and clinical practice.[4,5] To date, no clinical practice guideline has been developed in Australia or internationally that addresses any area of IP management. This *Technical Report* details the information required by the NHMRC in accordance with the requirements of the NHMRC Standards for Clinical Practice Guidelines 2011.[215]

6.1.2 GUIDELINE PURPOSE

The purpose of the IP Guideline is to utilise the best available evidence while considering the views and preferences from a multidisciplinary group of stakeholders and consumers. The IP Guideline aims to provide clinicians and consumers with a transparent evidence-based guidance for the management of altered IP to optimise patient care, improve health outcomes and reduce variation in care for Australian practitioners in private practice.

6.1.3 CLINICAL QUESTION LIST

The Working Group developed the following clinical questions while considering the clinical importance, the views, preferences and experiences of both consumers and clinicians.[4,5,219,220]

CQ.1). In Australian adults with increased intestinal permeability, what are the benefits of dietary choices for the treatment of increased IP?

CQ.2). In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for dietary choices?

CQ.3). In Australian adults with increased intestinal permeability, what are the benefits of oral probiotic, prebiotic or synbiotic supplementation for the treatment of increased intestinal permeability?

CQ.4). In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for oral probiotic, prebiotic or synbiotic supplementation use?

CQ.5). In Australian adults with increased intestinal permeability, what are the benefits of oral amino acid supplementation for the treatment of increased intestinal permeability?

CQ.6). In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for oral amino acid supplementation use?

CQ.7). In Australian adults with increased intestinal permeability, what are the benefits of oral plant-based medicine supplementation for the treatment of increased intestinal permeability?

CQ.8). In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for oral plant-based medicines use?

CQ.9).In Australian adults with increased intestinal permeability, what are the benefits of oral essential fatty acid supplementation for the treatment of increased intestinal permeability?

CQ.10). In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for oral essential fatty acid supplementation use?

CQ.11). In Australian adults with increased intestinal permeability, what are the benefits of oral mineral supplementation for the treatment of increased intestinal permeability?

CQ.12). In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for oral mineral supplementation use?

CQ.13). In Australian adults with increased intestinal permeability, what are the benefits of oral vitamin supplementation for the treatment of increased intestinal permeability?

CQ.14). In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for oral vitamin supplementation use?

CQ.15). In Australian adults with increased intestinal permeability, what are the benefits of oral colostrum supplementation for the treatment of increased intestinal permeability?

CQ.16). In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for oral colostrum use?

6.1.4 SEARCH METHODS

A single literature search was carried out by one researcher (BL) to identify the relevant articles for each of the clinical questions. All related articles were

identified and grouped according to clinical questions. Targeted searching was carried out when needed during the write up of the guideline.

6.1.5 SEARCH STRATEGY

The databases PubMed, Embase, CINAHL, and Scopus were searched for articles published between January 2000 up until July 2020 by the Working Group. A single-arm search strategy was used: 'intestinal permeability' OR 'intestinal integrity' OR 'intestinal barrier dysfunction' OR 'intestinal epithelial barrier dysfunction' OR 'gastrointestinal permeability' OR 'gut permeability' OR 'gut barrier' OR 'zonulin' OR 'dual sugar' OR 'lactulose AND mannitol' OR 'lactulose AND rhamnose' OR 'cellobiose AND mannitol' OR 'Intestinal fatty acid binding protein'. The human filter was applied to the search. A hand search of the reference list from the included articles and web search for any recently published articles was also be carried out.

6.1.6 INCLUSION AND EXCLUSION CRITERIA

Included articles were original research exploring topics relevant to the clinical questions published between January 2000 and July 2020. The primary focus of the included articles must be on adults; however, systematic reviews were not excluded if articles included involved participants under 18 years of age. Including young adults has been suggested as a method to improve the search strategy for IP.[221] Therefore, at least 80% of the enrolled study population must be over 18 years of age. Articles were excluded if the primary focus was on critically ill patients (i.e., in intensive care or palliative care) or includes patients with HIV,

acute appendicitis, receiving chemotherapy, undergoing dialysis or abdominal surgery as the IP Guideline is focused on private practice in the community. Articles were excluded if the primary focus is on genetic testing, polymorphism research or involve the treatment of exercise induced IP. Articles were included if participants had IP assessed using the dual sugar test, stool zonulin, serum zonulin, serum LPS or serum endotoxin. When inaccurate testing method are used, these articles were excluded. Examples of inaccurate testing methods includes the dual sugar urinary test where the collection is over 6 hours or measured in the serum. Furthermore, in studies assessing the effectiveness of an intervention for IP management where clear evidence suggest that the patients do not have IP, these studies were excluded. Case studies and case series were excluded. There was no exclusion based on geographical location. Only articles published in English were included.

6.1.7 RISK OF BIAS ASSESSMENT

All included articles were assessed for risk of bias by one researcher (BL) using the most appropriate tool identified by the Working Group. A full description of the risk of bias tools are detailed in the *Guideline Development Process*. Briefly, four different tools were used depending on study design: Risk Of Bias In Systematic reviews (ROBIS) assesses the risk of bias in systematic reviews and covers research questions relevant to interventions, diagnosis, prognosis and aetiology.[223] The Cochrane tool for assessing risk of bias in randomised trials (Cochrane RoB 2.0) was used for assessing randomised trials for risk of bias.[224] The Risk Of Bias In Non-randomised Studies—of Interventions (ROBINS-I) was

used to evaluate the risk of bias in intervention studies that are not randomised.[225] Risk of bias of observational studies were used to assess prevalence studies.[147]

6.1.8 SYSTEMATIC SEARCH RESULTS

A total of 18,011 articles were identified through the database searches, of which 7850 duplicates were removed. After title and abstract screening, 385 potentially relevant full-text articles were reviewed, of which 60 articles met the inclusion criteria. Hand searching the literature found an additional one article. A total of 61 articles were included (Figure 6.1).

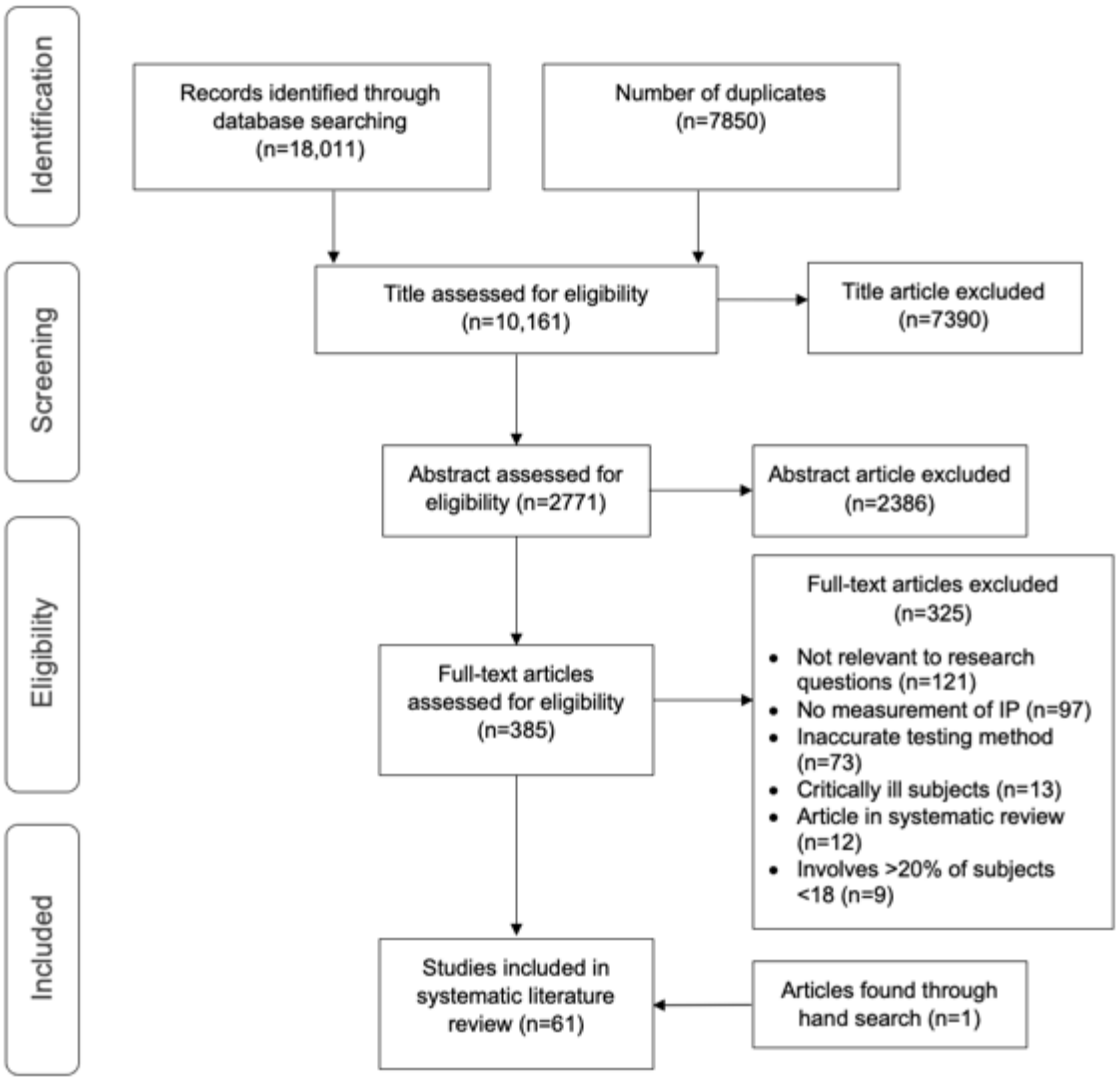

Figure 6.1 PRISMA flow diagram of study selection. Starting with 18,011 identified citations, 61 articles were included

6.1.9 FORMULATION OF RECOMMENDATIONS

6.1.9.1 DIETARY CHOICES

6.1.9.1.1 CLINICAL QUESTIONS

Clinical Question 1: In Australian adults with increased intestinal permeability, what are the benefits of dietary choices for the treatment of increased intestinal permeability?

Clinical Question 2: In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for dietary choices?

6.1.9.1.2 EVIDENCE SUMMARY AND OVERALL QUALITY OF EVIDENCE

6.1.9.1.2.1 ALCOHOL

There is limited (*n* = 4) low-quality evidence that alcohol consumption causes or contributes to the worsening of IP (Table 6.1 and Table 6.2).[221,271-273] There were no randomised controlled trials (RCT) on the impact of alcohol consumption on IP. One systematic review of observational studies evaluated the effect of alcohol on IP.[221] All included studies assessed short-term alcohol consumption on IP (0.5h – 1 week). Two studies saw mixed results, where 2 ml of vodka (40% ethanol)/kg body weight was given at one timepoint and assessed over 24h.[271,272] The consumption of <14 standard drinks per week of alcohol (OR=1.91; 95% CI: 1.01, 3.95; p=0.05) and above >15 standard drinks per week of alcohol (OR=1.56; 95% CI: 1.02, 2.67; p=0.05) were found to be associated with IP. Furthermore, two out of the four included studies were conducted in healthy adults.[271,272] The other studies assessed the effects of alcohol on individuals with inflammatory bowel disease (IBD) and moderate-to-severe fatty liver.[221,273] These studies suggest alcohol consumption may contribute to IP in people with Crohn's disease and fatty liver and not patients with ulcerative colitis. The impact of limiting or excluding alcohol consumption in the management of patients with IP remains unclear with a possible disease-specific effect.

6.1.9.1.2.2DIETARY FIBRE

There is (*n* = 5) moderate-quality evidence that dietary fibre consumption supports healthy intestinal integrity and improves IP.[199,274-277] No systematic reviews evaluating the effects of dietary fibre on IP were identified during the literature search. Three of the five identified articles were RCT,[274-276] with the other two studies[199,277] examining the impact of dietary fibre and IP. The included studies assessed the effects of dietary fibre for between one and six months. The type of dietary fibre used, the amount consumed, and the inclusion of gluten-containing products may influence the results. A diverse range of insoluble and soluble dietary fibre with prebiotic properties appears to benefit intestinal integrity.[199,275,277] The fortification of wheat-based products with prebiotics provides mixed results.[275,276] Two studies assessed prebiotic (inulin or beta-glucan) fortification with either wheat-based pasta or cake.[275,276] Inulin (11%) fortification was found to provide beneficial results,[275] whereas beta-glucan fortification was not.[276]

6.1.9.1.2.3MACRONUTRIENT RATIO AND ENERGY INTAKE

A total of 11 articles were included, with many being assessed as low-quality evidence.[277-286] Only one systematic review of observational studies evaluating the effect of energy intake and macronutrient distribution on intestinal integrity was found.[221] There were two RCT,[278,280] two cross-over clinical trials[282,283] and six non-randomised clinical trials included.[277,279,281,284-286] These articles assessed the effect of acute (<3 h) modified macronutrient distribution (*n* = 2),[281,283] and a combination of restricted energy intake (*n* = 2),[277,282] overfeeding

(n = 2)[280,286] and modified macronutrient distribution range (n = 6)[278-280,282,284,286] over a short-term study period (5 days – 3 months).

Increased energy intake (>10,945 kJ) per day (β=121.8; p=0.04) was found to be an independent risk factor for IP.[221] Kilojoule restriction (3,350kJ/day) in obese females over four weeks saw a significant decrease in serum zonulin compared to baseline, with zonulin levels returning to baseline values after two weeks on a balanced diet (7,500kJ/day).[285] Furthermore, a high fibre kilojoule restricted diet (4180-6700kJ/day) in overweight and obese adults saw a significant improvement in IP after nine and 23 weeks.[277] However, short-term overfeeding (116% of estimated energy requirements) with 25% estimated energy requirements from either fructose-sweetened beverages, high-fructose corn syrup-sweetened beverages or glucose-sweetened beverages lacked consistent results across IP markers.[280] Furthermore, a seven-day non-randomised clinical trial assessed the effects of overfeeding (estimated energy requirements plus 4,180kJ/day) in healthy men; results found no significant difference in intestinal integrity compared to baseline.[286]

Considering the literature on macronutrient distribution range, total fat percentage (β=0.23±0.11; p<0.05) was an independent risk factor for IP.[221] However, a high-fat diet saw mixed results, with the possibility of a detrimental effect on intestinal integrity. Two studies[281,283] assessed the effects of acute (<3 h) fat consumption. A slightly high fat breakfast (39%) saw no impact on serum zonulin in healthy adults,[283] while the consumption of 50g of fat (30% saturated) in morbid obesity adults found a significant increase in serum zonulin.[281] A further four studies

investigated the effects of a high-fat diet on IP.[278,279,284,286] Three of these studies found a high-fat diet consisting of 41-55% of estimated energy requirements from fats has no significant effect on lactulose/mannitol ratio and serum zonulin after 5-15 days.[278,279,286] However, a 12-week non-randomised clinical trial found that after a slightly high-fat diet (35% of estimated energy requirements from fats) serum and stool zonulin were significantly elevated compared to baseline.[284] Lastly, a significant increase in endotoxins was seen at five days after consuming a high-fat diet (55% of estimated energy requirements from fats).[279]

Mixed results were also seen in two studies investigating the effects of increased simple carbohydrate intake on intestinal integrity while remaining in normal macronutrient distribution range.[280,282] Both studies involved participants consuming 25% of their estimated energy requirements from fructose or glucose with one study also using high-fructose corn syrup.[280] Results showed the greatest impact on intestinal integrity was the consequence of fructose consumption rather than other simple carbohydrates. However, this was only seen with lactulose/mannitol ratio and endotoxin levels and not with serum zonulin.[280,282]

6.1.9.1.2.4 GLUTEN-FREE DIET

There is limited (*n* = 4) low to moderate-quality evidence that patients with confirmed IP should follow a gluten-free diet during IP treatment;[287-290] two RCTs,[287,288] one non-randomised clinical trial,[289] one case-control trial[290] and no systematic reviews. The included studies assessed the effects of dietary gluten on intestinal integrity. A gluten-containing diet for four weeks in individuals with positive and negative HLA-DQ2/8 genes and diarrhoea-predominant irritable

bowel syndrome found a significant increase in IP, especially patients with positive HLA-DQ2/8 compared to a gluten-free diet.[288] However, individuals with irritable bowel syndrome reported improvement in symptoms after the avoidance of gluten but found that the consumption of 16g of gluten daily for six weeks had no significant impact on IP compared to control or baseline.[287] Two of the studies[289,290] involved individuals diagnosed with coeliac disease, and therefore, results may not be transferable across other health conditions. These two studies demonstrated a gluten-free diet significantly decreased IP after two months with continued benefits to intestinal integrity after two years.[289,290]

Table 6.1 Evidence summary table for review articles

Author, Year	Study design; Level of Evidence (NHMRC)	Review aim	Type of evidence included	Year studies published	Main results from relevant studies	Authors' conclusions
Leech, 2019[221]	Systematic review; I	Summarise the known risk factors associated with IP	*n* = 5 Observational studies	2008-2018	Increased energy intake (>10,945 kilojoule) per day (β=121.8; p=0.04) and total fat percentage (β=0.23±0.11; p<0.05) were both reported to be independent risk factors for IP. Protein intake was found to be an independent risk factor (β=-0.139; 95% CI: -0.247, -0.031; p=0.01) for IP with a positive correlation reported for animal-deriver protein and IP (ρ=0.59; p=0.001). The consumption of <14 standard drinks per week of alcohol (OR=1.91; 95% CI: 1.01, 3.95; p=0.05) and above >15 standard drinks per week of alcohol (OR=1.56; 95% CI: 1.02, 2.67; p=0.05) were found to be associated with IP.	A high energy, nutrient-depleted diet with either inadequate protein intake or excess animal-derived protein in combination and excessive alcohol consumption are potential risk factors for IP. The consumption of a Western-style diet may increase the risk of IP.

Abbreviations: IP, increased intestinal permeability.

Table 6.2 Evidence summary table for clinical trials

Author, Year	Study design; Level of Evidence NHMRC	Country; Setting	Sample size; age in years M ± SD; gender; health condition	Intervention; control	Outcome measures; duration	Main results
Alcohol						
Bala, 2014[271]	Non-randomised, control clinical trial; III-2	USA; university research centre	*n* = 25; age range 21-56; 56% female; healthy adults	Intervention: 2 ml of vodka (40% ethanol)/kg body weight in total volume of 300 ml orange or strawberry juice Control: 300 ml of orange juice	Serum endotoxin at 0.5h, 1h, 1.5h, 2h, 2.5h, 3h, 3.5h, 4h and 24h	Binge drinking in health individuals' significantly increases endotoxin levels within 30 minutes to 3 hours after alcohol consumption ($p<0.05$). Females were found to have a higher level of endotoxin compared to men with a significant difference reported at 4 hours ($p<0.05$)
Stadlbauer, 2019[272]	Cohort study; III-2	Austria; university research centre	*n* = 15; 26±4; 27% female; healthy adults	Intervention: 2 ml of vodka (40% ethanol)/kg body weight in total volume of 300 ml orange or strawberry juice	Serum zonulin and endotoxin at 1h, 2h, 3h and 4h	Serum zonulin and endotoxin: n.s between timepoints

					Faecal zonulin and LMR at 24h	Faecal zonulin and LMR: n.s compared to baseline
Swanson, 2011[273]	Cohort study; III-2	USA; outpatient clinic	*n* = 23 (*n* = 21 analysed) Intervention: *n* = 8; median years 45; 50% female; ulcerative colitis; *n* = 6; median years 31; 34% female; Crohn's disease Control: *n* = 7; median years 24; 57% female; healthy adults	Intervention: 0.4 g of alcohol (red wine)/kg of body weight daily, 1 week Control: No intervention	LMR at 1 week	LMR: significantly increased in Crohn's disease compared to baseline (p=0.028) however, n.s in ulcerative colitis and healthy control compared to baseline.
Dietary fibre						
Machado, 2020[274]	Randomised placebo-controlled, double-blinded trial; II	Brazil; university research centre	*n* = 26 (*N* = 24 analysed) Intervention: *n* = 13; 31.3±8.8; 58% female; overweight or obese adults (BMI 31.3±8.5 kg/m^2) Control: *n* = 13; 31.3±8.8; 58% female; overweight or obese	Intervention: breakfast drink containing yacon flour (25g) plus an energy restricted diet (-500 kcal/day) consisting of carbohydrate 51%; fat 28%; protein 21% for 6 weeks Control: breakfast drink plus an energy restricted diet (-500 kcal/day) consisting of	LMR at 6 weeks	LMR: n.s between intervention and control or compared to baseline at 6 weeks (p>0.05)

			adults (BMI 31.3±8.5 kg/m^2)	carbohydrate 51%; fat 28%; protein 21% for 6 weeks		
Russo, 2012[275]	Randomised controlled, double-blinded crossover trial; III-1	Italy; university research centre	*n* = 20; 18.8±0.7; 100% male; healthy adults	Intervention: 100g of pasta containing wheat and fortified with inulin (11%), daily for 5 weeks Control: 100g of pasta containing wheat daily for 5 weeks	LMR at 5 weeks Serum and faecal zonulin at 5 weeks	LMR: significant decrease in fortified inulin pasta compared to control pasta group (p=0.001) Serum zonulin: significant decrease in fortified inulin pasta compared to control pasta group (p=0.013) Faecal zonulin: n.s between intervention and control (p>0.05)
Skouroliakou, 2016[276]	Randomised controlled, double-blinded trial; III-1	Greece; university research centre	*n* = 23 Intervention: *n* = 12; 48.5±8.8; 67% female; healthy adults Control: *n* = 11; 49.1±4.6; 82% female; healthy adults	Intervention: One portion of cake containing wheat and fortified with 0.75g beta-glucan from Barley, daily at breakfast for 1 month Control: One portion of cake containing wheat, daily at breakfast for 1 month	LMR at 1 month	LMR: n.s between fortified beta-glucan cake consumption and control cake consumption at 1 month (p=0.95)

Xiao, 2014[277]	Non-randomised, clinical trial; III-2	China; outpatient clinic	*n* = 123 (*n* = 76 analysed); age range 25-55; 64% female; overweight and obese adults (BMI >28 kg/m^2)	Intervention: High prebiotic (oat, buckwheat, guar gum, pectin, konjac flour, resistant starch, oligosaccharides), low meat (< 50 g/day), kilojoule restricted diet, 23 weeks.	LMR at 9 and 23 weeks	LMR significantly decreased compared to baseline (0.026 (0.020–0.031 IQR)) at 9 weeks 0.022 (0.019–0.026 IQR; p<0.01) and 23 weeks 0.023 (0.019–0.026 IQR; p<0.05)
Krawczyk, 2018[199]	Non-randomised, clinical trial; III-2	Poland; university research centre	*n* = 166 (*n* = 110 completed study with *N* = 32 analysed); 48.0±13.1; 31% female; NAFLD	Intervention: NIOR diet consisting of carbohydrate 55-65%; fat 20-30%; protein 15%; fibre 29.2±10.9 g/day (dietary sources included wheat-based products) for 6 months	Serum zonulin at 6 months	Serum zonulin: significant decrease compared to baseline at 6 months (p=0.001) Negative correlation between fibre intake and serum zonulin -0.30 ρ (p=0.043)
Macronutrient ratio and energy intake						
Kuzma, 2016[280]	Randomised controlled, double-blinded crossover trial; II	USA; research centre	*n* = 24 Intervention: *n* = 12; 33.0±11.0; 25% female; normal weight healthy adults (BMI 23.7±1.0); *n* = 12; 39.0±12.0; 50%	Intervention A: 4 servings of fructose-sweetened beverages daily (25% of EER) plus 116% standard USA diet consisting of carbohydrate 50%; fat 34%; protein 16%, 8 days	Serum zonulin at day 9 LMR at day 9	Serum zonulin: n.s between each sweetened beverage at day 9 (p=0.366) LMR: significant increase in glucose

			female; overweight/obese adults (BMI 31.0±4.3)	Intervention B: 4 servings of high-fructose corn syrup-sweetened beverages daily (25% of EER) plus 116% standard USA diet consisting of carbohydrate 50%; fat 34%; protein 16%, 8 days Intervention C: 4 servings of glucose-sweetened beverages daily (25% of EER) plus 116% standard USA diet consisting of carbohydrate 50%; fat 34%; protein 16%, 8 days		and fructose sweetened beverage groups compared to high-fructose corn syrup group at day 9 ($p<0.05$) LMR: n.s between glucose and fructose sweetened beverage groups ($p>0.05$)
Boers, 2014[278]	Randomised controlled, single-blinded trial; II	Netherlands; university research centre	*n* = 34 Intervention: *n* = 18; 52.0±10.2; 72% female; metabolic syndrome Control: *n* = 16; 55.4±9.0; 75% female; metabolic syndrome	Intervention: Palaeolithic diet consisting of carbohydrate 32%; fat 41% (10% saturated fat); protein 24%, 2 weeks Control: healthy reference diet consisting of carbohydrate 50%; fat 29% (9% saturated fat); protein 17% based on the Dutch Health Council guidelines, 2 weeks	LMR at 2 weeks	LMR: n.s between palaeolithic diet and control diet at 2 weeks ($p=0.35$)

Ohlsson, 2016[283]	Non-randomised, clinical crossover trial; III-2	Sweden; Hospital	*n* = 20 (*n* = 10 analysed); 46.0±14.5; 60% female; healthy adults	Intervention: breakfast based on Okinawan diet consisting of carbohydrate 34%; fat 39%; protein 23%; fibre 3% Control: breakfast based on Swedish National Nutrition Recommendations consisting of carbohydrate 54%; fat 27%; protein 15%; fibre 2%	Serum zonulin at 0.5h, 1h, 1.5h, 2h, 2.5h and 3h	Serum zonulin: n.s between intervention and control (p=0.211)
Ohlsson, 2017[284]	Non-randomised, clinical trial; III-2	Sweden; hospital	*n* = 30 (*n* = 28 analysed); 57.5±8.2; 57% female; type 2 diabetes	Intervention: Okinawan style diet consisting of carbohydrate 42%; fat 35%; protein 23%, 12 weeks	Serum and faecal zonulin at 12 and 28 weeks	Serum zonulin: significant increase compared to baseline at 12 weeks (p=0.019) remained elevated at 28 weeks compared to baseline (p=0.014) Faecal zonulin: significant increase compared to baseline at 12 weeks (p<0.001)
Molina-Vega, 2020[281]	Non-randomised, clinical trial; III-2	Spain; hospital	*n* = 39; 43.4±9.2; 67% female; morbid obesity adults (BMI >40 kg/m^2) awaiting bariatric surgery	Intervention: 50g fat consisting of 30% saturated, 49% monounsaturated, and 21% polyunsaturated fatty acids, 3h	Serum zonulin at 3h	Serum zonulin: significant increase compared to baseline at 3h (p=0.040)

Bowser, 2020[279]	Non-randomised, clinical trial; III-2	USA; university research centre	*n* = 13; 22.2±0.4; 100% male; healthy adults	Intervention: A high fat diet consisting of carbohydrate 30%; fat 55% (25% saturated fat); protein 15%, 5 days Control: A lead-in diet consisting of carbohydrate 55%; fat 30% (9% saturated fat); protein 15%, 2 weeks	LMR and endotoxin at 5 days	Endotoxin: significantly increased after 5 days of high fat diet compared to control diet (p=0.04) LMR: n.s between high fat and control diet at 5 days (p=0.084)
Ott, 2018[286]	Non-randomised, clinical trial; III-2	Germany; university research centre	*n* = 24; 23.0±2.8; 100% male; healthy adults	Intervention: A high fat diet consisting of carbohydrate 34%; fat 48%; protein 18% plus 1000kcal/d of whipping cream, 7 days	LMR at 7 and 15 days Serum zonulin at 7 and 15 days	LMR: n.s between baseline and day 7 or day 15 (p>0.05) Serum zonulin: n.s between baseline and day 7 or day 15 (p>0.05)
Ott, 2017[285]	Non-randomised, clinical trial; III-2	Germany; university research centre	*n* = 20; 46.8±11.5; 100% female; obesity with BMI >30kg/m^2	Intervention: Caloric restricted diet consisting of 800kcal/day, 4 weeks. Balanced diet of 1800kcal/day, 2 weeks	Serum zonulin at 4 and 6 weeks	Serum zonulin: significant decrease at 4 weeks compared to baseline (p<0.01). Returned to baseline values after 2 weeks on balanced diet.

Nier, 2019[282]	Non-randomised, crossover clinical trial; III-2	Germany; university research centre	*n* = 15 (*n* = 12 analysed); 26.3±1.2; 58% female; healthy adults	Intervention 1: fructose-enriched diet (25% of EER) consisting of carbohydrate 54%; fat 30%; protein 16%, 3 days Intervention 2: glucose-enriched diet (25% of EER) consisting of carbohydrate 54%; fat 30%; protein 16%, 3 days	Plasma endotoxin at day 3	Plasma endotoxin: significant increase (+1.3-fold) in fructose-enriched diet compared to baseline ($p<0.05$). N.s change in glucose-enriched diet compared to baseline ($p>0.05$).
Gluten-free diet						
Biesiekierski, 2011[287]	Randomised placebo-controlled, double-blinded trial; II	Australia; university research centre	*n* = 39 (*n* = 34 analysed) Intervention: *n* = 19; median years 40 (29-55 range); 84% female; IBS with improved symptoms on a gluten-free diet	Intervention: gluten-free diet and 16g of gluten (1 muffin and 2 slices of bread) daily for 6 weeks Control: gluten-free diet plus gluten-free muffin and 2 slices of bread daily for 6 weeks	LRR at 6 weeks	LRR: n.s compared to baseline at 6 weeks ($p>0.05$) LRR: n.s between intervention and control at 6 weeks ($p>0.05$)
Vazquez-Roque, 2013[288]	Randomised controlled, single-blinded trial; II	USA; outpatient clinic	*n* = 45 Intervention: *n* = 23; 41.8±2.5; 95% female; positive and negative HLA-DQ2/8 with IBS-D	Intervention: gluten containing diet consisting of carbohydrate 50%; fat 30%; protein 20% for 28 day Control: gluten-free diet consisting of carbohydrate	LMR at 28 days	LMR: significant increase in gluten containing diet compared to gluten-free diet ($p<0.05$). Significant increase in

			Control: *n* = 22; 43.4±2.7; 95% female; positive and negative HLA-DQ2/8 with IBS-D	50%; fat 30%; protein 20% for 28 days		HLA-DQ2/8 positive patients consuming a gluten containing diet compared to HLA-DQ2/8 negative patients (p=0.006)
Cummins, 2001[289]	Non-randomised, clinical trial; III-2	Australia; hospital	*n* = 36 (*n* = 26 analysed), 18-80 range; 55% female; newly diagnosed coeliac disease	Intervention: gluten-free diet for 24 months	LRR at 0.25, 0.5, 1, 2, 3, 6, 12 and 24 months	LRR: significant decrease compared to baseline at 2 months (p<0.001)
Duerksen, 2005[290]	Case-control; III-2	Canada; university research centre	*n* = 73 Intervention A: *n* = 3; median years 54.3 (30-70 range); 67% female; coeliac disease on a gluten-free diet for less than 1 month Intervention B: *n* = 9; median years 50.9 (33-74 range); 78% female; coeliac disease on a gluten-free diet for 1 month to 1 year	Intervention: gluten-free diet	LMR: at 4-12 weeks	LMR: significant increase in newly diagnosed coeliac disease and in those consuming a gluten-free diet for less than 1 year compared to control (P<0.05) Trace gluten consumption is associated with increased LMR in coeliac disease (p=0.048)

		Intervention C: *n* = 42; median years 49.9 (27-78 range); 90% female; coeliac disease on a gluten free diet for over 1 year Control: *n* = 19; median years 48.6 (24-72 range); 68% female; healthy adults

Abbreviations: BMI, body mass index; EER, estimated energy requirement; h, hour; IBS-D, diarrhoea-predominant irritable bowel syndrome; IQR, interquartile range; LMR, lactulose/mannitol ratio; LRR, lactulose/rhamnose ratio *n*, number of participants; n.s, not statistically significant; NAFLD, non-alcoholic fatty liver disease; NHMRC, National Health and Medical Research Council; NIOR, nutrient-induced insulin output ratio; USA, United States of America.

6.1.9.1.2.5 RISK OF BIAS ASSESSMENT

The following are the results from a risk of bias assessment (Table 6.3, Table 6.4 and Table 6.5). A summary of the results is found in the IP Guideline (Section 8.11).

Table 6.3 Risk of bias assessment in randomised trials

	Randomisation process	Deviations from intended interventions	Missing outcome data	Measurement of the outcome	Selection of the reported result	Overall
Biesiekierski, 2011[287]	Low	Low	Low	Low	Some	Some
Boers, 2014[278]	Some	Low	Low	Some	Low	Some
Kuzma, 2016[280]	Low	Low	Some	Low	Low	Some
Machado, 2020[274]	Low	Low	Low	Low	Low	Low
Russo, 2012[275]	Some	Low	Some	Low	Some	Some
Skouroliakou, 2016[276]	Some	Low	Low	Low	Some	Some
Vazquez-Roque, 2013[288]	Low	Low	Low	Low	Low	Low
Percentages						
Low risk	57%	100%	71%	86%	57%	29%
Some concerns	43%	0%	29%	14%	43%	71%
High risk	0%	0%	0%	0%	0%	0%

Abbreviations: Low = low risk of bias; Some = some concerns for risk of bias; High = high risk of bias.

Table 6.4 Risk of bias in systematic reviews assessment

Review	Phase 2				Phase 3
	1. Study eligibility criteria	2. Identification and selection of studies	3. Data collection and study appraisal	4. Synthesis and findings	Risk of bias in the review
Leech, 2019 [221]	Low	High	Low	High	High

Abbreviations: Low = low risk; High = high risk; Unclear = unclear risk.

Table 6.5 Risk of bias in non-randomised studies

Study	Confounding	Selection of participants	Classification of interventions	Deviations from intended interventions	Missing data	Measurement of outcome	Selection of reported results	Overall
Bala, 2014[271]	Moderate	Low	Moderate	Low	Serious	Moderate	Moderate	Serious
Bowser, 2020[279]	Moderate	Low	Low	Low	Low	Moderate	Low	Moderate
Cummins, 2001[289]	Serious	Serious	Moderate	Moderate	Critical	Serious	Critical	Critical
Duerksen, 2005[290]	Serious	Critical	Serious	Serious	Serious	Moderate	Critical	Critical
Krawczyk, 2018[199]	Serious	Serious	Moderate	Critical	Critical	Moderate	Serious	Critical
Molina-Vega, 2020[281]	Moderate	Low	Low	Low	Low	Low	Low	Moderate
Nier, 2019[282]	Moderate	Low	Moderate	Moderate	Moderate	Moderate	Moderate	Moderate
Ohlsson, 2016[283]	Moderate	Moderate	Low	Low	Low	Moderate	Moderate	Moderate
Ohlsson, 2017[284]	Moderate	Low	Low	Low	Low	Moderate	Moderate	Moderate
Ott, 2017[285]	Serious	Moderate	Moderate	Low	Low	Moderate	Low	Serious
Ott, 2018[286]	Serious	Moderate	Moderate	Low	Serious	Serious	Moderate	Serious
Stadlbauer, 2019[272]	Moderate	Moderate	Low	Low	Moderate	Moderate	Moderate	Moderate
Swanson, 2011[273]	Moderate	Serious	Low	Moderate	Serious	Moderate	Moderate	Serious
Xiao, 2014[277]	Moderate	Low	Low	Low	Moderate	Moderate	Low	Moderate

Abbreviations: Low = low risk of bias; Moderate = moderate risk of bias; Serious = serious risk of bias; Critical = critical risk of bias.

6.1.9.1.2.6NHMRC EVIDENCE STATEMENT

The following are the results from completing the NHMRC Evidence Statement (Table 6.6). Individual grade for each of the five domains are provided along with the overall grade for the evidence-based recommendation. Furthermore, consensus-based recommendations are listed below (Table 6.7).

Table 6.6 Summary of the NHMRC evidence statement for evidence-based recommendations: Dietary recommendations

Recommendations	Grade
Alcohol	
Recommendation 1.1: People with intestinal permeability should consider consuming no more than 10 standard drinks a week and no more than 4 standard drinks on any one day in accordance with the Australian Dietary Guidelines during the treatment of intestinal permeability.	**Grade: C**
Evidence base – number of studies, level of evidence, risk of bias in the included studies	**D**
Consistency – between studies if more than one study	**C**
Clinical impact – of the intervention, diagnosis, prognosis, aetiology, screening	**B**
Generalisability – how well the body of evidence matches the population and clinical setting	**B**
Applicability – relevance to Australian health care context in terms of services, delivery and cultural factors	**B**
Dietary fibre	
Recommendation 1.3: People with intestinal permeability should consider consuming a diet high in dietary fibre from a diverse range of sources.	**Grade: C**
Evidence base – number of studies, level of evidence, risk of bias in the included studies	**C**
Consistency – between studies if more than one study	**B**
Clinical impact – of the intervention, diagnosis, prognosis, aetiology, screening	**C**
Generalisability – how well the body of evidence matches the population and clinical setting	**B**
Applicability – relevance to Australian health care context in terms of services, delivery and cultural factors	**B**
Macronutrient ratio	

Recommendation 1.6: People with intestinal permeability should consider consuming the Acceptable Macronutrient Distribution Range of protein (15-25%), fats (20-35%) and carbohydrates (45-65%) in accordance with the Australian Dietary Guidelines.	**Grade: C**
Evidence base – number of studies, level of evidence, risk of bias in the included studies	**C**
Consistency – between studies if more than one study	**B**
Clinical impact – of the intervention, diagnosis, prognosis, aetiology, screening	**C**
Generalisability – how well the body of evidence matches the population and clinical setting	**B**
Applicability – relevance to Australian health care context in terms of services, delivery and cultural factors	**A**
Recommendation 1.7: People with intestinal permeability should consider NOT consuming a diet high in fat.	**Grade: C**
Evidence base – number of studies, level of evidence, risk of bias in the included studies	**C**
Consistency – between studies if more than one study	**C**
Clinical impact – of the intervention, diagnosis, prognosis, aetiology, screening	**B**
Generalisability – how well the body of evidence matches the population and clinical setting	**B**
Applicability – relevance to Australian health care context in terms of services, delivery and cultural factors	**A**
Recommendation 1.8: People with intestinal permeability should consider NOT consuming a diet high in fructose.	**Grade: C**
Evidence base – number of studies, level of evidence, risk of bias in the included studies	**C**
Consistency – between studies if more than one study	**A**
Clinical impact – of the intervention, diagnosis, prognosis, aetiology, screening	**B**
Generalisability – how well the body of evidence matches the population and clinical setting	**B**
Applicability – relevance to Australian health care context in terms of services, delivery and cultural factors	**A**
Energy intake	
Recommendation 1.9: People with intestinal permeability may consider consuming the estimated energy requirements in accordance with the Australian Dietary Guidelines.	**Grade: D**

Evidence base – number of studies, level of evidence, risk of bias in the included studies	**D**
Consistency – between studies if more than one study	**C**
Clinical impact – of the intervention, diagnosis, prognosis, aetiology, screening	**B**
Generalisability – how well the body of evidence matches the population and clinical setting	**B**
Applicability – relevance to Australian health care context in terms of services, delivery and cultural factors	**B**
Recommendation 1.10: Clinicians may consider using a kilojoule restricted diet in the short-term treatment of people with confirmed intestinal permeability.	**Grade: D**
Evidence base – number of studies, level of evidence, risk of bias in the included studies	**D**
Consistency – between studies if more than one study	**C**
Clinical impact – of the intervention, diagnosis, prognosis, aetiology, screening	**B**
Generalisability – how well the body of evidence matches the population and clinical setting	**B**
Applicability – relevance to Australian health care context in terms of services, delivery and cultural factors	**B**
Gluten-free diet	
Recommendation 1.11: Clinicians should only advise a strict gluten-free diet if clinical symptoms or pathology indicate a gluten intolerance, sensitivity or allergy.	**Grade: B**
Evidence base – number of studies, level of evidence, risk of bias in the included studies	**B**
Consistency – between studies if more than one study	**B**
Clinical impact – of the intervention, diagnosis, prognosis, aetiology, screening	**A**
Generalisability – how well the body of evidence matches the population and clinical setting	**A**
Applicability – relevance to Australian health care context in terms of services, delivery and cultural factors	**A**
Recommendation 1.12: Clinicians should only advise a gluten-free diet during the short-term treatment of people with confirmed intestinal permeability that report clinical symptoms in response to the consumption of gluten after the investigation for gluten intolerance, sensitivity or allergy has been carried out.	**Grade: B**

Evidence base – number of studies, level of evidence, risk of bias in the included studies	**B**
Consistency – between studies if more than one study	**B**
Clinical impact – of the intervention, diagnosis, prognosis, aetiology, screening	**A**
Generalisability – how well the body of evidence matches the population and clinical setting	**A**
Applicability – relevance to Australian health care context in terms of services, delivery and cultural factors	**A**
Recommendation 1.13: Clinicians should offer a low gluten diet for the management of people with confirmed intestinal permeability that report no clinical symptoms or pathology indicating a gluten intolerance, sensitivity or allergy.	**Grade: B**
Evidence base – number of studies, level of evidence, risk of bias in the included studies	**B**
Consistency – between studies if more than one study	**B**
Clinical impact – of the intervention, diagnosis, prognosis, aetiology, screening	**A**
Generalisability – how well the body of evidence matches the population and clinical setting	**A**
Applicability – relevance to Australian health care context in terms of services, delivery and cultural factors	**A**

Table 6.7 Summary of consensus-based recommendations and practice points: Dietary recommendations

No.	Recommendation
1.2	People with intestinal permeability may consider limiting or avoiding alcohol consumption during the short-term treatment of intestinal permeability.
1.4	People with intestinal permeability may consider consuming 38g for men and 28g for female of dietary fibre daily in accordance with the Australian Dietary Guidelines during the treatment of intestinal permeability.
1.5	People with intestinal permeability may consider prioritising for low gluten sources of dietary fibre during the treatment of intestinal permeability.

6.1.9.2 PROBIOTIC, PREBIOTIC AND SYNBIOTIC SUPPLEMENTATION

6.1.9.2.1 CLINICAL QUESTIONS

Clinical Question 3: In Australian adults with increased intestinal permeability, what are the benefits of oral probiotic, prebiotic or synbiotic supplementation for the treatment of increased intestinal permeability?

Clinical Question 4: In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for oral probiotic, prebiotic or synbiotic supplementation use?

6.1.9.2.2 EVIDENCE SUMMARY AND OVERALL QUALITY OF EVIDENCE

6.1.9.2.2.1PROBIOTIC

There are high quality research articles (*n* = 19) investigating the effects of probiotics on intestinal integrity (Table 6.8 and 6.9).[84,291-308] Two systematic reviews[295,301] and 16 randomised controlled trials[84,291-294,296-300,302-308] (RCT) were identified during the literature search with two[291,301] of these identified from hand searching the literature. One study was reported across two articles.[296,298] The included studies assessed the effects of probiotics on intestinal integrity for between two weeks and three months with most conducted over 12 weeks. Participants involved in the included articles were diagnosed with overweight/obesity (*n* = 7),[291,292,296,298,299,302,303,307] functional gastrointestinal disorders (n = 3),[300,304,308] inflammatory bowel disease (IBD) (*n* = 2),[84,297] liver disease (*n* = 2)[293,306] and migraine (*n* = 1).[305] Most studies (*n* = 11)[84,291,293,294,299,300,303-307] used a diverse range of multi-strain probiotics with no study using the same combination of probiotic strains. Two studies used single-

strains (*Saccharomyces boulard*[297] and *Akkermansia muciniphila*[292]). Four studies investigated fermented dairy-based drinks (kefir,[302] fermented milk[308] and Yakult light®[296,298,306]).

Multi-strain probiotics were found to have mixed results on intestinal integrity, with most studies finding no significant difference in stool zonulin, serum zonulin and dual sugar after probiotic supplementation. Although a trend for improvement in IP was seen in many studies, results were non-significant. A meta-analysis found probiotic and synbiotic supplementation significantly reduced serum zonulin compared to placebo (WMD = -10.55 [95% CI: -17.76, -3.34]; p=0.004), with a significant reduction in serum zonulin observed in probiotic supplementation compared to symbiotic supplementation.[301] One study that reported an improvement in IP also found that increasing the amount of multi-strain probiotic from $1x10^{10}$ CFU twice daily to $2.5x10^{9}$ CFU twice daily saw a significant decrease in LPS (-0.99, standardised mean difference; p=0.001) with a 20.1% decrease seen in the high dose multi-strain probiotic compared to baseline after 12 weeks (13.0±5.2 vs 10.4±5.5; p=0.0008).[307]

The two single-strain probiotics were found to have a beneficial effect on intestinal integrity. Supplementation with 200mg of *Saccharomyces boulard*-17 for 12 weeks significantly improves LMR at four weeks (0.004±0.004; p=0.005) and 12 weeks (0.008±0.006; p=0.005) with IP decreasing by 33.33% at 12 weeks (p>0.001).[297] Heat-killed *Akkermansia muciniphila* (10^{10} CFU) taken for three months was found to have a significant effect on IP compared with baseline, with a decrease in IP (mean change -0.28±0.09; p=0.044) whereas no difference was

seen in alive *Akkermansia muciniphila* (10^{10} CFU) (mean change -0.24±0.47; p=0.29).[292]

Fermented dairy-based drinks were found to have mixed results, with the type of probiotic drink appearing to influence the effect on intestinal integrity. Kefir milk and fermented milk were both found to have a beneficial effect on IP.[302,308] Kefir consumption of 300 ml daily for three weeks had a significant effect on IP (p=0.018).[302] Furthermore, 200g of fermented milk twice daily for four weeks significantly decreased LMR from 0.038 at baseline to 0.023 at four weeks (p=0.004).[308] The consumption of fermented milk further results in a significant decrease in the total number of patients with IP before and after study intervention (64.3% vs 28.6%; p=0.023).[308] On the other hand, 65ml of milk drink (Yakult light®) containing *Lactobacillus casei* Shirota 10^8/ml (6.5 x 10^9 CFU) three times daily was found to have no significant effect on IP.[296,298,306] These were consistent across multiple assessment methods (stool zonulin, serum zonulin and dual sugar), time points (12 weeks and six months) and disease states (metabolic syndrome and liver cirrhosis).[296,298,306]

6.1.9.2.2.2PREBIOTIC

There is a limited number (*n* = 6) of studies exploring the effects of prebiotics on altered IP.[295,309-313] Many of the included articles were of high quality with one systematic review,[295] four RCT[309,310,312,313] and one non-randomised clinical trial[311] found to meet the inclusion criteria. The included studies assessed the effects of prebiotics for between one and six months. Participants involved in the included studies were overweight or obese (*n* = 3),[295,309,313] had a functional

gastrointestinal disorder (*n* = 2),[311,312] diagnosed with type two diabetes (*n* = 1)[295] or were healthy adults (*n* = 1).[310] The prebiotics used were pectin (*n* = 2),[310,311] arabinoxylan (*n* = 2),[309,312] inulin (*n* = 1),[295] inulin-type fructans (*n* = 1),[295] polydextrose (*n* = 1),[313] slippery elm (*n* = 1)[311] and guar gum (*n* = 1).[311] A systematic review found prebiotics containing inulin or inulin-type fructans decreased either LPS or exotoxin levels.[295] However, this beneficial effect does not appear to apply to all prebiotics, with some prebiotics suggested to have a negative effect of IP. The effects of polydextrose at 12g daily saw a non-significant increase in serum zonulin after six months (55.5±9.1 vs 58.4±12.0).[313] Two studies investigating the effect of arabinoxylan on IP found the use of 7.5g to 15g of arabinoxylan daily had no effect of IP after six or 12 weeks.[309,312] Another study investigated 7.5g of pectin twice daily for four weeks found no significant effect of IP in healthy adults or the elderly (p=0.861).[310] Furthermore, an Australian based study explored the effects of a mix of prebiotics (slippery elm 500mg, guar gum 100mg, pectin 100mg) and other intestinal supportive herbal medicine and nutrients on IP.[311] This study found a significant decrease between baseline and 12 weeks in LMR (0.04±0.004 vs 0.03±0.001; p<0.0001).[311]

6.1.9.2.2.3SYNBIOTIC

A total of eight articles, with a moderate quality of evidence were included.[301,313-319] One systematic review,[301] four RCTs[313,317-319] and three non-randomised clinical trials[294,315,316] were identified including one article located through hand searching the literature.[301] The included studies assessed the effects of synbiotic supplementation on intestinal integrity for between one and six months. The included studies involved participants that were overweight or obese (*n* = 2),[313,317]

had a functional gastrointestinal disorder (n = 1),[315] diagnosed with Alzheimer disease (*n* = 1),[294] non-alcoholic steatohepatitis (*n* = 1),[318] had a history of proton pump inhibitor use (*n* = 1)[316] or were healthy adults (*n* = 1).[319] Most studies (*n* = 5)[301,313,317-319] used a diverse range of probiotics and prebiotics, with only three studies identified as having the same or similar ingredients.[294,315,316]

A meta-analysis found probiotic and synbiotic supplementation significantly reduced serum zonulin compared to placebo (WMD = -10.55 [95% CI: -17.76, -3.34]; p=0.004), with a study duration of below 3 months identified to have a significant impact on the effects of synbiotic on serum zonulin, having the greatest outcome on IP (coefficient = 33.23 [95% CI: 0.30, 66.16]; p=0.048). The three studies identified as having the same or similar ingredients used a combination of inulin, corn starch and fructooligosaccharides with a multi-strain probiotic (7.5 x 10^9 CFU).[294,315,316] All three studies demonstrated a beneficial effect of symbiotic therapy on intestinal integrity. Specifically, synbiotic therapy had a significant effect on stool zonulin, with two studies reporting a significant decrease from baseline to 4 weeks (67 [38-92] vs 36 [20-48] ng/ml; p=0.035)[315] and (93.1±56.3µg/L vs 66.6±54.2µg/L; p=0.01).[294] Furthermore, a six month study using a similar symbiotic formula found a significant decrease after three months in participants with elevated (>50ng/mg) stool zonulin at baseline (−46.3 ng/mg; 95% CI: −71.4; −21.2; p<0.001).[316] Other synbiotic combinations such as 250mg of tara gum and *Streptococcus thermophilus* (1 x 10^7 CFU) over four weeks resulted in a significant difference between synbiotic and placebo (0.014±0.004 vs 0.019±0.007; p=0.045).[319] However, one study found no difference in IP after three months treatment with 2.4g of partially hydrolysed guar

gum and 1.6g of inulin, *Lactobacillus reuteri* (1 x 10^8 CFU) twice daily (p=0.737).[318] Furthermore, one study identified synbiotic supplementation containing 12g of polydextrose and *Bifidobacterium animalis ssp. lactis* 420 (10^{10} CFU) resulted in a significant increase in serum lipopolysaccharide compared to placebo after six months (+9.1±40 vs −26±108; p=0.007).[313]

6.1.9.2.2.4PROBIOTIC, PREBIOTIC AND SYNBIOTIC ON NSAIDS INDUCED IP

There is a moderate level (*n* = 4) of evidence investigating the effects of probiotic (*n* = 2), prebiotic (*n* = 1) and synbiotic (*n* = 1) supplementation on nonsteroidal anti-inflammatory drugs (NSAIDs)-induced IP.[314,320-322] All included articles were RCTs[314,320-322] with no systematic reviews identified from the literature search. The studies investigated the effectiveness of probiotics, prebiotics and synbiotics on preventing NSAIDs induced IP over five days to six weeks. Participants involved in these studies were all healthy adults, with one study also including healthy elderly adults.[320] Included studies used the same designs to induce IP with NSAIDs. This design involved participants taking 75mg of a NSAID nine hours before measuring IP and another 50mg one hour prior to measuring IP.

Two short-term studies investigating the effect of five probiotic formulations on NSAID induced IP found no significant change between each probiotic formula or compared to baseline.[321,322] A six-week trial of two different prebiotics (12g of arabinoxylan or 12g of oat β-glucan) found no significant difference in the arabinoxylan group or oat β-glucan in preventing NSAID induced IP at six-weeks.[320] Synbiotic supplement containing fructooligosaccharides and a multi-

strain probiotic resulted in similar outcomes as prebiotic and probiotic.[314] One study found no significant difference in the dual sugar results between baseline with NSAIDs compared with synbiotic treatment (0.064; 0.046–0.106 IQR vs 0.055; 0.037–0.072 IQR; p=0.203).[314] Furthermore, no significant difference in serum zonulin was found between symbiotic and control after NSAIDs use at two weeks (13.2ng/ml vs 14.7ng/ml; p=0.650).[314]

Table 6.8 Evidence summary table for review articles

Author, Year	Study design; Level of Evidence (NHMRC)	Review aim	Type of evidence included; study length	Year studies published	Main results from relevant studies	Authors' conclusions
Ramezani Ahmadi, 2020[301]	Systematic review and meta-analysis; I	Influence of probiotics and synbiotic on serum zonulin	*n* = 9 Randomised clinical trials 2-24 weeks	2013-2018	Probiotic/synbiotic supplementation significantly reduced serum zonulin compared to placebo (WMD = -10.55 [95% CI: -17.76, -3.34]; p=0.004). Study duration (<3 months) was the only variable found to have a significant impact on the effects of probiotic/synbiotic on serum zonulin (coefficient = 33.23 [95% CI: 0.30, 66.16]; p=0.048). A significant reduction in serum zonulin was observed in studies less than 3 months in length (compared to more than 3 months), with participants > 45y (compared to < 45y), probiotic supplementation (compared to synbiotic supplementation), and in participants with	The use of probiotic/synbiotic has beneficial effects on reducing serum zonulin. The results should be interpreted with caution due to a single study influencing the pooled effect size.

					a health condition (compared to healthy participants).	
Moludi, 2020[295]	Systematic review; I	Role of prebiotic/probiotic in modulating the gut microbiota and host metabolism in metabolic endotoxemia	*n* = 16 Randomised clinical trials 1-28 weeks	2007-2019	Probiotics containing one or a combination of *Bifidobacterium bifidum, Bifidobacterium lactis, L. subtilis, L. acidophilus, L. casei, L. brevis, L. salivarius, Lactococcus lactis, Streptococcus faecium, E. coli* Nissle*, Bacillus indicus, Bacillus subtilis, Bacillus coagulans, Bacillus licheniformis, Bacillus clausii* or VSL#3 were found to decrease either LPS or exotoxin levels. Prebiotics containing inulin and inulin-type fructans were found to decrease either LPS or exotoxin levels.	The use of prebiotics/probiotics have potential benefits for reducing metabolic endotoxemia.

Abbreviations: CI, confidence interval; *Escherichia coli*, *E. coli*; LPS, lipopolysaccharide; *n*, number of participants; WMD, weighted mean difference; y, years.

Table 6.9 Evidence summary table for clinical trials

Author, Year	Study design; Level of Evidence NHMRC	Country; Setting	Sample size; age in years M ± SD; gender; health condition	Intervention; control	Outcome measures; duration	Main results
Probiotic						
Garcia Vilela, 2008[297]	Randomised, placebo controlled, single-blind trial; II	Brazil; outpatient clinic	*n* = 34 (*n* = 31 analysed); mean 37 years; 42% female Intervention: *n* = 15; Crohn's disease (CDAI 50.7±36.9) Control: *n* = 19; Crohn's disease (CDAI 62.8±44.6)	Intervention: 200mg of lyophilized *Saccharomyces boulard*-17 (4 x 10^8 CFU), 6mg sucrose and 2.4mg magnesium stearate (Floratil®) every 8h, 12 weeks Control: placebo capsule containing 200mg cellulose, 6mg sucrose and 2.4mg magnesium stearate every 8h, 12 weeks	LMR at 4 and 12 weeks	LMR intervention: significantly improved by 0.004±0.004 at 4 weeks and 0.008±0.006 at 12 weeks (p=0.005). *Saccharomyces boulard* decreased LMR by 33.33% at 12 weeks (p>0.001). LMR control: n.s increase by 0.004±0.010 at 12 weeks (p=0.12).
Szulińska, 2018[307]	Randomised, placebo controlled,	Poland; outpatient clinic	*n* = 81 (*n* = 71 analysed) Intervention high dose: *n* = 23;	Intervention high dose: 2g of multi-strain probiotic (Ecologic®	LPS at 12 weeks	LPS: significantly decreased by 20.1% in high dose probiotic compared with baseline

	double-blind trial; II	55.2±6.9 years; obese postmenopausal women (BMI 36.6±5.9 kg/m^2) Intervention low dose: *n* = 24; 56.4±6.5 years; obese postmenopausal women (BMI 36.0±5.2 kg/m^2) Control: *n* = 24; 58.7±7.2 years; obese postmenopausal women (BMI 36.1±4.4 kg/m^2)	Barrier) (1x10^{10} CFU) 2x/day, 12 weeks Intervention low dose: 2g of multi-strain probiotic (Ecologic® Barrier) (2.5x10^9 CFU) 2x/day, 12 weeks Control: placebo sachet containing maize starch and maltodextrins 2x/day, 12 weeks Ecologic® Barrier contains equal amounts of *Bifidobacterium bifidum* W23, *Bifidobacterium lactis* W51, *Bifidobacterium lactis* W52, *L. acidophilus* W37, *Lactobacillus brevis* W63, *L. casei* W56, *L. salivarius* W24, *Lactococcus lactis*	(13.0±5.2 vs 10.4±5.5) at 12 weeks (p=0.0008) LPS: significantly decreased in the high dose probiotic group decreased compared to control (-0.99, SMD; p=0.001) LPS: n.s decrease in low dose probiotic compared with baseline (12.3±6.7 vs 11.9±6.8) at 12 weeks (p=0.241).

				W19, and *Lactococcus lactis* W58		
Wegh, 2019[84]	Randomised, placebo controlled, double-blind trial; II	Netherlands; hospital	*n* = 25 Intervention: *n* = 13; 51.8±13.3 years; 54% female; UC or pancolitis in clinical remission Control: *n* = 12; 51.1±11.9 years; 42% female; UC or pancolitis in clinical remission	Intervention: *Bifidobacterium bifidum* W23, *Bifidobacterium lactis* W51, *Bifidobacterium lactis* W52, *L. acidophilus* W22, *L. casei* W56, *L. paracasei* W20, *L. plantarum* W62, *L. salivarius* W24, and *Lactococcus lactis* W19 (Ecologic® 825) $1.5x10^{10}$ CFU, 1x/day, 12 weeks Control: placebo containing maize starch and maltodextrins, 1x/day, 12 weeks.	Stool zonulin, serum zonulin and LRR at 6 and 12 weeks	Stool zonulin: n.s between probiotic group and placebo at 12 weeks (89.6±64.7 vs 118.4±91.9; p>0.05). Serum zonulin: n.s between probiotic group and placebo at 12 weeks (49.6±23.6 vs 51.8±17.9; p>0.005). LRR: n.s between probiotic group and placebo at 12 weeks (0.04±0.04 vs 0.04±0.03; p>0.05).
Kwak, 2014[293]	Randomised, placebo controlled,	Republic of Korea; Hospital	*n* = 53 (*n* = 50 analysed) Intervention: *n* = 25; 54.4±8.4 years; 28%	Intervention: *Bifidobacterium bifidum, Bifidobacterium lactis, Bifidobacterium*	LMR at 4 weeks	LMR: n.s difference in the improvement of LMR between intervention (50%) and

	double-blind trial; II		female; chronic liver disease. Control: *n* = 25; 53.3±9.8 years; 28% female; chronic liver disease.	*longum, L. acidophilus, L. rhamnosus,* and *Streptococcus thermophilus* (Duolac Gold probiotic, Cell Biotech Co) $5x10^9$ CFU, 2x/day, 4 weeks Control: placebo capsule, 2x/day, 4 weeks		placebo (31%) at 4 weeks (p=0.248).
de Roos, 2017[305]	Randomised, placebo controlled, double-blind trial; II	Netherlands; hospital and outpatient clinic	*n* = 63 (*n* = 60 analysed) Intervention: *n* = 31; 42 mean years; 90% female; migraine patients Control: *n* = 29; 38 mean years; 96% female; migraine patients	Intervention: *Bifidobacterium bifidum* W23, *Bifidobacterium lactis* W52, *L. acidophilus* W37, *L. brevis* W63, *L. casei* W56, *L. salivarius* W24, *L. salivarius* W24, *Lactococcus lactis* W19, *L. lactis* W58 $5x10^9$ CFU, 1x/day, 12 weeks Control: placebo containing maize starch	Stool zonulin, serum zonulin and LMR at 12 weeks	Stool zonulin: n.s difference between baseline and 12 weeks in intervention group (44.6ng/ml vs 44.0ng/ml; p=0.243). Serum zonulin: n.s difference between intervention and placebo at 12 weeks (9.1 [95% CI, 2.2-16.0ng/ml] vs 10.1 [95% CI, 1.7-18.6]; p=0.084).

				and maltodextrins, 1x/day, 12 weeks		LMR: n.s difference between baseline and 12 weeks in intervention group (0.035 vs 0.034; p=0.748).
Palacios, 2020[291]	Randomised, placebo controlled, double-blind, trial; II	Australia; research centre	*n* = 60 Intervention: *n* = 30; 61.4±8.9 years; 43% female; overweight and obese adults with prediabetes or T2D (BMI 35.5±6.2 kg/m^2) Control: *n* = 30; 56.1±12.3 years; 63% female; overweight and obese adults with prediabetes or T2D (BMI 35.5±6.2 kg/m^2)	Intervention: *L. plantarum* Lp-115, *L. bulgaricus* Lb-64, *L. gasseri* Lg-36, *Bifidobacterium breve* Bb-03, *Bifidobacterium animalis sbsp. lactis* Bi-07, *Bifidobacterium bifidum* Bb-06, *Streptococcus thermophilus* St-21, *Saccharomyces boulardii* DBVPG 6763 5x10^{10} CFU, 4x/day, 12 weeks Control: Microcrystalline cellulose, silica, and	Serum zonulin at 12 weeks	Serum zonulin: n.s difference between intervention and placebo at 12 weeks (p>0.05). In participants taking metformin, zonulin significantly decreased at 12 weeks (p=0.048).

				magnesium stearate, 4x/day, 12 weeks		
Kim, 2006[300]	Randomised, placebo controlled, double-blind trial; II	USA; research centre	*n* = 72 Intervention 1: *n* = 12; 46.8±13.9 years; 67% female; functional gastrointestinal disorder Intervention 2: *n* = 12; 48.6±18.1 years; 75% female; functional gastrointestinal disorder Intervention 3: *n* = 12; 41.1±12.5 years; 75% female; functional gastrointestinal disorder Intervention 4: *n* = 12; 43.8±13.6 years; 75% female; functional gastrointestinal disorder	Intervention 1: *L. acidophilus, Bifidobacterium bifidum, Bacillus subtilis, L. bulgaricus, L. lactis, and Bacillus lichenformis* (5 x 10^7 CFU), 820mg of barley grass and oat grass juice and 180 mg of ionic plant-based minerals. Intervention 2: *L. acidophilus, Bifidobacterium bifidum, L. bulgaricus, L. lactis, L. brevis, L. caucasicus, L. fermenti, L. leichmannii, L. caseii, L. plantarum, L. helveticus, and Saccharomyces*	LMR at 12 weeks	LMR: n.s difference between baseline and 12 weeks in intervention groups (p>0.05). Mean change at 12 weeks for probiotic group - 0.01±0.03.

	Intervention 5: *n* = 12; 47.4±17.3 years; 67% female; functional gastrointestinal disorder Control: *n* = 12; 41.5±15.8 years; 67% female; functional gastrointestinal disorder	*boulardii* (5 x 10^7 CFU), 820mg of barley grass and oat grass juice and 180 mg of ionic plant-based minerals. Intervention 3: *L. acidophilus, Bifidobacterium bifidum, Bacillus subtilis, L. bulgaricus, L. lactis, and Bacillus lichenformis* (5 x 10^7 CFU). Intervention 4: 820mg of barley grass and oat grass juice and 180 mg of ionic plant-based minerals. Intervention 5: *Bacillus coagulans, Saccharomyces boulardii, Bacillus subtilis, L. salivarius,*

				and L. plantarum (5 x 10^7 CFU), *Lentinula edodes*, *Grifola frondosa*, *Agaricus Blazei Murrill*, *Trametes versicolor*, *Chlorella Pyrensoida, Ptilota plumosa*, *Spirulina maxima*, and *Aphanizomenon flos-aquae.* Control: Inert ingredients Stepwise dosage: week 1: 1x/day, week 2: 3x/day, week 3: 6x/day, week 4: 9x/day, week 5 for 8 weeks: 12x/day.		
Lee, 2014[303]	Randomised, placebo controlled, double-blind trial; II	Republic of Korea; outpatient clinic	*n* = 50 Intervention: *n* = 25; 19-65 years; overweight females (BMI 28.3±1.3 kg/m^2)	Intervention: Bofutsushosan (3g) and *Streptococcus thermophiles* (KCTC 11870BP), *L. plantarum* (KCTC 10782BP), *L.*	LMR at 5 weeks	LMR: n.s difference between baseline and 5 weeks in intervention groups (2.7±1.9 vs 2.2±1.5; p=0.391).

			Control: *n* = 25; 19-65 years; overweight females (BMI 28.5±1.7 kg/m^2)	*acidophilus* (KCTC11906BP), *L. rhamnosus* (KCTC 12202BP), *Bifidobacterium lactis* (KCTC 11904BP), *Bifidobacterium longum* (KCTC 12200BP), and *Bifidobacterium breve* (KCTC 12201BP) (Duolac 7) 5x10^7 CFU, 2x/day, 8 weeks Control: Bofutsushosan (3g) and placebo, 2x/day, 8 weeks		
Bonfrate, 2020[304]	Randomised, placebo controlled, double-blind, cross-over trial; II	Italy; outpatient clinic	*n* = 30 (*n* = 25 analysed) Intervention: *n* = 15; 50.0±11.0 years; 80% female; IBS. Control: *n* = 10; 46.0±10.0 years; 60% female; IBS.	Intervention: *Bifidobacterium longum* BB536 and *L. rhamnosus* HN001 5×10^7 CFU, 1x/day, 30 days. Control: placebo containing maize starch	LMR at 30 days	LMR: n.s difference between intervention and placebo at 30 days (p>0.05).

				and maltodextrins, 1x/day, 30 days. Wash-out of 15 days.		
Mokkala, 2018[299]	Randomised, placebo controlled, double-blind trial; II	Finland; outpatient clinic	*n* = 200 (*n* = 199 analysed) Intervention 1: *n* = 51; 30.5±4.9 years; overweight and obese pregnant woman (BMI 30.3±5.1 kg/m^2) Intervention 2: *n* = 49; 30.7±5.5 years; overweight and obese pregnant woman (BMI 30.3±4.4 kg/m^2) Intervention 3: *n* = 49; 30.1±5.3 years; overweight and obese pregnant woman (BMI 30.0±4.1 kg/m^2) Control: *n* = 51; 30.2±3.9 years; overweight and obese	Intervention 1: B420 10^{10} CFU and *L. rhamnosus* HN001 10^{10} CFU 1x/day, 21.8±2.6 weeks Intervention 2: 1.2g of omega 3 consisting of 79.6% DHA and 9.7% EPA 2x/day, 21.5±2.5 weeks Intervention 3: B420 10^{10} CFU and *L. rhamnosus* HN001 10^{10} CFU 1x/day and 1.2g of omega 3 consisting of 79.6% DHA and 9.7% EPA 2x/day, 21.3±2.5 weeks Control: placebo containing microcrystalline	Serum zonulin at late pregnancy, 21 weeks LPS at late pregnancy, 21 weeks	Serum zonulin: n.s difference between early and late pregnancy with probiotic (mean change: +6.5±12.3ng/ml; 95%CI +3.0, +10.0; p>0.05) LPS: n.s difference between early and late pregnancy with probiotic (mean change: +0.03±0.05EU/ml; 95%CI +0.018, +0.041; p>0.05).

			pregnant woman (BMI 29.8±4.5 kg/m^2)	cellulose 1x/day and capric acid and caprylic 2x/day, 21.3±2.3 weeks		
Depommier, 2019[292]	Randomised, placebo controlled, double-blind trial; II	Belgium; research centre	*n* = 40 (*n* = 32 analysed) Intervention 1: *n* = 12; 52.7±7.2 years; 67% female; overweight and obese adults with insulin-resistance (BMI 39.8±4.8 kg/m^2) Intervention 2: *n* = 9; 52.9±8.6 years; 33% female; overweight and obese adults with insulin-resistance (BMI 36.8±3.7 kg/m^2) Control: *n* = 11; 49.5±9.7 years; 55% female; overweight and obese adults with insulin-resistance (BMI 37.6±5.8 kg/m^2)	Intervention 1: Heat-killed *Akkermansia muciniphila* 10^{10} CFU, 1x/day, 3 months Intervention 2: Alive *Akkermansia muciniphila* 10^{10} CFU, 1x/day, 3 months Control: placebo	LPS at 3 months	LPS: significant decrease between baseline and 3 months in heat-killed group (mean change -0.28±0.09; p=0.044). n.s difference between baseline and 3 months in the alive group (mean change -0.24±0.47; p=0.29).
Probiotic drinks						

Pražnikar, 2020[302]	Randomised, placebo controlled, cross-over trial; II	Slovenia; hospital	*n* = 28 (*n* = 27 analysed); 46.0±8.4 years; 54% female; overweight adults (BMI 29.1±4.6 kg/m^2)	Intervention: 300 ml of kefir milk 1x/day, 3 weeks Control: 300 ml of unfermented milk, 1x/day, 3 weeks between intervention and control phase 1-week washout period Kefir culture: commercially available kefir containing *L. parakefiri, L. kefiri, L. kefiranofaciens, Kluyveromyces marxianus, Kazachstania exigua, Rhodosporidium kratochvilovae*	Serum zonulin at 3 weeks	Serum zonulin: significantly decreased compared to baseline in the kefir group after 3 weeks (p=0.018).
Zeng, 2008[308]	Randomised, placebo controlled, single-blind trial; II	China; outpatient clinic	*n* = 30 (*n* = 29 analysed) Intervention: *n* = 14; 44.6±12.4 years; 29% female; IBS-D	Intervention: 200g of fermented milk 2x/day before meals, 4 weeks	LMR at 4 weeks	LMR: significantly decreased from 0.038 at baseline to 0.023 after 4 weeks in the

			Control: *n* = 15; 45.8±9.2 years; 40% female; IBS-D	Control: 200ml of milk 2x/day before meals, 4 weeks Fermented milk culture: *Streptococcus thermophilus, L. bulgaricus, L. acidophilus* and *Bifidobacterium longum.*		fermented milk group (p=0.004). The proportion of patients with IP significantly decreased from 64.3% at baseline to 28.6% after 4 weeks in the fermented milk group (p=0.023). No comparison with placebo group, however, no significant change in placebo group between baseline and 4 weeks.
Stadlbauer, 2015[296] Leber, 2012[298]	Randomised, controlled, trial; II	Austria; outpatient clinic	*n* = 30 (*n* = 28 analysed) Intervention: *n* = 13; 51.5±11.4 years; 30% female; metabolic syndrome Control: *n* = 15; 54.5±8.9 years; 40%	Intervention: 65ml of milk drink (Yakult light®) containing *L. casei* Shirota 10^8/ml (6.5 x 10^9 CFU) 3x/day, 12 weeks Control: standard therapy	Serum and stool zonulin at 12 weeks. LMR at 12 weeks	Serum and stool zonulin: n.s difference between baseline and 12 weeks (p>0.05). LMR: n.s difference between probiotic drink and standard therapy after 12 weeks

			female; metabolic syndrome			(0.030±0.016 vs 0.037±0.029; p=0.522).
Macnaughtan, 2020[306]	Randomised, placebo controlled, double-blind, trial; II	United Kingdom; hospital	*n* = 87 (*N* = 68 analysed) Intervention: *n* = 44; 56.2±8.5 years; 27% female; liver cirrhosis Control: *n* = 43; 58.2±9.2 years; 30% female; liver cirrhosis	Intervention: 65ml of milk drink (Yakult light®) containing *L. casei* Shirota 10^8/ml (6.5 x 10^9 CFU) 3x/day, 6 months. Control: placebo drink without bacteria	LRR at 6 months	LRR: n.s difference between probiotic drink and placebo after 6 months (0.03 [0.02-0.05] vs 0.03 [0.03-0.03]; p=0.76).
Prebiotic						
Wilms, 2019[310]	Randomised, placebo controlled, double-blind trial; II	Netherlands; research centre	*n* = 100 Intervention 1: *n* = 25; 23.4±4.5 years; 68% female; healthy young adults. Control 1: *n* = 27; 22.8±4.1 years; 48% female; healthy young adults. Intervention 2: *n* = 24; 69.5±3.1 years; 38% female; healthy elderly adults.	Intervention: Pectin derived from sugar beet (GENU® BETA pectin) 7.5g 2x/day, 4 weeks. Control: placebo containing 7.5g of maltodextrin 2x/day, 4 weeks.	LMR at 4 weeks	LMR: n.s difference between prebiotic and placebo in young adults or the elderly adults after 4 weeks (p=0.861).

			Control 2: *n* = 24; 69.8±2.4 years; 50% female; healthy elderly adults.			
Salden, 2018[309]	Randomised, placebo controlled, double-blind trial; II	Netherlands; research centre	*n* = 47 Intervention 1: *n* = 16; 49.0±17.0 years; 38% female; overweight and obese adults (BMI 30.2±1.9 kg/m^2). Intervention 2: *n* = 17; 47.0±15.0 years; 59% female; overweight and obese adults (BMI 31.5±2.2 kg/m^2). Control: *n* = 14; 49.0±17.0 years; 43% female; overweight and obese adults (BMI 31.4±3.1 kg/m^2).	Intervention 1: 3.75g of arabinoxylan and 3.75g of maltodextrin 2x/day, 6 weeks. Intervention 2: 7.5g of arabinoxylan 2x/day, 6 weeks. Control: placebo containing 7.5g of maltodextrin 2x/day, 6 weeks.	LRR at 6 weeks	LRR: n.s difference between low dose (0.060 vs 0.065; p=0.464) or high dose (0.065 vs 0.065; p=0.219) of arabinoxylan compared to placebo at 6 weeks.
Müller, 2020[312]	Randomised, placebo controlled,	Netherlands; research centre	*n* = 48 Intervention: *n* = 24; 36.1±12.9 years; 75% female; slow transit	Intervention: 5g of arabinoxylan 3x/day, 12 weeks.	LRR at 12 weeks	LRR: n.s difference between prebiotic and placebo after 12 weeks (p>0.05).

	double-blind trial; II		time without constipation. Control: *n* = 24; 35.7±11.0 years; 75% female; slow transit time without constipation.	Control: placebo containing 5g of maltodextrin 3x/day, 12 weeks.		
Ried, 2020[311]	Non-randomised clinical trial; III-3	Australia; outpatient clinic	*n* = 50 (*n* = 42 analysed); mean age of 50 years; 76% female; moderate gastrointestinal problems	Intervention: curcumin 6.38mg, glutamine 2.5g, quercetin 200mg, glucosamine 415mg, aloe vera 2.5mg, slippery elm 500mg, guar gum 100mg, pectin 100mg, peppermint oil 3mg, dibasic sodium diphosphate 260mg (Nutrition Care Gut Relief Formula) 5g/day for 4 weeks followed by 10g/day for 4 weeks followed by either 5g	LMR at 12 weeks	LMR: significantly decreased between baseline and 12 weeks (0.04±0.004 vs 0.03±0.001; p<0.0001).

				(30%) or 10g (65%) per day.		
Synbiotic						
Stenman, 2016[313]	Randomised, placebo controlled, double-blind trial; II	Finland; research centre	*n* = 225 (*n* = 134 analysed) Intervention 1: *n* = 25; 49.1±11.9 years; 72% female; overweight and obese adults (BMI 30.9±1.9 kg/m^2) Intervention 2: *n* = 36; 48.6±10.9 years; 77% female; overweight and obese adults (BMI 31.2±1.6 kg/m^2) Intervention 3: *n* = 37; 47.1±10.9 years; 84% female; overweight and obese adults (BMI 31.2±2.0 kg/m^2) Control: *n* = 36; 48.3±8.6 years; 72% female; overweight	Intervention 1: B420 10^{10} CFU in 12g of microcrystalline cellulose 1x/day, 6 months Intervention 2: 12g of polydextrose 1x/day, 6 months Intervention 3: B420 10^{10} CFU in 12g of polydextrose 1x/day, 6 months Control: placebo containing 12g of microcrystalline cellulose 1x/day, 6 months	Serum zonulin and LPS at 2, 4, 6 and 7 months	Serum zonulin: n.s difference between baseline and 6 months in B420 group (58.4±11.4 vs 57.1±8.3), polydextrose group (55.5±9.1 vs 58.4±12.0) or B420 + polydextrose group (64.6±14.2 vs 63.4±13.0) (p=0.10). Serum LPS: significant increase in B420 + polydextrose group compared to placebo (+9.1±40 vs −26±108) at 6 months (p=0.007).

			and obese adults (BMI 31.0±2.2 kg/m^2)			
Del Piano, 2014[319]	Randomised, placebo controlled, double-blind trial; II	Italy; research centre	*n* = 25 Intervention: *n* = 13; 37.7±11.2 years; 56% female; healthy adults. Control: *n* = 12; 37.7±11.2 years; 56% female; healthy adults.	Intervention: 250mg of tara gum and *Streptococcus thermophilus* 1 x 10^7 CFU 1x/day, 4 weeks. Control: placebo containing 2.5g of maltodextrin 1x/day, 4 weeks.	LMR at 4 and 6 weeks.	LMR: significant difference between symbiotic and placebo at 4 weeks (0.014±0.004 vs 0.019±0.007; p=0.045) and 6 weeks (0.015±0.006 vs 0.021±0.007; p=0.033).
Horvath, 2019[317]	Randomised, placebo controlled, double-blind trial; II	Austria; outpatient clinic	*n* = 41 (*n* = 26 analysed) Intervention: *n* = 12; 61 mean years (95% CI: 56-65); 8% female; diabesity (BMI 33 mean; 95% CI: 31-34 kg/m^2). Control: *n* = 14; 59 mean years (95% CI: 54-63); 43% female; diabesity (BMI 34	Intervention: *Bifidobacterium bifidum* W23, *Bifidobacterium lactis* W51, *Bifidobacterium lactis* W52, *L. acidophilus* W37, *Lactobacillus brevis* W63, *L. casei* W56, *L. salivarius* W24, *Lactococcus lactis* W19, and *Lactococcus lactis* W58 (Ecologic® Barrier) 1.5x10^{10} CFU	Serum zonulin at 3 and 6 months	Serum zonulin: significant difference between symbiotic and placebo at 3 months (-0.04 [-0.2; 0.1] vs +0.3 [-0.05; 0.6] ng/ml; p=0.004).

			mean; 95% CI: 32-36 kg/m^2).	and FOS 3.46g, GOS 2.54g, konjac 2g, vitamin D 0.66mcg, vitamin B2 0.18mg, calcium 106.66mg, zinc 1.34mg (Omnilogic Plus) 1x/day, 6 months. Control: placebo containing maize starch, maltodextrins, vegetable protein, potassium chloride, magnesium sulphate, amylases, and manganese sulphate 1x/day. 6 months.		
Ferolla, 2016[318]	Randomised, controlled, trial; III-1	Brazil; outpatient clinic	*n* = 50 (*n* = 49 analysed); 57 median years (25-74); 76% female; NASH	Intervention: 2.4g of PHGG and 1.6g of inulin, *L. reuteri* 1 x 10^8 CFU, 2x/day, plus healthy diet, 3 months. Control: healthy diet	LMR at 3 months	LMR: n.s difference between baseline and 3 months (p=0.737).

Moser, 2019[315]	Non-randomised clinical trial; III-3	Austria; outpatient clinic	*n* = 10; 46 (37-53) median years; 50% female; IBS-D	Intervention: *L. casei W56, Lactococcus lactis W19, L. acidophilus W22, Bifidobacterium lactis W52, L. paracasei W20, L. plantarum W62, Bifidobacterium lactis W51, Bifidobacterium bifidum W23 and L. salivarius W24* (7.5 x 10^9 CFU), corn starch, inulin, and FOS (Omni-biotic® Stress Repair) 2x/day, 4 weeks.	Stool zonulin at 4 weeks	Stool zonulin: significantly decreased between baseline and 4 weeks (67 [38-92] vs 36 [20-48] ng/ml; p=0.035).
Leblhuber, 2018[294]	Non-randomised clinical trial; III-3	Austria; outpatient clinic	*n* = 20; 76.7±9.7 years; 45% female; Alzheimer's disease	Intervention: *L. casei W56, Lactococcus lactis W19, L. acidophilus W22, Bifidobacterium lactis W52, L. paracasei W20, L. plantarum W62, Bifidobacterium lactis W51, Bifidobacterium*	Stool zonulin at 4 weeks	Stool zonulin: significantly decreased between baseline and 4 weeks (93.1±56.3µg/L vs 66.6±54.2µg/L; p=0.01).

				bifidum W23 and L. salivarius W24 (7.5 x 10^9 CFU), corn starch, inulin, and FOS (Omni-biotic® Stress Repair) 1x/day, 4 weeks.		
Horvath, 2020[316]	Non-randomised clinical trial; III-3	Austria; outpatient clinic	*n* = 49 (*n* = 36 analysed); 63 mean years (95% CI: 59-67); 47% female; history of PPI use (63 mean months (95% CI: 44-82).	Intervention: corn starch, maltodextrin, FOS, inulin, *Bacillus coagulans* W183, *Bacillus subtilis* W201, *Bifidobacterium bifidum* W23, *Bifidobacterium lactis* W52, *Bifidobacterium lactis* W51, *L. acidophilus* W37, *L. acidophilus* W22, *L. casei* W56, *L. rhamnosus* W71, *L. salivarius* W24, *Lactococcus lactis* W19, *Propionibacterium freudenreichii* W200 2 x	Stool zonulin at 3 and 6 months	Stool zonulin: significant decrease after 3 months in participants with elevated (>50ng/mg) zonulin at baseline (−46.3 ng/mg; 95% CI: −71.4; −21.2; p<0.001).

				10^9 CFU/g, 4g 1x/day, 3 months.		
Probiotic and prebiotic on NSAIDs induced IP						
Mujagic, 2017[321]	Randomised, placebo controlled, cross-over, double-blind trial; II	Netherlands; research centre	*n* = 10; 26.3±10.1 years; 70% female; healthy adults	Intervention 1: *L. plantarum* WCFS1 2.6 x 10^{10} CFU 2x/day, 7 days Intervention 2: *L. plantarum* CIP48 2.4 x 10^{10} CFU 2x/day, 7 days Intervention 3: *L. plantarum* TIFN101 5.9 x 10^{10} CFU 2x/day, 7 days Control: Maltodextrin and glucose 2x/day, 7 days NSAIDs: 75mg 9h prior and 50mg 1h prior to LRR	LRR at 7 days	LRR: n.s difference between baseline with NSAIDs compared with any *L. plantarum* at 7 days (WCFS1: 0.047 vs 0.076; CIP48: 0.069 vs 0.075; TIFN101: 0.057 vs 0.065; p>0.05).

				Washout of 4 weeks between each test period		
Gotteland, 2001[322]	Randomised, controlled, trial; III-1	Chile; research centre	*n* = 18; 23.1±4.3 years; 61% female; healthy adults	Intervention 1: Dairy based product containing alive *L. rhamnosus* GG (> 10^7 CFU/mL), *L. helveticus* (> 10^7 CFU/mL), and *L. acidophilus* (> 10^7 CFU/mL) 80ml 3x/day, 5 days Intervention 2: Dairy based product containing heat-killed *L. rhamnosus* GG (> 10^7 CFU/mL), *L. helveticus* (> 10^7 CFU/mL), and *L. acidophilus* (> 10^7 CFU/mL) 80ml 3x/day, 5 days Control: NSAID only	LMR at 5 days	LMR: n.s difference between baseline with NSAIDs (2.93%; 1.96-3.90) compared with either alive (2.43%; 1.25-3.61) or heat-killed (2.02%; 1.53-2.51) probiotics (p>0.05).

				NSAIDs: 75mg 9h prior and 50mg 1h prior to LRR Washout of 3 weeks between each test period		
Ganda Mall, 2020[320]	Randomised, placebo controlled, trial; II	Sweden; research centre	*n* = 51 (*n* = 49 analysed) Intervention 1: *n* = 17; 69.0 mean years; 47% female; healthy elderly adults. Intervention 2: *n* = 15; 69.0 mean years; 40% female; healthy elderly adults. Control: *n* = 17; 70.5 mean years; 44% female; healthy elderly adults.	Intervention 1: 12g of arabinoxylan 1x/day, 6 weeks. Intervention 2: 12g of oat β-glucan 1x/day, 6 weeks. Control: placebo containing 12g of maltodextrin 1x/day, 6 weeks. NSAIDs: 75mg 9h prior and 50mg 1h prior to LRR	LRR at 6 weeks	LRR: n.s between baseline and 6 weeks arabinoxylan (0.055; 0.045-0.125 IQR) or oat β-glucan (0.057; 0.042-0.090 IQR) and placebo (0.064; 0.028-0.098) (p>0.05).
Wilms, 2016[314]	Randomised, controlled, double-blind trial; II	Netherlands; research centre	*n* = 20 Intervention: *n* = 10; 19.7 median years;	Intervention: 10g of FOS and *Bifidobacterium bifidum* W23, *Bifidobacterium*	LRR at 2 weeks	LRR: n.s difference between baseline with NSAIDs compared with synbiotic treatment and

	20% female; healthy adults. Control: *n* = 10; 21.7 median years; 70% female; healthy adults.	*lactis* W51, *Bifidobacterium lactis* W52, *L. acidophilus* W22, *L. casei* W56, *L. paracasei* W20, *L. plantarum* W62, *L. salivarius* W24, and *Lactococcus lactis* W19 (Ecologic® 825) $1.5x10^{10}$ CFU, 2x/day, 2 weeks. Control: 10g of maltodextrin 2x/day, 2 weeks. NSAIDs: 75mg 9h prior and 50mg 1h prior to LRR	Serum zonulin at 2 weeks	NSAIDs at 2 weeks (0.064; 0.046–0.106 IQR vs 0.055; 0.037–0.072 IQR; p=0.203). Serum zonulin: n.s difference between symbiotic and control after NSAIDs use at 2 weeks (13.2ng/ml vs 14.7ng/ml; p=0.650).

Abbreviations: B420, Bifidobacterium animalis ssp. lactis 420; BMI, body mass index; CDAI, Crohn's disease activity index; CFU, colony forming units; CI, confidence interval; DHA, docosahexaenoic acid; EPA, eicosapentaenoic acid; FOS, fructooligosaccharides; GOS, galactooligosaccharides; h, hour; IBS-D, diarrhoea-predominant irritable bowel syndrome; IBS, irritable bowel syndrome; IP, increased intestinal permeability; IQR, median interquartile range; L, Lactobacillus; LMR, lactulose/mannitol ratio; LPS, lipopolysaccharide; LRR, lactulose/rhamnose ratio; *n*, number of participants; n.s, not statistically significant; NASH, nonalcoholic steatohepatitis; NSAIDs, nonsteroidal anti-inflammatory drugs; PHGG, partially hydrolyzed guar gum; PPI, proton pump inhibitor; SMD, standardised mean difference; T2D, type 2 diabetes; UC, ulcerative colitis.

6.1.9.2.2.5RISK OF BIAS ASSESSMENT

The following are the results from a risk of bias assessment (Table 6.10, Table 6.11 and Table 6.12). A summary of the results is found in the IP Guideline (Section 8.12).

Table 6.10 Risk of bias assessment in randomised trials

	Randomisation process	Deviations from intended interventions	Missing outcome data	Measurement of the outcome	Selection of the reported result	Overall
Bonfrate, 2020[304]	Some	Low	High	High	Low	High
de Roos, 2017[305]	Low	Low	Low	Low	Low	Low
Del Piano, 2014[319]	Low	Low	Low	Low	Low	Low
Depommier, 2019[292]	Some	Low	High	High	Some	High
Ferolla, 2016[318]	High	High	Some	Low	Low	High
Ganda Mall, 2020[320]	Some	Low	Low	Low	Low	Some
Garcia Vilela, 2008[297]	Low	Low	Low	Some	Some	Some
Gotteland, 2001[322]	Some	High	Low	Low	High	High
Horvath, 2019[317]	Some	Low	High	High	High	High
Kim, 2006[300]	Low	Low	Low	Low	Low	Low
Kwak, 2014[293]	Low	Low	Low	Low	Some	Some
Leber, 2012[298]	Some	Some	High	High	Some	High
Lee, 2014[303]	Low	High	Low	Low	Low	High
Macnaughtan, 2020[306]	Low	Low	High	Low	Low	High

Mokkala, 2018[299]	Low	Low	Low	High	Low	High
Mujagic, 2017[321]	Low	Low	Low	Low	Low	Low
Müller, 2020[312]	Low	Low	Low	Low	Low	Low
Palacios, 2020[291]	Low	Low	High	High	High	High
Pražnikar, 2020[302]	Some	Some	Low	Low	Some	Some
Salden, 2018[309]	Low	Low	Low	Low	Low	Low
Stadlbauer, 2015[296]	Some	Some	High	High	Some	High
Stenman, 2016[313]	Some	Low	High	High	Low	High
Szulińska, 2018[307]	Low	Low	Low	High	Low	High
Wegh, 2019[84]	Some	High	Low	Some	Low	High
Wilms, 2019[310]	Low	Some	High	Low	Low	High
Wilms, 2016[314]	Some	Low	Low	Low	Low	Some
Zeng, 2008[308]	Low	Some	Low	Low	Low	Some
Percentages						
Low risk	55.6%	70.4%	63%	59.3%	66.9%	22.2%
Some concerns	40.7%	14.8%	3.7%	7.4%	22.6%	22.2%
High risk	3.7%	14.8%	33.3%	33.3%	10.5%	55.6%

Abbreviations: Low = low risk of bias; Some = some concerns for risk of bias; High = high risk of bias.

Table 6.11 Risk of bias in systematic reviews assessment

Review	Phase 2				Phase 3
	1. Study eligibility criteria	2. Identification and selection of studies	3. Data collection and study appraisal	4. Synthesis and findings	Risk of bias in the review
Moludi, 2020[295]	Low	Unclear	High	High	High
Ramezani Ahmadi, 2020[301]	Low	Low	Low	Low	Low

Abbreviations: Low = low risk; High = high risk; Unclear = unclear risk.

Table 6.12 Risk of bias in non-randomised studies

Study	Confounding	Selection of participants	Classification of interventions	Deviations from intended interventions	Missing data	Measurement of outcome	Selection of reported results	Overall
Horvath, 2020[316]	Moderate	Serious	Low	Low	Serious	Moderate	Serious	Serious
Leblhuber, 2018[294]	Moderate	Moderate	Low	Low	Low	Moderate	Low	Moderate
Moser, 2019[315]	Moderate	Low	Low	Low	Low	Moderate	Low	Moderate
Ried, 2020[311]	Moderate	Moderate	Moderate	Low	Moderate	Moderate	Moderate	Moderate

Abbreviations: Low = low risk of bias; Moderate = moderate risk of bias; Serious = serious risk of bias; critical = Critical risk of bias.

6.1.9.2.3 NHMRC EVIDENCE STATEMENT

The following are the results from completing the NHMRC Evidence Statement (Table 6.13). Individual grade for each of the five domains are provided along with the overall grade for the evidence-based recommendation. Furthermore, consensus-based recommendations are listed below (Table 6.14).

Table 6.13 Summary of the NHMRC evidence statement for evidence-based recommendations: Probiotic, prebiotic and synbiotic

Recommendations	Grade
Probiotic	
Recommendation 2.1: There is insufficient evidence to form a recommendation on the use of probiotics as a collective group for the treatment of people with intestinal permeability.	**Grade: D**
Evidence base – number of studies, level of evidence, risk of bias in the included studies	**D**
Consistency – between studies if more than one study	**D**
Clinical impact – of the intervention, diagnosis, prognosis, aetiology, screening	**C**
Generalisability – how well the body of evidence matches the population and clinical setting	**C**
Applicability – relevance to Australian health care context in terms of services, delivery and cultural factors	**C**
Recommendation 2.2: Clinicians may consider using *Saccharomyces boulardii* supplementation in the treatment of people with intestinal permeability.	**Grade: C**
Evidence base – number of studies, level of evidence, risk of bias in the included studies	**C**
Consistency – between studies if more than one study	**N/A**
Clinical impact – of the intervention, diagnosis, prognosis, aetiology, screening	**B**
Generalisability – how well the body of evidence matches the population and clinical setting	**C**
Applicability – relevance to Australian health care context in terms of services, delivery and cultural factors	**B**

Recommendation 2.3: Clinicians may consider the use of effective probiotics for a period of 3 months when treating people with intestinal permeability.	**Grade: C**
Evidence base – number of studies, level of evidence, risk of bias in the included studies	**A**
Consistency – between studies if more than one study	**N/A**
Clinical impact – of the intervention, diagnosis, prognosis, aetiology, screening	**B**
Generalisability – how well the body of evidence matches the population and clinical setting	**A**
Applicability – relevance to Australian health care context in terms of services, delivery and cultural factors	**B**
Probiotic drinks	
Recommendation 2.6: People with intestinal permeability should consider the consumption of fermented milk products such as kefir.	**Grade: B**
Evidence base – number of studies, level of evidence, risk of bias in the included studies	**C**
Consistency – between studies if more than one study	**A**
Clinical impact – of the intervention, diagnosis, prognosis, aetiology, screening	**B**
Generalisability – how well the body of evidence matches the population and clinical setting	**B**
Applicability – relevance to Australian health care context in terms of services, delivery and cultural factors	**A**
Recommendation 2.7: People with intestinal permeability may consider NOT consuming Yakult light®.	**Grade: C**
Evidence base – number of studies, level of evidence, risk of bias in the included studies	**D**
Consistency – between studies if more than one study	**A**
Clinical impact – of the intervention, diagnosis, prognosis, aetiology, screening	**C**
Generalisability – how well the body of evidence matches the population and clinical setting	**B**
Applicability – relevance to Australian health care context in terms of services, delivery and cultural factors	**A**
Prebiotic	
Recommendation 2.8: There is insufficient evidence to form a recommendation on the use of prebiotics as a collective group for the treatment of people with intestinal permeability.	**Grade: D**

Evidence base – number of studies, level of evidence, risk of bias in the included studies	**D**
Consistency – between studies if more than one study	**D**
Clinical impact – of the intervention, diagnosis, prognosis, aetiology, screening	**C**
Generalisability – how well the body of evidence matches the population and clinical setting	**C**
Applicability – relevance to Australian health care context in terms of services, delivery and cultural factors	**C**
Synbiotic	
Recommendation 2.12: Clinicians may consider the use of effective synbiotic in the treatment of people with intestinal permeability.	**Grade: C**
Evidence base – number of studies, level of evidence, risk of bias in the included studies	**D**
Consistency – between studies if more than one study	**B**
Clinical impact – of the intervention, diagnosis, prognosis, aetiology, screening	**C**
Generalisability – how well the body of evidence matches the population and clinical setting	**B**
Applicability – relevance to Australian health care context in terms of services, delivery and cultural factors	**B**
Recommendation 2.13: Clinicians may consider the use of effective synbiotic for a period of 3 months when treating people with intestinal permeability.	**Grade: C**
Evidence base – number of studies, level of evidence, risk of bias in the included studies	**A**
Consistency – between studies if more than one study	**N/A**
Clinical impact – of the intervention, diagnosis, prognosis, aetiology, screening	**B**
Generalisability – how well the body of evidence matches the population and clinical setting	**A**
Applicability – relevance to Australian health care context in terms of services, delivery and cultural factors	**B**
Recommendation 2.14: Clinicians may consider NOT using polydextrose and *Bifidobacterium animalis ssp. lactis* 420 in the treatment of people with intestinal permeability.	**Grade: C**
Evidence base – number of studies, level of evidence, risk of bias in the included studies	**D**
Consistency – between studies if more than one study	**B**

Clinical impact – of the intervention, diagnosis, prognosis, aetiology, screening	**C**
Generalisability – how well the body of evidence matches the population and clinical setting	**C**
Applicability – relevance to Australian health care context in terms of services, delivery and cultural factors	**C**
NSAID induced intestinal permeability	
Recommendation 2.16: Clinicians should consider NOT using probiotics for the treatment of people with nonsteroidal anti-inflammatory drug induced intestinal permeability.	**Grade: C**
Evidence base – number of studies, level of evidence, risk of bias in the included studies	**C**
Consistency – between studies if more than one study	**A**
Clinical impact – of the intervention, diagnosis, prognosis, aetiology, screening	**B**
Generalisability – how well the body of evidence matches the population and clinical setting	**B**
Applicability – relevance to Australian health care context in terms of services, delivery and cultural factors	**A**
Recommendation 2.17: Clinicians should consider NOT using prebiotics for the treatment of people with nonsteroidal anti-inflammatory drug induced intestinal permeability.	**Grade: C**
Evidence base – number of studies, level of evidence, risk of bias in the included studies	**C**
Consistency – between studies if more than one study	**N/A**
Clinical impact – of the intervention, diagnosis, prognosis, aetiology, screening	**B**
Generalisability – how well the body of evidence matches the population and clinical setting	**B**
Applicability – relevance to Australian health care context in terms of services, delivery and cultural factors	**A**
Recommendation 2.18: Clinicians should consider NOT using synbiotics for the treatment of people with nonsteroidal anti-inflammatory drug induced intestinal permeability.	**Grade: C**
Evidence base – number of studies, level of evidence, risk of bias in the included studies	**C**
Consistency – between studies if more than one study	**N/A**
Clinical impact – of the intervention, diagnosis, prognosis, aetiology, screening	**B**

Generalisability – how well the body of evidence matches the population and clinical setting	**B**
Applicability – relevance to Australian health care context in terms of services, delivery and cultural factors	**A**

Table 6.14 Summary of consensus-based recommendations and practice points: Dietary recommendations

No.	Recommendation
2.4	Clinicians may consider researching probiotic strains for their effectiveness before using them to treat people with intestinal permeability.
2.5	Clinicians may consider the use of probiotics which are supported by pre-clinical research in conjunction with other treatment interventions for the management people with intestinal permeability.
2.9	Clinicians may consider researching prebiotic for their effectiveness before using them in the treatment of people with intestinal permeability.
2.10	Clinicians may consider the use of prebiotic which are supported by pre-clinical research in conjunction with other treatment interventions for the management people with intestinal permeability.
2.11	Clinicians may consider NOT using polydextrose in the treatment of people with intestinal permeability.
2.15	Clinicians may consider the use of synbiotic which are supported by pre-clinical research in conjunction with other treatment interventions for the management people with intestinal permeability.

6.1.9.3 AMINO ACID SUPPLEMENTATION

6.1.9.3.1 CLINICAL QUESTIONS

Clinical Question 5: In Australian adults with increased intestinal permeability, what are the benefits of oral amino acid supplementation for the treatment of increased intestinal permeability?

Clinical Question 6: In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for oral amino acid supplementation use?

6.1.9.3.2 EVIDENCE SUMMARY AND OVERALL QUALITY OF EVIDENCE

6.1.9.3.2.1 GLUTAMINE

There is a limited number (n = 3) of studies exploring the effects of glutamine on altered IP (Table 6.15).[192,311,323] Two RCT[192,323] and one non-randomised clinical trial[311] met the inclusion criteria and were included. The included studies assessed the effect of glutamine on intestinal integrity for between two and three months. Participants involved in the articles were diagnosed with IBS-D (n = 1),[192] Crohn's disease in remission (n = 1)[323] or had a functional gastrointestinal disorder (n = 1).[311] The dosage of glutamine varied between 2.5g to 15g per day, with one study using 0.5g/kg of glutamine of ideal body weight per day.[192,311,323]

The use of glutamine supplementation in people with IP resulted in consistently beneficial effects on intestinal integrity. A randomised, placebo-controlled, double-blind trial used 5g of glutamine three times daily in people with IBS-D.[192] After two months, IP had a significant decrease compared to baseline (0.11±0.03

vs. 0.04±0.01; p<0.0001).[192] Furthermore, in people taking the glutamine supplement, there was a significant correlation between irritable bowel syndrome severity and improvement of IP (r=0.72; p<0.001).[192] Another RCT investigated the effect of 0.5g/kg of glutamine on ideal body weight per day in Crohn's disease patients in remission.[323] The two-month study found both the glutamine and control group (whey protein) significantly improved IP.[323] Specifically, glutamine supplementation reduced the median value of LMR from 0.071 (0.041–0.254, range) to 0.029 (0.006–0.090, range) after two months, and whey protein also reduced the median value of LMR from 0.067 (0.040–0.136, range) to 0.033 (0.009–0.077, range) in the same period.[323] No significant difference was found between the glutamine group and whey protein group after two months (0.029 vs. 0.033; p>0.05).[323] An Australian based study of patients with a functional gastrointestinal disorder explored the effects of 2.5g of glutamine in combination with prebiotics, other intestinal supportive herbal medicine, and nutrients on IP.[311] This study found a significant decrease between baseline and 12 weeks in LMR (0.04±0.004 vs. 0.03±0.001; p<0.0001).[311]

6.1.9.3.2.2LACTOFERRIN

One article investigated the effects of lactoferrin on NSAID induced intestinal integrity. This RCT induced IP in healthy, non-smoking males consuming 75mg NSAID 9 hours prior and 50mg one hour before undertaking the dual sugar test. The intervention involved participants consuming 5g of recombinant human lactoferrin three times (24, 9 and 1 hour before the dual sugar test). Lactoferrin supplementation was found to significantly decrease NSAID-induced IP compared to NSAIDs and placebo (0.028 vs. 0.036; p<0.05).

Table 6.15 Evidence summary table for clinical trials

Author, Year	Study design; Level of Evidence HMRC	Country; Setting	Sample size; age in years M ± SD; gender; health condition	Intervention; control	Outcome measures; duration	Main results
Glutamine						
Zhou, 2019[192]	Randomised, placebo controlled, double-blind trial; II	USA; university research centre	*n* = 115 (n = 106 analysed) Intervention: n = 54; 32.4±9.5 years; 68% female; IBS-D (Rome III criteria) patients with IP Control: *n* = 52; 30.9±7.1 years; 71% female; IBS-D (Rome III criteria) patients with IP	Intervention: 5g of glutamine 3x/day, 2 months. Control: placebo containing 5g of whey protein 3x/day, 2 months.	LMR at 2 months	LMR: significant decrease in mean per-post change in intervention compared to control at 8 weeks (-0.06±0.03 vs. -0.0004±0.03; p<0.0001). LMR: significant decrease between baseline and 2 months (0.11±0.03 vs. 0.04±0.01; p<0.0001). Intervention group: IBS-SS correlates with

						improvement of LMR (*r*=0.72; p<0.001).
Benjamin, 2012[323]	Randomised, controlled, open-label trial; II	India; university research centre	*n* = 30 (n = 28 analysed) Intervention: n = 14; 35.1±10.8 years; 33% female; Crohn's disease patients in remission. Control: *n* = 14; 33.9±10.4 years; 33% female; Crohn's disease patients in remission.	Intervention: 0.5g/kg of glutamine of ideal body weight/day, 2 months. Control: 0.5g/kg of whey protein of ideal body weight/day, 2 months.	LMR at 2 months	LMR: n.s between intervention and control after 2 months (0.029 vs. 0.033; p>0.05). LMR: significant difference between baseline and 2 months in the intervention group (0.071 vs. 0.029; p=0.001). LMR: significant difference between baseline and 2 months in the control group (0.067 vs. 0.033; p=0.006).
Ried, 2020[311]	Non-randomised clinical trial; III-3	Australia; outpatient clinic	*n* = 50 (n = 42 analysed); mean age of 50 years; 76% female; moderate	Intervention: curcumin 6.38mg, glutamine 2.5g, quercetin 200mg, glucosamine 415mg, aloe vera 2.5mg, slippery elm 500mg, guar gum 100mg, pectin 100mg, peppermint oil 3mg,	LMR at 3 months	LMR: significantly decreased between baseline and 3 months (0.04±0.004 vs.

			gastrointestinal problems	dibasic sodium diphosphate 260mg (Nutrition Care Gut Relief Formula) 5g/day for 4 weeks followed by 10g/day for 4 weeks followed by either 5g (30%) or 10g (65%) per day.		0.03±0.001; p<0.0001).
Lactoferrin						
Troost, 2003[324]	Randomised, placebo controlled, double-blind, cross-over trial; II	Netherlands; hospital	*n* = 15; 23.9±2.2 years; 100% male; healthy non-smoking males.	Intervention: 5g recombinant human lactoferrin three times (24, 9 and 1 hour before the LRR) and NSAIDs. Control: placebo drink three times (24, 9 and 1 hour before the LRR) and NSAIDs. Wash-out: 2 weeks NSAIDs: 75mg 9h prior and 50mg 1h prior to LRR	LRR at 1 day	LRR: significantly decrease between NSAIDs and intervention compared to NSAIDs and placebo (0.028 vs. 0.036; p<0.05).

Abbreviations: IBS-D, diarrhoea-predominant irritable bowel syndrome; IBS-SS, irritable bowel syndrome severity scoring system; IP, increased intestinal permeability; LMR, lactulose/mannitol ratio; LRR, lactulose/rhamnose ratio; *n*, number of participants; n.s, not statistically significant; NSAIDs, nonsteroidal anti-inflammatory drugs.

6.1.9.3.2.3RISK OF BIAS ASSESSMENT

The following are the results from a risk of bias assessment (Table 6.16 and Table 6.17). A summary of the results is found in the IP Guideline (Section 8.13).

Table 6.16 Risk of bias assessment in randomised trials

	Randomisation process	Deviations from intended interventions	Missing outcome data	Measurement of the outcome	Selection of the reported result	Overall
Zhou, 2019[192]	Low	Low	Low	Low	Low	Low
Benjamin, 2012[323]	Low	Low	Low	Low	Low	Low
Troost, 2003[324]	Low	Low	Low	Low	Low	Low
Percentages						
Low risk	100.0%	100.0%	100.0%	100.0%	100.0%	100.0%
Some concerns	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%
High risk	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%

Abbreviations: Low = low risk of bias; Some = some concerns for risk of bias; High = high risk of bias.

Table 6.17 Risk of bias in non-randomised studies

Study	Confounding	Selection of participants	Classification of interventions	Deviations from intended interventions	Missing data	Measurement of outcome	Selection of reported results	Overall
Ried, 2020[311]	Moderate	Moderate	Moderate	Low	Moderate	Moderate	Moderate	Moderate

Abbreviations: Low = low risk of bias; Moderate = moderate risk of bias.

6.1.9.3.3 NHMRC EVIDENCE STATEMENT

The following are the results from completing the NHMRC Evidence Statement (Table 6.18). Individual grade for each of the five domains are provided along with the overall grade for the evidence-based recommendation. Furthermore, consensus-based recommendations are listed below (Table 6.19).

Table 6.18 Summary of the NHMRC evidence statement for evidence-based recommendations: Amino Acid

Recommendations	Grade
Glutamine	
Recommendation 3.1: Clinicians should offer glutamine supplementation for the treatment of people with intestinal permeability.	**Grade: B**
Evidence base – number of studies, level of evidence, risk of bias in the included studies	**B**
Consistency – between studies if more than one study	**B**
Clinical impact – of the intervention, diagnosis, prognosis, aetiology, screening	**B**
Generalisability – how well the body of evidence matches the population and clinical setting	**B**
Applicability – relevance to Australian health care context in terms of services, delivery and cultural factors	**A**
NSAID induced intestinal permeability	
Recommendation 3.3: Clinicians should consider the use of short-term lactoferrin supplementation for the treatment of people with nonsteroidal anti-inflammatory drug induced intestinal permeability.	**Grade: B**
Evidence base – number of studies, level of evidence, risk of bias in the included studies	**B**
Consistency – between studies if more than one study	**N/A**
Clinical impact – of the intervention, diagnosis, prognosis, aetiology, screening	**B**
Generalisability – how well the body of evidence matches the population and clinical setting	**B**
Applicability – relevance to Australian health care context in terms of services, delivery and cultural factors	**B**

Table 6.19 Summary of consensus-based recommendations and practice points: Amino Acid

No.	Recommendation
3.2	Clinicians may consider the use of glutamine supplementation in conjunction with other treatment interventions for the management people with intestinal permeability.

6.1.9.4 PLANT-BASED MEDICINE SUPPLEMENTATION

6.1.9.4.1 CLINICAL QUESTIONS

Clinical Question 7: In Australian adults with increased intestinal permeability, what are the benefits of oral plant-based medicine supplementation for the treatment of increased intestinal permeability?

Clinical Question 8: In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for oral plant-based medicines use?

6.1.9.4.2 EVIDENCE SUMMARY AND OVERALL QUALITY OF EVIDENCE

6.1.9.4.2.1 PLANT-BASED MEDICINE

There is a limited number (*n* = 5) of studies exploring the effects of plant-based medicine supplementation on altered IP (Table 6.20).[300,303,311,325,326] Four RCTs[300,303,325,326] and one non-randomised clinical trial[311] were identified and included. The included studies assessed the effects of herbal medicine for between three and 12 weeks. Participants involved in the included studies were overweight (*n* = 2),[303,325] had a functional gastrointestinal disorder (*n* = 2)[300,311] or were healthy adults (*n* = 1).[326] All studies used a diverse range of plant-based medicines, with no studies using a similar combination of herbal medicines.

The use of plant-based medicines in people with IP resulted in mixed outcomes with three out of the five studies reporting no significant effect. Of the two studies that found a potential positive impact of plant-based therapies, one study used pomegranate extract[325] and the other used a combination of gastrointestinal supporting herbs and amino acids.[311] Firstly, a randomised, placebo-controlled,

double-blind, crossover trial assessed the effect of two dosages of the pomegranate extract (450mg and 1.8g) in overweight and obese adults.[325] After three weeks, only the higher dosage of pomegranate extract significantly reduced lipopolysaccharide-binding protein compared to placebo ($p<0.001$).[325] The other study reporting a beneficial effect of a plant-based therapy was an Australian based study that explored the effects of a mix of herbal medicines (aloe vera 2.5mg, slippery elm 500mg, guar gum 100mg, pectin 100mg and peppermint oil 3mg) and amino acids in patients with a functional gastrointestinal disorder.[311] This study found a significant decrease between baseline and 12 weeks in lactulose/mannitol ratio (0.04 ± 0.004 vs 0.03 ± 0.001; $p<0.0001$).[311] Considering the three studies that report no significant effect of plant-based medicines on IP, a randomised, placebo-controlled, double-blind trial investigated the effects of a multi-herbal formula, with aloe vera as the main ingredient in healthy adults.[326] After eight weeks there were no significant difference in serum zonulin between placebo and the intervention group.[326] Similar results were seen in randomised, placebo controlled, double-blind trial of patients with a functional gastrointestinal disorder.[300] The supplementation containing 820mg of barley grass and oat grass juice over a 12 week period was found to have no significant effect on lactulose/mannitol ratio between baseline and 12 weeks ($p>0.05$).[300] The last study used a randomised, placebo-controlled, double-blind study design to explore the effects of a traditional Japanese formula known as Bofutsushosan.[303] The study found no significant effect between baseline and five week in lactulose/mannitol ratio (2.7 ± 1.9 vs 2.2 ± 1.5; $p=0.391$).[303]

Table 6.20 Evidence summary table for clinical trials

Author, Year	Study design; Level of Evidence NHMRC	Country; Setting	Sample size; age in years M ± SD; gender; health condition	Intervention; control	Outcome measures; duration	Main results
Bloomer, 2020[326]	Randomised, placebo controlled, double-blind trial; II	USA; research centre	*n* = 75 Intervention 1: *n* = 15; 26.9±5.4 years; healthy adults. Intervention 2: *n* = 15; 26.3±5.7 years; healthy adults. Intervention 3: *n* = 15; 30.2±11.4 years; healthy adults. Intervention 4: *n* = 15; 28.1±7.4 years; healthy adults. Control: *n* = 15; 29.5±11.6 years; healthy adults.	Intervention 1: 2g of Advanced Ambrotose® containing aloe vera extract inner leaf gel (containing acemannan), arabinogalactan, ghatti gum, glucosamine HCL, gum tragacanth, vitamin A, beta carotene, wakame algae extract, and rice starch 1x/day, 8 weeks. Intervention 2: 4g Advanced Ambrotose® containing of aloe vera extract inner leaf gel (containing acemannan), arabinogalactan, ghatti gum, glucosamine HCL, gum tragacanth, vitamin A, beta carotene, wakame algae	Serum zonulin at 4 and 8 weeks	Serum zonulin: n.s difference between placebo and any intervention group. Serum zonulin: significantly increased between baseline and 4 weeks in intervention 4 group (p=0.04), returned to baseline levels at 8 weeks.

	extract, and rice starch 1x/day, 8 weeks. Intervention 3: 4g of Ambrotose LIFE® containing aloe vera extract inner leaf gel (containing acemannan), arabinogalactan, ghatti gum, glucosamine HCL, gum tragacanth, vitamin A, beta carotene, wakame algae extract, rice starch, RiFiber (rice bran) and modified citrus pectin with sodium Alginate 1x/day, 8 weeks. Intervention 4: 2g Ambrotose LIFE® containing of aloe vera extract inner leaf gel (containing acemannan), arabinogalactan, ghatti gum, glucosamine HCL, gum tragacanth, vitamin A, beta carotene, wakame algae extract, rice starch, RiFiber (rice bran) and modified citrus

				pectin with sodium Alginate 1x/day, 8 weeks. Control: placebo containing maltodextrin 1x/day, 8 weeks.		
Kim, 2006[300]	Randomised, placebo controlled, double-blind trial; II	America; research centre	*n* = 72 Intervention 1: *n* = 12; 46.8±13.9 years; 67% female; functional gastrointestinal disorder Intervention 2: *n* = 12; 48.6±18.1 years; 75% female; functional gastrointestinal disorder Intervention 3: *n* = 12; 41.1±12.5 years; 75% female; functional gastrointestinal disorder Intervention 4: *n* = 12; 43.8±13.6 years; 75% female; functional gastrointestinal disorder Intervention 5: *n* = 12; 47.4±17.3 years; 67%	Intervention 1: *L. acidophilus, Bifidobacterium bifidum, Bacillus subtilis, L. bulgaricus, L. lactis, and Bacillus lichenformis* (5 x 10^7 CFU), 820mg of barley grass and oat grass juice and 180 mg of ionic plant-based minerals. Intervention 2: *L. acidophilus, Bifidobacterium bifidum, L. bulgaricus, L. lactis, L. brevis, L. caucasicus, L. fermenti, L. leichmannii, L. caseii, L. plantarum, L. helveticus, and Saccharomyces boulardii* (5 x 10^7 CFU), 820mg of barley grass and oat grass juice and 180 mg of ionic plant-based minerals.	LMR at 12 weeks	LMR: n.s difference between baseline and 12 weeks in intervention groups (p>0.05). Mean change at 12 weeks for plant-based group 0.00±0.05.

	female; functional gastrointestinal disorder Control: *n* = 12; 41.5±15.8 years; 67% female; functional gastrointestinal disorder	Intervention 3: *L. acidophilus, Bifidobacterium bifidum, Bacillus subtilis, L. bulgaricus, L. lactis, and Bacillus lichenformis* (5 x 10^7 CFU). Intervention 4: 820mg of barley grass and oat grass juice and 180 mg of ionic plant-based minerals. Intervention 5: *Bacillus coagulans, Saccharomyces boulardii, Bacillus subtilis, L. salivarius, and L. plantarum* (5 x 10^7 CFU), *Lentinula edodes*, *Grifola frondosa*, *Agaricus Blazei Murrill*, *Trametes versicolor*, *Chlorella Pyrensoida, Ptilota plumosa*, *Spirulina maxima*, and *Aphanizomenon flos-aquae.* Control: Inert ingredients Stepwise dosage: week 1: 1x/day, week 2: 3x/day, week

				3: 6x/day, week 4: 9x/day, week 5 for 8 weeks: 12x/day.		
Lee, 2014[303]	Randomised, placebo controlled, double-blind trial; II	Republic of Korea; outpatient clinic	*n* = 50 Intervention: *n* = 25; 19-65 years; overweight females (BMI 28.3±1.3 kg/m^2) Control: *n* = 25; 19-65 years; overweight females (BMI 28.5±1.7 kg/m^2)	Intervention: Bofutsushosan containing 3g of *Scutellaria baicalensis*, *Glycyrrhiza uralensis*, *Platycodon grandiflorum*, *Gypsum Fibrosum*, *Atractylodes japonica*, *Rheum palmatum*, *Schizonepeta tenuifolia*, *Gardenia jasminoides*, *Paeonia lactiflora*, *Cnidium officinale*, *Angelica acutiloba*, *Mentha arvensis*, *Ledebouriella seseloides*, *Ephedra sinica*, *Forsythia suspensa*, *Zingiber officinale*, *Talcum Crystallinum, Natrii Sulfas* and probiotic containing *Streptococcus thermophiles* (KCTC 11870BP), *L. plantarum* (KCTC 10782BP), *L. acidophilus* (KCTC11906BP), *L. rhamnosus* (KCTC	LMR at 5 weeks	LMR: n.s difference between baseline and 5 weeks in intervention groups (2.7±1.9 vs. 2.2±1.5; p=0.391) or the control group (2.8±1.9 vs. 3.4±2.5; p=0.555).

			12202BP), *Bifidobacterium lactis* (KCTC 11904BP), *Bifidobacterium longum* (KCTC 12200BP), and *Bifidobacterium breve* (KCTC 12201BP) (Duolac 7) $5x10^7$ CFU, 2x/day, 8 weeks Control: Bofutsushosan (3g) and placebo, 2x/day, 8 weeks		
González-Sarrias, 2018[325]	Randomised, placebo controlled, double-blind, crossover trial; II	*n* = 50 (*n* = 49 analysed) Overweight group: *n* = 29; 43.7±3.4 years; 31% female; healthy overweight adults (BMI 28.5±1.1 kg/m^2) Obese group: *n* = 20; 48.6±7.4 years; 40% female; healthy overweight adults (BMI 33.2±3.3 kg/m^2)	Study arm 1: 450mg of pomegranate extract 1x/day, 3 weeks; 3 weeks washout; placebo 1x/day, 3 weeks; 3 weeks washout; 450mg of pomegranate extract 4x/day, 3 weeks; 3 weeks washout; placebo 4x/day, 3 weeks. Study arm 2: placebo 1x/day, 3 weeks; 3 weeks washout; 450mg of pomegranate extract 1x/day, 3 weeks; 3 weeks washout; placebo 4x/day, 3 weeks; 3 weeks washout;	LBP before and after each 3-week study period.	LBP: significant decrease between placebo and pomegranate extract taken 4x/day ($p<0.001$).

				450mg of pomegranate extract 4x/day, 3 weeks.		
Ried, 2020[311]	Non-randomised clinical trial; III-3	Australia; outpatient clinic	*n* = 50 (*n* = 42 analysed); mean age of 50 years; 76% female; moderate gastrointestinal problems	Intervention: curcumin 6.38mg, glutamine 2.5g, quercetin 200mg, glucosamine 415mg, aloe vera 2.5mg, slippery elm 500mg, guar gum 100mg, pectin 100mg, peppermint oil 3mg, dibasic sodium diphosphate 260mg (Nutrition Care Gut Relief Formula) 5g/day for 4 weeks followed by 10g/day for 4 weeks followed by either 5g (30%) or 10g (65%) per day.	LMR at 12 weeks	LMR: significantly decreased between baseline and 12 weeks (0.04±0.004 vs. 0.03±0.001; p<0.0001).

Abbreviations: BMI, body mass index; CFU, colony forming units; L, Lactobacillus; LMR, lactulose/mannitol ratio; LBP, lipopolysaccharide-binding protein; *n*, number of participants; n.s, not statistically significant.

6.1.9.4.2.2RISK OF BIAS ASSESSMENT

The following are the results from a risk of bias assessment (Table 6.21 and Table 6.22). A summary of the results is found in the IP Guideline (Section 8.14).

Table 6.21 Risk of bias assessment in randomised trials

	Randomisation process	Deviations from intended interventions	Missing outcome data	Measurement of the outcome	Selection of the reported result	Overall
Bloomer, 2020[326]	Some	High	High	Low	Low	High
González-Sarrias, 2018[325]	Some	Low	Low	High	High	High
Kim, 2006[300]	Low	Low	Low	Low	Low	Low
Lee, 2014[303]	Low	High	Low	Low	Low	High
Percentages						
Low risk	50.0%	50.0%	75.0%	75.0%	75.0%	25.0%
Some concerns	50.0%	0.0%	0.0%	0.0%	0.0%	0.0%
High risk	0.0%	50.0%	25.0%	25.0%	25.0%	75.0%

Abbreviations: Low = low risk of bias; Some = some concerns for risk of bias; High = high risk of bias.

Table 6.22 Risk of bias in non-randomised studies

Study	Confounding	Selection of participants	Classification of interventions	Deviations from intended interventions	Missing data	Measurement of outcome	Selection of reported results	Overall
Ried, 2020[311]	Moderate	Moderate	Moderate	Low	Moderate	Moderate	Moderate	Moderate

Abbreviations: Low = low risk of bias; Moderate = moderate risk of bias.

6.1.9.4.3 NHMRC EVIDENCE STATEMENT

The following are the results from completing the NHMRC Evidence Statement (Table 6.23). Individual grade for each of the five domains are provided along with the overall grade for the evidence-based recommendation. Furthermore, consensus-based recommendations are listed below (Table 6.24).

Table 6.23 Summary of the NHMRC evidence statement for evidence-based recommendations: Plant-based medicine supplementation

Recommendation	Grade
Recommendation 4.1: There is insufficient evidence to form a recommendation on the use of plant-based medicines as a collective group for the treatment of people with intestinal permeability.	**Grade: D**
Evidence base – number of studies, level of evidence, risk of bias in the included studies	**D**
Consistency – between studies if more than one study	**C**
Clinical impact – of the intervention, diagnosis, prognosis, aetiology, screening	**D**
Generalisability – how well the body of evidence matches the population and clinical setting	**C**
Applicability – relevance to Australian health care context in terms of services, delivery and cultural factors	**B**

Table 6.24 Summary of consensus-based recommendations and practice points: Plant-based medicine supplementation

NO	Recommendation
4.2	Clinicians may consider the use of plant-based medicines which are supported by pre-clinical research in conjunction with other treatment interventions for the management people with intestinal permeability.

6.1.9.5 ESSENTIAL FATTY ACID SUPPLEMENTATION

6.1.9.5.1 CLINICAL QUESTIONS

Clinical Question 9: In Australian adults with increased intestinal permeability, what are the benefits of oral essential fatty acid supplementation for the treatment of increased intestinal permeability?

Clinical Question 10: In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for oral essential fatty acid use?

6.1.9.5.2 EVIDENCE SUMMARY AND OVERALL QUALITY OF EVIDENCE

6.1.9.5.2.1ESSENTIAL FATTY ACID

There is a limited number (n = 1)[299] of studies exploring the effects of essential fatty acid supplementation on altered IP (Table 6.25). This randomised, double-blind placebo-controlled trial assessed the effects of four study arms: omega-3, probiotic, omega-3 and probiotic or placebo over 21 weeks in pregnant women.[299] The omega-3 supplement contained 2g of omega-3 (79.6% DHA and 9.7% EPA) twice daily. The study found no significant effect in serum zonulin between early and late pregnancy with omega-3 supplementation (mean change: +5.2±11.2ng/ml; 95%CI +2.0, +8.5; p>0.05). Furthermore, LPS had no significant change between early and late pregnancy with omega-3 supplementation (mean change: +0.06±0.11EU/ml; 95%CI +0.023, +0.088; p>0.05).

Table 6.25 Evidence summary table for clinical trials

Author, Year	Study design; Level of Evidence NHMRC	Country; Setting	Sample size; age in years M ± SD; gender; health condition	Intervention; control	Outcome measures; duration	Main results
Mokkala, 2018[299]	Randomised, placebo controlled, double-blind trial; II	Finland; outpatient clinic	*n* = 200 (*n* = 199 analysed) Intervention 1: *n* = 51; 30.5±4.9 years; overweight and obese pregnant woman (BMI 30.3±5.1 kg/m^2) Intervention 2: *n* = 49; 30.7±5.5 years; overweight and obese pregnant woman (BMI 30.3±4.4 kg/m^2) Intervention 3: *n* = 49; 30.1±5.3 years; overweight and obese pregnant woman (BMI 30.0±4.1 kg/m^2)	Intervention 1: B420 10^{10} CFU and *L. rhamnosus* HN001 10^{10} CFU 1x/day, 21.8±2.6 weeks Intervention 2: 1.2g of omega 3 consisting of 79.6% DHA and 9.7% EPA 2x/day, 21.5±2.5 weeks Intervention 3: B420 10^{10} CFU and *L. rhamnosus* HN001 10^{10} CFU 1x/day and 1.2g of omega 3 consisting of 79.6% DHA and 9.7% EPA 2x/day, 21.3±2.5 weeks Control: placebo containing microcrystalline cellulose 1x/day and capric acid and	Serum zonulin at late pregnancy, 21 weeks LPS at late pregnancy, 21 weeks	Serum zonulin: n.s difference between early and late pregnancy with omega 3 (mean change: +5.2±11.2ng/ml; 95%CI +2.0, +8.5; p>0.05) LPS: n.s difference between early and late pregnancy with omega 3 (mean change: +0.06±0.11EU/ml; 95%CI +0.023, +0.088; p>0.05).

	Control: *n* = 51; 30.2±3.9 years; overweight and obese pregnant woman (BMI 29.8±4.5 kg/m^2)	caprylic 2x/day, 21.3±2.3 weeks

Abbreviations: B420, Bifidobacterium animalis ssp. lactis 420; BMI, body mass index; CFU, colony forming units; DHA, docosahexaenoic acid; EPA, eicosapentaenoic acid; L, Lactobacillus; LPS, lipopolysaccharide; *n*, number of participants; n.s, not statistically significant.

6.1.9.5.2.2RISK OF BIAS ASSESSMENT

The following are the results from a risk of bias assessment (Table 6.26). A summary of the results is found in the IP Guideline (Section 8.15).

Table 6.26 Risk of bias assessment in randomised trials

	Randomisation process	**Deviations from intended interventions**	**Missing outcome data**	**Measurement of the outcome**	**Selection of the reported result**	**Overall**
Mokkala, 2018[299]	Low	Low	Low	High	Low	High
Percentages						
Low risk	100.0%	100.0%	100.0%	0.0%	100.0%	0.0%
Some concerns	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%
High risk	0.0%	0.0%	0.0%	100.0%	0.0%	100.0%

Abbreviations: Low = low risk of bias; High = high risk of bias.

6.1.9.5.3 NHMRC EVIDENCE STATEMENT

The following are the results from completing the NHMRC Evidence Statement (Table 6.27). Individual grade for each of the five domains are provided along with the overall grade for the evidence-based recommendation.

Table 6.27 Summary of the NHMRC evidence statement for evidence-based recommendations: Essential fatty acid supplementation

Recommendation	Grade
Recommendation 4.1: There is insufficient evidence to form a recommendation on the use of essential fatty acid supplementation for the treatment of people with intestinal permeability.	**Grade: D**
Evidence base – number of studies, level of evidence, risk of bias in the included studies	**D**
Consistency – between studies if more than one study	**N/A**
Clinical impact – of the intervention, diagnosis, prognosis, aetiology, screening	**D**
Generalisability – how well the body of evidence matches the population and clinical setting	**C**
Applicability – relevance to Australian health care context in terms of services, delivery and cultural factors	**C**

6.1.9.6 MINERAL SUPPLEMENTATION

6.1.9.6.1 CLINICAL QUESTIONS

Clinical Question 11: In Australian adults with increased intestinal permeability, what are the benefits of oral mineral supplementation for the treatment of increased intestinal permeability?

Clinical Question 12: In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for oral mineral supplementation use?

6.1.9.6.2 EVIDENCE SUMMARY AND OVERALL QUALITY OF EVIDENCE

6.1.9.6.2.1MINERAL

After systematically searching the literature, only one non-randomised clinical trial met the inclusion criteria and was included (Table 6.28).[327] This study explored the effects of zinc supplementation in 12 Crohn's disease patients in remission with a lactulose mannitol ratio >0.035. The study involved zinc supplementation containing 25mg of elemental zinc three times daily for eight weeks. After the study period, there was a significant decrease in IP from baseline to eight weeks (0.041±0.003 vs. 0.026±0.005; p= 0.0028). Furthermore, at the end of the eight weeks, the lactulose mannitol ratio normalised in 75% of participants.

Table 6.28 Evidence summary table for clinical trials

Author, Year	Study design; Level of Evidence NHMRC	Country; Setting	Sample size; age in years M ± SD; gender; health condition	Intervention; control	Outcome measures; duration	Main results
Sturniolo, 2001[327]	Non-randomised clinical trial; III-2	Italy, outpatient clinic	*n* = 12; 33.0±11.0 years; 33% female; Crohn's disease patients in remission with IP	Intervention: zinc sulfate containing 25mg of elemental zinc taken 3x/day, 8 weeks.	LMR at 8 weeks	LMR: Significantly decreased from baseline to 8 weeks (0.041±0.003 vs. 0.026±0.005; p= 0.0028).

Abbreviations: IP, increased intestinal permeability; LMR, lactulose mannitol ratio; *n*, number of participants.

6.1.9.6.2.2RISK OF BIAS ASSESSMENT

The following are the results from a risk of bias assessment (Table 6.29). A summary of the results is found in the IP Guideline (Section 8.16).

Table 6.29 Risk of bias in non-randomised studies

Study	Confounding	Selection of participants	Classification of interventions	Deviations from intended interventions	Missing data	Measurement of outcome	Selection of reported results	Overall
Sturniolo, 2001[327]	Moderate	Moderate	Moderate	Low	Moderate	Moderate	Moderate	Moderate

Abbreviations: Low = low risk of bias; Moderate = moderate risk of bias.

6.1.9.6.3 NHMRC EVIDENCE STATEMENT

The following are the results from completing the NHMRC Evidence Statement (Table 6.30). Individual grade for each of the five domains are provided along with the overall grade for the evidence-based recommendation.

Table 6.30 Summary of the NHMRC evidence statement for evidence-based recommendations: Mineral supplementation

Recommendation	Grade
Recommendation 6.1: Clinicians may consider using zinc supplementation in the treatment of people with intestinal permeability	**Grade: C**
Evidence base – number of studies, level of evidence, risk of bias in the included studies	**C**
Consistency – between studies if more than one study	**N/A**
Clinical impact – of the intervention, diagnosis, prognosis, aetiology, screening	**B**
Generalisability – how well the body of evidence matches the population and clinical setting	**B**
Applicability – relevance to Australian health care context in terms of services, delivery and cultural factors	**B**

6.1.9.7 VITAMIN SUPPLEMENTATION

6.1.9.7.1 CLINICAL QUESTIONS

Clinical Question 13: In Australian adults with increased intestinal permeability, what are the benefits of oral vitamin supplementation for the treatment of increased intestinal permeability?

Clinical Question 14: In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for oral vitamin supplementation use?

6.1.9.7.2 EVIDENCE SUMMARY AND OVERALL QUALITY OF EVIDENCE

6.1.9.7.2.1 VITAMINS

No research was found exploring the effects of vitamin supplementation on altered IP. Therefore, no recommendation was developed.

6.1.9.8 COLOSTRUM SUPPLEMENTATION

6.1.9.8.1 CLINICAL QUESTIONS

Clinical Question 15: In Australian adults with increased intestinal permeability, what are the benefits of oral colostrum supplementation for the treatment of increased intestinal permeability?

Clinical Question 16: In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for oral colostrum supplementation use?

6.1.9.8.2 EVIDENCE SUMMARY AND OVERALL QUALITY OF EVIDENCE

6.1.9.8.2.1 COLOSTRUM

No research was found exploring the effects of colostrum supplementation on altered IP. Therefore, no recommendation was developed.

6.2 CHAPTER SUMMARY

This chapter provides the *Technical Report* for the IP Guideline, a comprehensive evaluation of the literature and data extraction of the relevant information for each study used in the IP Guideline. A total of 61 principal articles were identified and used in the IP Guideline. The identified evidence can direct the development of clinical recommendations to support clinicians in managing patients with IP. The views of the individuals with suspected IP and the available literature were both considered when drafting the recommendations. However, whether these recommendations are clinically relevant or applicable to the end-users remain unknown. Obtaining stakeholder feedback on these recommendations would ensure they align with their views and preferences.

7. CLINICAL PRACTICE GUIDELINE FOR THE MANAGEMENT OF INCREASED INTESTINAL PERMEABILITY: STAKEHOLDER SURVEY

A comprehensive summary of the evidence, risk of bias assessment and a summary of the NHMRC evidence statement was discussed in the preceding chapter. It presents 38 recommendations based on the combined evidence and experience of the Working Group. The aim of this project is to develop evidence-based treatment recommendations that are consistent with the views and preferences of Australian adults with IP as well as key stakeholders. Therefore, this chapter provides stakeholders (n=8) views and feedback on the developed recommendations. Section 3.5 presents the methods used to evaluate stakeholders' agreement with each recommendation.

7.1 COLLECTIVE SUMMARY OF STAKEHOLDER FEEDBACK

7.1.1 Understanding

The developed recommendations were well understood, with 73.7% of the recommendations receiving a 'good' understanding and only 18.4% indicating a 'poor' understanding. The recommendations with the highest agreement rate were recommendations 2.4 (researching probiotics), 2.9 (researching prebiotics), 2.10 (use prebiotic with pre-clinical research), 3.3 (lactoferrin in NSAID-induced IP) and 6.1 (zinc supplementation), which received a 100% agreement from all stakeholders. Recommendations 1.1 (alcohol consumption pre the Australian

Dietary Guidelines), 2.16 (not to use probiotics in NSAID-induced IP), 2.17 (not to use prebiotics in NSAID-induced IP) and 2.18 (not to use synbiotics in NSAID-induced IP) only had a 20% agreement rate from stakeholders.

7.1.2 Appropriateness

Recommendations 1.2 (avoid or limit alcohol consumption), 2.9 (researching prebiotics), 2.10 (use prebiotic with pre-clinical research), and 6.1 (zinc supplementation) were identified as the most appropriate with an in-unison agreement between stakeholders. While the appropriateness of recommendation 2.17 (not to use prebiotics in NSAID-induced IP) saw a disagreement rate of 50%.

7.1.3 Importance

Recommendations 1.2 (avoid or limit alcohol consumption), 2.4 (researching probiotics), and 6.1 (zinc supplementation) were identified to have the highest importance, with these recommendations receiving 100% agreement from the stakeholders. On the contrary, the importance of four recommendations, namely 2.8 (insufficient evidence for prebiotic use), 2.16 (not to use probiotics in NSAID-induced IP), 2.17 (not to use prebiotics in NSAID-induced IP) and 5.1 (insufficient evidence for essential fatty acid use), saw a disagreement rate of 37.5%.

7.1.4 Overall consensus

Collectively, the recommendations with the highest consensus (>80%) for understanding, agreement, appropriateness, and importance were recommendations 1.2 (avoid or limit alcohol consumption), 2.4 (researching probiotics), 2.9 (researching prebiotics), 2.10 (use prebiotic with pre-clinical

research), 3.1 (use glutamine supplementation), 3.2 (use glutamine with other interventions), 3.3 (lactoferrin in NSAID-induced IP), 4.2 (use plant-based medicines with pre-clinical research) and 6.1 (zinc supplementation). While the recommendation with the lowest agreement rate (<25%) for agreement, appropriateness, and importance were recommendations 2.16 (not to use probiotics in NSAID-induced IP), 2.17 (not to use prebiotics in NSAID-induced IP) and 2.18 (not to use synbiotics in NSAID-induced IP).

7.2 STAKEHOLDERS' VIEWS AND PREFERENCES TOWARDS DIETARY RECOMMENDATIONS

Considering the individual dietary recommendations for managing patients with IP, stakeholders reported consensus among some recommendations and a disagreement with others (Table 7.1). Specifically, although well understood, stakeholders report a lack of consensus on the agreement, appropriateness, and importance of recommendation 1.1 (alcohol consumption pre the Australian Dietary Guidelines), with a disagreement rate of 62.5% found for the agreement with the recommendation. However, stakeholders were found to have a consensus on the agreement, appropriateness, and importance for recommendations 1.2 (avoid or limit alcohol consumption).

All fibre related recommendations (1.3, 1.4, 1.5) were understood by the stakeholder group and found to have a consensus (>75%) on the agreement, appropriateness, and importance of the recommendations to be followed when treating patients with IP (Table 7.1). Recommendation 1.6 had no consensus,

with stakeholders expressing mixed views towards the acceptable macronutrient distribution range for people to follow during IP treatment. The lack of consensus was also seen in recommendations 1.9 and 1.10, with diverse opinions from stakeholders on whether people with IP should follow the estimated energy requirements per the Australian Dietary Guidelines or follow a kilojoule restricted diet. Most stakeholders (>62.5%) agreed that recommendations 1.7 (avoiding a high-fat diet) and 1.8 (limiting fructose consumption) were understandable, appropriate, and important for clinicians to follow in clinical practice.

The stakeholders understood the gluten-related recommendations yet not all recommendations had a consensus. Recommendation 1.11 (not to use polydextrose) was found to have a consensus (>75%) on the recommendation's agreement, appropriateness, and importance to be followed when treating patients with IP. At the same time, most stakeholders (>62.5%) agreed that recommendations 1.12 (use synbiotics) and 1.13 (use synbiotics for 3 months) were understandable, appropriate, and important for clinicians to follow in clinical practice.

Table 7.1 Stakeholders' views and preferences towards dietary recommendations (n=8)

Recommendation	Response %
Recommendation 1.1 People with intestinal permeability should consider consuming no more than 10 standard drinks a week and no more than 4 standard drinks on any one day in accordance with the Australian Dietary Guidelines during the treatment of intestinal permeability.	
Understanding of recommendation	
Good/very good	100.0
Neutral	0.0
Poor/very poor	0.0

Agreement with recommendation	
Agree/strongly agree	25.0
Neutral	12.5
Disagree/strongly disagree	62.5
Appropriateness of recommendation	
Agree/strongly agree	62.5
Neutral	0.0
Disagree/strongly disagree	37.5
Importance of recommendation	
Agree/strongly agree	75.0
Neutral	0.0
Disagree/strongly disagree	25.0
Change anything about the recommendation	
Yes	75.0
No	25.0
Recommendation 1.2 People with intestinal permeability may consider limiting or avoiding alcohol consumption during the short-term treatment of intestinal permeability.	
Understanding of recommendation	
Good/very good	100.0
Neutral	0.0
Poor/very poor	0.0
Agreement with recommendation	
Agree/strongly agree	87.5
Neutral	12.5
Disagree/strongly disagree	0.0
Appropriateness of recommendation	
Agree/strongly agree	100.0
Neutral	0.0
Disagree/strongly disagree	0.0
Importance of recommendation	
Agree/strongly agree	100.0
Neutral	0.0
Disagree/strongly disagree	0.0
Change anything about the recommendation	
Yes	50.0
No	50.0
Recommendation 1.3 People with intestinal permeability should consider consuming a diet high in dietary fibre from a diverse range of sources.	
Understanding of recommendation	

Good/very good	100.0
Neutral	0.0
Poor/very poor	0.0
Agreement with recommendation	
Agree/strongly agree	75.0
Neutral	0.0
Disagree/strongly disagree	25.0
Appropriateness of recommendation	
Agree/strongly agree	75.0
Neutral	0.0
Disagree/strongly disagree	25.0
Importance of recommendation	
Agree/strongly agree	87.5
Neutral	0.0
Disagree/strongly disagree	12.5
Change anything about the recommendation	
Yes	37.5
No	62.5
Recommendation 1.4 Clinicians are advised to recommend patients to consume 38g for men and 28g for female of dietary fibre daily while treating patients with intestinal permeability**.**	
Understanding of recommendation	
Good/very good	100.0
Neutral	0.0
Poor/very poor	0.0
Agreement with recommendation	
Agree/strongly agree	75.0
Neutral	0.0
Disagree/strongly disagree	25.0
Appropriateness of recommendation	
Agree/strongly agree	75.0
Neutral	0.0
Disagree/strongly disagree	25.0
Importance of recommendation	
Agree/strongly agree	75.0
Neutral	0.0
Disagree/strongly disagree	25.0
Change anything about the recommendation	
Yes	50.0
No	50.0

Recommendation 1.5 Clinicians are encouraged to recommend gluten-free sources of dietary fibre to patients with confirmed intestinal permeability.	
Understanding of recommendation	
Good/very good	100.0
Neutral	0.0
Poor/very poor	0.0
Agreement with recommendation	
Agree/strongly agree	75.0
Neutral	0.0
Disagree/strongly disagree	25.0
Appropriateness of recommendation	
Agree/strongly agree	75.0
Neutral	0.0
Disagree/strongly disagree	25.0
Importance of recommendation	
Agree/strongly agree	75.0
Neutral	0.0
Disagree/strongly disagree	25.0
Change anything about the recommendation	
Yes	25.0
No	75.0
Recommendation 1.6 People with intestinal permeability should consider consuming the Acceptable Macronutrient Distribution Range of protein (15-25%), fats (20-35%) and carbohydrates (45-65%) in accordance with the Australian Dietary Guidelines.	
Understanding of recommendation	
Good/very good	100.0
Neutral	0.0
Poor/very poor	0.0
Agreement with recommendation	
Agree/strongly agree	50.0
Neutral	25.0
Disagree/strongly disagree	25.0
Appropriateness of recommendation	
Agree/strongly agree	50.0
Neutral	25.0
Disagree/strongly disagree	25.0
Importance of recommendation	
Agree/strongly agree	37.5

Neutral	37.5
Disagree/strongly disagree	25.0
Change anything about the recommendation	
Yes	25.0
No	75.0
Recommendation 1.7 People with intestinal permeability should consider NOT consuming a diet high in fat.	
Understanding of recommendation	
Good/very good	100.0
Neutral	0.0
Poor/very poor	0.0
Agreement with recommendation	
Agree/strongly agree	75.0
Neutral	0.0
Disagree/strongly disagree	25.0
Appropriateness of recommendation	
Agree/strongly agree	62.5
Neutral	12.5
Disagree/strongly disagree	25.0
Importance of recommendation	
Agree/strongly agree	62.5
Neutral	25.0
Disagree/strongly disagree	12.5
Change anything about the recommendation	
Yes	62.5
No	37.5
Recommendation 1.8 People with intestinal permeability should consider NOT consuming a diet high in fructose.	
Understanding of recommendation	
Good/very good	100.0
Neutral	0.0
Poor/very poor	0.0
Agreement with recommendation	
Agree/strongly agree	75.0
Neutral	12.5
Disagree/strongly disagree	12.5
Appropriateness of recommendation	
Agree/strongly agree	75.0
Neutral	12.5
Disagree/strongly disagree	12.5

Importance of recommendation	
Agree/strongly agree	62.5
Neutral	25.0
Disagree/strongly disagree	12.5
Change anything about the recommendation	
Yes	37.5
No	62.5
Recommendation 1.9 People with intestinal permeability may consider consuming the estimated energy requirements in accordance with the Australian Dietary Guidelines.	
Understanding of recommendation	
Good/very good	87.5
Neutral	0.0
Poor/very poor	12.5
Agreement with recommendation	
Agree/strongly agree	37.5
Neutral	50.0
Disagree/strongly disagree	12.5
Appropriateness of recommendation	
Agree/strongly agree	25.0
Neutral	50.0
Disagree/strongly disagree	25.0
Importance of recommendation	
Agree/strongly agree	25.0
Neutral	62.5
Disagree/strongly disagree	12.5
Change anything about the recommendation	
Yes	25.0
No	75.0
Recommendation 1.10 Clinicians may consider using a kilojoule restricted diet in the short-term treatment of people with confirmed intestinal permeability.	
Understanding of recommendation	
Good/very good	100.0
Neutral	0.0
Poor/very poor	0.0
Agreement with recommendation	
Agree/strongly agree	62.5
Neutral	12.5
Disagree/strongly disagree	25.0

Appropriateness of recommendation	
Agree/strongly agree	62.5
Neutral	12.5
Disagree/strongly disagree	25.0
Importance of recommendation	
Agree/strongly agree	50.0
Neutral	25.0
Disagree/strongly disagree	25.0
Change anything about the recommendation	
Yes	37.5
No	62.5
Recommendation 1.11 Clinicians should only advise a strict gluten-free diet if clinical symptoms or pathology indicate a gluten intolerance, sensitivity or allergy.	
Understanding of recommendation	
Good/very good	100.0
Neutral	0.0
Poor/very poor	0.0
Agreement with recommendation	
Agree/strongly agree	75.0
Neutral	0.0
Disagree/strongly disagree	25.0
Appropriateness of recommendation	
Agree/strongly agree	75.0
Neutral	0.0
Disagree/strongly disagree	25.0
Importance of recommendation	
Agree/strongly agree	87.5
Neutral	0.0
Disagree/strongly disagree	12.5
Change anything about the recommendation	
Yes	25.0
No	75.0
Recommendation 1.12 Clinicians should only advise a gluten-free diet during the short-term treatment of people with confirmed intestinal permeability that report clinical symptoms in response to the consumption of gluten after the investigation for gluten intolerance, sensitivity or allergy has been carried out.	
Understanding of recommendation	
Good/very good	100.0

Neutral	0.0
Poor/very poor	0.0
Agreement with recommendation	
Agree/strongly agree	50.0
Neutral	12.5
Disagree/strongly disagree	37.5
Appropriateness of recommendation	
Agree/strongly agree	62.5
Neutral	12.5
Disagree/strongly disagree	25.0
Importance of recommendation	
Agree/strongly agree	62.5
Neutral	12.5
Disagree/strongly disagree	25.0
Change anything about the recommendation	
Yes	25.0
No	75.0
Recommendation 1.13 Clinicians should offer a low gluten diet for the management of people with confirmed intestinal permeability that report no clinical symptoms or pathology indicating a gluten intolerance, sensitivity or allergy.	
Understanding of recommendation	
Good/very good	100.0
Neutral	0.0
Poor/very poor	0.0
Agreement with recommendation	
Agree/strongly agree	62.5
Neutral	0.0
Disagree/strongly disagree	37.5
Appropriateness of recommendation	
Agree/strongly agree	62.5
Neutral	0.0
Disagree/strongly disagree	37.5
Importance of recommendation	
Agree/strongly agree	62.5
Neutral	12.5
Disagree/strongly disagree	25.0
Change anything about the recommendation	
Yes	12.5
No	87.5

7.3 STAKEHOLDERS' VIEWS AND PREFERENCES TOWARDS PROBIOTIC, PREBIOTIC AND SYNBIOTIC SUPPLEMENTATION RECOMMENDATIONS

7.3.1 Probiotic

Stakeholders reported consensus among some probiotic recommendations and disagreement with others (Table 7.2). Specifically, although well understood, stakeholders report a lack of consensus on the agreement, appropriateness, and importance of recommendation 2.1 (insufficient evidence for probiotic use), with a disagreement rate of 62.5% for the appropriateness of the recommendation. In contrast, the recommendation for a single probiotic supplement *Saccharomyces boulardii* (recommendation 2.2), was understood by the stakeholders and found to have a consensus (>75%) for the agreement, appropriateness, and importance of the recommendation to be followed when treating patients with IP. Recommendations regarding the length of use, pre-clinical research and researching specific strains of probiotics (2.3, 2.4, 2.5) were understood by the stakeholders and found to have a consensus (>75%) for the agreement, appropriateness, and importance. While recommendations for probiotic drinks were understood, stakeholders reported a lack of consensus for the agreement, appropriateness, and importance of recommendations 2.6 and 2.7, with a neutral response (>25%) frequently reported by stakeholders.

7.3.2 Prebiotic

Stakeholders reported consensus among some recommendations while disagreeing with others (Table 7.2). Stakeholders report a lack of consensus on the agreement, appropriateness, and importance of recommendation 2.8 (insufficient evidence for prebiotic use), with a disagreement rate of 37.5% for each domain. However, recommendations regarding the use of pre-clinical research and researching specific prebiotics (recommendations 2.9 and 2.10) were understood by the stakeholders and found to have a consensus (>87.5%) for the agreement, appropriateness, and importance of the recommendations. Most stakeholders (>62.5%) agreed that recommendation 2.1 (avoid polydextrose) was understandable, appropriate, and important for clinicians to follow in clinical practice.

7.3.3 Synbiotic

Recommendations regarding the length of use, pre-clinical research and using specific synbiotics (2.12, 2.13, 1.15) were understood by the stakeholders, with most (>62.5%) agreeing that the recommendations were appropriate and important for clinical practice. While recommendation 2.14 for a specific synbiotic, was found to lack a consensus on the agreement, appropriateness, and importance for the recommendation with a neutral response rate (>50%) was reported by stakeholders. Although well understood, stakeholders report a lack of consensus on the agreement, appropriateness, and importance of recommendations 2.16 (not to use probiotics in NSAID-induced IP), 2.17 (not to use prebiotics in NSAID-induced IP) and 2.18 (not to use synbiotics in NSAID-induced IP) with a low agreement rate (25%) found for each domain.

Table 7.2 Stakeholders' views and preferences towards the probiotic, prebiotic and synbiotic supplementation recommendations (n=8)

Recommendations	Response %
Recommendation 2.1 There is insufficient evidence to form a recommendation on the use of probiotics as a collective group for the treatment of people with intestinal permeability.	
Understanding of recommendation	
Good/very good	87.5
Neutral	0.0
Poor/very poor	12.5
Agreement with recommendation	
Agree/strongly agree	62.5
Neutral	12.5
Disagree/strongly disagree	25.0
Appropriateness of recommendation	
Agree/strongly agree	50.0
Neutral	12.5
Disagree/strongly disagree	37.5
Importance of recommendation	
Agree/strongly agree	62.5
Neutral	12.5
Disagree/strongly disagree	25.0
Change anything about the recommendation	
Yes	25.0
No	75.0
Recommendation 2.2 Clinicians may consider using *Saccharomyces boulardii* supplementation in the treatment of people with intestinal permeability.	
Understanding of recommendation	
Good/very good	100.0
Neutral	0.0
Poor/very poor	0.0
Agreement with recommendation	
Agree/strongly agree	87.5
Neutral	12.5
Disagree/strongly disagree	0.0
Appropriateness of recommendation	
Agree/strongly agree	87.5

Neutral	12.5
Disagree/strongly disagree	0.0
Importance of recommendation	
Agree/strongly agree	75.0
Neutral	25.0
Disagree/strongly disagree	0.0
Change anything about the recommendation	
Yes	50.0
No	50.0
Recommendation 2.3 Clinicians may consider the use of effective probiotics for a period of 3 months when treating people with intestinal permeability.	
Understanding of recommendation	
Good/very good	100.0
Neutral	0.0
Poor/very poor	0.0
Agreement with recommendation	
Agree/strongly agree	75.0
Neutral	25.0
Disagree/strongly disagree	0.0
Appropriateness of recommendation	
Agree/strongly agree	87.5
Neutral	12.5
Disagree/strongly disagree	0.0
Importance of recommendation	
Agree/strongly agree	87.5
Neutral	12.5
Disagree/strongly disagree	0.0
Change anything about the recommendation	
Yes	37.5
No	62.5
Recommendation 2.4 Clinicians may consider researching probiotic strains for their effectiveness before using them to treat people with intestinal permeability.	
Understanding of recommendation	
Good/very good	100.0
Neutral	0.0
Poor/very poor	0.0
Agreement with recommendation	
Agree/strongly agree	100.0

Neutral	0.0
Disagree/strongly disagree	0.0
Appropriateness of recommendation	
Agree/strongly agree	87.5
Neutral	12.5
Disagree/strongly disagree	0.0
Importance of recommendation	
Agree/strongly agree	100.0
Neutral	0.0
Disagree/strongly disagree	0.0
Change anything about the recommendation	
Yes	12.5
No	87.5
Recommendation 2.5 Clinicians may consider the use of probiotics which are supported by pre-clinical research in conjunction with other treatment interventions for the management people with intestinal permeability.	
Understanding of recommendation	
Good/very good	100.0
Neutral	0.0
Poor/very poor	0.0
Agreement with recommendation	
Agree/strongly agree	75.0
Neutral	12.5
Disagree/strongly disagree	12.5
Appropriateness of recommendation	
Agree/strongly agree	75.0
Neutral	12.5
Disagree/strongly disagree	12.5
Importance of recommendation	
Agree/strongly agree	75.0
Neutral	25.0
Disagree/strongly disagree	0.0
Change anything about the recommendation	
Yes	37.5
No	62.5
Recommendation 2.6 People with intestinal permeability should consider the consumption of fermented milk products such as kefir.	
Understanding of recommendation	
Good/very good	100.0
Neutral	0.0

Poor/very poor	0.0
Agreement with recommendation	
Agree/strongly agree	50.0
Neutral	25.0
Disagree/strongly disagree	25.0
Appropriateness of recommendation	
Agree/strongly agree	50.0
Neutral	25.0
Disagree/strongly disagree	25.0
Importance of recommendation	
Agree/strongly agree	50.0
Neutral	37.5
Disagree/strongly disagree	12.5
Change anything about the recommendation	
Yes	50.0
No	50.0
Recommendation 2.7 People with intestinal permeability may consider NOT consuming Yakult light®.	
Understanding of recommendation	
Good/very good	75.0
Neutral	12.5
Poor/very poor	12.5
Agreement with recommendation	
Agree/strongly agree	75.0
Neutral	25.0
Disagree/strongly disagree	0.0
Appropriateness of recommendation	
Agree/strongly agree	75.0
Neutral	25.0
Disagree/strongly disagree	0.0
Importance of recommendation	
Agree/strongly agree	50.0
Neutral	50.0
Disagree/strongly disagree	0.0
Change anything about the recommendation	
Yes	12.5
No	87.5
Recommendation 2.8 There is insufficient evidence to form a recommendation on the use of prebiotics as a collective group for the treatment of people with intestinal permeability.	

Understanding of recommendation	
Good/very good	100.0
Neutral	0.0
Poor/very poor	0.0
Agreement with recommendation	
Agree/strongly agree	50.0
Neutral	12.5
Disagree/strongly disagree	37.5
Appropriateness of recommendation	
Agree/strongly agree	50.0
Neutral	12.5
Disagree/strongly disagree	37.5
Importance of recommendation	
Agree/strongly agree	50.0
Neutral	12.5
Disagree/strongly disagree	37.5
Change anything about the recommendation	
Yes	25.0
No	75.0
Recommendation 2.9 Clinicians may consider researching prebiotic for their effectiveness before using them in the treat of people with intestinal permeability.	
Understanding of recommendation	
Good/very good	100.0
Neutral	0.0
Poor/very poor	0.0
Agreement with recommendation	
Agree/strongly agree	100.0
Neutral	0.0
Disagree/strongly disagree	0.0
Appropriateness of recommendation	
Agree/strongly agree	100.0
Neutral	0.0
Disagree/strongly disagree	0.0
Importance of recommendation	
Agree/strongly agree	87.5
Neutral	12.5
Disagree/strongly disagree	0.0
Change anything about the recommendation	
Yes	37.5

No	62.5
Recommendation 2.10 Clinicians may consider the use of prebiotic which are supported by pre-clinical research in conjunction with other treatment interventions for the management people with intestinal permeability.	
Understanding of recommendation	
Good/very good	100.0
Neutral	0.0
Poor/very poor	0.0
Agreement with recommendation	
Agree/strongly agree	100.0
Neutral	0.0
Disagree/strongly disagree	0.0
Appropriateness of recommendation	
Agree/strongly agree	100.0
Neutral	0.0
Disagree/strongly disagree	0.0
Importance of recommendation	
Agree/strongly agree	87.5
Neutral	12.5
Disagree/strongly disagree	0.0
Change anything about the recommendation	
Yes	0.0
No	100.00
Recommendation 2.11 Clinicians may consider NOT using polydextrose in the treatment of people with intestinal permeability.	
Understanding of recommendation	
Good/very good	75.0
Neutral	12.5
Poor/very poor	12.5
Agreement with recommendation	
Agree/strongly agree	62.5
Neutral	25.0
Disagree/strongly disagree	12.5
Appropriateness of recommendation	
Agree/strongly agree	62.5
Neutral	25.0
Disagree/strongly disagree	12.5
Importance of recommendation	
Agree/strongly agree	62.5
Neutral	37.5

Disagree/strongly disagree	0.0
Change anything about the recommendation	
Yes	25.0
No	75.0
Recommendation 2.12 Clinicians may consider the use of effective synbiotic in the treatment of people with intestinal permeability.	
Understanding of recommendation	
Good/very good	87.5
Neutral	12.5
Poor/very poor	0.0
Agreement with recommendation	
Agree/strongly agree	62.5
Neutral	37.5
Disagree/strongly disagree	0.0
Appropriateness of recommendation	
Agree/strongly agree	62.5
Neutral	37.5
Disagree/strongly disagree	0.0
Importance of recommendation	
Agree/strongly agree	62.5
Neutral	37.5
Disagree/strongly disagree	0.0
Change anything about the recommendation	
Yes	12.5
No	87.5
Recommendation 2.13 Clinicians may consider the use of effective synbiotic for a period of 3 months when treating people with intestinal permeability.	
Understanding of recommendation	
Good/very good	75.0
Neutral	25.0
Poor/very poor	0.0
Agreement with recommendation	
Agree/strongly agree	87.5
Neutral	12.5
Disagree/strongly disagree	0.0
Appropriateness of recommendation	
Agree/strongly agree	62.5
Neutral	37.5
Disagree/strongly disagree	0.0

Importance of recommendation	
Agree/strongly agree	62.5
Neutral	37.5
Disagree/strongly disagree	0.0
Change anything about the recommendation	
Yes	37.5
No	62.5
Recommendation 2.14 Clinicians may consider NOT using polydextrose and *Bifidobacterium animalis ssp. lactis* 420 in the treatment of people with intestinal permeability.	
Understanding of recommendation	
Good/very good	62.5
Neutral	12.5
Poor/very poor	25.0
Agreement with recommendation	
Agree/strongly agree	50.0
Neutral	50.0
Disagree/strongly disagree	0.0
Appropriateness of recommendation	
Agree/strongly agree	37.5
Neutral	62.5
Disagree/strongly disagree	0.0
Importance of recommendation	
Agree/strongly agree	37.5
Neutral	62.5
Disagree/strongly disagree	0.0
Change anything about the recommendation	
Yes	25.0
No	75.0
Recommendation 2.15 Clinicians may consider the use of synbiotic which are supported by pre-clinical research in conjunction with other treatment interventions for the management people with intestinal permeability.	
Understanding of recommendation	
Good/very good	100.0
Neutral	0.0
Poor/very poor	0.0
Agreement with recommendation	
Agree/strongly agree	75.0
Neutral	25.0

Disagree/strongly disagree	0.0
Appropriateness of recommendation	
Agree/strongly agree	75.0
Neutral	25.0
Disagree/strongly disagree	0.0
Importance of recommendation	
Agree/strongly agree	75.0
Neutral	12.5
Disagree/strongly disagree	12.5
Change anything about the recommendation	
Yes	37.5
No	62.5
Recommendation 2.16 Clinicians should consider NOT using probiotics for the treatment of people with nonsteroidal anti-inflammatory drug induced intestinal permeability.	
Understanding of recommendation	
Good/very good	75.0
Neutral	12.5
Poor/very poor	12.5
Agreement with recommendation	
Agree/strongly agree	25.0
Neutral	25.0
Disagree/strongly disagree	50.0
Appropriateness of recommendation	
Agree/strongly agree	25.0
Neutral	37.5
Disagree/strongly disagree	37.5
Importance of recommendation	
Agree/strongly agree	25.0
Neutral	37.5
Disagree/strongly disagree	37.5
Change anything about the recommendation	
Yes	25.0
No	75.0
Recommendation 2.17 Clinicians should consider NOT using prebiotics for the treatment of people with nonsteroidal anti-inflammatory drug induced intestinal permeability.	
Understanding of recommendation	
Good/very good	87.5
Neutral	0.0

Poor/very poor	12.5
Agreement with recommendation	
Agree/strongly agree	25.0
Neutral	12.5
Disagree/strongly disagree	62.5
Appropriateness of recommendation	
Agree/strongly agree	25.0
Neutral	25.0
Disagree/strongly disagree	50.0
Importance of recommendation	
Agree/strongly agree	25.0
Neutral	25.0
Disagree/strongly disagree	50.0
Change anything about the recommendation	
Yes	12.5
No	87.5
Recommendation 2.18 Clinicians should consider NOT using synbiotics for the treatment of people with nonsteroidal anti-inflammatory drug induced intestinal permeability.	
Understanding of recommendation	
Good/very good	75.0
Neutral	12.5
Poor/very poor	12.5
Agreement with recommendation	
Agree/strongly agree	25.0
Neutral	37.5
Disagree/strongly disagree	37.5
Appropriateness of recommendation	
Agree/strongly agree	25.0
Neutral	50.0
Disagree/strongly disagree	25.0
Importance of recommendation	
Agree/strongly agree	25.0
Neutral	50.0
Disagree/strongly disagree	25.0
Change anything about the recommendation	
Yes	12.5
No	87.5

7.4 STAKEHOLDERS' VIEWS AND PREFERENCES TOWARDS AMINO ACID SUPPLEMENTATION RECOMMENDATIONS

All amino acid recommendations (3.1, 3.2, 3.3) were understood by the stakeholders and found to have a consensus on the agreement, appropriateness, and importance of the recommendations to be followed when treating patients with IP (Table 7.3).

Table 7.3 Stakeholders' views and preferences towards amino acid supplementation recommendations (n=8)

Recommendations	Response %
Recommendation 3.1 Clinicians should offer glutamine supplementation for the treatment of people with intestinal permeability.	
Understanding of recommendation	
Good/very good	100.0
Neutral	0.0
Poor/very poor	0.0
Agreement with recommendation	
Agree/strongly agree	87.5
Neutral	12.5
Disagree/strongly disagree	0.0
Appropriateness of recommendation	
Agree/strongly agree	87.5
Neutral	12.5
Disagree/strongly disagree	0.0
Importance of recommendation	
Agree/strongly agree	87.5
Neutral	12.5
Disagree/strongly disagree	0.0
Change anything about the recommendation	
Yes	25.0
No	75.0

Recommendation 3.2 Clinicians may consider the use of glutamine supplementation in conjunction with other treatment interventions for the management of people with intestinal permeability.	
Understanding of recommendation	
Good/very good	100.0
Neutral	0.0
Poor/very poor	0.0
Agreement with recommendation	
Agree/strongly agree	87.5
Neutral	12.5
Disagree/strongly disagree	0.0
Appropriateness of recommendation	
Agree/strongly agree	87.5
Neutral	12.5
Disagree/strongly disagree	0.0
Importance of recommendation	
Agree/strongly agree	87.5
Neutral	12.5
Disagree/strongly disagree	0.0
Change anything about the recommendation	
Yes	12.5
No	87.5
Recommendation 3.3 Clinicians should consider the use of short-term lactoferrin supplementation for the treatment of people with nonsteroidal anti-inflammatory drug induced intestinal permeability.	
Understanding of recommendation	
Good/very good	100.0
Neutral	0.0
Poor/very poor	0.0
Agreement with recommendation	
Agree/strongly agree	100.0
Neutral	0.0
Disagree/strongly disagree	0.0
Appropriateness of recommendation	
Agree/strongly agree	87.5
Neutral	12.5
Disagree/strongly disagree	0.0
Importance of recommendation	
Agree/strongly agree	87.5
Neutral	12.5

Disagree/strongly disagree	0.0
Change anything about the recommendation	
Yes	12.5
No	87.5

7.5 STAKEHOLDERS' VIEWS AND PREFERENCES TOWARDS PLANT-BASED MEDICINE SUPPLEMENTATION RECOMMENDATIONS

Although well understood, stakeholders report a lack of consensus on the agreement, appropriateness, and importance of recommendation 4.1 (insufficient evidence for plant-based medicine use) (Table 7.4). While recommendation 4.2 (use plant-based medicines with pre-clinical research) received an agreed consensus for the agreement, appropriateness, and importance of the recommendation to be followed in clinical practice.

Table 7.4 Stakeholders' views and preferences towards plant-based medicine supplementation recommendations (n=8)

Recommendations	**Response %**
Recommendation 4.1 There is insufficient evidence to form a recommendation on the use of plant-based medicines as a collective group for the treatment of people with intestinal permeability.	
Understanding of recommendation	
Good/very good	100.0
Neutral	0.0
Poor/very poor	0.0
Agreement with recommendation	
Agree/strongly agree	37.5
Neutral	37.5
Disagree/strongly disagree	25.0
Appropriateness of recommendation	
Agree/strongly agree	37.5

Neutral	37.5
Disagree/strongly disagree	25.0
Importance of recommendation	
Agree/strongly agree	25.0
Neutral	50.0
Disagree/strongly disagree	25.0
Change anything about the recommendation	
Yes	37.5
No	62.5
Recommendation 4.2 Clinicians may consider the use of plant-based medicines which are supported by pre-clinical research in conjunction with other treatment interventions for the management people with intestinal permeability.	
Understanding of recommendation	
Good/very good	100.0
Neutral	0.0
Poor/very poor	0.0
Agreement with recommendation	
Agree/strongly agree	87.5
Neutral	12.5
Disagree/strongly disagree	0.0
Appropriateness of recommendation	
Agree/strongly agree	87.5
Neutral	12.5
Disagree/strongly disagree	0.0
Importance of recommendation	
Agree/strongly agree	87.5
Neutral	12.5
Disagree/strongly disagree	0.0
Change anything about the recommendation	
Yes	12.5
No	87.5

7.6 STAKEHOLDERS' VIEWS AND PREFERENCES TOWARDS ESSENTIAL FATTY ACID SUPPLEMENTATION RECOMMENDATIONS

Although well understood, stakeholders report a lack of consensus on the agreement, appropriateness, and importance of recommendation 5.1 (insufficient evidence for essential fatty acid use), with a disagreement rate of 37.5% found in all three domains (Table 7.5).

Table 7.5 Stakeholders' views and preferences towards essential fatty acid supplementation recommendations (n=8)

Recommendations	Response %
Recommendation 5.1 There is insufficient evidence to form a recommendation on the use of essential fatty acid supplementation for the treatment of people with intestinal permeability.	
Understanding of recommendation	
Good/very good	100.0
Neutral	0.0
Poor/very poor	0.0
Agreement with recommendation	
Agree/strongly agree	50.0
Neutral	12.5
Disagree/strongly disagree	37.5
Appropriateness of recommendation	
Agree/strongly agree	50.0
Neutral	12.5
Disagree/strongly disagree	37.5
Importance of recommendation	
Agree/strongly agree	50.0
Neutral	12.5
Disagree/strongly disagree	37.5
Change anything about the recommendation	
Yes	25.0
No	75.0

7.7 STAKEHOLDERS' VIEWS AND PREFERENCES TOWARDS MINERAL SUPPLEMENTATION RECOMMENDATIONS

Stakeholders had a consensus on the understanding, agreement, appropriateness, and importance of zinc supplementation for IP management, with recommendation 6.1 identified as the most agreed-upon recommendation (Table 7.6).

Table 7.6 Stakeholders' views and preferences towards mineral supplementation recommendations (n=8)

Recommendations	Response %
Recommendation 6.1 Clinicians may consider using zinc supplementation in the treatment of people with intestinal permeability.	
Understanding of recommendation	
Good/very good	100.0
Neutral	0.0
Poor/very poor	0.0
Agreement with recommendation	
Agree/strongly agree	100.0
Neutral	0.0
Disagree/strongly disagree	0.0
Appropriateness of recommendation	
Agree/strongly agree	100.0
Neutral	0.0
Disagree/strongly disagree	0.0
Importance of recommendation	
Agree/strongly agree	100.0
Neutral	0.0
Disagree/strongly disagree	0.0
Change anything about the recommendation	
Yes	12.5
No	87.5

7.8 RECOMMENDATION WORDING AND STRENGTH

From the 38 recommendations, stakeholder feedback resulted in modification of 19 recommendations. Table 7.7 provides the changes made based on stakeholders' feedback. Three recommendations 2.16 (not to use probiotics in NSAID-induced IP), 2.17 (not to use prebiotics in NSAID-induced IP) and 2.18 (not to use synbiotics in NSAID-induced IP) had the strength reduced from *recommendation* to *option* as stakeholder feedback suggested the number of explored prebiotics, probiotics and synbiotic were limited and the results may not be applicable to all therapies. Furthermore, recommendation 2.11 (avoid polydextrose) was downgraded from a *consensus-based recommendation* to a *practice point* as stakeholder feedback suggested the strength of recommendation was too strong to reflect the level of evidence.

The feedback on four recommendations 1.3 (consume high fibre diet), 1.4 (meet suggested dietary intake for dietary fibre), 2.6 (consume kefir) and 2.10 (use prebiotic with pre-clinical research), was to indicate a trial of the intervention before continued use. Therefore, these recommendations were modified to include "*trialling and if tolerated, consume".*

Further clarification on nine recommendations (1.1, 1.7, 1.8, 1.10, 1.11, 1.12, 1.13, 2.9, 2.15) was suggested by stakeholders. These clarifications included specifying the term 'alcoholic' drinks (recommendation 1.1), the type of fat (recommendation 1.7) and fructose (recommendation 1.8). While others were

regarding the clarification on when the recommendations should be followed; in patients with obesity (recommendation 1.10), during the treatment of IP (recommendation 1.11), as a treatment aim (recommendations 1.12 and 1.13). Clarification was further suggested to indicate specific treatment rather than broad intervention in recommendations 2.9 and 2.15. Changes to recommendations 2.3 (probiotic use for 3 months) and 2.13 (synbiotic use for 3 months) were suggested with feedback from stakeholders indicating a longer period would be required for clinical effectiveness. The wording of one recommendation (2.2) was modified to reflect the correct scientific naming of *Saccharomyces boulardii.*

Table 7.7 Changes to recommendations based on stakeholder feedback

No.	Recommendation	Strength	Recommendation	Strength
Dietary based recommendations				
Alcohol recommendations				
1.1	People with intestinal permeability should consider consuming no more than 10 standard drinks a week and no more than 4 standard drinks on any one day in accordance with the Australian Dietary Guidelines during the treatment of intestinal permeability.	⊕⊕⊕⊕	People with intestinal permeability should consider consuming no more than 10 standard **alcoholic** drinks a week and no more than 4 standard **alcoholic** drinks on any one day in accordance with the Australian Dietary Guidelines during the treatment of intestinal permeability.	⊕⊕⊕⊕
1.2	People with intestinal permeability may consider limiting or avoiding alcohol consumption during the short-term treatment of intestinal permeability.	⊕⊕	People with intestinal permeability may consider limiting or avoiding alcohol consumption during the short-term treatment of intestinal permeability.	⊕⊕
Dietary fibre recommendations				
1.3	People with intestinal permeability should consider consuming a diet high in dietary fibre from a diverse range of sources.	⊕⊕⊕⊕	People with intestinal permeability should consider **trialling and if tolerated, consume** a diet high in dietary fibre from a diverse range of sources.	⊕⊕⊕⊕
1.4	Clinicians are advised to recommend patients to consume 38g for men and 28g for female of dietary fibre daily while treating patients with intestinal permeability.	⊕⊕	Clinicians are advised to **trial and if tolerated,** recommend patients to consume 38g for men and 28g for female of dietary fibre daily while treating patients with intestinal permeability.	⊕⊕

1.5	Clinicians are encouraged to recommend gluten-free sources of dietary fibre to patients with confirmed intestinal permeability.	⊕⊕	Clinicians are encouraged to recommend gluten-free sources of dietary fibre to patients with confirmed intestinal permeability.	⊕⊕
Macronutrient ratio recommendations				
1.6	People with intestinal permeability should consider consuming the Acceptable Macronutrient Distribution Range of protein (15-25%), fats (20-35%) and carbohydrates (45-65%) in accordance with the Australian Dietary Guidelines.	⊕⊕⊕⊕	People with intestinal permeability should consider consuming the Acceptable Macronutrient Distribution Range of protein (15-25%), fats (20-35%) and carbohydrates (45-65%) in accordance with the Australian Dietary Guidelines.	⊕⊕⊕⊕
1.7	People with intestinal permeability should consider NOT consuming a diet high in fat.	⊕⊕⊕⊕	People with intestinal permeability should consider **consuming a diet moderate in fat and limit high consumption of long-chain saturated fatty acids.**	⊕⊕⊕⊕
1.8	People with intestinal permeability should consider NOT consuming a diet high in fructose.	⊕⊕⊕⊕	People with intestinal permeability should consider NOT consuming a diet high in **free** fructose.	⊕⊕⊕⊕
Energy intake recommendations				
1.9	People with intestinal permeability may consider consuming the estimated energy requirements in accordance with the Australian Dietary Guidelines.	⊕⊕⊕	People with intestinal permeability may consider consuming the estimated energy requirements in accordance with the Australian Dietary Guidelines.	⊕⊕⊕
1.10	Clinicians may consider using a kilojoule restricted diet in the short-term treatment of people with confirmed intestinal permeability.	⊕⊕⊕	Clinicians may consider using a kilojoule restricted diet in the short-term treatment of people with confirmed intestinal permeability **when clinically appropriate (e.g., obesity).**	⊕⊕⊕
Gluten-free diet recommendations				

1.11	Clinicians should only advise a strict gluten-free diet if clinical symptoms or pathology indicate a gluten intolerance, sensitivity or allergy.	⊕⊕⊕⊕⊕	Clinicians should only advise a strict gluten-free diet **during the treatment of people with confirmed intestinal permeability** if clinical symptoms or pathology indicate a gluten intolerance, sensitivity or allergy.	⊕⊕⊕⊕⊕
1.12	Clinicians should only advise a gluten-free diet during the short-term treatment of people with confirmed intestinal permeability that report clinical symptoms in response to the consumption of gluten after the investigation for gluten intolerance, sensitivity or allergy has been carried out.	⊕⊕⊕⊕⊕	Clinicians **should aim to** advise a gluten-free diet during the short-term treatment of people with confirmed intestinal permeability that report clinical symptoms in response to the consumption of gluten after the investigation for gluten intolerance, sensitivity or allergy has been carried out	⊕⊕⊕⊕⊕
1.13	Clinicians should offer a low gluten diet for the management of people with confirmed intestinal permeability that report no clinical symptoms or pathology indicating a gluten intolerance, sensitivity or allergy.	⊕⊕⊕⊕⊕	Clinicians should **aim to** offer a low gluten diet for the management of people with confirmed intestinal permeability that report no clinical symptoms or pathology indicating a gluten intolerance, sensitivity or allergy.	⊕⊕⊕⊕⊕
Probiotic, prebiotic and synbiotic supplementation recommendations				
Probiotics				
2.1	There is insufficient evidence to form a recommendation on the use of probiotics as a collective group for the treatment of people with intestinal permeability.	∅	There is insufficient evidence to form a recommendation on the use of probiotics as a collective group for the treatment of people with intestinal permeability.	∅
2.2	Clinicians may consider using *Saccharomyces boulardii* supplementation in the treatment of people with intestinal permeability.	⊕⊕⊕	Clinicians may consider using ***Saccharomyces cerevisiae var boulardii*** **(***Saccharomyces boulardii***)** supplementation in the treatment of people with intestinal permeability.	⊕⊕⊕

2.3	Clinicians may consider the use of effective probiotics for a period of 3 months when treating people with intestinal permeability.	⊕⊕⊕	Clinicians may consider the use of effective probiotics for a **minimum** of 3 months when treating people with intestinal permeability.	⊕⊕⊕
2.4	Clinicians may consider researching probiotic strains for their effectiveness before using them to treat people with intestinal permeability.	⊕	Clinicians may consider researching probiotic strains for their effectiveness before using them to treat people with intestinal permeability.	⊕
2.5	Clinicians may consider the use of probiotics which are supported by pre-clinical research in conjunction with other treatment interventions for the management people with intestinal permeability.	⊕	Clinicians may consider the use of probiotics which are supported by pre-clinical research in conjunction with other treatment interventions for the management people with intestinal permeability.	⊕
Probiotic drink				
2.6	People with intestinal permeability should consider the consumption of fermented milk products such as kefir.	⊕⊕⊕⊕	People with intestinal permeability should consider **trialling and if tolerated, consume** fermented milk products such as kefir.	⊕⊕⊕⊕
2.7	People with intestinal permeability may consider NOT consuming Yakult light®.	⊕⊕⊕	People with intestinal permeability may consider NOT consuming Yakult light®.	⊕⊕⊕
Prebiotics				
2.8	There is insufficient evidence to form a recommendation on the use of prebiotics as a collective group for the treatment of people with intestinal permeability.	∅	There is insufficient evidence to form a recommendation on the use of prebiotics as a collective group for the treatment of people with intestinal permeability.	∅
2.9	Clinicians may consider researching prebiotic for their effectiveness before using them in the treatment of people with intestinal permeability.	⊕	Clinicians may consider researching **specific** prebiotic for their effectiveness before using them in the treatment of people with intestinal permeability.	⊕

2.10	Clinicians may consider the use of prebiotic which are supported by pre-clinical research in conjunction with other treatment interventions for the management people with intestinal permeability.	⊕	Clinicians may consider **trialling and if tolerated, recommend patients to** use prebiotic which are supported by pre-clinical research in conjunction with other treatment interventions for the management people with intestinal permeability.	⊕
2.11	Clinicians may consider NOT using polydextrose in the treatment of people with intestinal permeability.	⊕⊕	Clinicians may consider NOT using polydextrose in the treatment of people with intestinal permeability.	⊕
Synbiotic				
2.12	Clinicians may consider the use of effective synbiotic in the treatment of people with intestinal permeability.	⊕⊕⊕	Clinicians may consider the use of effective synbiotic in the treatment of people with intestinal permeability.	⊕⊕⊕
2.13	Clinicians may consider the use of effective synbiotic for a period of 3 months when treating people with intestinal permeability.	⊕⊕⊕	Clinicians may consider the use of effective synbiotic for a **minimum** of 3 months when treating people with intestinal permeability.	⊕⊕⊕
2.14	Clinicians may consider NOT using polydextrose and *Bifidobacterium animalis ssp. lactis* 420 in the treatment of people with intestinal permeability.	⊕⊕⊕	Clinicians may consider NOT using polydextrose and *Bifidobacterium animalis ssp. lactis* 420 in the treatment of people with intestinal permeability.	⊕⊕⊕
2.15	Clinicians may consider the use of synbiotic which are supported by pre-clinical research in conjunction with other treatment interventions for the management people with intestinal permeability.	⊕	Clinicians may consider the use of **specific** synbiotic which are supported by pre-clinical research in conjunction with other treatment interventions for the management people with intestinal permeability.	⊕
NSAID induced intestinal permeability				
2.16	Clinicians should consider NOT using probiotics for the treatment of people with nonsteroidal anti-inflammatory drug induced intestinal permeability.	⊕⊕⊕⊕	Clinicians **may** consider NOT using probiotics for the treatment of people with nonsteroidal anti-inflammatory drug induced intestinal permeability.	⊕⊕⊕

2.17	Clinicians should consider NOT using prebiotics for the treatment of people with nonsteroidal anti-inflammatory drug induced intestinal permeability.	⊕⊕⊕⊕	Clinicians **may** consider NOT using prebiotics for the treatment of people with nonsteroidal anti-inflammatory drug induced intestinal permeability.	⊕⊕⊕
2.18	Clinicians should consider NOT using synbiotics for the treatment of people with nonsteroidal anti-inflammatory drug induced intestinal permeability.	⊕⊕⊕⊕	Clinicians **may** consider NOT using synbiotics for the treatment of people with nonsteroidal anti-inflammatory drug induced intestinal permeability.	⊕⊕⊕
Amino acid supplementation recommendations				
Glutamine				
3.1	Clinicians should offer glutamine supplementation for the treatment of people with intestinal permeability.	⊕⊕⊕⊕⊕	Clinicians should offer glutamine supplementation for the treatment of people with intestinal permeability.	⊕⊕⊕⊕⊕
3.2	Clinicians may consider the use of glutamine supplementation in conjunction with other treatment interventions for the management of people with intestinal permeability.	⊕⊕	Clinicians may consider the use of glutamine supplementation in conjunction with other treatment interventions for the management of people with intestinal permeability.	⊕⊕
NSAID-induced intestinal permeability				
3.3	Clinicians should consider the use of short-term lactoferrin supplementation for the treatment of people with nonsteroidal anti-inflammatory drug induced intestinal permeability.	⊕⊕⊕⊕	Clinicians should consider the use of short-term lactoferrin supplementation for the treatment of people with nonsteroidal anti-inflammatory drug induced intestinal permeability.	⊕⊕⊕⊕
Plant-based medicine supplementation recommendations				
4.1	There is insufficient evidence to form a recommendation on the use of plant-based medicines as a collective group for the treatment of people with intestinal permeability.	∅	There is insufficient evidence to form a recommendation on the use of plant-based medicines as a collective group for the treatment of people with intestinal permeability.	∅

4.2	Clinicians may consider the use of plant-based medicines which are supported by pre-clinical research in conjunction with other treatment interventions for the management people with intestinal permeability.	⊕	Clinicians may consider the use of plant-based medicines which are supported by pre-clinical research in conjunction with other treatment interventions for the management people with intestinal permeability.	⊕
Essential fatty acid supplementation recommendations				
5.1	There is insufficient evidence to form a recommendation on the use of essential fatty acid supplementation for the treatment of people with intestinal permeability.	∅	There is insufficient evidence to form a recommendation on the use of essential fatty acid supplementation for the treatment of people with intestinal permeability.	∅
Mineral supplement recommendations				
6.1	Clinicians may consider using zinc supplementation in the treatment of people with intestinal permeability.	⊕⊕⊕	Clinicians may consider using zinc supplementation in the treatment of people with intestinal permeability.	⊕⊕⊕

7.9 CHAPTER SUMMARY

This chapter explored the views and preferences of key stakeholders known to represent users of the IP Guideline. As a result, the 38 recommendations were revised to reflect the necessary changes to ensure the IP Guideline was based on evidence and were clinically relevant. The recommendations identified as having a low agreement rate or suggested a change was required were modified to ensure they are relevant and reflect clinical practice. Collectively, the IP Guideline, with the input from stakeholders, is suggested to align with the views and practices of clinicians supporting patients with IP in clinical practice.

8. CLINICAL PRACTICE GUIDELINE FOR THE MANAGEMENT OF INCREASED INTESTINAL PERMEABILITY: IP GUIDELINE

In the preceding chapter, views and preferences of stakeholders were explored to gain feedback on the recommendations in the IP Guideline. This project aims to develop evidence-based treatment recommendations that align with the views and preferences of Australian adults with IP and essential stakeholders. Therefore, this chapter provides the IP Guideline and presents the clinical need for the clinical question, a summary of the evidence and justification for the recommendations.

8.1 REPORTING OF IP GUIDELINE

The following section contains the *IP Guideline*, which has been formatted based on the NHMRC *Guidelines for Guidelines* Handbook to meet the 2016 *NHMRC Standards for Guidelines.*[215] Details are as followed:

> Leech, B, McIntyre, E, Steel, A, Sibbritt, D (2022) "Clinical practice guideline for the management of increased intestinal permeability: IP Guideline", University of Technology Sydney.

8.2 EXECUTIVE SUMMARY

Increased intestinal permeability (IP), also known as 'leaky gut', has gained researchers attention in recent years, with research linking the integrity of the intestine to health and disease.[328] IP can be defined as the loss of integrity between the cells of the small intestine caused by the disassembling of the proteins holding the cells together. The idea of IP was first mentioned in the literature during the 1960s[128] however, it was not until the 2000s where evidence emerged describing the potential mechanism of action.[130] Although the consequence of IP remains unclear, preliminary evidence suggest Australian adults with suspected IP experience disease burden.[220] This disease burden includes increased healthcare costs associated with the management of IP, lower subjective wellbeing compared to the Australian population and poor health related quality of life.[220]

The need to develop a *clinical practice guideline for the management of increased intestinal permeability* (IP Guideline) was identified after health services research revealed gaps in both the published literature and clinical practice[4,5] with discrepancy between what practitioners are using and what patients desire.[4,5,219,220] This clinical practice guideline serves as the first guideline for IP as no guideline surrounding any part of the management of IP has been developed in Australia or internationally.

The Working Group undertook a structured and evidence-based approach based on the NHMRC *Guidelines for Guidelines* Handbook to meet the 2016 *NHMRC Standards for Guidelines* in the development of the IP Guideline.[215] A total of 16

clinical questions were addressed, producing 38 recommendations: 27 evidence based recommendations, seven practice points and four consensus based recommendations. Each recommendation was reviewed and assessed by key stakeholders.

8.3 GUIDELINE PURPOSE AND AIM

The purpose of the IP Guideline is to utilise the best available evidence while considering the views and preferences from a multidisciplinary group of stakeholders and consumers. The IP Guideline aims to provide practitioners and consumers with a transparent evidence-based guidance for the management of altered IP to optimise patient care, improve health outcomes and reduce variation in care for Australian practitioners in private practice.

The IP Guideline aims to ensure Australian adults with IP receive, optimal evidence-based care by:

1. identifying any dietary choices available for the management of altered IP in Australian adults;
2. identifying any probiotic, prebiotic and synbiotic supplementation available for the management of altered IP in Australian adults;
3. identifying any amino acid supplementation available for the management of altered IP in Australian adults;
4. identifying any plant-based medicine supplementation available for the management of altered IP in Australian adults;
5. identifying any essential fatty acid supplementation available for the management of altered IP in Australian adults;

6. identifying any mineral supplementation available for the management of altered IP in Australian adults;
7. identifying any vitamin supplementation available for the management of altered IP in Australian adults;
8. identifying any colostrum supplementation available for the management of altered IP in Australian adults.

8.4 SETTING AND AUDIENCE

The IP Guideline and recommendations are designed to inform the care provided by clinicians in private clinical practice to Australian adults with suspected or confirmed IP. Confirmed IP is classified as an elevation in the commercially available lactulose/mannitol ratio urine test or elevation of stool zonulin.

8.5 GUIDELINE DEVELOPMENT GROUP

The Guideline Development Group was comprised of two sub-groups: the *Working Group* and *Stakeholder Group*. These groups were formed from a multidisciplinary background of health professionals representing all potential clinicians which may see patients with IP, content experts, consumers and other major stakeholders. The Working was involved in the development of the IP Guideline. Whereas the Stakeholder Group provided their views and preferences on the drafted recommendations. A detailed description of the recruitment strategy, responsibilities of each member, and the method used to assess the conflict of interest of each member are in the *Guideline Development Process* (Section 3.4).

8.6 GUIDELINE DEVELOPMENT METHODS

The IP Guideline was based on the NHMRC *Guidelines for Guidelines* Handbook to meet the 2016 *NHMRC Standards for Guidelines*.[215] The level of evidence for each recommendation was determined based on both the NHMRC grades for recommendations and NHMRC Evaluation of Evidence process.[217] The reporting of the IP Guideline followed the RIGHT statement.[218] A total of 12 steps were undertaken to develop the IP Guideline:

1. Create multidisciplinary guideline development group;
2. Identify scope and topics for guideline;
3. Develop a structured clinical question;
4. Perform a systematic review;
5. Summarise the relevant data;
6. Risk of bias assessment;
7. Assess the body of evidence and formulate recommendations;
8. Grade recommendation according to NHMRC;
9. Write the content narrative;
10. Stakeholders review recommendations;
11. Finalise guideline content;
12. Disseminate and implement IP Guideline.

8.7 CONSUMER INVOLVEMENT

The views and preferences of consumers were continuously integrated into the development of the IP Guideline from the initial scoping and planning through to

the implementation of the IP Guideline according to the NHMRC requirement A.4. A survey study design was used to involve the consumers (target population) in the development of the IP Guideline. This method ensured the views and preference of the target population were incorporated in the IP Guideline, ensuring it is both relevant and appropriate.

A cross-sectional survey (*The Leaky Gut Survey*) of 589 Australian adults with suspected IP was undertaken during the initial stages of the IP Guideline development process.[219,220] *The Leaky Gut Survey* was designed to capture the health-seeking behaviours, views and preference of Australian adults identifying as having suspected or confirmed IP concerning the management and assessment of IP. Although the Stakeholder Group did not include a representative from Aboriginal and Torres Strait Islander Peoples, the views and preferences of the community were incorporated during *The Leaky Gut Survey* as participants included in this study identified as Aboriginal and Torres Strait Islander.

8.8 FUNDING

The development of the IP Guideline was funded by the Australian Research Centre in Complementary and Integrative Medicine (ARCCIM), providing a total of $4470 in support of guideline development, publication and dissemination. The Australian Government Research Training Program Scholarship provided Bradley Leech with a scholarship. The scholarship funding had no influence on the development or content of the guidelines.

8.9 INTEROPERATING THE RECOMMENDATIONS

A detailed method of guideline development including the process used to evaluate and form the recommendations can be found in the *Guideline Development Process* (Section 3.4). To assist in interpreting the IP Guideline each recommendation has been categorised according to the type of available evidence (Table 8.1) and classified according to the strength of the recommendation (Table 8.2).

Table 8.1 Categories of the IP Guideline recommendations

EBR	Evidence-based recommendations: A recommendation formulated after a systematic review of the evidence, with supporting references.
CBR	Consensus-based recommendations: A recommendation formulated in the absence of quality evidence, with the guideline development group forming a consensus.
PP	Practice points: A recommendation on a subject that is outside the scope of the search strategy for the systematic review, based on expert opinion and formulated by a consensus process

Table 8.2 Strength of recommendations

Strong recommendation	⊕⊕⊕⊕	Clinicians should follow a strong recommendation unless a clear and compelling rationale for an alternative approach is present.
Recommendation	⊕⊕⊕	Clinicians should also generally follow a recommendation but should remain alert to new information and sensitive to patient preferences.
Option	⊕⊕	Clinicians should be flexible in their decision making regarding appropriate practice, although they may set bounds on alternatives; patient preference should have a substantial influencing role.
Consensus-based recommendation	⊕	
Practice point	∅	
No recommendation		Clinicians should feel little constraint in their decision making and be alert to new published evidence that clarifies the balance of benefit versus harm; patient preference should have a substantial influencing role.

8.9.1 RECOMMENDATION WORDING ACCORDING TO STRENGTH OF EVIDENCE

The IP Guideline utilises the National Institute for Health and Care Excellence methodology for the wording of the recommendations.[226] Wording for a strong recommendation uses terms such as “offer”, “advise”, “do NOT offer”, or “do NOT advise”, while the wording for a recommendation contains “consider” or “consider NOT”. For options, consensus-based recommendations and practice points, the key terminology contains “may consider” to reflect the strength of the recommendation and evidence.

8.10 RECOMMENDATION SUMMARY

Table 8.3 Recommendation Summary

No.	Category	Recommendation	Strength
Dietary based recommendations			
Alcohol recommendations			
1.1	EBR	People with intestinal permeability should consider consuming no more than 10 standard alcoholic drinks a week and no more than 4 standard alcoholic drinks on any one day in accordance with the Australian Dietary Guidelines during the treatment of intestinal permeability.	⊕⊕⊕⊕
1.2	CBR	People with intestinal permeability may consider limiting or avoiding alcohol consumption during the short-term treatment of intestinal permeability.	⊕⊕
Dietary fibre recommendations			
1.3	EBR	People with intestinal permeability should consider trialling and if tolerated, consume a diet high in dietary fibre from a diverse range of sources.	⊕⊕⊕⊕
1.4	CBR	Clinicians are advised to trial and if tolerated, recommend patients to consume 38g for men and 28g	⊕⊕

		for female of dietary fibre daily while treating patients with intestinal permeability.	
1.5	CBR	Clinicians are encouraged to recommend gluten-free sources of dietary fibre to patients with confirmed intestinal permeability.	⊕⊕
Macronutrient ratio recommendations			
1.6	EBR	People with intestinal permeability should consider consuming the Acceptable Macronutrient Distribution Range of protein (15-25%), fats (20-35%) and carbohydrates (45-65%) in accordance with the Australian Dietary Guidelines.	⊕⊕⊕⊕
1.7	EBR	People with intestinal permeability should consider consuming a diet moderate in fat and limit high consumption of long-chain saturated fatty acids.	⊕⊕⊕⊕
1.8	EBR	People with intestinal permeability should consider NOT consuming a diet high in free fructose.	⊕⊕⊕⊕
Energy intake recommendations			
1.9	EBR	People with intestinal permeability may consider consuming the estimated energy requirements in accordance with the Australian Dietary Guidelines.	⊕⊕⊕
1.10	EBR	Clinicians may consider using a kilojoule restricted diet in the short-term treatment of people with confirmed intestinal permeability when clinically appropriate (e.g., obesity).	⊕⊕⊕
Gluten-free diet recommendations			
1.11	EBR	Clinicians should only advise a strict gluten-free diet during the treatment of people with confirmed intestinal permeability if clinical symptoms or pathology indicate a gluten intolerance, sensitivity or allergy.	⊕⊕⊕⊕⊕
1.12	EBR	Clinicians should aim to advise a gluten-free diet during the short-term treatment of people with confirmed intestinal permeability that report clinical symptoms in response to the consumption of gluten after the investigation for gluten intolerance, sensitivity or allergy has been carried out	⊕⊕⊕⊕⊕
1.13	EBR	Clinicians should aim to offer a low gluten diet for the management of people with confirmed intestinal permeability that report no clinical symptoms or	⊕⊕⊕⊕⊕

		pathology indicating a gluten intolerance, sensitivity or allergy.	
Probiotic, prebiotic and synbiotic supplementation recommendations			
Probiotics			
2.1	EBR	There is insufficient evidence to form a recommendation on the use of probiotics as a collective group for the treatment of people with intestinal permeability.	∅
2.2	EBR	Clinicians may consider using *Saccharomyces cerevisiae var boulardii (Saccharomyces boulardii)* supplementation in the treatment of people with intestinal permeability.	⊕⊕⊕
2.3	EBR	Clinicians may consider the use of effective probiotics for a minimum of 3 months when treating people with intestinal permeability.	⊕⊕⊕
2.4	PP	Clinicians may consider researching probiotic strains for their effectiveness before using them to treat people with intestinal permeability.	⊕
2.5	PP	Clinicians may consider the use of probiotics which are supported by pre-clinical research in conjunction with other treatment interventions for the management people with intestinal permeability.	⊕
Probiotic drink			
2.6	EBR	People with intestinal permeability should consider trialling and if tolerated, consume fermented milk products such as kefir.	⊕⊕⊕⊕
2.7	EBR	People with intestinal permeability may consider NOT consuming Yakult light®.	⊕⊕⊕
Prebiotics			
2.8	EBR	There is insufficient evidence to form a recommendation on the use of prebiotics as a collective group for the treatment of people with intestinal permeability.	∅
2.9	PP	Clinicians may consider researching specific prebiotic for their effectiveness before using them in the treatment of people with intestinal permeability.	⊕
2.10	PP	Clinicians may consider trialling and if tolerated, recommend patients to use prebiotic which are supported by pre-clinical research in conjunction with	⊕

		other treatment interventions for the management people with intestinal permeability.	
2.11	PP	Clinicians may consider NOT using polydextrose in the treatment of people with intestinal permeability.	⊕
Synbiotic			
2.12	EBR	Clinicians may consider the use of effective synbiotic in the treatment of people with intestinal permeability.	⊕⊕⊕
2.13	EBR	Clinicians may consider the use of effective synbiotic for a minimum of 3 months when treating people with intestinal permeability.	⊕⊕⊕
2.14	EBR	Clinicians may consider NOT using polydextrose and *Bifidobacterium animalis ssp. lactis* 420 in the treatment of people with intestinal permeability.	⊕⊕⊕
2.15	PP	Clinicians may consider the use of specific synbiotic which are supported by pre-clinical research in conjunction with other treatment interventions for the management people with intestinal permeability.	⊕
NSAID induced intestinal permeability			
2.16	EBR	Clinicians may consider NOT using probiotics for the treatment of people with nonsteroidal anti-inflammatory drug induced intestinal permeability.	⊕⊕⊕
2.17	EBR	Clinicians may consider NOT using prebiotics for the treatment of people with nonsteroidal anti-inflammatory drug induced intestinal permeability.	⊕⊕⊕
2.18	EBR	Clinicians may consider NOT using synbiotics for the treatment of people with nonsteroidal anti-inflammatory drug induced intestinal permeability.	⊕⊕⊕
Amino acid supplementation recommendations			
Glutamine			
3.1	EBR	Clinicians should offer glutamine supplementation for the treatment of people with intestinal permeability.	⊕⊕⊕⊕⊕
3.2	CBR	Clinicians may consider the use of glutamine supplementation in conjunction with other treatment interventions for the management of people with intestinal permeability.	⊕⊕
NSAID-induced intestinal permeability			
3.3	EBR	Clinicians should consider the use of short-term lactoferrin supplementation for the treatment of people	⊕⊕⊕⊕

		with nonsteroidal anti-inflammatory drug induced intestinal permeability.	
Plant-based medicine supplementation recommendations			
4.1	EBR	There is insufficient evidence to form a recommendation on the use of plant-based medicines as a collective group for the treatment of people with intestinal permeability.	∅
4.2	PP	Clinicians may consider the use of plant-based medicines which are supported by pre-clinical research in conjunction with other treatment interventions for the management people with intestinal permeability.	⊕
Essential fatty acid supplementation recommendations			
5.1	EBR	There is insufficient evidence to form a recommendation on the use of essential fatty acid supplementation for the treatment of people with intestinal permeability.	∅
Mineral supplement recommendations			
6.1	EBR	Clinicians may consider using zinc supplementation in the treatment of people with intestinal permeability.	⊕⊕⊕

Abbreviations: CBR = Consensus-based recommendation; EBR = evidence-based recommendation; NSAID = nonsteroidal anti-inflammatory drug; PP = practice point.

8.11 DIETARY CHOICES

8.11.1 CLINICAL QUESTIONS

Clinical Question 1: In Australian adults with increased intestinal permeability, what are the benefits of dietary choices for the treatment of increased intestinal permeability?

Clinical Question 2: In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for dietary choices?

8.11.2 CLINICAL NEED FOR THE QUESTION

The consumption of some food products such as alcohol,[52,329-331] gluten,[2,332-334] and dairy[335,336] are suggested to negatively affect intestinal integrity. In contrast, other food products such as dietary fibre, including prebiotics, display a beneficial action on intestinal integrity.[337] Considering the opinions of clinicians that commonly treat patients with IP, dietary modification is the most frequently used treatment intervention used in the management of IP.[5] These clinicians were found to employ diverse dietary interventions while treating patients with IP, including the reduced consumption of alcohol, gluten, dairy while incorporating organic foods, apple cider vinegar, bone broth, lemon water and fermented foods. The opinions of clinicians appear to align with opinions of Australian adults with suspected IP. Dietary products are the preferred method for treating IP; over 80% of Australian adults with suspected IP report they prefer to be treated using dietary modification.[220] This population also choose to allocate their finances to dietary interventions regardless of their income manageability, with many of them prescribing the dietary change themselves. To further support the need for dietary

recommendations, patients expect that clinicians should have a comprehensive understanding of dietary treatment interventions to manage IP.[219]

8.11.3 SUMMARY OF EVIDENCE

A review of the literature found 22 studies addressing the two clinical questions (CQ.1 and CQ.2) regarding the impact of dietary intake on intestinal integrity. The risk of bias (RoB) assessment found the identified systematic review had a high RoB (*n* = 1),[221] while the seven randomised control trials were found to have some RoB; *n* = 5 with moderate RoB[275,276,278,280,287] and n = 2 with low RoB.[274,288] The 14 non-randomised clinical trials had moderate (*n* = 7),[272,277,279,281-284] serious (*n* = 4),[271,272,285,286] or critical (*n* = 3) RoB.[199,289,290] A summary of the evidence is provided here with a full review of the literature found in the *Technical Report*.

Four studies reported on the impact of alcohol consumption on IP, five studies explored the effect of dietary fibre on intestinal integrity, ten studies were included evaluating energy intake and macronutrient distribution on intestinal integrity, and four studies evaluating the effects of a gluten-free diet on IP. There is sufficient evidence to suggest dietary modifications can influence intestinal integrity, with some changes supporting health and integrity and others causing a detrimental effect to IP. However, a major limitation is the lack of high-quality research, even though the included articles utilising a non-randomised clinical trial study design. Studies on alcohol consumption most notably saw a lack of high-quality research, as such there is insufficient evidence to suggest complete avoidance of alcohol is necessary during the treatment of IP.[221,271-273] There is potential benefit in limiting alcohol consumption in specific health conditions, however the

combination of low-quality evidence and mixed results suggests further randomised controlled trials and longitudinal research is required before evidence-based recommendations can be made.[221,271-273]

There is moderate-quality evidence to suggest the consumption of dietary fibre supports intestinal integrity and improves IP.[199,274-277] Although limited evidence is available on the type and amount of dietary fibre, consuming a diverse range of dietary fibre with prebiotic properties appears to benefit intestinal integrity.[199,275,277] However, the fortification of wheat-based products with prebiotics (inulin or beta-glucan) results in mixed effects on IP.[275,276] There is limited low to moderate-quality evidence that patients at risk of IP or have confirmed IP should follow a gluten-free diet during IP treatment.[287-290] Instead, evidence suggests that patients at risk of IP could tolerate a low amount of gluten (<16g/day).[287] However, patients with positive HLA-DQ2/8, the genetic predisposition for coeliac disease, may not tolerate the consumption of gluten-containing products with a more significant impact on IP found in patients with positive HLA-DQ2/8 compared to negative HLA-DQ2/8 after the consumption of a gluten-containing diet.[288]

The evidence evaluating energy intake and macronutrient distribution was of low-quality.[277-286] In terms of energy intake, kilojoule restriction of 3,350-6700kJ/day has been found to improve IP[277,285] while overfeeding with either estimated energy requirements plus 4,180kJ/day[286] or 116% estimated energy requirements saw no significant effect on IP.[280] In contrast with this latter finding, increased energy intake (>10,945 kJ/day) was found to be an independent risk

factor for IP.[221] Although the distribution of macronutrients was found to have mixed results, a trend suggests a high-fat diet may have a detrimental effect on intestinal integrity. Total fat percentage was found to be an independent risk factor for IP.[221] Although short-term (<15 days) consumption of a high-fat diet (41-55% of estimated energy requirements from fats) does not appear to significantly impact intestinal integrity,[278,279,286] a slightly high-fat diet (35% of estimated energy requirements from fats) over a longer period (12 weeks) saw a significant impact on IP.[284] The only other macronutrient distribution ratio identified to influence intestinal integrity potentially was simple carbohydrates.[280,282] Although increased simple carbohydrate consumption saw mixed results, the most significant impact on intestinal integrity was the consequence of fructose consumption rather than other simple carbohydrates.[280,282]

Table 8.4 Recommendations: Dietary Based

No.	Category	Recommendation	Strength
Alcohol recommendations			
1.1	EBR	People with intestinal permeability should consider consuming no more than 10 standard alcoholic drinks a week and no more than 4 standard alcoholic drinks on any one day in accordance with the Australian Dietary Guidelines during the treatment of intestinal permeability.	⊕⊕⊕⊕
1.2	CBR	People with intestinal permeability may consider limiting or avoiding alcohol consumption during the short-term treatment of intestinal permeability.	⊕⊕
Dietary fibre recommendations			

1.3	EBR	People with intestinal permeability should consider trialling and if tolerated, consume a diet high in dietary fibre from a diverse range of sources.	⊕⊕⊕⊕
1.4	CBR	Clinicians are advised to trial and if tolerated, recommend patients to consume 38g for men and 28g for female of dietary fibre daily while treating patients with intestinal permeability.	⊕⊕
1.5	CBR	Clinicians are encouraged to recommend gluten-free sources of dietary fibre to patients with confirmed intestinal permeability.	⊕⊕
Macronutrient ratio recommendations			
1.6	EBR	People with intestinal permeability should consider consuming the Acceptable Macronutrient Distribution Range of protein (15-25%), fats (20-35%) and carbohydrates (45-65%) in accordance with the Australian Dietary Guidelines.	⊕⊕⊕⊕
1.7	EBR	People with intestinal permeability should consider consuming a diet moderate in fat and limit high consumption of long-chain saturated fatty acids.	⊕⊕⊕⊕
1.8	EBR	People with intestinal permeability should consider NOT consuming a diet high in free fructose.	⊕⊕⊕⊕
Energy intake recommendations			
1.9	EBR	People with intestinal permeability may consider consuming the estimated energy requirements in accordance with the Australian Dietary Guidelines.	⊕⊕⊕

1.10	EBR	Clinicians may consider using a kilojoule restricted diet in the short-term treatment of people with confirmed intestinal permeability when clinically appropriate (e.g., obesity).	⊕⊕⊕
Gluten-free diet recommendations			
1.11	EBR	Clinicians should only advise a strict gluten-free diet during the treatment of people with confirmed intestinal permeability if clinical symptoms or pathology indicate a gluten intolerance, sensitivity or allergy.	⊕⊕⊕⊕⊕
1.12	EBR	Clinicians should aim to advise a gluten-free diet during the short-term treatment of people with confirmed intestinal permeability that report clinical symptoms in response to the consumption of gluten after the investigation for gluten intolerance, sensitivity or allergy has been carried out	⊕⊕⊕⊕⊕
1.13	EBR	Clinicians should aim to offer a low gluten diet for the management of people with confirmed intestinal permeability that report no clinical symptoms or pathology indicating a gluten intolerance, sensitivity or allergy.	⊕⊕⊕⊕⊕

Abbreviations: CBR = Consensus-based recommendation; EBR = evidence-based recommendation; PP = practice point.

8.11.4 JUSTIFICATION

Recommendations were informed by the best available evidence on the impact dietary intake can have on intestinal integrity. The guideline development group

carefully considered the available literature and the importance patients with IP place of dietary treatments for the management of IP while forming each recommendation. As Australian adults with suspected IP report the desire to use dietary modifications as the primary treatment interventions in the management of IP, recommendations were formulated regardless of the low grade identified. As diet is not a short-term fix but rather a long-term solution, recommendations considered the length of included studies and the possible long-term health outcomes. While many studies involved healthy adults without IP or associated conditions, the recommendations also considered the potential effect diet might have on population groups with IP or disease associated with impaired IP.

With the available evidence and the current advice clinicians are currently providing patients with IP, the recommendation regarding alcohol consumption should follow the Australian Dietary Guidelines. However, to align with clinician's current views and preclinical research, a consensus-based recommendation was developed whereby patients with IP may limit or avoid alcohol consumption during the treatment period for IP.

Whilst limited evidence was found on the precise amount of dietary fibre required to provide a beneficial effect, the available evidence supports the consumption of a diverse range of dietary fibre. There is moderate-quality evidence indicating no harmful effects after the consumption of dietary fibre among individuals with suspected IP. Therefore, the best available recommendation is to follow the suggested dietary target for dietary fibre according to the Australian Dietary Guidelines. Prioritising the consumption of low gluten sources of dietary fibre was

included to complement the recommendations related to gluten consumption. Guiding patients to consume a strict gluten-free diet limits the consumption of major food groups and increases social stress involved in gluten avoidance. Therefore, evidence and consensus suggest a low gluten diet rather than a strict gluten-free diet may provide the best outcomes for individuals with IP.

The recommendation to follow the Acceptable Macronutrient Distribution Range in accordance with the Australian Dietary Guidelines is based on the findings that deviations from these reference ranges may results in worsening of IP. Furthermore, many of the included studies used a variation of the Acceptable Macronutrient Distribution Range as the control diet while exploring macronutrients ratio. Although there is conflicting evidence on the effect of a high-fat diet in the short-term (<15 days), the advice not to follow a high-fat diet is supported by the long-term (>12 weeks) effect of a slightly high-fat diet on both serum and stool zonulin. Limiting the consumption of excess fructose is supported by the available literature and the Australian Dietary Guidelines. Although total kilojoule intake is suggested to influence intestinal integrity, following the Australian Dietary Guidelines regarding kilojoule intake remains the best option for people with IP. However, while under the care of a clinician, there may be benefits for a short-term kilojoule restricted diet.

8.12 PROBIOTIC, PREBIOTIC AND SYNBIOTIC SUPPLEMENTATION

8.12.1 CLINICAL QUESTIONS

Clinical Question 3: In Australian adults with increased intestinal permeability, what are the benefits of oral probiotic, prebiotic or synbiotic supplementation for the treatment of increased intestinal permeability?

Clinical Question 4: In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for oral probiotic, prebiotic or synbiotic supplementation use?

8.12.2 CLINICAL NEED FOR THE QUESTION

Probiotics and prebiotics may influence intestinal integrity by changing the environment of the gastrointestinal system.[338,339] Mechanistic research indicate probiotics may increase gene expression of ZO-1, ZO-2, claudin-1, occluding, which are involved in tight junction modulation in the small intestine.[339] Other probiotics may stabilise the mucosal barrier by increasing mucin expression.[339] While gliadin-induced IP has been suggested to be reduced by the use of probiotics increasing ZO-1, claudin-1, and occluding gene expression.[340] Some probiotics can produce bacteriocins, antimicrobial peptides that inhibit pathogenic bacteria, thereby modifying the microbiome so that pathogenic bacteria are unable to stimulate IP.[341] While less is known about prebiotic effect on intestinal integrity, mechanistic research suggests prebiotics may mitigate the impact of

lipopolysaccharide on intestinal integrity and protect the mucosa from inflammation.[338]

Australian adults with suspected IP preferred the use of dietary supplements such as probiotics and prebiotics managing IP.[219] Specifically, the use of dietary supplements is the third most preferred treatment method for IP, with many people with IP frequently using dietary supplements (73%) for the management of IP.[219,220] Dietary supplements are most frequently prescribed by a naturopath, with over 70% of probiotic and prebiotic supplementation prescribed by a clinician.[220] This population group reports spending an average of AUD $2,175 on dietary supplements annually.[219] The economic burden of using dietary supplements does not appear to prohibit this populations' use of dietary supplements.[219] Almost 90% of Australian adults with suspected IP perceive that it is important for clinicians to be knowledgeable about dietary supplements.[219] The most frequently used dietary supplement in people with IP are probiotics, with prebiotics as a close third.[220] *Saccharomyces boulardii* is the most frequently prescribed probiotic for IP management by clinicians.[5] From the *Saccharomyces boulardii* supplements used by this population group, 85% is reported to be prescribed by a clinician.[220] Supplementation with *Saccharomyces boulardii* is reported as an independent predictor of a greater number of days each week that IP affects daily living, suggesting patients with more severe IP use *Saccharomyces boulardii.*[220] At the same time, Australian adults with suspected IP that report an improved IP are more likely to use dietary supplements.[220] Clinicians frequently to treat IP, with almost 80% of clinicians reporting they always prescribe probiotics to patients with IP.[5] Although limited evidence is

available on the type of probiotics clinicians use, multi-strain probiotics are more frequently used than single-strain probiotics.[5] Clinicians also use prebiotic fibres such as resistant starch, pectin and slippery elm in the treatment of patients with IP.[5] The use of synbiotic (a combination of prebiotic and probiotic) by individuals with IP have not been investigated in the literature.

The mechanistic evidence suggests potential benefits of probiotic and prebiotic supplementations and a collective summary of the available literature is required. Furthermore, as both clinicians and individuals with suspected IP report the frequent use of probiotics and prebiotics, structured recommendations are necessary to ensure optimal care is provided.

8.12.3 SUMMARY OF EVIDENCE

A literature review found 33 studies addressing the two clinical questions (CQ.3 and CQ.4) regarding the effect of probiotic, prebiotic and synbiotic on intestinal integrity. The risk of bias (RoB) assessment found the identified systematic reviews had a high RoB (*n* = 1)[295] and low RoB (*n* = 1),[301] while the 27 randomised control trials were found to have high (*n* = 15),[84,291,292,296,298,299,303,304,306,307,310,313,317,318,322] some (*n* = 6)[293,297,302,308,314,320] and low (*n* = 6)[300,305,309,312,319,321] RoB. The four non-randomised clinical trials had moderate RoB (*n* = 3)[294,311,315] and serious RoB (*n* = 1).[316] A summary of the evidence is provided here with a full review of the literature found in the *Technical Report*.

Although there is substantial evidence (n = 19)[84,291-308] investigating the effect of probiotics on intestinal integrity, the heterogeneity of these studies presents difficulties synthesising the research. The research on multi-strain probiotics their effect on intestinal integrity was mixed. Most studies found no significant difference in stool zonulin, serum zonulin, and dual sugar test after probiotic supplementation. The quantity of each probiotic strain may influence the effectiveness of supporting IP; however, further studies are required.[307] One commercially available probiotic, *Saccharomyces boulardii* was found to decrease IP by 33.33% at 12 weeks.[297] Furthermore, intervention duration of less than three months had a significant impact on the effects of probiotics on serum zonulin, with greater improvement seen in studies lasting for three months then any longer (coefficient = 33.23 [95% CI: 0.30, 66.16]; p=0.048). One type of probiotic with a sufficient amount of studies with similar designs was probiotic drinks. Kefir milk and fermented milk were both found to have a beneficial effect on IP.[302,308] Three further studies examined 65ml of milk drink (Yakult light®) containing *Lactobacillus casei* Shirota 10^8/ml (6.5 x 10^9 CFU) three times daily and found no significant effect on IP.[296,298,306] These results were consistent across multiple assessment methods (stool zonulin, serum zonulin and dual sugar), time points (12 weeks and six months) and disease states (metabolic syndrome and liver cirrhosis).[296,298,306]

The use of prebiotics in the treatment of IP was met with mixed evidence in the limited studies found (*n* = 6).[295,309-313]. Synthesis of the available evidence was not possible due to the diverse prebiotics fibres used in the identified studies, including pectin (*n* = 2),[310,311] arabinoxylan (*n* = 2),[309,312] inulin (*n* = 1),[295] inulin-

type fructans (*n* = 1),[295] polydextrose (*n* = 1),[313] slippery elm (*n* = 1)[311] and guar gum (*n* = 1).[311] However, the effect of prebiotics on intestinal integrity appears to be influenced by the type of prebiotic used. For instance, inulin or inulin-type fructans were found to decreased either LPS or exotoxin levels[295] while others such as arabinoxylan[309,312] and pectin[310] were found to have no effect on IP.

The evidence evaluating synbiotic supplementation found moderate quality of evidence.[301,313-319] Most studies (*n* = 5)[301,313,317-319] used a diverse range of probiotics and prebiotics with only three studies identified as having the same or similar ingredients.[294,315,316] The variety of synbiotic combinations used in the included studies made synthesising the evidence difficult. a meta-analysis found synbiotic supplementation significantly reduced serum zonulin compared to placebo (weighted mean difference = -10.55 [95% CI: -17.76, -3.34]; p=0.004), with a study duration of less than three months identified to have a significant impact on the effects of synbiotic on serum zonulin (coefficient = 33.23 [95% CI: 0.30, 66.16]; p=0.048). The three studies identified as having the same or similar ingredients used a combination of inulin, corn starch and fructooligosaccharides with a multi-strain probiotic (7.5×10^9 CFU).[294,315,316] All three studies demonstrated a beneficial effect of symbiotic therapy on intestinal integrity. Mixed results were found for other synbiotic combinations, with some such as 250mg of tara gum and *Streptococcus thermophilus* (1×10^7 CFU) found to improve IP after 4 weeks[319] while other combinations of 2.4g of partially hydrolysed guar gum and 1.6g of inulin, *Lactobacillus reuteri* (1×10^8 CFU) twice daily round to have no effect after 3 months (p=0.737).[318] Furthermore, one study identified synbiotic supplementation containing 12g of polydextrose and *Bifidobacterium animalis*

ssp. lactis 420 (10^{10} CFU) resulted in a significant increase in serum lipopolysaccharide compared to placebo after six months (+9.1±40 vs −26±108; p=0.007).[313]

Four high-quality studies investigated the effects of probiotic (*n* = 2), prebiotic (*n* = 1) and synbiotic (*n* = 1) supplementation on nonsteroidal anti-inflammatory drugs (NSAIDs) induced IP.[314,320-322] All included studies used the same designs to induce IP with NSAIDs. This study design involved participants taking 75mg of a NSAID nine hours before measuring IP and another 50mg one hour prior to measuring IP. Five probiotic formulations measured across two short-term studies found no significant change in NSAID induced IP.[321,322] A six-week trial of two different prebiotics (12g of arabinoxylan or 12g of oat β-glucan) found no significant difference in arabinoxylan group or oat β-glucan in preventing NSAID induced IP.[320] Synbiotic supplement containing fructooligosaccharides and a multi-strain probiotic resulted in similar outcomes as prebiotic and probiotic with no significant difference.[314]

Table 8.5 Recommendations: Probiotic, prebiotic and synbiotic supplementation

No.	Category	Recommendation	Strength
Probiotics			
2.1	EBR	There is insufficient evidence to form a recommendation on the use of probiotics as a collective group for the treatment of people with intestinal permeability.	∅
2.2	EBR	Clinicians may consider using *Saccharomyces cerevisiae var boulardii (Saccharomyces boulardii)* supplementation in the treatment of people with intestinal permeability.	⊕⊕⊕
2.3	EBR	Clinicians may consider the use of effective probiotics for a minimum of 3 months when treating people with intestinal permeability.	⊕⊕⊕

2.4	PP	Clinicians may consider researching probiotic strains for their effectiveness before using them to treat people with intestinal permeability.	⊕
2.5	PP	Clinicians may consider the use of probiotics which are supported by pre-clinical research in conjunction with other treatment interventions for the management people with intestinal permeability.	⊕
Probiotic drink			
2.6	EBR	People with intestinal permeability should consider trialling and if tolerated, consume fermented milk products such as kefir.	⊕⊕⊕⊕
2.7	EBR	People with intestinal permeability may consider NOT consuming Yakult light®.	⊕⊕⊕
Prebiotics			
2.8	EBR	There is insufficient evidence to form a recommendation on the use of prebiotics as a collective group for the treatment of people with intestinal permeability.	∅
2.9	PP	Clinicians may consider researching specific prebiotic for their effectiveness before using them in the treatment of people with intestinal permeability.	⊕
2.10	PP	Clinicians may consider trialling and if tolerated, recommend patients to use prebiotic which are supported by pre-clinical research in conjunction with other treatment interventions for the management people with intestinal permeability.	⊕
2.11	PP	Clinicians may consider NOT using polydextrose in the treatment of people with intestinal permeability.	⊕
Synbiotic			
2.12	EBR	Clinicians may consider the use of effective synbiotic in the treatment of people with intestinal permeability.	⊕⊕⊕

2.13	EBR	Clinicians may consider the use of effective synbiotic for a minimum of 3 months when treating people with intestinal permeability.	⊕⊕⊕
2.14	EBR	Clinicians may consider NOT using polydextrose and *Bifidobacterium animalis ssp. lactis* 420 in the treatment of people with intestinal permeability.	⊕⊕⊕
2.15	PP	Clinicians may consider the use of specific synbiotic which are supported by pre-clinical research in conjunction with other treatment interventions for the management people with intestinal permeability.	⊕
NSAID induced intestinal permeability			
2.16	EBR	Clinicians may consider NOT using probiotics for the treatment of people with nonsteroidal anti-inflammatory drug induced intestinal permeability.	⊕⊕⊕
2.17	EBR	Clinicians may consider NOT using prebiotics for the treatment of people with nonsteroidal anti-inflammatory drug induced intestinal permeability.	⊕⊕⊕
2.18	EBR	Clinicians may consider NOT using synbiotics for the treatment of people with nonsteroidal anti-inflammatory drug induced intestinal permeability.	⊕⊕⊕

Abbreviations: CBR = Consensus-based recommendation; EBR = evidence-based recommendation; NSAID = nonsteroidal anti-inflammatory drug; PP = practice point.

8.12.4 JUSTIFICATION

Recommendations were informed by the best available evidence on the impact probiotics, prebiotics and synbiotics may have on intestinal integrity. The guideline development group carefully considered the available literature and the importance patients with IP place on supplementation for the management of IP while forming each recommendation. The heterogeneity of the available evidence impacted the grade and the ability to form evidence-based recommendations.

As probiotics are unique, with each strain able to have a different clinical and physiological effect, no collective recommendation was formed for all probiotics. Instead, two practice points were developed to provide clinicians with direction when prescribing probiotics in treating IP. As the evidence on probiotics is an expanding area of research, the two practice points were designed to safeguard the longevity of the IP Guideline. The focus of these two recommendations were for clinicians to undertake their own research for beneficial strains and use pre-clinical research when choosing a probiotic. In response to the evidence supporting the use of *Saccharomyces boulardii* and clinicians frequently prescribing this probiotic, an evidence-based recommendation was developed. Another single-strain probiotic *Akkermansia muciniphila* demonstrated promising results, however, no recommendation was developed as this strain is not available in the Australian market.

Each type of prebiotic has a unique structure and associated action; therefore, no collective recommendation was formed for all prebiotics as a collective group. As prebiotics are known to have a beneficial effect on many health conditions associated with IP, advising practitioners not to prescribe a prebiotic supplement could be met with resistance. Therefore, three practice points were created to provide clinicians with direction for the prescription of prebiotics in the treatment of IP. Two of these practice points encourage clinicians to research beneficial types of prebiotics and use pre-clinical research where necessary. As the evidence on prebiotics and the effect on the microbiome is an expanding area of research, these two practice points can ensure the IP Guideline is relevant for

clinicians. The other practice point was developed to address the potential adverse effects of supplementing with polydextrose. Although polydextrose is not a frequently prescribed prebiotic, this recommendation may further direct clinicians until further research confirms the safety in people with IP.

Unlike probiotic and prebiotic supplementation, the evidence supporting the use of synbiotic therapy for the treatment of IP suggests a beneficial effect. Therefore, an evidence-based recommendation was created to reflect the evidence. Furthermore, like probiotic and prebiotic supplementation, a practice point was developed, endorsing clinicians to prescribe synbiotic supplementation based on pre-clinical research if they were to use synbiotic in clinical practice. According to the research, a synbiotic formula containing polydextrose and *Bifidobacterium animalis ssp. lactis* 420 has the potential to exacerbate IP. This led to the recommendation for clinicians to avoid this combination until further research confirms the safety in people with IP.

Regarding NSAID induced IP, there is consistent research across different probiotics and prebiotics intervention studies providing evidence to recommend clinicians not to use probiotics, prebiotics, or synbiotics to prevent NSAID induced IP. Therefore, this resulted in three practice points to support clinicians and reflect the level of research.

8.13 AMINO ACID

8.13.1 CLINICAL QUESTIONS

Clinical Question 5: In Australian adults with increased intestinal permeability, what are the benefits of oral amino acid supplementation for the treatment of increased intestinal permeability?

Clinical Question 6: In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for oral amino acid supplementation use?

8.13.2 CLINICAL NEED FOR THE QUESTION

Amino acids, especially glutamine, may have potential benefits on the intestinal integrity.[342] Glutamine is a major energy source for intestinal epithelial, promoting enterocyte proliferation and protects the epithelium from apoptosis.[342] Glutamine may increase the expression of zonula occludens-1 (ZO-1), ZO-2, ZO-3, and claudin-1, therefore, enhancing tight junction integrity.[343,344] Caco-2 cell monolayer *in vitro* study design suggests glutamine may prevent alcohol-induced IP.[331] Glutamine has been suggested to enhances the transactivation of heat shock factor-1 and induces heat shock factor-1 expression, therefore protect the intestinal epithelium against oxidative stress and inflammation.[345] Another amino acid with potential clinical application is lactoferrin is an iron-binding glycoprotein that is naturally found in human breast milk.[346] Mechanistic research on lactoferrin indicates a potential antibacterial and anti-inflammatory action with the supplementation used an adjuvant therapy in gastrointestinal disorders.[346]

Australian adults with suspected IP report preferring dietary supplements including amino acids such as glutamine managing IP.[219] Specifically, the use of dietary supplements is the third most preferred treatment method for IP, with many people with IP frequently using dietary supplements (73%) for the management of IP.[219,220] Dietary supplements are most frequently prescribed by a naturopath, with over 70% of glutamine supplementation prescribed by a clinician and the other 30% self-prescribed.[220] In addition, almost 90% of Australian adults with suspected IP perceive that it is important for clinicians to be knowledgeable about dietary supplement.[219] This population group reports spending an average of AUD $2,175 on dietary supplements annually.[219] The economic burden of using dietary supplements does not appear to prohibit this populations' use of dietary supplements.[219] Glutamine is the fifth most frequently used dietary supplement in people with IP, with amino acid complex also used to a lesser extent.[220] While clinicians frequently use glutamine in the management of patients with IP, with 73% of clinicians always using glutamine.[5]

The mechanistic evidence indicates a potential benefit of amino acids, especially glutamine.[342] As both clinicians and people with suspected IP report the frequent use of amino acids, a collective summary of the available literature and structured recommendations are necessary to ensure optimal care is provided.

8.13.3 SUMMARY OF EVIDENCE

A literature review found four studies addressing the two clinical questions (CQ.5 and CQ.6) regarding the effect of amino acids on intestinal integrity. The risk of bias (RoB) assessment found the three randomised control trials to have low RoB

(*n* = 3).[192,323,324] The one non-randomised clinical trials had moderate RoB.[311] A summary of the evidence is provided here with a full review of the literature found in the Technical Report.

A total of three studies exploring the effects of glutamine on altered IP were included.[192,311,323] The use of glutamine supplementation in people with IP saw consistent beneficial effects on intestinal integrity. All studies involved participants with a gastrointestinal disorder, and they were assessed for IP through the dual sugar test. The supplementation of glutamine in people with diarrhoea-predominant irritable bowel syndrome demonstrated a significant decrease in IP compared to baseline after two months (0.11±0.03 vs. 0.04±0.01; $p<0.0001$).[192] Furthermore, there is a significant correlation between irritable bowel syndrome severity and improvement of IP in people taking glutamine ($r=0.72$; $p<0.001$).[192] Another RCT investigated the effect of 0.5g/kg of glutamine on ideal body weight per day in Crohn's disease patients currently in remission.[323] This study found glutamine supplementation reduced lactulose/mannitol ratio (LMR) median value from 0.071 (0.041–0.254, range) to 0.029 (0.006–0.090, range) after two months.[323] Furthermore, the control group also had a significant improvement in IP from a median value of 0.067 (0.040–0.136, range) to 0.033 (0.009–0.077, range), possibly influenced by the placebo supplement containing a large amount of whey protein. No significant difference was found between the glutamine group and whey protein group after two months (median value 0.029 vs. 0.033; $p>0.05$).[323] An Australian based study explored the effects of 2.5g of glutamine in combination with prebiotics, other intestinal supportive herbal

medicine, and nutrients on IP.[311] This study found a significant decrease between baseline and 12 weeks in LMR (0.04±0.004 vs. 0.03±0.001; p<0.0001).[311]

One article investigated the effects of lactoferrin on NSAID induced intestinal integrity. Healthy males had IP induced by consuming 75mg NSAID 9 hours prior and 50mg 1 hour prior to undertaking the dual sugar test. The intervention involved participants consuming 5g of lactoferrin three times (24, 9 and 1 hour before the dual sugar test). Lactoferrin supplementation was found to significantly decrease NSAID-induced IP compared to NSAIDs and placebo (0.028 vs. 0.036; p<0.05).

Table 8.6 Recommendations: Amino acid supplementation

No.	Category	Recommendation	Strength
Glutamine			
3.1	EBR	Clinicians should offer glutamine supplementation for the treatment of people with intestinal permeability.	⊕⊕⊕⊕⊕
3.2	CBR	Clinicians may consider the use of glutamine supplementation in conjunction with other treatment interventions for the management of people with intestinal permeability.	⊕⊕
NSAID-induced intestinal permeability			
3.3	EBR	Clinicians should consider the use of short-term lactoferrin supplementation for the treatment of people with nonsteroidal anti-inflammatory drug induced intestinal permeability.	⊕⊕⊕⊕

Abbreviations: CBR = Consensus-based recommendation; EBR = evidence-based recommendation; NSAID = nonsteroidal anti-inflammatory drug.

8.13.4 JUSTIFICATION

Recommendations were informed by the best available evidence on the impact amino acids may have on IP. The guideline development group carefully considered the available literature and the importance patients with IP place on supplementation for the management of IP while forming each recommendation.

There is consistent evidence supporting the use of glutamine in people with IP. Although one study found a significant improvement in the glutamine group from baseline to two months, this same study found no significant difference between the control and glutamine supplementation. Key characteristics in the study design such as demographic and control supplement (a large amount of whey protein) could potentially explain this finding and therefore did not affect the grading of the recommendation. Although whey protein, a complex source of amino acids, could be considered as a potential therapeutic intervention based on this study,[323] the conflicting result seen with Zhou et al,[192] resulted in no recommendation considered. A consensus-based recommendation was developed considering the whole system approach clinicians follow in managing people with IP. This recommendation suggests glutamine be considered as a part of other treatment interventions rather than the sole ingredient. Although the grade for the recommendation for lactoferrin was allocated a B, the Working Group downgraded the strength of the recommendation from a strong recommendation to a recommendation considering this intervention is less used in clinical practice, and only one study was included.

8.14 PLANT-BASED MEDICINE

8.14.1 CLINICAL QUESTIONS

Clinical Question 7: In Australian adults with increased intestinal permeability, what are the benefits of oral plant-based medicine supplementation for the treatment of increased intestinal permeability?

Clinical Question 8: In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for oral plant-based medicines use?

8.14.2 CLINICAL NEED FOR THE QUESTION

Plant-based medicines are reported to influence the function of the gastrointestinal system and may modulate the integrity of the small intestine.[347] Herbal therapy is suggested to support IP by reducing the effect of endotoxins, changing the expression of tight junction proteins and contributing to the mucus layer of the gastrointestinal tract.[348,349] Mechanistic research on individual herbal therapies such as *Aloe barbadensis Mill* (aloe vera) suggest that it may potentially enhance the expression of intestinal zonula occludens (ZO)-1.[350] While other herbal medicines, including *Curcuma longa* (turmeric) is reported to regulate the expression of ZO-1 and claudin-1.[351]

Australian adults with suspected IP report preferring dietary supplements such as herbal medicine for managing IP.[219] Specifically, the use of dietary supplements is the third most preferred treatment method for IP, with many people with IP frequently using dietary supplements (73%) for the management of IP.[219,220] Dietary supplements are most frequently prescribed by a naturopath,

with over 70% of herbal mixtures prescribed by a clinician.[220] Clinicians frequently prescribe herbal medicine to treat IP, with many reporting they always prescribe *Curcuma longa* (73%), *Allium sativum* (52%), *Ulmus rubra* (51%), *Zingiber officinale* (50%), *Aloe barbadensis Mill* (48%), *Althaea officinalis* (44%) and *Gentiana lutea* (44%) in patients with IP.[5] Clinicians treating people with IP will generally use a combination of herbal products as part of a whole system treatment approach.[5]

People with IP report spending an average of $2,175 AUD on dietary supplements annually.[219] The financial cost of dietary supplements and peoples financial status, does not appear to affect this populations' decision in using dietary supplements.[219] The second most frequently used dietary supplement in people with IP are herbal mixtures.[220] The herbal products most commonly used by people with IP are *Curcuma longa*, *Ulmus rubra* (slippery elm) and *Aloe barbadensis Mill*.[220] Australian adults with suspected IP that report an improved IP are more likely to use dietary supplements.[220] Almost 90% of Australian adults with suspected IP report that the knowledge and understanding of dietary supplements are important for clinicians to understand.[219] The mechanistic evidence indicates a potential benefit of herbal medicine.[347,350] As clinicians and individuals with suspected IP report the frequent use of herbal medicine, a collective summary of the available literature and structured recommendations are necessary to the provision of optimal care.

8.14.3 SUMMARY OF EVIDENCE

A literature review found five studies addressing the two clinical questions (CQ.7 and CQ.8) regarding the effect of plant-based medicines on intestinal integrity. The risk of bias (RoB) assessment found the four randomised control trials to have high (*n* = 3)[303,325,326] and low (*n* = 1)[300] RoB. The one non-randomised clinical trials had moderate RoB.[311] A summary of the evidence is provided here with a full review of the literature found in the *Technical Report*.

A total of five studies exploring the effects of a diverse range of plant-based medicines on intestinal integrity were included.[300,303,311,325,326] The use of plant-based medicines in people with IP resulted in mixed outcomes with three out of the five studies reporting no significant effect. One study exploring the effects of pomegranate extract in overweight and obese adults found after three weeks, pomegranate extract significantly reduced lipopolysaccharide-binding protein compared to placebo ($p<0.001$).[325] While an Australian based study exploring the effects of a mix of herbal medicines (aloe vera 2.5mg, slippery elm 500mg, guar gum 100mg, pectin 100mg and peppermint oil 3mg) and amino acids in patients with a functional gastrointestinal disorder found a significant decrease between baseline and 12 weeks in lactulose/mannitol ratio (0.04±0.004 vs 0.03±0.001; $p<0.0001$).[311] Three other studies investigating the effects of plant-based medicines on IP found no significant impact. These studies used a combination of herbal ingredients. One of these studies used multi-herbal formula in healthy adults over eight weeks and found a significant difference in serum zonulin between placebo and the intervention group.[326] Another herbal combination containing barley grass and oat grass juice had no significant impact on

lactulose/mannitol ratio between baseline and 12 weeks (p>0.05).[300] Similar results were seen with a traditional Japanese formula known as Bofutsushosan.[303] The study found no significant effect between baseline and five week in lactulose/mannitol ratio (2.7±1.9 vs 2.2±1.5; p=0.391).[303]

Table 8.7 Recommendations: Plant-based medicine supplementation

No.	Category	Recommendation	Strength
4.1	EBR	There is insufficient evidence to form a recommendation on the use of plant-based medicines as a collective group for the treatment of people with intestinal permeability.	∅
4.2	PP	Clinicians may consider the use of plant-based medicines which are supported by pre-clinical research in conjunction with other treatment interventions for the management people with intestinal permeability.	⊕

Abbreviations: EBR = evidence-based recommendation; PP = practice point.

8.14.4 JUSTIFICATION

Recommendations were informed by the best available evidence on the impact plant-based medicine may have on intestinal integrity. The guideline development group carefully considered the available literature and the importance patients with IP place on supplementation for the management of IP while forming each recommendation. The limited available literature and heterogeneity of the available evidence impacted the grade and the ability to develop evidence-based recommendations.

Plant-based medicines are unique, with every herbal ingredient able to have a different clinical and physiological effect. Therefore, no collective

recommendation was developed for all herbal therapies. Instead, the development of one practice point to provide clinicians with direction when prescribing plant-based medicines in treating IP was formulated. This practice point was developed as plant-based medicines are frequently used in clinical practice and no safety concerns were identified. Furthermore, as the evidence on herbal therapies is an expanding area of research, the practice point was designed to provide confidence in the IP Guideline. The focus of this practice point is for clinicians to undertake their own research for beneficial herbal therapies and use pre-clinical research when prescribing to patients with IP. Although one research study identified pomegranate extract as a potential therapy for the management of IP, important characteristics such as the high RoB and only using lipopolysaccharide-binding protein as a marker for IP, no recommendation could be developed.

8.15 ESSENTIAL FATTY ACID

8.15.1 CLINICAL QUESTIONS

Clinical Question 9: In Australian adults with increased intestinal permeability, what are the benefits of oral essential fatty acid supplementation for the treatment of increased intestinal permeability?

Clinical Question 10: In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for oral essential fatty acid use?

8.15.2 CLINICAL NEED FOR THE QUESTION

Essential fatty acids are a group of polyunsaturated fatty acids, including two types of omega-3 docosahexaenoic acid (DHA) and eicosapentaenoic acid (EPA) with the other being omega-6. These essential fatty acids, especially omega-3, are thought to prevent changes to IP by inhibiting the production of pro-inflammatory cytokines.[352] Mechanistic research suggests omega-3 may normalise the expression of zonula occludens (ZO)-1 and occluding in the intestine.[353] Australian adults with suspected IP report preferring dietary supplements such as omega 3 for managing IP.[219] Specifically, the use of dietary supplements is the third most preferred treatment method for IP, with many people with IP frequently using dietary supplements (73%) for the management of IP.[219,220] Dietary supplements are most frequently prescribed by a naturopath, with over 66% of omega-3 prescribed by a clinician.[220] People with IP report spending an average of $2,175 AUD on dietary supplements annually.[219] The financial cost of dietary supplements and peoples financial status, does not appear to affect this populations' decision in using dietary supplements.[219] One

third of people with IP report using omega-3 in the treatment of IP.[220] Australian adults with suspected IP that report an improved IP are more likely to use dietary supplements.[220] Almost 90% of Australian adults with suspected IP report that it is important for clinicians to understand dietary supplements in the context of IP management and treatment.[219] Due to this early mechanistic evidence and the frequent use of omega-3 supplements reported by both patients and clinicians, exploration of the evidence regarding essential fatty acid supplementation for people with IP is needed.

8.15.3 SUMMARY OF EVIDENCE

A literature review found one study addressing the two clinical questions (CQ.9 and CQ.10) regarding the effect of essential fatty acid supplementation on intestinal integrity. The risk of bias (RoB) assessment found this trial to have high RoB.[299] A summary of the evidence is provided here with a full review of the literature found in the *Technical Report*.

A total of one study exploring the effects of essential fatty acid supplementation on intestinal integrity was included.[299] This randomised, double-blind placebo-controlled trial assessed the effects of four study arms: omega-3, probiotic, omega-3 and probiotic or placebo over 21 weeks in pregnant women.[299] The omega-3 supplement contained 2g of omega-3 (79.6% DHA and 9.7% EPA) twice daily. The study found no significant effect in serum zonulin between early and late pregnancy with omega-3 supplementation (mean change: +5.2±11.2ng/ml; 95%CI +2.0, +8.5; $p>0.05$). Furthermore, lipopolysaccharide (LPS) had no significant change between early and late pregnancy with omega-

3 supplementation (mean change: +0.06±0.11EU/ml; 95%CI +0.023, +0.088; p>0.05).

Table 8.8 Recommendations: Essential fatty acid supplementation

No.	Category	Recommendation	Strength
5.1	EBR	There is insufficient evidence to form a recommendation on the use of essential fatty acid supplementation for the treatment of people with intestinal permeability.	∅

Abbreviations: EBR = evidence-based recommendation.

8.15.4 JUSTIFICATION

Recommendations were informed by the best available evidence on the impact essential fatty acid supplementation may have on intestinal integrity. The guideline development group carefully considered the available literature and the importance patients with IP place on supplementation for the management of IP while forming the recommendation. There is insufficient evidence to recommend the use of essential fatty acid supplementation in people with IP, with only included study finding no significant change in markers of IP.

8.16 MINERAL SUPPLEMENTATION

8.16.1 CLINICAL QUESTIONS

Clinical Question 11: In Australian adults with increased intestinal permeability, what are the benefits of oral mineral supplementation for the treatment of increased intestinal permeability?

Clinical Question 12: In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for oral mineral supplementation use?

8.16.2 CLINICAL NEED FOR THE QUESTION

Minerals, especially zinc, are essential for tight junction maintenance, with mechanistic research suggesting zinc may prevent the breakdown of tight junction proteins and enhance the expression of zonula occludens (ZO)-1.[354-356] Australian adults with suspected IP report preferring dietary supplements such as minerals, including zinc and magnesium for managing IP.[219] Specifically, the use of dietary supplements is the third most preferred treatment method for IP, with many people with IP frequently using dietary supplements (73%) for the management of IP.[219,220] Dietary supplements are most frequently prescribed by a naturopath, with over 80% of zinc prescribed by a clinician.[220] Furthermore, zinc is the most prescribed dietary supplement in the treatment of people with IP.[5] People with IP report spending an average of $2,175 AUD on dietary supplements annually.[219] The financial cost of dietary supplements and peoples financial status, does not appear to affect this populations' decision in using dietary supplements.[219] Over 20% of people with IP report using zinc in the

treatment of IP.[220] Australian adults with suspected IP that report an improved IP are more likely to use dietary supplements.[220] Almost 90% of Australian adults with suspected IP report that it is important for clinicians to understand dietary supplements in the context of IP management and treatment.[219] Due to this early mechanistic evidence and the frequent use of zinc supplementation reported by both patients and clinicians, exploration of the evidence regarding mineral supplementation for people with IP is needed.

8.16.3 SUMMARY OF EVIDENCE

A literature review found two studies addressing the two clinical questions (CQ.11 and CQ.12) regarding the effect of oral mineral supplementation on intestinal integrity. The risk of bias (RoB) assessment found the one non-randomised clinical trial to have a moderate RoB.[299] A summary of the evidence is provided here with a full review of the literature found in the *Technical Report*.

Only one non-randomised clinical trial met the inclusion criteria and was included.[327] This study explored the effects of zinc supplementation in 12 Crohn's disease patients currently in remission with a lactulose mannitol ratio >0.035. The study involved zinc supplementation containing 25mg of elemental zinc three times daily for eight weeks. After the study period, there was a significant decrease in IP from baseline to eight weeks (0.041±0.003 vs. 0.026±0.005; p= 0.0028). Furthermore, at the end of the eight weeks, the lactulose-mannitol ratio normalised in 75% of participants.

Table 8.9 Recommendations: Mineral supplementation

No.	Category	Recommendation	Strength
6.1	EBR	Clinicians may consider using zinc supplementation in the treatment of people with intestinal permeability.	⊕⊕⊕

Abbreviations: EBR = evidence-based recommendation.

8.16.4 JUSTIFICATION

Recommendations were informed by the best available evidence on the impact mineral supplementation may have on intestinal integrity. The guideline development group carefully considered the available literature and the importance patients with IP place on supplementation for the management of IP while forming the recommendation. One evidence-based recommendation was developed to reflect the available literature and the importance clinicians place on zinc supplementation.

8.17 VITAMIN SUPPLEMENTATION

8.17.1 CLINICAL QUESTIONS

Clinical Question 13: In Australian adults with increased intestinal permeability, what are the benefits of oral vitamin supplementation for the treatment of increased intestinal permeability?

Clinical Question 14: In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for oral vitamin supplementation use?

8.17.2 SUMMARY OF EVIDENCE

A literature review found no studies addressing the two clinical questions (CQ.13 and CQ.14) regarding the effect of vitamin supplementation on intestinal integrity. Therefore, no recommendation was developed.

8.18 COLOSTRUM SUPPLEMENTATION

8.18.1 CLINICAL QUESTIONS

Clinical Question 15: In Australian adults with increased intestinal permeability, what are the benefits of oral colostrum supplementation for the treatment of increased intestinal permeability?

Clinical Question 16: In Australian adults with increased intestinal permeability, what are the harms, cautions and contraindications for oral colostrum supplementation use?

8.18.2 SUMMARY OF EVIDENCE

A literature review found no studies addressing the two clinical questions (CQ.15 and CQ.16) regarding the effect of colostrum supplementation on intestinal integrity. Therefore, no recommendation was developed

8.19 CHAPTER SUMMARY

This *clinical practice guideline for the management of IP* serves as the first guideline for IP both nationally and internationally. The recommendations developed utilised the best available evidence while considering the views and preferences of consumers and clinicians. In response to the research question outlined in this thesis, 38 recommendations consisting of 27 evidence-based recommendations, seven practice points and four consensus-based recommendations were created to optimise patient care, improve health outcomes and reduce variation in care for Australian practitioners in private practice.

9. DISCUSSION

This thesis is the first body of work to develop a clinical practice guideline for managing IP, which also considers the views and preferences of Australian adults with suspected IP. This project provides a foundation for clinicians to follow for treating IP in their clinical practice. Recommendations developed as part of the IP Guideline may offer a foundation for future clinical research in this emerging area of research. Major findings briefly discussed in Chapters 4, 5, 6, 7 and 8 will be further discussed in detail throughout this chapter, along with other important findings reported in this body of work.

9.1 STAKEHOLDERS VIEWS ON RECOMMENDATIONS

The IP Guideline, with the involvement of stakeholders, is suggested to align with the views and practices of clinicians that treat patients with IP in clinical practice. The 38 recommendations were revised based on stakeholders' feedback to ensure the IP Guideline was clinically appropriate and based on evidence. The views and perspectives of stakeholders provided clinical and industry feedback on the developed recommendations. Stakeholders' feedback directly influenced the recommendations to align with the target users of the IP Guideline.

Nine recommendations (1.2, 2.4, 2.9, 2.10, 3.1, 3.2, 3.3, 4.2 and 6.1) had a high consensus rate (>80%) among all three domains (agreement, appropriateness, and importance) and thereby acknowledged by stakeholders as well accepted recommendations. An explanation for consistent consensus may be multifaceted.

It may involve alignment seen in clinical practice, the frequency of use by clinicians and patients or consistent with clinical perspectives, regardless of whether there is substantial supportive research. For instance, in recommendation 6.1, the use of zinc supplementation in people with IP was found to have the highest consensus among stakeholders, with all members reporting they agree with the use in clinical practice. This agreement is most likely contributed to zinc supplementation being the most prescribed dietary supplement when treating people with IP.[5] The alignment between published evidence and clinicians use of zinc supplementation strengthens the acceptance in clinical practice. Another example of where stakeholders had consistently high consensus was with the prebiotic recommendations 2.9 and 2.10 (research specific prebiotics and use of pre-clinical evidence). Pre-clinical research suggests prebiotics may mitigate the impact of lipopolysaccharide on intestinal integrity and protect the mucosa from inflammation.[338] Stakeholders appear to support the concepts of researching specific prebiotics while also using pre-clinical research to guide treatment interventions.

On the contrary, recommendations surrounding alcohol consumption had conflicting results. Stakeholders disagreed with recommendation 1.1 (consume alcohol as per the Australian dietary guidelines), while recommendation 1.2 (limit or avoid alcohol consumption) had a consistent agreement. The inconsistency results from developing a recommendation (1.1) according to the available literature compared to forming a consensus-based recommendation that aligns with clinical practice (recommendation 1.2). This inconsistency highlights the

importance of stakeholder involvement in developing clinical practice guidelines and indicates that further research is required to confirm clinicians' notions.

The recommendations identified as having a low agreement rate or stakeholders suggesting a change were modified to ensure they reflect clinical practice. The feedback from stakeholders resulted in 19 recommendations obtaining some level of modification. A common theme noted throughout the feedback was the concept of personalised medicine or individualising the treatment interventions, where stakeholders expressed that some patients may not tolerate the treatment interventions. Following the notion that the target users of the IP Guideline frequently use personalised medicine, modifications were made to the recommendations to reflect this critical consideratio.[240,357] Such changes applied to some fermented foods and prebiotics recommendations, as a small percentage of people with IP may have additional health conditions that may have adverse reactions, such as bloating to some prebiotics and fermented foods.[358,359] To overcome the possibility of consumers not tolerating such interventions, the concept of trialling the recommendation and, if tolerated, follow was incorporated into the identified recommendations.

By making the changes suggested by the stakeholders, it is expected that the recommendations will better align with clinicians. Incorporating stakeholder's feedback and modifying the recommendations is suggested to increase uptake of guideline use in practice.[360] Obtaining stakeholder's feedback on drafted recommendations has previously been demonstrated to balance the scientific process and provide practical insight, translating to more appropriate

recommendations.[361] Although some recommendations were initially identified as a lack of agreement, these changes to the strength and wording allowed the recommendations to be more clinically relevant when treating people with IP.

9.2 IP GUIDELINE IN CLINICAL PRACTICE

The IP Guideline in clinical practice is expected to influence how clinicians manage people with suspected IP. Specifically, the IP Guideline provides clinicians with new treatment interventions and directions for managing IP, while highlighting the treatment methods without sufficient evidence. The clinicians who most frequently treat IP in clinical practice include naturopaths, integrative medicine practitioners, nutritionists and general practitioners.[219] These clinicians, especially integrative medicine practitioners and general practitioners, use and follow clinical practice guidelines to inform disease management.[126] On the other hand, naturopaths and nutritionists rely less on clinical practice guidelines with their use mainly informing care rather than treatment interventions.[127] This may be due to multiple factors such as the lack of guidelines that involve these clinicians as key stakeholders and the shortage of naturopathic specific treatment interventions.[127] To overcome these barriers, the IP Guideline followed a structured process to ensure the view and perspectives of clinicians that treat people with IP were considered during the development of the recommendations. Therefore, the IP Guideline is expected to be utilised by these clinicians in managing IP.

The IP Guideline provides a new direction for clinicians in prescribing a gluten-free diet to manage IP. The recommendations surrounding the use of a gluten-free diet have the strongest level of evidence from all the diet-related recommendations in the IP Guideline. Clinicians frequently recommend that people with IP follow a strictly gluten-free diet.[5] However, three strong evidence-based recommendations were developed to guide the clinical prescription of a gluten-free diet in people with suspected IP (recommendations 1.1, 1.12 and 1.13). Correctly identifying patients with gluten intolerance, sensitivity or allergy is essential as to not miss a more serious health condition. Many gluten-related conditions go undiagnosed partly due to people avoiding gluten before the proper investigation has taken place.[362,363] Avoiding gluten in some individuals may prolong the diagnosis as gluten consumption is required as part of a gluten challenge to diagnose coeliac disease.[362] The emphasis of the developed recommendation is for clinicians to assess the patient for gluten intolerance, sensitivity or allergy before recommending they follow a low gluten diet. Furthermore, a change of mindset is required for clinicians as a gluten-free diet is not advised for people with IP but rather a low gluten diet. A driving factor that may increase the uptake of the gluten relating dietary recommendations is that these recommendations are the strongest with the highest level of evidence and that clinicians pride themselves on evidence-based practice with the greatest barrier being a lack of clinical evidence in their profession.[364]

9.3 CONSUMERS AND THE IP GUIDELINE

Many of the dietary habits that people with IP follow, such as the consumption of red meat, organic foods, and dairy products while avoiding gluten-containing products and alcohol, conflict with the evidence-based recommendations created in the IP Guideline.[220] The IP Guideline provides consumers with evidence-based recommendations to follow in regards to macronutrient distribution range, estimated energy requirements and suggested dietary target for dietary fibre while advising the avoidance of high fat (long-chain saturated fatty acids) or high free fructose diets (recommendations 1.3, 1.6, 1.7, 1.8 and 1.9). These recommendations suggest consumers should follow the macronutrient distribution range, energy intake and dietary fibre target in accordance with the Australian Dietary Guidelines. Although no direct recommendations were created for red meat or dairy products, these five recommendations can be used to quantify the amount of food products and energy intake. Many other clinical practice guidelines for digestive related conditions are centred on general dietary advice rather than specific interventions.[365] The IP Guideline provides consumers with directions for dietary intake which aligns with the advice from the Australian Dietary Guidelines.

There is mixed evidence on the dietary supplements that people with IP use to manage their IP and the developed recommendations in the IP Guideline.[220] Specifically, some of the most used dietary supplements such as probiotics, plant-based medicines and prebiotics are frequently used by consumers, yet no evidence-based recommendations were developed to support their use. For each of these dietary supplements, an evidence-based recommendation advised

clinicians that there is insufficient evidence to support their clinical use (recommendations 2.1, 2.8 and 4.1). However, other frequently used dietary supplements such as zinc and glutamine both have evidence-based recommendations for their use in managing IP (recommendations 3.1 and 6.1). Alternatively, *Saccharomyces boulardii is* not often used by people with IP yet is supported by evidence. The research resulted in an evidence-based recommendation for the use of *Saccharomyces boulardii* supplementation in managing people with IP (recommendation 2.2). Collectively, the IP Guideline recommendations for dietary supplements may be different from what consumers are currently using, however align with clinicians' treatment methods.[5]

The IP Guideline brings new treatment strategies, many of which people with IP are currently not following. Although these recommendations are different to their current methods, the acceptance and utilisation is anticipated to be well received. By incorporating consumers in the development of the IP Guideline, the developed recommendations align with their views and perspectives. The early involvement of consumers is shown to produce more patient-centric recommendations, thereby increasing consumer uptake of recommendations.[366] Furthermore, as the IP Guideline involved the consumers' views and perspectives, the recommendations are more likely to be relevant, readable and understandable to this population group.[365] Especially as many of the recommendations are centred on dietary interventions and this population group reports the use of dietary interventions as the preferred method of managing IP.

9.4 IP RISK FACTORS AND TREATMENT INTERVENTIONS

The systematic review in Chapter 2 identified several risk factors associated with IP. These were dyslipidaemia, poor glycaemic control, inflammation, anthropometric measurements that resemble obesity, the consumption of a Western-style diet, comorbidity of chronic diseases and advanced disease severity. Correcting many of these risk factors is suggested to improve IP. For instance, reducing BMI from 43 to 36 has been demonstrated to significantly improve IP.[75] Furthermore, a recent systematic review and meta-analysis explored the pooled standardised mean differences in IP after a weight loss intervention.[367] The review found weight loss was associated with a significant reduction in IP, with each kg of body weight loss associated with a 0.017 reduction in IP. Some of the recommendations developed in this thesis may further support weight management in patients with IP. For instance, recommendations 1.3 and 1.4 promote the consumption of a high fibre diet which has been demonstrated to reduce body weight.[368] Other recommendations that may reduce IP through supporting weight management include recommendations 1.6, 1.7 and 1.8, which promote clinicians to prescribe a macronutrient balanced diet while limiting the consumption of a high fat and free fructose diet, both of which play a contributing factor in weight gain.[369,370] The last set of recommendations that may have a beneficial action on IP via weight management are recommendations 1.9 and 1.10. These recommendations advise clinicians to prescribe an energy-balanced diet for the long-term management of IP with the option of short-term energy restriction also advised. The use of a hypocaloric diet is widely used as an effective method of weight reduction.[371]

Glycaemic control is another area where the improvement of IP may be influenced by glycaemic status. For instance, the improvement of IP has been shown to have a strong correlation with the reduction of HbA1c.[187] The IP Guideline may support glycaemic control via several recommendations intended to treat IP. Recommendation 2.6 suggests clinicians use fermented milk products such as kefir in managing IP. Kefir consumption has been demonstrated to decrease fasting insulin and improve insulin resistance.[372] Furthermore, recommendation 6.1 may also improve IP via glycaemic control, with zinc supplementation also known to reduce blood glucose and insulin resistance while improving β-cell function.[373] The previous mentioned dietary recommendations are also suggested to influence metabolic status with dietary fibre (recommendations 1.3 and 1.4),[374] macronutrient balance (recommendations 1.6, 1.7 and 1.8),[374] and energy balanced diet (recommendations 1.9 and 1.10)[375] are all treatment interventions indicated for balancing metabolic status.

Given the relationship between the identified risk factors and the developed recommendations, further treatment objectives may be proposed. Clinicians may consider addressing the risk factors identified in Section 2, in addition to following the recommendations developed in the IP Guideline. Using risk factors as treatment objectives is frequently seen in clinical practice, especially by integrative medicine practitioners.[376] Therefore, clinicians may consider prescribing treatment interventions that are anti-inflammatory, controls serum glucose, improve lipid profile, supports healthy weight and reduce disease severity. Essentially, a whole system approach where clinicians treat the whole

person rather than an isolated reaction or condition, may have additional benefits for managing IP.

9.5 INSUFFICIENT RESEARCH

Through the development of the IP Guideline, several clinical questions could not be answered or provide an evidence-based recommendation due to insufficient research. The clinical questions were based on the treatment interventions currently used in clinical practice by clinicians or self-prescribed by people with IP. The lack of evidence found may reduce the strength of recommendations. Based on previous research, clinicians may be relying on pre-clinical research to inform their clinical practice rather than conclusive clinical trials.[5] However, a possible explanation may involve clinicians using research that does not directly relate to IP but rather indirectly addressing the risk factors for IP. For instance, there is substantial evidence indicating the use of *curcuma longa* in the treatment of inflammatory conditions.[377,378] Clinicians may be addressing inflammation as a treatment objective known to amend a risk factor for IP, thereby indirectly supporting IP via an anti-inflammatory action. Another explanation as to why clinicians may be using *curcuma longa* in treating IP and why the IP Guideline was unable to identify relevant studies may be due to the tight inclusion criteria of the IP Guideline. Specifically, studies that involved exercise-induced IP were excluded as the results of these studies are not applicable to general clinical practice. Thereby, clinicians may be utilising research that may not directly apply to their patient population group. Further understanding of these interventions, especially colostrum, vitamin D, omega 3, *Ulmus rubra, curcuma longa* and *Aloe*

barbadensis Mill is required to determine whether they are therapeutic in people with IP.

9.6 STUDY LIMITATIONS

This thesis is the first clinical practice guideline for managing IP, which also considers the views and preferences of Australian adults with suspected IP. The creation of the IP Guideline employed multiple study designs to develop recommendations that were the most accurate and relevant for managing IP.

As there is a lack of gold standards in the assessment of IP, this thesis has several limitations. Firstly, the results of this thesis are most applicable to people with suspected IP. Furthermore, the systematic review in Chapter 2 included studies that used LPS as the marker for IP, these results require considerations in the application of IP in clinical practice. Most notably, LPS is suggested to be an exacerbator of IP, and is increased in late-stage disease or advanced IP.[142,143] However, less weighting was given to studies that used LPS alone when interpreting the results. Furthermore, as discussed in section 2.2.4.4, the most significant risk factors for IP had multiple markers confirming the association. This strengthens the association as the correlation is not dependent on LPS alone.

Considering the results from the cross-sectional survey of Australian adults with suspected IP, a few limitations need to be considered when interpreting the results. The study employed a cross-sectional survey design, which prevents any causation or etiological relationships, only associations can be made.[204]

However, this limitation does not impact the current research objectives due to the exploratory aim of the research project. The sample group had a larger percentage (93%) of females than the Australian population. This may be due to the vast majority of health conditions associated with IP being more prevalent in females.[1] Nonetheless, some caution is required when generalising the findings to the wider Australian community, with the results more relevant to females who suspect they have IP. The other limitation of surveying this population group is the accuracy of reporting their IP status and the reported symptoms correlating to poor health rather than IP alone. Participants were asked to report if they have IP, with many self-diagnosing their IP. As a result, the conformation of IP status is unknown, and some symptoms may be the result of poor health. This limitation was addressed by referring to the population group as having suspected IP rather than diagnosed IP. This reporting method does not appear to affect the accuracy of reported health-seeking behaviours as previous research has shown that people with self-reported IBS have similar health care utilisation and QoL as those with diagnosed IBS.[270]

The IP Guideline was developed based on the NHMRC *Guidelines for Guidelines* Handbook to meet the 2016 *NHMRC Standards for Guidelines.*[215] As outlined in the NHMRC guidelines, HMRC standard 6.2 requires guidelines to be appropriately peer-reviewed. Other guidelines have the guideline appraised by two independent reviewers using the Appraisal of Guidelines for Research and Evaluation II (AGREE II) instrument. This process was not undertaken as the guideline was not intended to be submitted to the NHMRC for approval or to be registered on the Australian Clinical Practice Guideline Register. However, to

ensure agreement with major stakeholders, after the completion of the recommendations, the stakeholder survey served as a method to capture the agreement and public opinion on the developed recommendations.

Due to the inclusion and exclusion criteria in the literature search, some potential treatment interventions were missed. Articles were excluded if they involved participants undertaking some form of physical activity. Excluding these studies was on the basis that exercise-induced IP may have a different aetiology than the IP seen in clinical practice. However, upon evaluating the literature, a few possible treatment interventions were identified as having a beneficial impact on intestinal integrity in patients with exercise-induced IP. Of note, there were six studies using colostrum that were identified.[379-384] As described in the IP Guideline, no evidence was found on the use of colostrum in patients with IP as all were excluded as they were performed in exercise-induced IP. Therefore, there may be other treatment interventions clinicians may consider using that were not mentioned in this thesis.

9.7 FUTURE DIRECTION IN RESEARCH

The results of this study provide novel information and understanding for the growing area of IP research. This thesis has identified critical areas where further research should focus its attention. Attending to the following areas of research is thought to address major knowledge gaps, advance the understanding of IP and improve the management of IP in clinical practice.

9.7.1 CLINICAL TRIALS FOR THE MANAGEMENT OF IP

Ultimately, further research can use the results of this thesis as the basis and justification for the development of clinical trials to explore the treatment strategies used by clinicians and consumers that has limited evidence. There are several possible treatment interventions that have no high-level evidence, or the study designs are inadequate to answer the research question. These interventions included vitamin D, *Curcuma longa,* omega-3 fatty acid and colostrum supplementation. Alternatively, some treatment options had a large body of evidence yet still produced a lack of clarity in treatment recommendations. This was most notable for prebiotics and probiotics as many interventions used multi-strain probiotics or a mix of prebiotic fibres. One area where future research can draw on our results is the use of strain-specific probiotics and intervention studies using a single prebiotic. This would enable the development of more targeted treatment interventions.

One area where future research may consider changing the study design is alcohol consumption. All studies used to create the recommendations in the IP Guideline were between 30 minutes and one week in length. Designing an observation or clinical trial with a study length of four weeks or more may provide the evidence to change the recommendation associated with alcohol consumption. Other considerations surrounding study design that future clinical trials should consider following are the method of evaluating IP and inclusion criteria. Specifically, combined use of stool zonulin and the dual sugar test should be used to produce the most accurate assessment of IP. Secondly, participants

should be screened for IP before entering the study to ensure the population has confirmed IP.

9.7.2 DISEASE BURDEN ASSOCIATED WITH IP

This thesis provides the first indication that a relationship exists between IP and disease burden, namely SWB and HRQoL. The results of this research project offer only a preliminary understanding due to the inability to confirm the diagnosis of IP. Future research should expand on our findings by undertaking a cross-sectional study that employs a validated method of IP assessment to confirm the presence of IP and explore the relationship with the same patient report outcome measures (PROMs) used in this thesis. This would allow other research to confirm our findings or give an indication of any significance. Other preliminary studies may consider investigating extra types of disease burden in people with suspected IP. Specifically, are mortality and morbidity associated with the diagnosis of IP? This type of research may shed light on the long-term impact of IP and quantify the rate of disability-adjusted life years (DALYs) in this population group.

9.7.3 CLINICAL PRACTICE GUIDELINE FOR THE ASSESSMENT OF IP

This thesis has focused on the treatment of IP in clinical practice. However, in our survey of Australian adults with suspected IP, the exploration of health-seeking behaviours for the assessment of IP was also undertaken. These results highlighted major inconsistency in the diagnosis of IP in clinical practice. In conjunction with the vast number of assessment methods available to clinicians

in clinical practice, further guidance is needed to address these knowledge gaps. The development of a clinical practice guideline for the assessment of IP would provide clinicians with the guidance to improve the identification of IP. This guideline should focus on the following objectives: (1) explore the circumstances and health conditions that clinicians should measure patients for IP; and (2) identify the most accurate, accessible, and affordable methods to measure altered IP. The combination of this proposed guideline and the developed guideline as part of this thesis provides clinicians with the best available evidence for the assessment and management of IP in clinical practice.

9.7.4 DISSEMINATION AND IMPLEMENTATION OF THE IP GUIDELINE

The dissemination and implementation of the IP Guideline remain an area for further consideration. Applying new knowledge to clinical decision-making is faced with many barriers, including access to information and time required to implement effectively.[385] Furthermore, the methodology used to inform clinicians of new treatment recommendations can impact adherence and utilisation of guidelines.[386] Therefore, correctly informing clinicians of the new recommendations and implementing them into clinical practice is an additional step required to increase the utilisation of the IP Guideline in clinical practice. Researchers can employ knowledge mobilisation frameworks involving clinicians and patients to effectively disseminate and implement the IP Guideline into clinical practice.[387] The most appropriate methods of disseminating and implementing the IP Guideline may involve a multi-strategy knowledge translation approach involving educational materials, seminars, podcasts and publications.[386] This implementation research would benefit from following

previously established checklists such as the *Guideline Implementation Planning Checklist* to ensure the most appropriate methods are applied.[388] After the implementation of the IP Guideline, further updates for this guideline should take place every five years to ensure the recommendation reflect the latest evidence and support clinicians in clinical practice. As emerging evidence becomes available for lifestyle and pharmaceutical interventions, updates of the IP Guideline should consider incorporating these treatment options. Future updates should also consider registering and submitting the guideline with the NHMRC to further support with the implementation and dissemination.

10. CONCLUSION

This thesis presents a clinical practice guideline for managing Australian adults with IP, which considers the views and preferences of major stakeholders and consumers. This research project incorporates the best available published literature while considering the health-seeking behaviours of the target population group. In addition to producing evidence-based recommendations, this thesis also provides a comprehensive summary of risk factors associated with IP and a novel understanding of the burden associated with IP. Through analysing both published literature and the results of two cross-sectional surveys, this thesis provides an understanding to the research objectives while offering novel insights into the advancement of IP management.

Principally, in response to Research Objective 1, this thesis identified the most significant risk factors associated with IP. The strongest risk factors for IP were elevated levels of pro-inflammatory markers, dyslipidaemia, hyperglycaemia, insulin resistance, anthropometric measurements resembling obesity, advanced disease severity, comorbidity, and consuming a Western-style diet. Most notably, the risk of IP increases when coupled with a multiple disease state or combined with other environmental risk factors.

Secondly, through exploring the views, preferences, and health-seeking behaviours of adults with suspected IP, Research Objective 2 was addressed. Specifically, most Australian adults with suspected IP are self-diagnosing their condition, with the majority of these individuals preferring to be assessed using

an accurate method by a general practitioner or naturopath. Most Australian adults with suspected IP reported using dietary products, dietary supplements and lifestyle therapies for managing IP. This research project also identified that most people that believe they have IP experienced a considerable length of time, approximately 11 years, between first suspecting IP and receiving a diagnosis of IP. Following on from these findings, the out-of-pocket expenditure associated with the management of IP suggests a financial burden, especially for Australian adults with suspected IP that report struggling financially.

In response to Research Objective 3, the findings suggest IP may pose a greater health burden than previously thought, with poor SWB and HRQoL reported in Australian adults with self-reported IP. These results have provided the first known indication that suspected loss of intestinal integrity may be associated with an increase in disease burden. Especially as our study found self-reported improvement in IP is suggested to be a significant predictor of SWB and HRQoL and that there is a correlation between both SWB and HRQoL and the number of days IP affects daily living. Collectively, these findings contribute to a much-needed clinical understanding of the consequence and clinical relevance of altered IP.

Thirdly, the development of the IP Guideline has addressed Research Objective 4 and 5. The IP Guideline serves as the first guideline for the management of IP here in Australia and internationally. The 38 recommendations created utilised the best available evidence while considering the views and preferences of consumers and clinicians. These recommendations provide clinicians with

beneficial dietary choices and dietary supplements while also suggesting interventions that are not effective and should be avoided. Furthermore, most of the developed recommendations were accepted and acknowledged to be important and appropriate for clinicians to follow according to key stakeholders. Collectively, the IP Guideline may optimise patient care, improve health outcomes, and reduce variation in care for clinicians treating IP in a primary care setting.

Finally, as a collective body of work, this thesis provides a foundation to support clinicians in the management of IP in clinical practice while directing future research to expand the current scientific understanding of IP. The created recommendations align with consumer and stakeholder views and values, enabling clinicians to follow confidently. This thesis provides a comprehensive insight into the need of this under-investigated population group while laying the foundations for multiple research opportunities, especially in the exploration of disease burden and IP. Ultimately, further research can use these results as the basis and justification in developing clinical trials to explore the treatment strategies used by clinicians and consumers yet have no supporting evidence.

11. APPENDICES

APPENDIX 1.1: MANUSCRIPT OF SYSTEMATIC REVIEW

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SYSTEMATIC REVIEW

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Risk factors associated with intestinal permeability in an adult population: A systematic review

Bradley Leech | Erica McIntyre | Amie Steel | David Sibbritt

Faculty of Health, Australian Research Centre in Complementary and Integrative Medicine, University of Technology Sydney, Ultimo, New South Wales, Australia

Correspondence
Bradley Leech, Faculty of Health, Australian Research Centre in Complementary and Integrative Medicine, University of Technology Sydney, 15 Broadway, Ultimo, NSW 2007, Australia
Email: bradgleech@gmail.com

Abstract

Background: Increased intestinal permeability (IP) involves the loss of integrity between the cells of the small intestine. IP has been suggested to contribute to the pathogenesis and exacerbation of many chronic diseases. Many potential risk factors for IP are proposed in contemporary literature. The purpose of this review is to identify the most significant risk factors for IP.

Methods: A systematic search of literature published up until September 2018 in the PubMed, EMBASE, CINAHL, and Scopus databases was conducted.

Results: A total of 47 articles met the inclusion criteria. Elevated levels of proinflammatory markers, dyslipidaemia, hyperglycaemia, insulin resistance, anthropometric measurements resembling obesity, advanced disease severity, comorbidity and the consumption of a Western-style diet were identified as the strongest risk factors for altered intestinal integrity. The risk of IP increases when coupled with a multiple disease state or combined with other environmental risk factors. Furthermore, many of the identified risk factors such as anthropometric measurements and biomarkers were external from intestinal health and rather resembled a metabolic-like condition.

Conclusions: This review identified a number of potential risk factors for IP, ranging from biomarkers to anthropometric measurements, demographics, dietary intake and chronic diseases. These risk factors warrant the attention of clinicians and other healthcare providers to aid the identification of potential patients at risk of altered IP. Further research needs to examine whether the identified risk factors are homogeneous with the diagnosis of IP or whether the disease state influences the association.

1 | INTRODUCTION

Increased intestinal permeability (IP) involves the loss of integrity between the cells of the small intestine.[1] The prevalence of altered IP is estimated to be 10%-87%[2] in diseases with a known association compared to about 5% in healthy subjects.[3,4] Furthermore, approximately one in three individuals are suggested to experience IP when diagnosed with a disease associated with IP.[2] Although the concept of IP was first mentioned in the literature during the 1960s[5] and further explored in relation to disease during the 1970s[6] it was not until the 2000s that the mechanism of action for IP development was discovered, providing further clarification into the role IP plays in health and disease.[7] While IP may be considered an emerging health condition that clinicians should be aware of, the consequence of impaired barrier function remains underexamined.[8]

The loss of intestinal integrity occurs when the transmembrane proteins connecting the cells of the small intestine disassemble in response to a cascade of events involving the protein zonulin.[3] As a result of altered IP, particular aspects of disease such as clinical symptoms, severity and activity have been found to be exacerbated in the presence of IP.[9,10] In addition, preliminary evidence suggests that IP may be involved in the pathogenesis of type 1 diabetes,[11,12] Crohn's disease,[13] coeliac disease[14] and diarrhoea-predominant irritable bowel syndrome (IBS-D).[15,16] Altered IP has also

been associated with many autoimmune conditions, liver diseases, gastrointestinal conditions and metabolic conditions.[2] Although the pathogenesis is not clearly defined, inflammation appears to be involved both as a driving factor for and consequence of altered IP.[2,17] Furthermore, the aetiology of IP is poorly understood, with early research indicating that two aspects, namely pathogenic bacteria and gliadin from gluten, are responsible for triggering IP.[18] Although, recent research suggests that the pathogenesis of IP is multifactorial and different for each individual.[19]

There are two tests primarily used for the clinical diagnosis of IP, namely the dual sugar test and serum zonulin; with many others used in a research setting. However, there remains controversy surrounding the gold standard of IP testing and the consistency between measurement methods.[20] The dual sugar test involves the oral consumption of two sugars after an overnight fast followed by the collection of urine for a given period of time. The fundamental principle behind the dual sugar test is the different molecule size of monosaccharide and disaccharide. When the integrity of the intestine is healthy the monosaccharide (mannitol) is easily absorbed whereas the disaccharide (lactulose) is poorly absorbed and remains in the intestine. During altered IP the disaccharide is readily absorbed resulting in an increased ratio between lactulose and mannitol in the urine.[21] Whereas zonulin, the protein responsible for the disassembling of the tight junctions, can be measured in either the serum or stool.[1] Zonulin is considered to be the only measurable biomarker that reflects an impairment of the intestinal barrier.[1,22] However, zonulin has been reported to be released from many tissues including adipose tissue and proposed to be a biomarker of metabolic syndrome, obesity, inflammation and poor health more so than IP.[23] Nevertheless, zonulin is recognised as an accurate measurement of IP.[22] Another method of measuring IP is the level of lipopolysaccharide (LPS) found within the blood. LPS is suggested to be an exacerbator and marker of IP and is mostly increased at the later stage of disease or in advanced IP.[24,25] Collectively, these markers of IP provide healthcare practitioners with a method to measure and assess IP in clinical practice.

Correctly identifying patients at risk of IP may allow for timely testing to determine the potential severity of IP and facilitate access to appropriate treatment interventions if required. Although the full extent of untreated IP remains underexamined, there is a considerable amount of research linking the health and integrity of the intestine to chronic disease.[19] The purpose of this review is to summarise the known risk factors for IP and identify the most significant of these risk factors.

Review criteria

- Observational studies reporting on risk factors associated with intestinal permeability in an adult population were identified through a systematic search on PubMed, EMBASE, CINAHL and Scopus up until September 2018. The reporting of this systematic review followed the PRISMA statement and MOOSE checklist.

Message for the clinic

- This review identified over 100 potential risk factors associated with intestinal permeability with the strongest risk factors being inflammation, dyslipidaemia, hyperglycaemia, anthropometric measurements resembling obesity, advanced disease severity with comorbidity and the consumption of a Western-style diet. These risk factors warrant the attention of clinicians to aid in the identification of potential patients at risk of altered intestinal permeability.

2 | METHODS

The reporting of this systematic review followed the Preferred Reporting Items for Systematic Reviews and Meta-Analyses[26] statement and the Meta-analysis Of Observational Studies in Epidemiology checklist.[27] The protocol was registered with the International Prospective Register of Systematic Reviews (PROSPERO) (#CRD42018109384).

2.1 | Search strategy

The databases PubMed, EMBASE, CINAHL and Scopus were searched for articles published up until September 2018 by the lead researcher (BL). The single-arm search terms used were: "intestinal permeability" OR "intestinal integrity" OR "intestinal barrier dysfunction" OR "gastrointestinal permeability" OR "gut permeability" OR "zonulin" OR "dual sugar" OR "lactulose AND mannitol" OR "lactulose AND rhamnose" OR "cellobiose AND mannitol." A hand search of the reference list from the included articles was also carried out.

2.2 | Eligibility criteria

Included articles were original observational studies reporting on risk factors associated with IP in an adult population. These risk factors are in relation to low-grade chronic IP rather than acutely induced IP caused by sepsis in critically ill patients. Articles were excluded if subjects were under the age of 18, were critically ill (ie in intensive care or palliative care), involved an experimental design or used a method of diagnosing IP other than zonulin (serum, plasma, stool), dual sugar urinary test (lactulose/mannitol, lactulose/rhamnose, cellobiose/mannitol) and serum LPS. These methods were selected to ensure clinical relevance of the review. There was no exclusion based on language, geographical location or publication date.

2.3 | Study selection and data extraction

All identified citations were imported to Endnote (Version X9) and duplicates removed. The citations were independently screened for eligibility by the lead author (BL). A sample (20%) of the eligibility citations was reviewed by a second author (EM). When uncertainty

of eligibility criteria arose the corresponding author of the article in question was contacted for clarification.

2.4 | Critical appraisal analysis and risk of bias assessment

The quality of the included articles was assessed (by BL) and reviewed (by EM) using the Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) statement.[28] In addition, the included articles were assessed for risk of bias using a previously established tool for prevalence studies. The assessment tool is composed of 10 items covering four main domains of bias including external validity, internal validity, measurement bias and bias relating to analysis.[29]

2.5 | Statistical analysis

A thematic synthesis of the association between risk factors of IP and altered IP was carried out. Three categories of association namely odds ratio (OR), beta coefficient and correlation coefficient were collectively assessed for associated risk factors with IP. Only statistically significant risk factors were extracted from the included articles, along with the confidence interval (CI). Furthermore, only ORs and beta coefficients that adjusted for confounders were extracted. Unadjusted correlation coefficients were extracted; however, precedence was given to adjusted correlation coefficients when available. Interpretation of both Spearman's (ρ) and Pearson's (r) correlation coefficient were as followed: little (0.00-0.29), weak (0.30-4.9), moderate (0.50-0.69), high (0.70-0.89) and very high (0.90-1.00) correlation.[30] Variables with a little correlation coefficient were omitted from the results to minimise misinterpretation where results remain uncertain. However, these variables were still reported in study characteristics and considered in the discussion as the articles met the inclusion criteria. When associations were determined by a coefficient of determination (R^2), this value was converted to a correlation coefficient by taking the square root of the R^2 value.

3 | RESULTS

A total of 22 118 articles were identified through the key database searches, of which 10 914 duplicates were removed. After title and abstract screening 149 potentially relevant full-text articles were reviewed, of which 42 articles were considered eligible. Hand searching the reference list of the 42 eligible articles identified an additional five articles. A total of 47 articles were included in this systematic review (Figure 1). From the sample of eligible studies reviewed (by EM), a strong agreement (Kappa score 0.90) was achieved.

The sample size of each study varied from 21 to 1015[31,32] (mean = 155) with the majority of the studies carried out in Europe (n = 34) followed by Asia (n = 5), America (n = 5), Africa (n = 2) and Australia (n = 1). The laboratory markers of IP used in each study

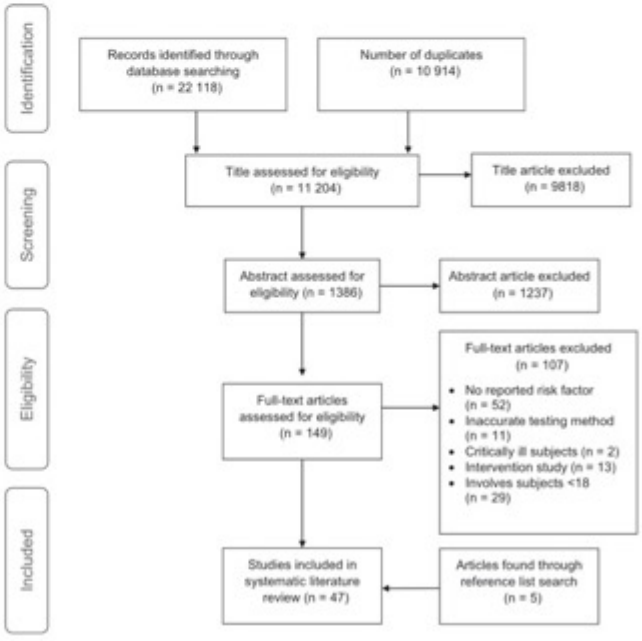

FIGURE 1 PRISMA flow diagram of study selection. Starting with 22 118 identified citations, 47 articles were included in the final systematic review

were zonulin (n = 24), dual sugar (n = 13), LPS (n = 10) and stool zonulin (n = 3). A total of 30 different study populations were measured for IP with findings suggesting 101 statistically significant risk factors associated with IP. Risk factors were identified in study populations with glucose metabolism disorders (n = 57), body mass index (BMI) >29 (n = 42), pregnancy (n = 39), liver conditions (n = 34), general population (n = 29), polycystic ovarian syndrome (PCOS) (n = 14), digestive conditions (n = 13), kidney disease (n = 12), obstructive sleep apnoea (n = 12), respiratory conditions (n = 6), pain conditions (n = 4), alcohol use disorder (n = 2), Parkinson's disease (n = 2), ankylosing spondylitis (n = 1) and systemic sclerosis (n = 1). These risk factors were grouped into five major domains; *medical history and disease*, *dietary factors*, *anthropometric measurements*, *biomarkers* and *demographic factors*.

3.1 | Critical appraisal and risk of bias assessment

STROBE evaluation identified that the majority of the included articles provided an inadequate indication of study design, methods of addressing bias, study size calculation or consider the use of a flowchart (Table 1). Three articles were recognised as low-quality.[33-35] During risk of bias assessment, no articles were identified as high risk of bias; although, 27 of the 47 articles were classified as having a moderate risk of bias. This moderate risk of bias was primarily because of the articles demonstrating large gaps in the external validity criteria. Internal validity assessment showed a low risk of bias with a large degree of consistency between articles. Results from the risk of bias assessment are presented in Table 2.

3.2 | Medical history and disease risk factors

Twenty studies reported a statistically significant association between IP and 19 medical history attributes.[10,23,25,33,34,36-50] The diagnosis of particular health conditions such as diabetes,[44,46,50] liver disease[25,45,47] and gastrointestinal conditions[33,39,48,49] were reported to be associated with altered IP. First, the likelihood of altered IP in type 2 diabetes ranges from OR = 1.080 (95% CI: 1.005, 1.161; $P = 0.037$) to OR = 2.888 (95% CI: 1.553, 5.370; $P < 0.001$) with the severity of IP associated with the odds of type 2 diabetes[44] (Table 3). Gestational diabetes was also reported to have a similar association with altered IP (OR = 1.08; 95% CI: 1.02, 1.15; $P = 0.009$).[46] Furthermore, the age of type 1 diabetes onset was reported to correlate with IP ($\beta = -0.14$; $P < 0.001$)[50] (Table 4). The degree of liver damage in non-alcoholic fatty liver disease was reported to have a moderate positive correlation with IP ($r = 0.69$; $P = 0.01$)[25] while the diagnosis of moderate-to-severe fatty liver was associated with altered IP (OR = 1.77; 95% CI: 1.13, 2.76; $P = 0.015$).[47] Altered IP was reported to be associated with underlying organic digestive diseases (OR = 1.56; 95% CI: 1.32, 1.85; $P < 0.0001$)[48]; although, altered IP was also reported to be an independent risk factors for diarrhoea-predominant irritable bowel syndrome (IBS-D) ($\beta = 0.63$; 95% CI: 0.09, 1.16; $P = 0.022$)[49] and has a moderate positive correlation with Crohn's disease relapse ($r = 0.48$; $P = 0.008$) (Table 5).[39] The association between disease duration of Parkinson's disease, inflammatory bowel disease and systemic sclerosis and IP were reported to have a weak to high correlation in three studies ($r = 0.73$; $P < 0.011$) with altered IP reported in the early stages of disease manifestation.[36-38]

The strength of association between IP and disease severity varied depending on the study population and the nature of disease severity classification. Pain, as measured by numeric rating scale, was reported to have a weak positive correlation with IP.[40] Whereas conflicting evidence was reported for blood pressure and the strength of association with altered IP. The association between portal hypertension and altered IP was only reported for second and third-degree portal hypertension (OR = 3.1; 95% CI: 1.1, 4.2; $P < 0.01$).[45] A moderate positive correlation was reported between IP and systolic blood pressure in obesity ($r = 0.40$; $P = 0.009$).[42] Whereas diastolic blood pressure (66-72 mmHg) was reported to be an independent risk factor for altered IP (OR = 2.82; 95% CI: 1.43, 5.58; $P = 0.003$) in the general population.[23]

3.3 | Dietary risk factors

Five studies reported nine statistically significant dietary factors that were associated with IP.[25,31,47,51,52] Intake of >2616 kcal/day was reported as an independent risk factor for altered IP ($\beta = 121.8$; $P = 0.04$) as measured by LPS[31] (Table 4). Total fat percentage in the diet was also reported as an independent risk factor for altered IP ($\beta = 0.23$; 95% CI: ±0.11; $P < 0.05$).[51] One study reported protein intake as an independent risk factor for altered IP ($\beta = -0.139$; 95% CI: −0.247, −0.031; $P = 0.01$).[52] While one other study[25] reported a moderate positive correlation between total protein intake and IP ($\rho = 0.59$; $P = 0.001$) with sub-analysis on protein source reporting that animal-derived protein intake had a moderate positive correlation with altered IP ($\rho = 0.54$; $P = 0.002$)[25] (Table 5). One study reported alcohol consumption to be a predictive risk factor for altered IP, with <14 standard drinks per week (OR = 1.91; 95% CI: 1.01, 3.95; $P = 0.05$) and above >15 standard drinks per week (OR = 1.56; 95% CI: 1.02, 2.67; $P = 0.05$) associated with altered IP.[47]

3.4 | Anthropometric risk factors

Ten studies reported a statistically significant association between 12 anthropometric measurements and IP.[10,23,42,51,53-58] The correlation between BMI and IP ranged from a weak to moderate positive correlation, of which most were reported to have a weak positive correlation[23,44,51,55,56,59] (Table 5). Two studies report BMI as an independent risk factor for altered IP as measured by zonulin levels ($\beta = 0.26$; ±0.10; $P < 0.05$, $\beta = 1.507$; 0.34 SEM; $P < 0.01$)[51,54] (Table 4). Furthermore, it was reported in the general population that a BMI of >25.0 and BMI of >30.0 were associated with altered IP OR = 4.10 (95% CI: 1.87, 8.97; $P < 0.001$) and OR = 4.90 (95% CI: 1.49,

31.65; $P = 0.047$), respectively as measured by zonulin (>64 ng/mL)[23] (Table 3). Two studies reported the strength of association between IP and both waist circumference and waist to hip ratio.[10,53] Although only a weak positive correlation was reported between waist circumference and IP[53] one study reported an association between altered IP and waist circumference >97 cm (OR = 7.03; 95% CI: 1.97, 25.11; $P = 0.003$).[23]

3.5 | Biomarker risk factors

Twenty-four studies reported on 29 statistically significant biomarkers and association with altered IP.[10,23,25,32,35,40-44,50-53,55-72] Two studies reported that fasting glucose had a weak positive correlation with IP[44,53] (Table 5). Moreover, an additional study reported that a plasma glucose level >5.7 nmol/L is associated with a greater odds of having altered IP (OR = 2.09; 95% CI: 2.09, 4.18; $P = 0.036$) in the general population[23] (Table 3). In addition, fasting glucose was reported to be an independent risk factor associated with altered IP as measured by zonulin ($\beta = 0.38$; ±0.12; $P < 0.05$)[51] (Table 4). In contrast, a 120-minute glucose tolerance test was reported in three studies to have a weak positive correlation with IP.[10,44,60] Three studies reported a weak positive correlation between glycated haemoglobin A1c (HbA1c) and IP.[42,44,60]

From the four studies that reported a statistically significant association between fasting insulin and IP a weak to moderate positive correlation was reported ($\rho = 0.616$; $P < 0.001$)[44,53,55,61] (Table 5). Furthermore, one study reported fasting insulin to be associated with IP as measured by zonulin levels ($\beta = 0.015$; 95% CI: 0.007, 0.022; $P < 0.001$).[61] Four slightly different methods were used to measure insulin sensitivity with the strength of association varying from a weak to moderate correlation between insulin sensitivity and IP ($r = 0.605$; $P < 0.05$).[10,44,56,61] Moreover, insulin sensitivity was reported to be associated with IP as measured by zonulin ($\beta = -0.263$; $P = 0.004$, $\beta = -0.002$; 95% CI: −0.003, −0.001; $P < 0.001$)[56,61] (Table 4). Five studies found a similar strength of association between markers of insulin resistance and altered IP; with a weak to moderate positive correlation reported ($\rho = 0.616$; $P < 0.001$).[10,44,55,58,61]

Serum lipids and lipoproteins were measured in 10 studies with a varying degree of strength of association with IP.[10,42,44,50,53,55,58,60-62] Total cholesterol was reported to have a statistically significant association with IP in four studies, these studies report a weak to moderate positive correlation between total cholesterol and IP ($\rho = 0.566$; $P < 0.001$)[44,53,61,62] (Table 5). Furthermore, total cholesterol was reported to correlate with IP as measured by zonulin ($\beta = 0.004$; 95% CI: 0.000, 0.007; $P = 0.032$)[61] (Table 4); whereas, low-density lipoprotein cholesterol was reported to have a weak positive correlation with IP.[44,61] Five studies reported high-density lipoprotein (HDL) cholesterol to have a weak negative correlation with zonulin and the dual sugar test.[10,42,44,55,58,60] Seven studies reported triglycerides to have a weak to high positive correlation with IP ($r = 0.73$; $P < 0.001$).[10,42,44,50,60-62] Triglycerides were further reported as an independent risk factor for IP as measured by zonulin ($\beta = 0.009$; 95% CI: 0.003, 0.015; $P = 0.003$).[61]

Numerous inflammatory markers were measured in a total of 11 studies and were reported to have an association with IP.[41,44,51,53,56,57,59-61,63,72] High-sensitivity C-Reactive protein (hsCRP) was reported to have a weak positive correlation IP[53,61,63] (Table 5) with two studies also reporting that hsCRP correlates with IP as measured by zonulin ($\beta = 0.013$; 95% CI: 0.003, 0.023; $P = 0.015$, $\beta = 0.075$; 95% CI: 0.008, 0.158; $P = 0.046$)[53,61] (Table 4). In contrast, another study reported CRP to be an independent risk factor for IP ($\beta = 3.28$; 95% CI: 1.28, 5.46; $P < 0.01$).[72] Circulating interleukin-6 (IL-6) was reported to have a weak to moderate positive correlation with IP according to four studies ($r = 0.542$; $P < 0.001$).[56,57,59,60] Furthermore, two studies reported circulating IL-6 to independently correlate with levels of zonulin ($\beta = 0.23$; $P = 0.04$)[56] and LPS ($\beta = 0.171$; $P = 0.04$)[64]; whereas, tumour necrosis factor alpha (TNF-α)—the other major inflammatory marker measured in the included studies—was reported to have a weak to moderate positive correlation with IP ($r = 0.647$; $P < 0.05$).[44,57,60]

Three studies report the liver enzyme alanine transaminase (ALT) to have a statistically significant weak to moderate positive correlation with IP ($\rho = 0.50$; $P = 0.005$)[25,53,55] (Table 5), with one study reporting ALT to correlate with IP as measured by zonulin ($\beta = 0.014$; 95% CI: 0.001, 0.028; $P = 0.04$)[53] (Table 4). Two other liver enzymes aspartate transaminase (AST) and gamma glutamyltransferase were reported to have a weak positive correlation with IP, with only AST reported as an independent risk factor for IP ($\beta = 0.02$; 95% CI: 0.002, 0.037; $P = 0.04$).[53] One study reported that microbial diversity had a weak negative correlation with serum zonulin and stool zonulin.[70]

The strongest association among biomarkers and IP were markers of IP themselves. Two studies used both zonulin and the dual sugar test and reported a high positive correlation between the two tests ($\rho = 0.891$; $P = 0.05$)[32,66] (Table 5). However, mixed evidence was reported for the strength of association between zonulin and LPS with a weak to moderate positive correlation reported ($\rho = 0.545$; $P < 0.001$)[63,67-69]; although, one study reported LPS to independently correlate with zonulin levels ($\beta = 0.002$; 95% CI: 0.001, 0.003; $P = 0.002$)[61] (Table 4). The dual sugar test was also reported to have a moderate to high positive correlation with gastroduodenal permeability according to one study ($r = 0.68$; $P < 0.0001$, $r = 0.88$; $P < 0.0001$).[40]

3.6 | Demographic risk factors

Although most studies evaluated basic demographic characteristics only age was reported to have a statistically significant association with IP in three studies.[45,51,73] Study populations that were diagnosed with a health condition reported a weak positive correlation between age and IP according to both zonulin and the dual sugar test[45,51,73] (Table 5). Age was reported an independent risk factor for altered IP[51] with the increase in IP more likely over the age of 50 (OR = 1.9; 95% CI: 1.1, 2.3; $P < 0.001$)[45] (Table 3).

TABLE 1 Results from critical appraisal according to STROBE guidelines for included articles

STROBE critical appraisal tool	Title, abstract and Introduction				Methods											
Study	Title and Abstract		Background	Objectives	Study design	Setting	Participants		Variables	Data sources	Bias	Study size	Quantitative variables	Statistical methods		
	1a	1b	2	3	4	5	6a	6b	7	8	9	10	11	12a	12b	12c
Amar et al[35]	x	x	x	x	x	x	x	/	x	x	-	-	x	x	x	x
Barcelo et al[12]	x	x	x	x	x	x	x	x	x	x	x	-	x	x	x	-
Cangemi et al[49]	x	x	x	x	x	x	x	x	x	x	-	-	x	x	x	x
Cariello et al[45]	x	x	x	x	x	x	x	x	x	x	-	-	x	x	x	x
Carnevale et al[47]	x	x	x	x	x	x	x	/	x	x	-	x	x	x	x	x
Caserta et al[36]	-	x	x	x	x	x	x	x	x	x	-	x	-	x	x	x
Caviglia et al[37]	x	x	x	x	x	x	-	-	x	x	-	-	-	x	x	-
Ciccia et al[44]	-	x	x	x	x	x	x	/	x	x	-	-	x	x	x	x
Di Leo et al[48]	x	x	x	x	x	x	x	/	x	x	x	-	x	x	x	x
Donnadieu-Rigole et al[34]	-	x	x	x	x	x	x	-	x	x	-	-	x	x	x	-
Du Plessis et al[42]	-	x	x	x	x	x	x	x	x	x	-	-	x	x	x	x
Duerksen et al[32]	-	x	x	x	x	x	x	x	x	x	x	-	x	x	-	-
Ficek et al[64]	-	x	x	x	x	-	x	x	x	x	-	-	x	x	x	x
Goebel et al[40]	-	x	x	x	x	x	x	x	x	x	-	-	x	x	x	x
Hendy et al[33]	-	x	x	x	x	x	x	x	x	x	-	-	x	x	x	x
Hilsden et al[39]	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	-
Jayashree et al[60]	-	x	x	x	x	x	x	x	x	x	-	-	x	x	x	x
Johnston et al[71]	x	x	x	x	x	x	x	x	x	x	x	-	x	x	-	x
Karthikeyan et al[53]	-	x	x	x	x	x	x	/	x	x	-	-	x	x	x	x
Kim et al[67]	-	x	x	x	x	x	x	/	x	x	-	-	x	x	x	-
Kvehaugen et al[72]	x	x	x	x	x	x	x	/	x	x	x	-	x	x	x	x
Lassenius et al[50]	-	x	x	x	x	x	x	x	x	x	x	-	x	x	x	x
Lindheim et al[70]	x	x	x	x	x	x	x	x	x	x	-	-	x	x	-	x
Lukaszyk et al[55]	-	x	x	x	x	-	x	/	x	x	-	-	x	x	-	-
Malickova et al[65]	-	x	x	x	x	x	x	x	x	x	-	-	x	x	x	x
Malyszko et al[43]	-	x	x	x	x	x	x	/	x	x	-	-	x	x	x	x
Mokkala et al[61]	x	x	x	x	x	x	x	/	x	x	x	x	x	x	x	x
Mokkala et al[62]	x	x	x	x	x	x	x	/	x	x	x	x	x	x	x	x
Mokkala et al[66]	x	x	x	x	x	x	x	/	x	x	x	x	x	x	-	-

		Results										Discussion & other information					
		Participants			Descriptive data			Outcome data	Main results			Other analyses	Key results	Limitations	Interpretation	Generalisability	Funding
12d	12e	13a	13b	13c	14a	14b	14c	15	16a	16b	16c	17	18	19	20	21	22
x	x	x	x	-	x	x	/	x	x	x	/	x	x	x	x	x	x
x	x	x	x	-	x	x	/	x	x	x	/	x	x	x	x	x	x
/	x	-	-	-	x	x	-	x	x	x	/	x	x	x	x	x	x
x	x	x	x	-	x	x	/	x	x	x	/	x	x	-	x	x	x
x	x	x	-	-	x	x	/	x	x	x	/	x	x	x	x	x	x
x	x	x	x	-	x	x	/	x	x	x	/	x	x	-	x	x	-
-	x	x	x	-	x	x	/	x	x	x	/	x	x	x	x	x	x
x	x	x	x	-	x	x	/	x	x	x	/	x	x	-	x	-	x
-	x	x	x	-	x	x	x	x	x	x	x	x	x	-	x	x	x
-	x	x	x	x	x	x	x	x	x	x	/	x	x	-	x	x	x
x	x	-	-	-	x	x	/	x	x	x	/	x	x	x	x	x	x
-	-	x	x	-	x	x	/	x	x	x	/	x	x	x	x	x	x
x	x	x	x	x	x	x	/	x	x	x	/	x	x	x	x	x	x
x	x	x	x	-	x	x	/	x	x	x	/	x	x	x	x	x	x
x	x	x	x	-	x	x	/	x	x	x	/	x	x	-	x	x	x
x	x	x	x	-	x	x	x	x	x	x	/	x	x	-	x	x	x
x	x	x	x	-	x	x	/	x	x	x	/	x	x	-	x	x	x
x	x	x	-	-	-	x	-	x	x	x	/	x	x	-	x	x	x
-	x	x	x	-	x	x	/	x	x	x	/	x	x	-	x	x	x
-	x	x	x	-	x	x	/	x	x	x	x	x	x	x	x	x	x
x	x	x	x	x	x	x	/	x	x	x	x	x	x	x	x	x	x
x	x	x	x	-	x	x	/	x	x	x	/	x	x	-	x	x	x
x	x	x	x	-	x	x	/	x	x	x	/	x	x	x	x	x	x
x	x	-	-	-	x	x	/	x	x	x	/	x	x	x	x	x	x
x	x	x	x	-	x	x	/	x	x	x	/	x	x	x	x	x	x
-	x	x	-	-	x	x	/	x	x	x	/	x	x	-	x	x	-
x	x	x	x	-	-	x	/	x	x	x	x	x	x	x	x	x	x
x	x	x	x	-	-	x	/	x	x	x	x	x	x	x	x	x	x
x	x	x	x	-	x	x	/	x	x	x	/	x	x	x	x	x	x

(Continues)

TABLE 1 (Continued)

STROBE critical appraisal tool	Title, abstract and introduction				Methods											
Study	Title and Abstract		Background	Objectives	Study design	Setting	Participants		Variables	Data sources	Bias	Study size	Quantitative variables	Statistical methods		
	1a	1b	2	3	4	5	6a	6b	7	8	9	10	11	12a	12b	12c
Moreno-Navarrete et al[54]	-	x	x	x	x	x	x	/	x	x	x	-	x	x	x	x
Morkl et al[59]	-	x	x	x	x	x	x	/	x	x	-	-	x	x	x	x
Mujagic et al[49]	-	x	x	x	x	x	x	/	x	x	x	-	x	x	x	x
Nymark et al[42]	x	x	x	x	x	x	x	x	x	x	-	-	x	x	x	x
Ohlsson et al[23]	-	x	x	x	x	x	x	/	x	x	x	x	x	x	x	x
Qi et al[57]	-	x	x	x	x	x	x	/	x	x	-	-	x	x	x	-
Raparelli et al[48]	x	x	x	x	x	x	x	/	x	x	-	-	x	x	x	x
Riordan et al[34]	-	x	x	x	x	-	x	/	x	x	-	-	x	x	-	-
Rutten et al[43]	-	x	x	x	x	x	x	x	x	x	x	x	x	x	-	-
Schwiertz et al[58]	x	x	x	x	x	x	-	x	x	x	x	-	x	x	x	x
Swanson et al[71]	-	x	x	x	x	x	x	x	x	x	x	x	x	x	-	-
Teixeira et al[50]	-	x	x	x	x	x	x	/	x	x	-	x	x	x	x	x
Troseid et al[62]	x	x	x	x	x	x	x	x	x	x	-	-	x	x	-	x
Volynets et al[72]	-	x	x	x	x	x	x	/	x	x	-	-	x	x	x	-
Wyatt et al[73]	x	-	-	x	x	x	x	x	x	x	-	-	x	x	-	-
Zak-Golab et al[51]	-	x	x	x	x	x	x	/	x	x	-	-	x	x	x	-
Zhang et al[60]	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x
Zhang et al[44]	-	x	x	x	x	x	x	/	x	x	-	-	x	x	x	-

"x," found within study; "-," not found within study; "/," not applicable.

4 | DISCUSSION

This is the first systematic review to explore the potential risk factors associated with IP in an adult population. This review identified over 100 potential risk factors associated with IP that had a varying degree of strength of association. The majority of the identified risk factors were only found to have a weak association with IP; however, there were similarities with many of the risk factors measured and reported to be associated with IP in numerous instances. This similarity further strengthens the identified risk factors as valuable clinical features healthcare professionals may consider as part of their differential diagnosis. Many of the risk factors identified have previously been reported as major risk factors for morbidity and mortality in chronic diseases worldwide.[74,75] Therefore, IP may be considered a feature of chronic disease rather than merely a digestive health issue.

4.1 | Strongest risk factors for altered intestinal permeability

Elevated levels of proinflammatory markers, dyslipidaemia, hyperglycaemia, anthropometric measurements resembling obesity, advanced disease severity with comorbidity and the consumption of a Western-style diet were identified as the strongest risk factors for altered IP (Figure 2). An unexpected finding of our review was the paucity of digestive health symptoms reported to be associated with IP alongside the magnitude of risk factors that resemble a metabolic-like condition. Although digestive health symptoms such as bloating, abdominal cramps and pain, heartburn, reflux, nausea and flatulent were measured in a few of the included studies, none were reported to be significantly correlated with the risk of IP.[23,40,49] The digestive health issues that were reported to be associated with IP were diseases situated primarily within the

		Results										Discussion & other information					
		Participants			Descriptive data			Outcome data	Main results			Other analyses	Key results	Limitations	Interpretation	Generalisability	Funding
12d	12e	13a	13b	13c	14a	14b	14c	15	16a	16b	16c	17	18	19	20	21	22
x	x	x	x	-	x	x	/	x	x	x	/	x	x	x	x	x	x
x	x	x	x	-	x	x	/	x	x	x	/	x	x	x	x	x	x
x	x	x	x	-	x	x	/	x	x	x	/	x	x	x	x	x	x
x	x	x	x	-	x	x	-	x	x	x	x	x	x	-	x	x	x
x	x	x	x	-	x	x	/	x	x	x	/	x	x	x	x	x	x
-	x	x	-	-	x	x	/	x	x	x	/	x	x	x	x	x	x
x	x	x	x	-	x	x	/	x	x	x	/	x	x	x	x	-	x
-	x	x	x	-	-	x	/	x	x	x	/	x	x	-	x	x	x
-	x	x	x	x	x	x	/	x	x	x	/	x	x	x	x	x	x
-	x	x	x	-	x	x	/	x	x	x	/	x	x	x	x	x	x
x	x	x	x	x	x	x	/	x	x	x	/	x	x	-	x	x	x
-	x	x	x	-	x	x	/	x	x	x	/	x	x	-	x	x	x
x	x	x	-	-	x	x	/	x	x	x	-	x	x	x	x	x	x
x	x	x	x	-	x	x	/	x	x	x	/	x	x	-	x	x	x
-	x	x	x	x	x	-	x	x	x	x	x	x	-	-	x	x	-
x	x	x	x	x	x	x	/	x	x	x	/	x	x	x	x	x	x
x	x	x	x	-	x	x	/	x	x	x	/	x	x	x	x	x	x
x	x	x	x	-	x	x	/	x	x	x	/	x	x	x	x	x	x

gastrointestinal system such as inflammatory bowel disease,[33,37] diarrhoea predominant irritable bowel syndrome (IBS-D),[49] intestinal dysbiosis,[51,52,70] symptoms like diarrhoea (especially from an organic disease)[48] and indigestion syndrome.[49] However, these digestive health symptoms were not found to be associated with IP in the general population.[23] Although digestive health symptoms appear to lack association with IP, this should not undermine the association between gastrointestinal conditions and IP, especially provided the high correlation between the improvement of altered IP and a reduction in postinfectious IBS disease severity purported in the literature.[76] Conversely, many of the risk factors that resemble a metabolic-like condition were found to be associated with IP in the general population.[23,44,56] However, risk factors such as waist-to-hip ratio, waist circumference and elevated triglycerides, were less associated with IP in the general population when compared to a disease state. It appears that the identified risk factors have a stronger association with altered IP within a disease state rather than in the general population.

Many hypotheses exist detailing the mechanism of action linking the health and integrity of the digestive system to inflammation, obesity, poor glycaemic control and dyslipidaemia.[47,52,60] One of the most prominent theories suggests IP is both a cause and consequence of LPS absorption. The translocation of LPS as the result of IP may contribute to the cascade of events that is responsible for the metabolic-like risk factors.[77,78] First, LPS have been shown to trigger inflammation that may alter glucose metabolism resulting in poor glycaemic control and insulin resistance.[77] The occurrence of dyslipidaemia may contribute to the loss of intestinal integrity as HDL is in part responsible for neutralising LPS, whereby low levels of HDL may result in inflammation and LPS exacerbating IP.[79] The link between metabolic factors and altered IP is further strengthened as improvement in some of the identified risk factors such as BMI,[80]

TABLE 2 Risk of bias assessment of the included articles

	External validity				Internal validity
Risk of bias assessment	Representativeness to national population	True representation of the target population	Random sampling methods	Likelihood of nonresponse bias minimal	Data directly collected from participants
Amar et al[31]	-	-	-	x	-
Barcelo et al[53]	x	x	x	x	-
Cangemi et al[69]	x	-	x	x	-
Cariello et al[45]	x	x	x	x	-
Carnevale et al[67]	x	x	x	x	-
Caserta et al[36]	x	x	x	x	-
Caviglia et al[37]	x	x	x	x	-
Ciccia et al[66]	x	x	x	x	-
Di Leo et al[48]	x	-	x	x	-
Donnadieu-Rigole et al[54]	x	-	x	x	-
Du Plessis et al[82]	x	x	x	x	-
Duerksen et al[32]	x	-	x	x	-
Ficek et al[64]	x	x	x	x	-
Goebel et al[40]	x	-	x	x	-
Hendy et al[55]	x	-	x	x	-
Hilsden et al[39]	x	-	x	x	-
Jayashree et al[60]	-	-	-	x	-
Johnston et al[73]	-	-	-	x	-
Karthikeyan et al[63]	x	x	x	x	-
Kim et al[47]	x	-	x	x	-
Kvehaugen et al[72]	x	-	x	x	-
Lassenius et al[50]	x	-	x	x	-
Lindheim et al[70]	x	x	x	x	-
Lukaszyk et al[35]	x	x	x	x	-
Malickova et al[65]	x	-	x	x	-
Malyszko et al[41]	x	x	x	x	-
Mokkala et al[61]	x	-	x	x	-
Mokkala et al[52]	x	-	x	x	-
Mokkala et al[66]	x	x	x	x	-
Moreno-Navarrete et al[56]	x	-	-	x	-
Morkl et al[59]	x	x	x	x	-
Mujagic et al[49]	-	-	x	x	-
Nymark et al[62]	-	-	x	x	-
Ohlsson et al[23]	-	-	-	x	-
Qi et al[57]	x	x	x	x	-
Raparelli et al[68]	x	x	x	x	-
Riordan et al[34]	x	x	x	x	-
Rutten et al[43]	x	x	x	x	-
Schwiertz et al[38]	x	-	-	x	-

Acceptable case definition	Validated study tool used	Consistent data collections methods	Appropriate parameters of numerator & denominator	Summary of overall risk of study bias	Score
-	-	-	-	-	1
-	-	-	-	-	4
-	-	-	-	-	3
-	-	-	-	-	4
-	-	-	-	-	4
-	-	-	-	-	4
-	-	x	-	x	6
-	-	-	-	-	4
-	-	-	-	-	3
-	-	-	-	-	3
-	-	-	-	-	4
-	-	-	-	x	4
-	-	-	-	-	4
-	-	-	-	x	4
-	-	-	-	-	3
-	-	-	-	-	3
-	-	-	-	-	1
-	-	-	-	x	2
-	-	-	-	-	4
-	-	-	-	-	3
-	-	-	-	-	3
-	-	-	-	x	4
-	-	-	-	-	4
-	-	-	-	x	5
-	-	-	-	-	3
-	-	-	-	x	5
-	-	-	-	-	3
-	-	-	-	-	3
-	-	-	-	-	4
-	-	-	-	-	2
-	-	-	-	-	4
-	-	-	-	-	2
-	-	-	-	-	2
-	-	-	-	-	1
-	-	-	-	x	5
-	-	-	-	-	4
x	-	-	-	x	6
-	-	-	x	x	6
-	-	-	-	x	3

(Continues)

TABLE 2 (Continued)

	External validity				Internal validity
Risk of bias assessment	Representativeness to national population	True representation of the target population	Random sampling methods	Likelihood of nonresponse bias minimal	Data directly collected from participants
Swanson et al[71]	x	x	x	x	-
Teixeira et al[58]	x	x	x	x	-
Troseid et al[42]	x	x	x	x	-
Volynets et al[25]	x	x	x	x	-
Wyatt et al[53]	x	x	x	x	-
Zak-Golab et al[51]	x	x	x	x	-
Zhang et al[30]	x	-	x	x	-
Zhang et al[44]	x	-	x	x	-

Note: "x," risk of bias; "-," low risk of bias.

HbA1c[42] and inflammation[81] have been shown to be associated with the improvement of IP.

4.2 | Chronic disease and multiple risk factors

Comorbidity of chronic diseases such as diabetes, liver disease, metabolic syndrome, kidney disease and obesity were identified to increase the risk of IP.[45,47,50,62] Moreover, the severity and activity of chronic health conditions including liver disease,[25,45,47,68,82] metabolic syndrome,[50] PCOS,[30] coeliac disease[32] and BMI[54] were reported to correlate with the degree of IP. The notion that IP correlates with disease severity is further supported as the severity of particular risk factors also increase alongside the degree of IP.[23,44,50,52,59,61] The involvement of IP in chronic disease, especially with advanced disease severity, highlights the potential importance of intestinal integrity in health and disease. This review also suggests a synergistic effect is possible when more than one risk factor is experienced. In particular, BMI, age, alcohol consumption and inflammation were all identified as having some degree of synergistic effect[47,51,56,57,61]; although, inflammation appears to be the driving factor in many of the risk factors.[51]

4.3 | Dietary and lifestyle habits and intestinal permeability

It appears that a high energy, nutrient-depleted diet with either inadequate protein intake or excess animal-derived protein in combination with alcohol consumption is a potential risk factor for IP. This dietary pattern closely resembles that of the Western diet, which has been suggested to increase the risk of chronic disease[83] and metabolic disease such as obesity.[79] Dietary intervention studies are limited; however, one study suggests an increase in dietary protein is associated with elevated zonulin and inflammation.[84] Furthermore, it has been demonstrated that an increase in dietary fibre reduces zonulin.[85] While alcohol withdrawal is associated with a reduction of IP with a greater result seen in patients with a high BMI.[54] Based on these findings, dietary and lifestyle habits may present a key clinical feature that healthcare professionals may utilise in identifying patients at risk of altered IP.

4.4 | Similarities and differences between markers of intestinal permeability

Both zonulin and the dual sugar test were reported to highly correlated with each other. However, only two risk factors namely HDL cholesterol and insulin resistance were shown to be associated with both zonulin and the dual sugar. This finding may be because of the limited number of studies using the dual sugar test included in this review. Previous research has suggested that zonulin is a biomarker of metabolic syndrome, obesity, inflammation and poor health rather than an indicator of IP[23]; although, zonulin is associated with many of the risk factors that resemble a metabolic-like condition after adjusting for metabolic syndrome, obesity and inflammation, implicating zonulin as a true marker of IP. However, the mixed evidence surrounding the association between serum and stool zonulin could be explained by zonulin being described as an acute phase biomarker of coeliac disease[86] and IBD.[87] This feature of zonulin being an acute phase biomarker may also explain the lack of consistency between the dual sugar test and zonulin. For example, IBS-D is a condition known to be related to IP and has recently been shown to be associated with the dual sugar test but not zonulin.[88] Another potential factor influencing the results is the accuracy of the commercial zonulin assay; with recent research advocating caution in using the commercial zonulin assay as a means of evaluating intestinal integrity.[89] Limited studies have used both the zonulin and dual sugar test; however, our review found that these two tests have the highest association with each other compared to all the risk factors identified. Whether zonulin is a more sensitive marker of IP for particular risk factors compared to the dual sugar test is yet to be investigated. Moreover, the ideal

Acceptable case definition	Validated study tool used	Consistent data collections methods	Appropriate parameters of numerator & denominator	Summary of overall risk of study bias	Score
-	-	-	-	-	4
-	-	-	-	-	4
-	-	-	-	x	5
-	-	-	-	-	4
x	-	-	-	x	6
-	-	-	-	-	4
-	-	-	-	-	3
-	-	-	-	-	3

test for specific disease diagnosis and the stage and activity of the disease requires further investigation. Healthcare professionals may find clinical benefit from using both the serum zonulin and dual sugar test for an accurate diagnosis of IP when patients present with the risk factors for IP.

4.5 | Limitations

This systematic review has some limitations worth mentioning. As a result of limited research examining risk factors associated with IP, this review consisted of a heterogeneous range of health conditions, preventing cumulative statistical meta-analysis. The target population for our review was adults 18 years and over; however, many large cohort studies involved adolescents. A number of articles were included when age range was unknown yet evaluated to be adults 18 years and over. Future similar reviews are suggested to incorporate adolescents into the target population. Numerous studies were excluded as they did not use measurable and comparable analysis of IP risk factors. In addition, risk factors that were not statically significant were not included potentially increasing selection bias. Other potential risk factors may have been missed because of the nature of the risk factor only being included in experimental research designs.

4.6 | Clinical significance

The clinical relevance of the identified risk factors warrants the attention of healthcare practitioners in their differential diagnosis. IP has previously been recognised by healthcare practitioners to be associated with gastrointestinal conditions more so than any other group of diseases, including metabolic conditions.[90] In our review digestive health symptoms were not identified as a major risk factor for IP. In contrast, many conditions such as food sensitivities[9] and histamine intolerance,[91] were found to be clinically relevant in the identification of patients at risk of IP. Lastly, until there is a comprehensive understanding of the clinical diagnosis of IP healthcare professionals are advised to consider multiple methods of IP testing, and to account for the identified risk factors to ensure the most accurate diagnosis of intestinal integrity.

4.7 | Further research

Further research needs to examine whether the identified risk factors are solely linked with the diagnosis of IP or whether the disease state influences the association. Further evidence is necessary to distinguish which marker of IP is most appropriate and accurate for measuring IP in different conditions and at different stages of disease manifestation. Longitudinal studies measuring the identified risk factors may provide increased understanding of the cause or consequence of IP. Lastly, the validation of serum zonulin, stool zonulin and the dual sugar test as markers for altered IP is necessary to be undertaken for the advancement of IP research.

5 | CONCLUSION

Dyslipidaemia, poor glycaemic control, inflammation, anthropometric measurements that resemble obesity, and Western-style dietary habits have the strongest association with altered IP—which amplify when combined. In addition, comorbidity of chronic diseases and advanced disease severity are also strong risk factors of altered IP. These risk factors warrant the attention of clinicians and other healthcare providers to aid in the identification of potential patients at risk of altered IP.

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TABLE 3 Risk factors associated with intestinal permeability according to adjusted odds ratio

Author	Study population	Sample size	Age (range)	Country	Test	Cut-off	Risk factor	Odds ratio	95% CI	P value
Medical history and disease										
Zhang et al[44]	Type 2 diabetes	102	47 ± 12	China	Zonulin	6 ± 4 ng/mL	Type 2 diabetes	1.140[a]	1.076, 1.208	$P < 0.001$
								1.117[b]	1.051, 1.187	$P < 0.001$
								1.080[c]	1.005, 1.161	$P = 0.037$
Zhang et al[44]	Various glucose tolerance	130	47 ± 12	China	Zonulin	4.3-7.6 ng/mL	Type 2 diabetes	1.966[c]	1.068, 3.618	$P < 0.001$
Zhang et al[44]	Various glucose tolerance	120	47 ± 12	China	Zonulin	7.6-27.2 ng/mL	Type 2 diabetes	2.888[c]	1.553, 5.370	$P < 0.001$
Mokkala et al[46]	Pregnancy	88	30.1 ± 4.9	Finland	Zonulin	45 ± 10 ng/mL	Gestational diabetes	1.08[c]	1.02, 1.15	$P = 0.009$
Kim et al[47]	Moderate-to-severe fatty liver	34	44.7 ± 9.2 (30-60)	South Korea	Zonulin	6-21 ng/mL	Moderate-to-severe fatty liver	1.83[b]	1.18, 2.84	$P = 0.007$
								1.77[c]	1.13, 2.76	$P = 0.015$
Carielo et al[45]	Chronic liver disease	83	54.1 (28-78)	Italy	L/M	0.030%	Portal hypertension degree 2-3	3.1[a]	1.1, 4.2	$P < 0.01$
Ohlsson et al[23]	General population	363	43 (28-53)	Sweden	Zonulin	34 ± 14 ng/mL	Diastolic blood pressure 66-72 mm Hg	2.82[d]	1.43, 5.58	$P = 0.003$
Di Leo et al[48]	Chronic diarrhoea	261	37 ± 15 (18-83)	Italy	L/M	0.030%	Underlying organic disease	1.56[c]	1.32, 1.85	$P < 0.0001$
Dietary factors										
Kim et al[47]	Moderate-to-severe fatty liver	34	44.7 ± 9.2 (30-60)	South Korea	Zonulin	6-21 ng/mL	<14 standard drinks per week	1.91[c]	1.01, 3.95	$P = 0.05$
							>15 standard drinks per week/>5 standard drinks in one setting	1.56[c]	1.02, 2.67	$P = 0.05$
Anthropometric measurements										
Ohlsson et al[23]	General population	363	43 (28-53)	Sweden	Zonulin	54-64 ng/mL	Overweight, BMI > 25	2.36[e]	1.07, 5.21[b]	$P = 0.033^b$
Ohlsson et al[23]	General population	363	43 (28-53)	Sweden	Zonulin	>64 ng/mL	Overweight, BMI > 25	4.10[e]	1.87, 8.97[b]	$P < 0.001^b$
							Obesity, BMI > 30	4.90[e]	1.49, 31.65[b]	$P = 0.047^b$
Ohlsson et al[23]	General population	363	43 (28-53)	Sweden	Zonulin	34 ± 14 ng/mL	Waist circumference > 97cm	7.03[d]	1.97, 25.11	$P = 0.003$
Biomarkers										
Ohlsson et al[23]	General population	363	43 (28-53)	Sweden	Zonulin	34 ± 14 ng/mL	Plasma glucose levels (mmol/L) >5.7	2.09[d]	1.05, 4.18	$P = 0.036$
Demographic factors										
Carielo et al[45]	Chronic liver disease	83	54.1 (28-78)	Italy	L/M	0.030%	Age > 50 y	1.9[a]	1.1, 2.3	$P < 0.001$

BMI, body mass index; CI, confidence interval; L/M, lactulose/mannitol.

[a]Adjusted for age, alcohol use, associated diabetes, and degree of portal hypertension.

[b]Adjusted for HbA1c, LDL, HDL and BMI.

[c]Adjusted for HbA1c, LDL, HDL, BMI, age, smoking status, alcohol consumption, physical activity and the use of diabetes, and dyslipidaemia medication.

[d]Adjusted for weight, BMI, waist and hip circumference, systolic and diastolic blood pressure, and fasting glucose levels.

[e]Adjusted for systolic and diastolic blood pressure, and fasting glucose levels.

[f]Adjusted for BMI, previous gestational diabetes, original intervention group.

[g]Adjusted for age and gender.

[h]Adjusted for age, gender, BMI and waist to hip ratio.

[i]Adjusted for age, gender, BMI, waist to hip ratio, LDL, HDL, triglycerides and total cholesterol.

[j]Adjusted for age.

TABLE 4 Risk factors associated with intestinal permeability according to beta correlation coefficient

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	β	95% CI	*P* value
Medical history and disease									
Mujagic et al[49]	IBS	91	44.4 ± 1.6 (18-75)	Netherlands	L/R	IBS-D	0.63[f]	0.09, 1.16	$P = 0.022$
Wyatt et al[33]	Crohn's disease	72	37 (>18)	Austria	L/M	Crohn's disease relapse	3.54[n]	–	$P < 0.0001$
Lassenius et al[50]	Type 1 diabetes	911	46 (36-56)	Finland	LPS	Age of type 1 diabetes onset	−0.14[q]	–	$P < 0.001$
Lassenius et al[50]	Type 1 diabetes	911	46 (36-56)	Finland	LPS	Diastolic blood pressure	0.10[q]	–	$P = 0.004$
Dietary factors									
Amar et al[31]	General population	201	53.9 ± 6.1 (45-64)	France	LPS	Total energy intake	132.2[o] 121.8[o]	62.7 (SE) 57.7 (SE)	$P = 0.04$ $P = 0.04$
Mokkala et al[52]	Pregnancy (BMI 30)	100	29.4 ± 4.9 (18-45)	Finland	Zonulin	Dietary protein	−0.139[d]	−0.247, 0.031	$P = 0.01$
Zak-Golab et al[51]	Various BMI	80	48 (32-63)	Poland	Zonulin	Fat percentage in diet	0.23[j]	±0.11	$P < 0.05$
Anthropometric factors									
Zak-Golab et al[51]	Various BMI	80	48 (32-63)	Poland	Zonulin	BMI	0.26[h]	±0.10	$P < 0.05$
Donnadieu-Rigole et al. 2018[54]	Alcohol use disorder	41	48.2 ± 8.7 (>18)	France	Zonulin	BMI	1.507[m]	0.34 (SEM)	$P < 0.01$
Biomarkers									
Kvehaugen et al. 2017[72]	Obesity (BMI 35-55)	140	43.1 (>18)	Norway	Zonulin	CRP	3.28[c]	1.10, 5.46	$P < 0.01$
Mokkala et al. 2017[41]	Pregnancy (BMI 30)	100	29.4 ± 4.9 (18-45)	Finland	Zonulin	hsCRP	0.013[b]	0.003, 0.023	$P = 0.015$
Barcelo et al. 2016[53]	Obstructive sleep apnoea	38	50 ± 12 (18-74)	Spain	Zonulin	hsCRP	0.075[r]	0.008, 0.158	$P = 0.046$
Moreno-Navarrete et al. 2012[56]	Various glucose tolerance	123	52 ± 11.7	Spain	Zonulin	Circulating IL-6	0.23[j]	–	$P = 0.04$
Ficek et al. 2017[74]	Haemodialysis patients	150	62 (59-64)	Poland	LPS	Circulating IL-6	0.171[s]	–	$P = 0.04$
Mokkala et al[41]	Pregnancy (BMI 30)	100	29.4 ± 4.9 (18-45)	Finland	Zonulin	GlycA	0.004[b]	0.002, 0.006	$P < 0.001$

(Continues)

TABLE 4 (Continued)

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	ρ	95% CI	P value
Mokkala et al[41]	Pregnancy (BMI 30)	100	29.4 ± 4.9 (18–45)	Finland	Zonulin	Insulin	0.015[b]	0.007, 0.022	$P < 0.001$
Mokkala et al[41]	Pregnancy (BMI 30)	100	29.4 ± 4.9 (18–45)	Finland	Zonulin	Insulin resistance (HOMA)	0.015[b]	0.007, 0.022	$P < 0.001$
Zhang et al[44]	Type 2 diabetes	102	47 ± 12	China	Zonulin	Insulin resistance (HOMA)	0.024[e]	0.009 (SE)	$P = 0.005$
Mokkala et al[41]	Pregnancy (BMI 30)	100	29.4 ± 4.9 (18–45)	Finland	Zonulin	Insulin sensitivity (QUICKI)	−0.002[b]	−0.003, −0.001	$P < 0.001$
Moreno-Navarrete et al[56]	Various glucose tolerance	123	52 ± 11.7	Spain	Zonulin	Insulin sensitivity	−0.263[b]	–	$P = 0.004$
Zak-Golab et al[51]	Various BMI	80	48 (32-63)	Poland	Zonulin	Glucose	0.38[j]	±0.12	$P < 0.05$
Mokkala et al[41]	Pregnancy (BMI 30)	100	29.4 ± 4.9 (18–45)	Finland	Zonulin	Triglycerides	0.009[b]	0.003, 0.015	$P = 0.003$
Lassenius et al[50]	Type 1 diabetes	911	46 (36-56)	Finland	LPS	Triglycerides	0.69[a]	–	$P < 0.001$
Mokkala et al[41]	Pregnancy (BMI 30)	100	29.4 ± 4.9 (18–45)	Finland	Zonulin	Total cholesterol	0.004[b]	0.000, 0.007	$P = 0.032$
Barcelo et al[53]	Obstructive sleep apnoea	38	50 ± 12 (18-74)	Spain	Zonulin	Alanine transaminase	0.014[f]	0.001, 0.028	$P = 0.04$
Barcelo et al[53]	Obstructive sleep apnoea	38	50 ± 12 (18-74)	Spain	Zonulin	Aspartate transaminase	0.02[f]	0.002, 0.037	$P = 0.04$
Cangemi et al[49]	Pneumonia	278	70 ± 16	Italy	LPS	sP-selection	0.415[g]	–	$P < 0.001$
Raparelli et al[48]	Liver cirrhosis	69	62.6 ± 13.5	Italy	LPS	sCD40L	0.43[a]	–	$P < 0.0001$
Mokkala et al[41]	Pregnancy (BMI 30)	100	29.4 ± 4.9 (18–45)	Finland	LPS	Zonulin	0.002[b]	0.001, 0.003	$P = 0.002$
Małyszko et al[45]	Kidney transplant recipients	72	45.5 ± 12.2	Poland	Zonulin	Total serum protein	−0.51[a]	–	$P = 0.014$
Małyszko et al[45]	Kidney transplant recipients	72	45.5 ± 12.2	Poland	Zonulin	Thyroglobulin-binding protein	0.47[e]	–	$P = 0.03$
Zak-Golab et al[51]	Various BMI	80	48 (32-63)	Poland	Zonulin	Microbiota bacteria count	0.33[h]	±0.13	$P < 0.05$

(Continues)

TABLE 4 (Continued)

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	β	95% CI	P value
Lassenius et al[10]	Type 1 diabetes	911	46 (36-56)	Finland	LPS	uMCP1/ creatinine ratio	0.10[a]	—	$P = 0.003$
Demographic factors									
Zak-Golab et al[11]	Various BMI	80	48 (32-63)	Poland	Zonulin	Age	0.31[b]	±0.06	$P < 0.05$

BMI, body mass index; CI, confidence interval; GlycA, glycoprotein acetylation; hsCRP, high-sensitivity C-reactive protein; HOMA-IR, homeostatic model assessment; IBS-D, diarrhoea predominant irritable bowel syndrome; IBS, irritable bowel syndrome; IL-6, interleukin 6; L/M, lactulose/mannitol; LPS, lipopolysaccharides; L/R, lactulose/rhamnose; QUICKI, quantitative insulin-sensitivity check index; SE, standard error; sP-selectin, plasma soluble P-selectin; SEM, structural equation modelling; sCD40L, soluble cluster of differentiation 40 ligand; uMCP1, urinary monocyte chemoattractant protein-1.

[a]Multivariate analysis adjusted for sex, age, Child Pugh score and LPS.

[b]Multiple linear regression analysis adjusted for log-transformed BMI and gestational weeks.

[c]Linear regression adjusting for age, sex and BMI.

[d]Multiple linear regression analysis model including protein and polyunsaturated fatty acids.

[e]Multiple stepwise linear regression analysis adjusted for age, BMI, waist to hip ratio, triglycerides, total cholesterol, HbA1c, HDL, LDL, IL-6, TNF-α, uric acid and zonulin.

[f]Linear regression analysis model including demographical factors, medication, psychological symptoms and lifestyle.

[g]Multiple regression analysis model including systolic and diastolic blood pressure, haemoglobin, erythrocyte count, fasting glucose, thyroglobulin-binding protein, total protein and treatment with angiotensin-converting enzyme inhibitors.

[h]Multiple regression analysis model including age, BMI, total bacterial, Bacteroides and Firmicutes counts.

[i]Multiple regression analysis model including energy intake and macronutrients content.

[j]Multiple regression analysis model including parameters of carbohydrate and lipid metabolism.

[k]Multiple regression analysis model including age, BMI, log insulin sensitivity and log fasting triglycerides.

[l]Multiple regression analysis model including age, BMI, log insulin sensitivity, log fasting triglycerides and IL-6.

[m]Multiple regression analysis model including factors known to influence bacteria translocation and associated factors.

[n]Multivariate analysis adjusted for age, physical activity, BMI and residuals from linear regression of energy on protein, carbohydrates and alcohol in subjects with LPS > 39 U/L.

[o]Multivariate analysis adjusted for age, physical activity, BMI and residuals from linear regression of energy on protein carbohydrates and alcohol in subjects with LPS > 39 U/L excluding energy from alcohol.

[p]Multivariable regression analysis adjusting for clinical characteristics.

[q]Multivariate linear regression analysis adjusted for clinical variables.

[r]Multiple regression analysis adjusted for age, sex, BMI and metabolic syndrome components.

[s]Multivariable regression analysis adjusting for zonulin, LPS and D-lactates.

TABLE 5 Risk factors associated with intestinal permeability according to correlation coefficient

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	Correlation	*P* value
Medical history and disease								
Caserta et al[36]	Systemic sclerosis	32	45.7 ± 10.9	Italy	C/M	Disease duration	$r = 0.73$	$P < 0.011$
Caviglia et al[37]	Inflammatory bowel disease	118	49 (18-77)	Italy	Zonulin	Disease duration	$\rho = -0.30$	$P = 0.001$
Schwiertz et al[38]	Parkinson's disease	36	65.5 (44-78)	Germany	Stool zonulin	Disease duration	$r = -0.34$	$P = 0.042$
Hilsden et al[39]	Crohn's disease	61	36 (18-66)	Canada	L/M	Crohn's disease relapse	$r = 0.48$	$P = 0.008$
Goebel et al[40]	Fibromyalgia	40	48 ± 11 (18-65)	Germany	L/M	Pain (NRS)	$r = -0.3$	$P < 0.05$
Mahyszko et al[41]	Kidney transplant recipients	72	45.5 ± 12.2	Poland	Zonulin	Systolic blood pressure	$r = -0.33$	$P < 0.05$
Troseid et al[42]	Obesity (BMI 45)	49	42.9 ± 9.2 (28-55)	Norway	LPS	Systolic blood pressure	$r = 0.40$	$P = 0.009$
Rutten et al[43]	COPD	18	63.6 ± 1.3	Netherlands	L/R	COPD	$r = 0.67$	$P < 0.01$
Riordan et al[34]	SIBO	34	64 (22-95)	Australia	L/M	SIBO	$r = 0.61$	$P < 0.0005$
Zhang et al[10]	PCOS	78	29 ± 5	China	Zonulin	Number of menstrual cycles	$\rho = -0.401^a$	$P < 0.001$
Schwiertz et al[38]	Parkinson's disease	36	65.5 (44-78)	Germany	Zonulin	Levodopa dose	$r = -0.39$	$P = 0.019$
Volynets et al[25]	NAFLD	20	41.9 ± 2.3	Germany	LPS	Degree of liver damage	$r = 0.69$	$P = 0.01$
Dietary factors								
Volynets et al[25]	NAFLD	20	41.9 ± 2.3	Germany	LPS	Protein intake	$\rho = 0.59$	$P = 0.001$
Volynets et al[25]	NAFLD	20	41.9 ± 2.3	Germany	LPS	Animal-derived protein intake	$\rho = 0.54$	$P = 0.002$
Mokkala et al[52]	Pregnancy (BMI 30)	95	29.4 ± 4.9 (18-45)	Finland	Zonulin	Potassium intake	$\rho = -0.343$	$P = 0.001$
Anthropometric measurements								
Barceló et al[53]	Overweight (BMI 29)	38	49 ± 12 (18-73)	Spain	Zonulin	Waist circumference	$\rho = 0.382$	$P = 0.04$
Barceló et al[53]	Obstructive sleep apnoea	38	50 ± 12 (18-74)	Spain	Zonulin	Waist circumference	$\rho = 0.442$	$P = 0.004$
Zhang et al[10]	PCOS	78	29 ± 5	China	Zonulin	Waist-to-hip ratio	$r = 0.401$	$P = 0.015$
Teixeira et al[54]	Various BMI	40	28.5 ± 7.6/30.7 ± 6.5	Brazil	L/M	Abdomen circumference	$\rho = 0.30$	$P = 0.05$
Hendy et al[55]	NAFLD	56	37.2 ± 6.8 (29-46)	Egypt	Zonulin	BMI	$r = 0.378$	$P < 0.05$
Zak-Golab et al[51]	Various BMI	80	48 (32-63)	Poland	Zonulin	BMI	$\rho = 0.41$	$P < 0.001$
Moreno-Navarrete et al[56]	Glucose intolerance	41	55.9 ± 10.3	Spain	Zonulin	BMI	$r = 0.42$	$P = 0.007$
Zhang et al[10]	PCOS	78	29 ± 5	China	Zonulin	BMI	$r = 0.535$	$P < 0.05$

(Continues)

TABLE 5 (Continued)

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	Correlation	P value
Troseid et al[42]	Obesity (BMI 45)	49	42.9 ± 9.2 (28-55)	Norway	LPS	BMI	$r = 0.37$	$P = 0.017$
Zak-Golab et al[51]	Various BMI	80	48 (32-63)	Poland	Zonulin	Weight	$\rho = 0.34$	$P < 0.01$
Zak-Golab et al[51]	Various BMI	80	48 (32-63)	Poland	Zonulin	Fat mass	$\rho = 0.42$	$P < 0.001$
Zak-Golab et al[51]	Various BMI	80	48 (32-63)	Poland	Zonulin	Fat percentage	$\rho = 0.40$	$P < 0.001$
Troseid et al[42]	Obesity (BMI 45)	49	42.9 ± 9.2 (28-55)	Norway	LPS	Subcutaneous fat	$r = 0.33$	$P = 0.038$
Zhang et al[10]	PCOS	78	29 ± 5	China	Zonulin	Visceral adiposity index	$r = 0.432$	$P = 0.011$
Troseid et al[42]	Obesity (BMI 45)	49	42.9 ± 9.2 (28-55)	Norway	LPS	Intra-abdominal fat	$r = 0.61$	$P < 0.001$
Qi et al[57]	General population	37	23.1 ± 3.9/76.7 ± 5.2 (18->70)	America	Zonulin	Muscle strength	$r = -0.332$	$P = 0.048$
Qi et al[57]	General population	37	23.1 ± 3.9/76.7 ± 5.2 (18->70)	America	Zonulin	Steps per day	$r = -0.410$	$P = 0.016$
Biomarkers								
Glucose metabolism								
Barcelo et al[53]	Obstructive sleep apnoea	38	50 ± 12 (18-74)	Spain	Zonulin	Fasting glucose	$\rho = 0.321$	$P = 0.04$
Barcelo et al[53]	Overweight (BMI 29)	38	49 ± 12 (18-73)	Spain	Zonulin	Fasting glucose	$\rho = 0.343$	$P = 0.035$
Zhang et al[44]	Various glucose tolerance	290	40 ± 19	China	Zonulin	Fasting glucose	$r = 0.300^{b}$	$P = 0.001$
Zhang et al[10]	PCOS	78	29 ± 5	China	Zonulin	Glucose tolerance at 0 min	$r = 0.351^{b}$	$P = 0.045$
Zhang et al[10]	PCOS	78	29 ± 5	China	Zonulin	Glucose tolerance at 120 min	$r = 0.347^{b}$	$P = 0.045$
Zhang et al[44]	Glucose intolerance	92	39 ± 13	China	Zonulin	Glucose tolerance at 120 min	$r = 0.325^{b}$	$P < 0.05$
Zhang et al[44]	Type 2 diabetes	102	47 ± 12	China	Zonulin	Glucose tolerance at 120 min	$r = 0.342^{b}$	$P < 0.05$
Jayashree et al[60]	Type 2 diabetes	45	51 ± 6 (30-60)	India	LPS	Glucose tolerance at 120 min	$r = 0.341$	$P < 0.001$
Jayashree et al[60]	Type 2 diabetes	45	51 ± 6 (30-60)	India	LPS	HbA1c	$r = 0.334$	$P < 0.001$
Troseid et al[42]	Obesity (BMI 45)	49	42.9 ± 9.2 (28-55)	Norway	LPS	HbA1c	$r = 0.56$	$P = 0.001$
Zhang et al[44]	Various glucose tolerance	290	40 ± 19	China	Zonulin	HbA1c	$r = 0.302^{b}$	$P = 0.002$
Barcelo et al[53]	Obstructive sleep apnoea	38	50 ± 12 (18-74)	Spain	Zonulin	Fasting insulin	$\rho = 0.351$	$P = 0.03$

(Continues)

TABLE 5 (Continued)

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	Correlation	P value
Barcelo et al[53]	Overweight (BMI 29)	38	49 ± 12 (18-73)	Spain	Zonulin	Fasting insulin	$\rho = 0.328$	$P = 0.041$
Hendy et al[55]	NAFLD	56	37.2 ± 6.8 (29-46)	Egypt	Zonulin	Fasting insulin	$r = 0.305$	$P < 0.05$
Zhang et al[44]	Type 2 diabetes	102	47 ± 12	China	Zonulin	Fasting insulin	$r = 0.325^b$	$P = 0.005$
Mokkala et al[61]	Pregnancy (BMI 30)	75	29.4 ± 4.9 (18-45)	Finland	Zonulin	Fasting insulin	$\rho = 0.616^f$	$P < 0.001$
Zhang et al[10]	PCOS	78	29 ± 5	China	Zonulin	Insulin sensitivity (OGTT at 0 min)	$r = 0.605$	$P < 0.05$
Zhang et al[10]	PCOS	78	29 ± 5	China	Zonulin	Insulin sensitivity (OGTT at 120 min)	$r = 0.527$	$P = 0.001$
Moreno-Navarrete et al[56]	Glucose intolerance	41	55.9 ± 10.3	Spain	Zonulin	Insulin sensitivity	$r = -0.36$	$P = 0.02$
Mokkala et al[61]	Pregnancy (BMI 30)	75	29.4 ± 4.9 (18-45)	Finland	Zonulin	Insulin sensitivity (QUICKI)	$\rho = -0.600^c$	$P < 0.001$
Zhang et al[44]	Glucose intolerance	92	39 ± 13	China	Zonulin	Insulin sensitivity (QUICKI)	$r = -0.311^b$	$P = 0.001$
Teixeira et al[58]	Various BMI	40	28.5 ± 7.6/30.7 ± 6.5	Brazil	L/M	Insulin resistance (HOMA)	$\rho = 0.3$	$P = 0.014$
Hendy et al[55]	NAFLD	56	37.2 ± 6.8 (29-46)	Egypt	Zonulin	Insulin resistance (HOMA)	$r = 0.413$	$P < 0.01$
Mokkala et al[61]	Pregnancy (BMI 30)	75	29.4 ± 4.9 (18-45)	Finland	Zonulin	Insulin resistance (HOMA)	$\rho = 0.616^c$	$P < 0.001$
Zhang et al[10]	PCOS	78	29 ± 5	China	Zonulin	Insulin resistance (HOMA)	$r = 0.315^b$	$P = 0.044$
Zhang et al[44]	Type 2 diabetes	102	47 ± 12	China	Zonulin	Insulin resistance (HOMA)	$r = 0.434^b$	$P < 0.05$
Cholesterol and triglycerides								
Barcelo et al[53]	Obstructive sleep apnoea	38	50 ± 12 (18-74)	Spain	Zonulin	Total cholesterol	$\rho = 0.397$	$P = 0.011$
Mokkala et al[61]	Pregnancy (BMI 30)	75	29.4 ± 4.9 (18-45)	Finland	Zonulin	Total cholesterol	$\rho = 0.566^c$	$P < 0.001$
Zhang et al[44]	Various glucose tolerance	290	40 ± 19	China	Zonulin	Total cholesterol	$r = 0.333^b$	$P < 0.05$
Nymark et al[62]	Type 1 diabetes	477	36.5 ± 11	Finland	LPS	Total cholesterol	$r = 0.34$	$P < 0.001$
Mokkala et al[61]	Pregnancy (BMI 30)	75	29.4 ± 4.9 (18-45)	Finland	LPS	Total cholesterol	$\rho = 0.374^d$	$P = 0.001$
Mokkala et al[61]	Pregnancy (BMI 30)	75	29.4 ± 4.9 (18-45)	Finland	Zonulin	LDL cholesterol	$\rho = 0.458^c$	$P = 0.001$
Zhang et al[44]	Type 2 diabetes	102	47 ± 12	China	Zonulin	LDL cholesterol	$r = 0.362^b$	$P = 0.002$

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TABLE 5 (Continued)

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	Correlation	*P* value
Zhang et al[44]	Various glucose tolerance	290	40 ± 19	China	Zonulin	HDL cholesterol	$r = -0.342^{h}$	$P < 0.05$
Zhang et al[10]	PCOS	78	29 ± 5	China	Zonulin	HDL cholesterol	$r = -0.412$	$P = 0.031$
Zhang et al[44]	General population	95	34 ± 11	China	Zonulin	HDL cholesterol	$r = -0.390^{h}$	$P < 0.05$
Hendy et al[55]	NAFLD	56	37.2 ± 6.8 (29-46)	Egypt	Zonulin	HDL cholesterol	$r = -0.397$	$P < 0.01$
Troseid et al[42]	Obesity (BMI 45)	49	42.9 ± 9.2 (28-55)	Norway	LPS	HDL cholesterol	$r = -0.43$	$P = 0.006$
Jayashree et al[40]	Type 2 diabetes	45	51 ± 6 (30-60)	India	LPS	HDL cholesterol	$r = 0.531$	$P < 0.001$
Teixeira et al[58]	Various BMI	40	28.5 ± 7.6/30.7 ± 6.5	Brazil	L/M	HDL cholesterol	$\rho = -0.39$	$P = 0.01$
Mokkala et al[41]	Pregnancy (BMI 30)	75	29.4 ± 4.9 (18-45)	Finland	Zonulin	Triglycerides	$\rho = 0.529^{g}$	$P < 0.001$
Zhang et al[10]	PCOS	78	29 ± 5	China	Zonulin	Triglycerides	$r = 0.422$	$P = 0.031$
Zhang et al[44]	Various glucose tolerance	290	40 ± 19	China	Zonulin	Triglycerides	$r = 0.449^{h}$	$P < 0.05$
Zhang et al[44]	General population	95	34 ± 11	China	Zonulin	Triglycerides	$r = 0.329^{h}$	$P = 0.002$
Zhang et al[44]	Glucose intolerance	92	39 ± 13	China	Zonulin	Triglycerides	$r = 0.501^{h}$	$P < 0.05$
Lassenius et al[50]	Type 1 diabetes	904	46 (36-56)	Finland	LPS	Triglycerides	$r = 0.73$	$P < 0.001$
Troseid et al[42]	Obesity (BMI 45)	49	42.9 ± 9.2 (28-55)	Norway	LPS	Triglycerides	$r = 0.52$	$P = 0.001$
Nymark et al[62]	Type 1 diabetes	477	36.5 ± 11	Finland	LPS	Triglycerides	$r = 0.61$	$P < 0.001$
Jayashree et al[40]	Type 2 diabetes	45	51 ± 6 (30-60)	India	LPS	Triglycerides	$r = 0.353$	$P < 0.001$
Nymark et al[62]	Type 1 diabetes	477	36.5 ± 11	Finland	LPS	ApoB	$r = 0.34$	$P < 0.001$
Inflammatory markers								
Barceló et al[63]	Obstructive sleep apnoea	38	50 ± 12 (18-74)	Spain	Zonulin	hsCRP	$\rho = 0.372$	$P = 0.02$
Karthikeyan et al[63]	Liver cirrhosis	30	47.7 ± 1.4 (18-60)	India	Zonulin	hsCRP	$\rho = 0.482$	$P = 0.0063$
Mokkala et al[41]	Pregnancy (BMI 30)	75	29.4 ± 4.9 (18-45)	Finland	Zonulin	hsCRP	$\rho = 0.412^{g}$	$P = 0.004$
Moreno-Navarrete et al[56]	General population	82	48.3 ± 11.7	Spain	Zonulin	Circulating IL-6	$r = 0.31$	$P = 0.01$
Morkl et al[59]	Women (BMI 13-46)	102	24.6 ± 4.6	Austria	Zonulin	Circulating IL-6	$\rho = 0.317$	$P = 0.001$
Qi et al[57]	General population	37	23.1 ± 3.9/ 76.7 ± 5.2 (18->70)	America	Zonulin	Circulating IL-6	$r = 0.345$	$P = 0.043$
Jayashree et al[40]	Type 2 diabetes	45	51 ± 6 (30-60)	India	LPS	Circulating IL-6	$r = 0.542$	$P < 0.001$
Qi et al[57]	General population	37	23.1 ± 3.9/ 76.7 ± 5.2 (18->70)	America	Zonulin	TNF-α	$r = 0.357$	$P = 0.032$

(Continues)

TABLE 5 (Continued)

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	Correlation	P value
Zhang et al[44]	General population	95	34 ± 11	China	Zonulin	TNF-α	$r = 0.623^{a}$	$P < 0.05$
Zhang et al[44]	Glucose intolerance	92	39 ± 13	China	Zonulin	TNF-α	$r = 0.647^{a}$	$P < 0.05$
Zhang et al[44]	Type 2 diabetes	102	47 ± 12	China	Zonulin	TNF-α	$r = 0.352^{a}$	$P = 0.001$
Jayashree et al[40]	Type 2 diabetes	45	51 ± 6 (30-60)	India	LPS	TNF-α	$r = 0.407$	$P < 0.001$
Zak-Golab et al[21]	Various BMI	80	48 (32-63)	Poland	Zonulin	sTNFR1	$\rho = 0.34$	$P < 0.001$
Mokkala et al[41]	Pregnancy (BMI 30)	75	29.4 ± 4.9 (18-45)	Finland	LPS	GlycA	$\rho = 0.387^{d}$	$P = 0.001$
Mokkala et al[41]	Pregnancy (BMI 30)	75	29.4 ± 4.9 (18-45)	Finland	Zonulin	GlycA	$\rho = 0.616^{c}$	$P < 0.001$
Malickova et al[45]	Inflammatory bowel disease	40	(18-65)	Czech Republic	Stool zonulin	Stool calprotectin	$\rho = 0.430$	$P = 0.006$
Markers of intestinal permeability								
Ciccia et al[46]	Ankylosing spondylitis	20	47 (23-58)	Italy	L/M	Serum zonulin	$\rho = 0.851$	$P = 0.0177$
Duerksen et al[22]	Coeliac disease (Marsh type 3)	6	53.5	Canada	Zonulin	L/M	$\rho = 0.891$	$P = 0.05$
Carnevale et al[47]	Glucose intolerance	70	62.5 ± 13.2	Italy	Zonulin	LPS	$r = 0.529$	$P = 0.001$
Raparelli et al[48]	Liver cirrhosis (Child-Pugh B + C)	34	62.5 ± 13.4	Italy	Zonulin	LPS	$\rho = 0.48$	$P < 0.05$
Mokkala et al[41]	Pregnancy (BMI 30)	75	29.4 ± 4.9	Finland	Zonulin	LPS	$\rho = 0.458^{c}$	$P = 0.001$
Cangemi et al[49]	Pneumonia	278	70 ± 16	Italy	Zonulin	LPS	$\rho = 0.545$	$P < 0.001$
Goebel et al[40]	Fibromyalgia	40	48 ± 11 (18-65)	Germany	L/M	Gastroduodenal permeability	$r = 0.68$	$P < 0.0001$
Goebel et al[40]	Complex regional pain syndrome	17	43 ± 13 (18-65)	Germany	L/M	Gastroduodenal permeability	$r = 0.88$	$P < 0.0001$
Liver pathology								
Barcelo et al[53]	Obstructive sleep apnoea	38	50 ± 12 (18-74)	Spain	Zonulin	Alanine transaminase	$\rho = 0.484$	$P = 0.002$
Hendy et al[55]	NAFLD	56	37.2 ± 6.8 (29-46)	Egypt	Zonulin	Alanine transaminase	$r = 0.312$	$P < 0.05$
Volynets et al[25]	NAFLD	20	41.9 ± 2.3	Germany	LPS	Alanine transaminase	$\rho = 0.50$	$P = 0.005$
Barcelo et al[53]	Obstructive sleep apnoea	38	50 ± 12 (18-74)	Spain	Zonulin	Aspartate transaminase	$\rho = 0.426$	$P = 0.006$
Barcelo et al[53]	Obstructive sleep apnoea	38	50 ± 12 (18-74)	Spain	Zonulin	Gamma glutamyltransferase	$\rho = 0.444$	$P = 0.004$
Hendy et al[55]	NAFLD	56	37.2 ± 6.8 (29-46)	Egypt	Zonulin	Liver histopathology	$r = 0.518$	$P < 0.001$

(Continues)

TABLE 5 (Continued)

Author	Study population	Sample size	Age (range)	Country	Test	Risk factor	Correlation	*P* value
Intestinal microbiome markers								
Lindheim et al[70]	PCOS	24	27	Austria	Zonulin	Microbial diversity	$r = -0.334$	$P = 0.029$
Lindheim et al[70]	PCOS	24	27	Austria	Stool zonulin	Microbial diversity	$\rho = -0.366$	$P = 0.016$
Other biomarkers								
Barcelo et al[53]	Obstructive sleep apnoea	38	50 ± 12 (18-74)	Spain	Zonulin	Mean oxygen saturation	$\rho = -0.378$	$P = 0.019$
Lukaszyk et al[35]	Chronic kidney disease	35	73.9 ± 10.9	Poland	Zonulin	Hepcidin with hsCRP > 10mg/dL	$r = -0.37$	$P < 0.05$
Malyszko et al[41]	Kidney transplant recipients	72	45.5 ± 12.2	Poland	Zonulin	Haemoglobin	$r = 0.32$	$P < 0.01$
Malyszko et al[41]	Kidney transplant recipients	72	45.5 ± 12.2	Poland	Zonulin	Total protein	$r = -0.33$	$P < 0.05$
Raparelli et al[48]	Liver cirrhosis	69	62.6 ± 13.5	Italy	LPS	Platelet activation	$\rho = 0.55$	$P < 0.001$
Rutten et al[43]	COPD	18	63.6 ± 1.3	Netherlands	L/R	Plasma lactic acid	$r = 0.66$	$P = 0.01$
Swanson et al[71]	Alcohol use disorder	20	45.9 ± 12.2	America	L/M	Plasma melatonin	$r = -0.39$	$P = 0.03$
Volynets et al[25]	NAFLD	20	41.9 ± 2.3	Germany	LPS	Plasminogen activator inhibitor-1	$\rho = 0.46$	$P = 0.01$
Raparelli et al[48]	Liver cirrhosis	69	62.6 ± 13.5	Italy	LPS	sP-Selectin	$\rho = 0.32$	$P = 0.008$
Cangemi et al[69]	Pneumonia	278	70 ± 16 (>18)	Italy	LPS	sP-selectin	$\rho = 0.362$	$P < 0.001$
Cangemi et al[69]	Pneumonia	278	70 ± 16 (>18)	Italy	LPS	sNOX2-dp	$\rho = 0.455$	$P < 0.001$
Demographic factors								
Johnston et al[72]	Coeliac disease	77	35.0 (25-64)	Ireland	L/M	Age	$r = 0.34$	$P = 0.001$
Zak-Golab et al[51]	Various BMI	80	48 (32-63)	Poland	Zonulin	Age	$\rho = 0.43$	$P < 0.001$

Abbreviation: ApoB, apolipoprotein; C/M, cellobiose/mannitol; COPD, chronic obstructive pulmonary disease; GlycA, glycoprotein acetylation; HbA1c, glycated haemoglobin A1c; HDL, high-density lipoprotein; hsCRP, high-sensitivity C-reactive protein; HOMA, homeostatic model assessment; IBS-D, diarrhoea predominant irritable bowel syndrome; IBS, irritable bowel syndrome; IL-6, interleukin 6; LDL, low-density lipoprotein; L/M, lactulose/mannitol; LPS, lipopolysaccharides; L/R, lactulose/rhamnose; n-6, omega-6 fatty acid; NAFLD, non-alcoholic fatty liver disease; NRS, numeric rating scale; OGTT, oral glucose tolerance test; ρ, Spearman's correlation coefficient; PCOS, polycystic ovarian syndrome; PUFA, polyunsaturated fatty acids; QUICKI, quantitative insulin-sensitivity check index; r, Pearson's correlation coefficient for the sample; sNOX2-dp, soluble NOX2-derived peptide; sP-selectin, plasma soluble P-selectin; SIBO, small intestine bacteria overgrowth; sTNFR1, soluble tumour necrosis factor receptor-1; TNF-α, tumour necrosis factor alpha.

[a]Adjusted for BMI.

[b]Adjusted for BMI and age.

[c]Multiple linear regression adjusted for log-transformed BMI and gestational weeks in subjects with hsCRP > 3mg/L.

[d]Only including subjects with hsCRP > 3mg/L.

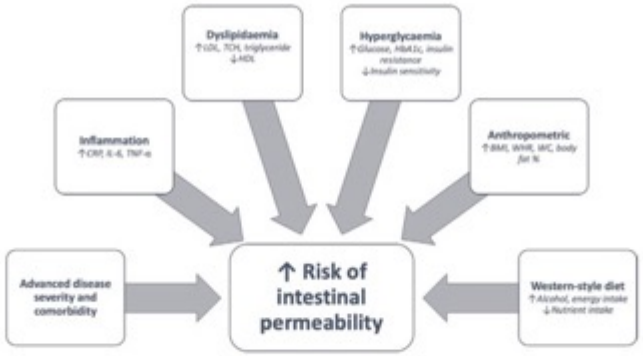

FIGURE 2 The strongest identified risk factors identified to be associated with intestinal permeability include advanced disease severity, comorbidity, inflammation, dyslipidaemia, hyperglycaemia, anthropometric measurements that resemble obesity, and Western-style dietary habits

DISCLOSURES

The authors declare that they have nothing to disclose.

ORCID

Bradley Leech https://orcid.org/0000-0003-4866-9638
Erica McIntyre https://orcid.org/0000-0002-3970-9979
Amie Steel https://orcid.org/0000-0001-6643-9444
David Sibbritt https://orcid.org/0000-0003-3561-9447

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APPENDIX 3.1: LEAKY GUT SURVEY

Page exit logic: Skip / Disqualify Logic
IF: #3 Question "How old are you?" is less than "18" **THEN:** Disqualify and display:
Sorry, you do not qualify to take this survey. You must be over the age of 18, living in Australia and think you have leaky gut.

Page exit logic: Skip / Disqualify Logic
IF: #2 Question "Do you believe you have leaky gut?" is one of the following answers ("No")
THEN: Disqualify and display:
Sorry, you do not qualify to take this survey. You must be over the age of 18, living in Australia and think you have leaky gut.

ID 279

Leaky gut involves the loss of integrity between the cells of the small intestine. Leaky gut is also known as 'intestinal permeability', 'leaky gut syndrome', 'intestinal hyperpermeability' and 'increased intestinal permeability'. Throughout this survey, the term leaky gut will be used.

LOGIC Show/hide trigger exists.
ID 2
1. Have you been diagnosed with leaky gut? *

- Yes
- No
- I'm not sure

LOGIC Hidden unless: #1 Question "Have you been diagnosed with leaky gut?" is one of the following answers ("No","I'm not sure")
ID 4
2. Do you believe you have leaky gut? *

- Yes
- No
- I'm not sure

VALIDATION %s format expected Using custom RegEx pattern

ID 300

3. How old are you? *

ID 7

4. Are you an Australian resident? *

- Yes
- No

Demographics

ID 8

5. What is your gender?

- Male
- Female
- I would rather not say
- Other

VALIDATION %s format expected Using custom RegEx pattern

ID 209

6. What is your height in centimetres?

cm

VALIDATION %s format expected Using custom RegEx pattern

ID 210

7. How much do you weigh in kilograms?

kg

ID 9

8. In which country were you born?

- Australia
- England
- New Zealand
- India
- Italy
- Vietnam
- Philippines
- Other

ID 383

9. Are you of Aboriginal and/or Torres Strait Islander origin?

- Yes
- No

ID 301

10. What state or territory do you live in?

- Australian Capital Territory
- New South Wales
- Northern Territory
- Queensland
- South Australia
- Tasmania
- Victoria
- Western Australia

ID 11

11. Where best describes your living location?

- Urban
- Rural
- Remote

Leaky Gut Diagnosis

LOGIC Show/hide trigger exists.

ID 15

12. Is leaky gut your primary health concern?

- Yes
- No

LOGIC Hidden unless: #12 Question "Is leaky gut your primary health concern?" is one of the following answers ("No")

ID 16

13. What is your primary health concern?

LOGIC Show/hide trigger exists.

ID 17

14. What method was used to **confirm** you have leaky gut?

- My practitioner advised me based on my symptoms and case history
- A urine test – lactulose/mannitol
- A blood test – zonulin
- A blood test - lactulose/rhamnose (from CSIRO)
- A stool/faecal test – zonulin
- Food intolerance/sensitivity test - IgG
- Looking into eyes - Iridology
- Looking at blood under a microscope – Hemaview (live blood analysis)
- Kinesiology
- I have not been diagnosed but think I have leaky gut
- I don't know
- Other

VALIDATION %s format expected Using custom RegEx pattern

LOGIC Hidden unless: #14 Question "What method was used to **confirm** you have leaky gut?" is one of the following answers ("My practitioner advised me based on my symptoms and case history","A urine test – lactulose/mannitol","A blood test – zonulin","A blood test - lactulose/rhamnose (from CSIRO)","A stool/faecal test – zonulin","Food intolerance/sensitivity test - IgG","Looking into eyes - Iridology","Looking at blood under a microscope – Hemaview (live blood analysis)","Kinesiology","Other")

ID 18

15. What **year** were you diagnosed with leaky gut?

VALIDATION %s format expected Using custom RegEx pattern

ID 19

16. What **year** do you believe your leaky gut first started?

LOGIC Hidden unless: #14 Question "What method was used to **confirm** you have leaky gut?" is one of the following answers ("A urine test – lactulose/mannitol","A blood test – zonulin","A blood test - lactulose/rhamnose (from CSIRO)","A stool/faecal test – zonulin","Food intolerance/sensitivity test - IgG","Looking into eyes - Iridology","Looking at blood under a microscope – Hemaview (live blood analysis)","Kinesiology","Other")

ID 20

17. At what point did you get tested for leaky gut? Select all that apply.

- ☐ Before receiving treatment for leaky gut
- ☐ During treatment for leaky gut
- ☐ After receiving treatment for leaky gut

LOGIC Show/hide trigger exists. Hidden unless: #14 Question "What method was used to **confirm** you have leaky gut?" is one of the following answers ("A urine test – lactulose/mannitol","A blood test – zonulin","A blood test - lactulose/rhamnose (from CSIRO)","A stool/faecal test – zonulin","Food intolerance/sensitivity test - IgG","Looking into eyes - Iridology","Looking at blood under a microscope – Hemaview (live blood analysis)","Kinesiology","Other")

ID 21

18. How many times have you been tested for leaky gut?

- 1
- 2
- 3
- 4
- 5 or more

LOGIC Hidden unless: #18 Question "How many times have you been tested for leaky gut?" is one of the following answers ("2","3","4","5 or more")

ID 23

19. How long after the **first** test did you get tested for leaky gut the **second** time?

- Between 1 and 3 months
- Between 3 and 6 months
- Between 6 and 9 months
- Between 9 and 12 months
- Between 12 and 18 months
- Between 18 and 24 months
- Over 2 years

24

20. When would you like your practitioner to **test you for leaky gut?** Select all that apply.

- [ ] Before treatment for leaky gut
- [ ] During treatment for leaky gut
- [ ] After treatment for leaky gut
- [ ] For monitoring of leaky gut
- [ ] For monitoring diseases related to leaky gut
- [ ] When I ask
- [ ] When the healthcare practitioner advises
- [ ] I do not want to be tested for leaky gut
- [ ] Other

302

21. Please indicate the extent to which you would **prefer** to be tested for leaky gut if your practitioner **suspects** leaky gut.

No preference to be tested for leaky gut	Slightly prefer to be tested for leaky gut	Prefer to be tested for leaky gut	Strongly prefer to be tested for leaky gut	Very strongly prefer to be tested for leaky gut
○	○	○	○	○

26

22. How **important** is it to you that your practitioner tests for leaky gut?

Not important	Slightly important	Moderately important	Important	Very important
○	○	○	○	○

27

23. How likely are you to **adhere to your practitioner's treatment prescription** if you were tested **positive** for leaky gut?

Very unlikely	Moderately unlikely	Neither likely nor unlikely	Moderately likely	Very likely
○	○	○	○	○

28

24. What factors are **important** to you when it comes to testing leaky gut? Rank the **importance** of **each** from 'not important' to 'very important'.

	Not important	Slightly important	Moderately important	Important	Very important
Affordability	○	○	○	○	○
Accuracy in measuring leaky gut	○	○	○	○	○
Easy to access the test	○	○	○	○	○
Not requiring a blood test	○	○	○	○	○
Time involved in performing the test	○	○	○	○	○

37

25. What **method** would you **prefer** to be used to test your leaky gut? Select all that apply.

- ☐ Blood sample at a pathology lab
- ☐ Urine collection at home
- ☐ Stool/faecal collection at home
- ☐ Signs and symptoms according to a practitioner

Practitioner Involvement

38

26. What type of practitioner **first diagnosed** you with leaky gut? If your practitioner has **multiple qualifications** select all that apply.

- [ ] Self-diagnosed
- [ ] Acupuncturist
- [ ] Ayurvedic practitioner
- [ ] Chinese medicine practitioner
- [ ] Chiropractor
- [ ] Dietitian
- [ ] Gastroenterologist
- [ ] General practitioner (GP)
- [ ] Herbalist
- [ ] Homeopath
- [ ] Integrative practitioner
- [ ] Kinesiologist
- [ ] Naturopath
- [ ] Nutritionist
- [ ] Nurse
- [ ] Nurse Practitioner
- [ ] Osteopath
- [ ] Pharmacist
- [ ] Other

303

27. What type of **practitioner/s** would you want to see for the **testing and treatment of leaky gut**? Select all that apply.

- [ ] Acupuncturist
- [ ] Ayurvedic practitioner
- [ ] Chinese medicine practitioner
- [ ] Chiropractor
- [ ] Dietitian
- [ ] Gastroenterologist
- [ ] General practitioner (GP)
- [ ] Herbalist
- [ ] Homeopath
- [ ] Integrative practitioner
- [ ] Kinesiologist
- [ ] Naturopath
- [ ] Nutritionist
- [ ] Nurse
- [ ] Nurse Practitioner
- [ ] Osteopath
- [ ] Pharmacist
- [ ] Other

*

ID 39

28. Who speculated you had leaky gut first?

- I first speculated I had leaky gut
- A practitioner first speculated I had leaky gut

LOGIC Show/hide trigger exists.

ID 177

29. To the best of your knowledge are you currently using any of the **following treatments specifically for the management of leaky gut?** Select all that apply.

** **Dietary changes:** any change to normal eating.*
** **Lifestyle changes:** any activity or routine such as exercise or breathing techniques that does NOT involve food, supplements or medicines.*
** **Health products/dietary supplements:** any vitamin, mineral, herbal medicine, probiotic, prebiotic, fish oil or amino acid.*
** **Medication** any over the counter medication or prescription drug used within conventional medicine.*

- ☐ Dietary changes
- ☐ Lifestyle changes
- ☐ Health products/dietary supplements
- ☐ Medications

Treatment for Leaky Gut

ID 280

PIPING Piped Values From Question 29. (To the best of your knowledge are you currently using any of the **following treatments specifically for the management of leaky gut?** Select all that apply.)

30. What type of **practitioner/s prescribed the following treatments**? Select multiple if your practitioner holds more than **one** qualification.

- Self-prescribed
- Acupuncturist
- Ayurvedic practitioner
- Chinese medicine practitioner
- Chiropractor
- Dietitian
- Gastroenterologist
- General practitioner
- Herbalist
- Homeopath
- Integrative practitioner
- Kinesiologist
- Naturopath
- Nutritionist
- Nurse
- Nurse Practitioner
- Osteopath
- Pharmacist

Enter another option

57

30. How often have you consumed the following **dietary products**? Rank the frequency of use **over the last 12 months**.

	Never	Less than once a month	1-3 times a month	Once a week	2-6 times a week	Every day
Alcoholic drinks	○	○	○	○	○	○
Gluten containing foods	○	○	○	○	○	○
Red meat	○	○	○	○	○	○
Dairy products	○	○	○	○	○	○
Refined sugar	○	○	○	○	○	○
Organic foods	○	○	○	○	○	○
Apple cider vinegar	○	○	○	○	○	○
Bone broth	○	○	○	○	○	○
Collagen products	○	○	○	○	○	○
Fermented foods	○	○	○	○	○	○
Enter another option	○	○	○	○	○	○
Enter another option	○	○	○	○	○	○

88

31. How often do you incorporate the following **lifestyle habits**? Rank the frequency of use **over the last 12 months**.

	Never	Less than once a month	1-3 times a month	Once a week	2-6 times a week	Every day
Vigorous exercise such as running, cycling or HIIT	○	○	○	○	○	○
Meditation	○	○	○	○	○	○
Yoga	○	○	○	○	○	○
Breathing exercises	○	○	○	○	○	○
Stress management	○	○	○	○	○	○
Stimulation of the vagus nerve	○	○	○	○	○	○
Enter another option	○	○	○	○	○	○
Enter another option	○	○	○	○	○	○

LOGIC Hidden unless: #29 Question "To the best of your knowledge are you currently using any of the **following treatments specifically for the management of leaky gut?** Select all that apply." is one of the following answers ("Health products/dietary supplements")

ID 304

33. What **health products/dietary supplements** are you currently taking for the treatment of leaky gut? **Health products/dietary supplements** include any vitamin, mineral, herbal medicine, probiotic, prebiotic, fish oil or amino acid.

	Brand Name	Product Name
Supplement 1		
Supplement 2		
Supplement 3		
Supplement 4		
Supplement 5		

LOGIC Hidden unless: #29 Question "To the best of your knowledge are you currently using any of the **following treatments specifically for the management of leaky gut?** Select all that apply." is one of the following answers ("Medications")

ID 312

34. What **medications** are you currently taking for the treatment of leaky gut? **Medications** are any over the counter medication or prescription drug used within conventional medicine.

	Brand Name	Product Name
Medication 1		
Medication 2		
Medication 3		
Medication 4		
Medication 5		

ID 193

35. How often have you taken the following **medications**? Rank the frequency of use **over the last 12 months**.

	Never	Less than once a month	1-3 times a month	Once a week	2-6 times a week	Every day
Antibiotics	○	○	○	○	○	○
Nonsteroidal anti-inflammatory drugs (NSAIDs) such as aspirin and ibuprofen	○	○	○	○	○	○
Methotrexate	○	○	○	○	○	○
Prednisone	○	○	○	○	○	○

200

36. Please indicate the extent to which **you would prefer** to use the following to **treat** a leaky gut.

	No preference	Slightly prefer	Prefer	Strongly prefer	Very strongly prefer
Dietary changes	○	○	○	○	○
Lifestyle changes	○	○	○	○	○
Health products/dietary supplements	○	○	○	○	○
Medications	○	○	○	○	○

Financial Cost of Leaky Gut

133

37. How well do you manage your **household income**?

- ○ Always difficult to manage on available income
- ○ Sometimes difficult to manage on available income
- ○ Managing on available income is not too bad
- ○ Easy to manage on available income

VALIDATION %s format expected Using custom RegEx pattern

LOGIC Hidden unless: #29 Question "To the best of your knowledge are you currently using any of the **following treatments specifically for the management of leaky gut?** Select all that apply." is one of the following answers ("Health products/dietary supplements")

ID 318

38. On average, how much do you spend each month on **health products/dietary supplements** for the treatment of leaky gut? **Health products/dietary supplements** include any vitamin, mineral, herbal medicine, probiotic, prebiotic, fish oil or amino acid).

$ _____ .00

VALIDATION %s format expected Using custom RegEx pattern

LOGIC Hidden unless: #14 Question "What method was used to **confirm** you have leaky gut?" is one of the following answers ("A urine test – lactulose/mannitol","A blood test – zonulin","A blood test - lactulose/rhamnose (from CSIRO)","A stool/faecal test – zonulin","Food intolerance/sensitivity test - IgG","Looking into eyes - Iridology","Looking at blood under a microscope – Hemaview (live blood analysis)","Kinesiology")

ID 365

PIPING Piped From Question 14. (What method was used to **confirm** you have leaky gut?)

How much did you spend on ***[question('value'), id='17']*** to measure leaky gut?

$ _____ .00

VALIDATION %s format expected Using custom RegEx pattern

ID 214

39. How much have you spent in the last **12 months** on practitioner **consultation fees** regarding the treatment of your leaky gut?

$ _____ .00

218

40. How **important** is cost in your **decision** to get tested for leaky gut?

Not important	Slightly important	Moderately important	Important	Very important
○	○	○	○	○

219

41. How much would you be **willing to spend** on a test to measure leaky gut?

- ○ Nothing
- ○ $0-50
- ○ $51-100
- ○ $101-150
- ○ $151-200
- ○ $201 or more

220

42. Which of the following would you **rather spend your money on**? Please indicate the **importance** of **each**.

	Not important	Slightly important	Important	Fairly important	Very important
Being tested for leaky gut	○	○	○	○	○
Dietary treatments	○	○	○	○	○
Lifestyle treatments	○	○	○	○	○
Health products/dietary supplements	○	○	○	○	○
Medications	○	○	○	○	○

Wellbeing and Quality of Life #1

229

43. Over the last **12 months**, do you believe your leaky gut is getting **better or worse**?

- Better
- Worse
- No change

230

44. How many days a week do you think your leaky gut **affects your daily living**?

- 0
- 1
- 2
- 3
- 4
- 5
- 6
- 7

Wellbeing and Quality of Life #2

319

45. In general, would you say your health is:

- Excellent
- Very good
- Good
- Fair
- Poor

320

46. For how long (if at all) has your **health limited you** in **each** of the following activities?

	Limited for more than 3 months	Limited for 3 months or less	Not limited at all
The kinds or amounts of vigorous activities you can do, like lifting heavy objects, running or participating in strenuous sports			
The kinds or amounts of moderate activities you can do, like moving a table, carrying groceries, or bowling			
Walking uphill or climbing a few flights of stairs			
Bending, lifting, or stooping			
Walking one block			
Eating, dressing, bathing, or using the toilet			

329

47. How much **bodily** pain have you had **during the past 4 weeks**:

- None
- Very mild
- Mild
- Moderate
- Severe
- Very severe

330

48. Does your health **keep** you from working at a job, doing work around the house, or going to school?

- YES, for more than 3 months
- YES, for 3 months or less
- NO

331

49. Have you been unable to do **certain kinds or amounts** of work, housework, or schoolwork because of your health?

- YES, for more than 3 months
- YES, for 3 months or less
- NO

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50. For **each** of the following questions, please mark the circle for the **one** answer that comes **closest** to the way you have been feeling **during the past month.**

	All of the time	Most of the time	A good bit of the time	Some of the time	A little of the time	None of the time
How much of the time, during the past month, has your health limited your social activities (like visiting with friends or close relatives)?	○	○	○	○	○	○
How much of the time, during the past month, have you been a very nervous person?	○	○	○	○	○	○
During the past month, how much of the time have you felt calm and peaceful?	○	○	○	○	○	○
How much of the time, during the past month, have you felt downhearted and blue?	○	○	○	○	○	○
During the past month, how much of the time have you been a happy person?	○	○	○	○	○	○
How often, during the past month, have you felt so down in the dumps that nothing could cheer you up?	○	○	○	○	○	○

341

51. Please mark the circle that **best** describes whether **each** of the following statements is **true** or **false** for you.

	Definitely true	Mostly true	Mostly false	Definitely false
I am somewhat ill	○	○	○	○
I am as healthy as anybody I know	○	○	○	○
My health is excellent	○	○	○	○
I have been feeling bad lately	○	○	○	○

Wellbeing and Quality of Life #3

Page description:
The following questions ask how satisfied you feel, on a scale from zero to 10. **Zero** means you feel no satisfaction at all and **10** means you feel completely satisfied.

349

52. How satisfied are you **with your standard of living**?

	0	1	2	3	4	5	6	7	8	9	10	
No satisfaction at all	○	○	○	○	○	○	○	○	○	○	○	**Completely satisfied**

350

53. How satisfied are you **with your health**?

	0	1	2	3	4	5	6	7	8	9	10	
No satisfaction at all	○	○	○	○	○	○	○	○	○	○	○	**Completely satisfied**

351

54. How satisfied are you **with what you are achieving in life**?

	0	1	2	3	4	5	6	7	8	9	10	
No satisfaction at all	○	○	○	○	○	○	○	○	○	○	○	**Completely satisfied**

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55. How satisfied are you **with your personal relationships**?

	0	1	2	3	4	5	6	7	8	9	10	
No satisfaction at all	○	○	○	○	○	○	○	○	○	○	○	**Completely satisfied**

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56. How satisfied are you **with how safe you feel**?

	0	1	2	3	4	5	6	7	8	9	10	
No satisfaction at all	○	○	○	○	○	○	○	○	○	○	○	**Completely satisfied**

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57. How satisfied are you **with feeling part of your community**?

	0	1	2	3	4	5	6	7	8	9	10	
No satisfaction at all	○	○	○	○	○	○	○	○	○	○	○	**Completely satisfied**

356

58. How satisfied are you **with your future security**?

	0	1	2	3	4	5	6	7	8	9	10	
No satisfaction at all	○	○	○	○	○	○	○	○	○	○	○	**Completely satisfied**

Research Topics

245

59. In your opinion, which of the following areas are **most important for practitioners to know**? Rank the importance of each area.

	Not important	Slightly important	Important	Fairly important	Very important
Which health conditions are associated with leaky gut	○	○	○	○	○
Who should be checked for leaky gut	○	○	○	○	○
How to accurately measure leaky gut	○	○	○	○	○
When to measure leaky gut	○	○	○	○	○
Signs and symptoms of leaky gut	○	○	○	○	○
Risk factors for leaky gut	○	○	○	○	○
Biomarkers to help identify leaky gut	○	○	○	○	○
Diet treatment for leaky gut	○	○	○	○	○
Lifestyle treatment for leaky gut	○	○	○	○	○
Health products/dietary supplements	○	○	○	○	○
Medications for leaky gut	○	○	○	○	○
Enter another option	○	○	○	○	○
Enter another option	○	○	○	○	○

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60. What form of educational material would you prefer to learn about leaky gut? Select all that apply.

- [ ] Reading material (e.g. books or articles)
- [ ] Website
- [ ] Patient handout
- [ ] Video content
- [ ] Visual diagram
- [ ] Health events
- [ ] Podcast
- [ ] Phone app
- [ ] Other

Understanding of Leaky Gut

243

61. How did you first hear about leaky gut?

- ○ Internet
- ○ Practitioner
- ○ Health event
- ○ Friend/family member
- ○ Other

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62. In your opinion, which of the following best describes leaky gut?

- ○ A syndrome – "Leaky gut syndrome"
- ○ A medical condition – A diagnosed medical condition
- ○ A reaction – A response to a stimulus (e.g. inflammation)

(untitled)

401

63. By selecting 'I understand' you acknowledge that the answers you have provided cannot be withdrawn once submitted and that consent is given.

- ☐ I understand

Thank You!

1

Thank you for taking our survey.

Want to participate in further research?

We are developing a guideline for the management of leaky gut which will guide practitioners in clinical practice. This guideline will contain recommendations for practitioners to follow on the assessment and management of leaky gut based on published research and stakeholder engagement.

We will require members of the public who have been diagnosed with leaky gut to help inform whether the recommendations developed are appropriate.

If you would like more information or would like to participate please fill out your contact details **> HERE <.**

ID 272

If you would like to receive a copy of the published research please enter your email address **> HERE <**.

ID 400

This survey aims to explore the health-seeking behaviour of adults with suspected leaky gut. The content of this survey is not intended to provide any medical advice for the assessment, treatment or management of leaky gut. If you need help or advice regarding the management of leaky gut it is recommended you speak with your general practitioner. Below are a few useful resources.

- Clinical Labs and NutriPATH
- The CSIRP Healthy Gut Diet Book
- Leaky Gut Support Group Australia

APPENDIX 3.2: PHASE ONE UTS HREC ETHICS APPROVAL

Dear Applicant

Thank you for your response to the Committee's comments for your project titled, "Health-seeking behaviour of adults with suspected increased intestinal permeability". The Committee agreed that this application now meets the requirements of the National Statement on Ethical Conduct in Human Research (2007) and has been approved on that basis. You are therefore authorised to commence activities as outlined in your application.

You are reminded that this letter constitutes ethics approval only. This research project must also be undertaken in accordance with all UTS policies and guidelines including the Research Management Policy (http://www.gsu.uts.edu.au/policies/research-management-policy.html).

Your approval number is UTS HREC REF NO. ETH19-4012.

Approval will be for a period of five (5) years from the date of this correspondence subject to the submission of annual progress reports.

The following standard conditions apply to your approval:

• Your approval number must be included in all participant material and advertisements. Any advertisements on Staff Connect without an approval number will be removed.

• The Principal Investigator will immediately report anything that might warrant review of ethical approval of the project to the Ethics Secretariat (Research.Ethics@uts.edu.au).

• The Principal Investigator will notify the UTS HREC of any event that requires a modification to the protocol or other project documents, and submit any required amendments prior to implementation. Instructions can be found at https://staff.uts.edu.au/topichub/Pages/Researching/Research%20Ethics%20and%20Integrity/Human%20research%20ethics/Post-approval/post-approval.aspx#tab2.

• The Principal Investigator will promptly report adverse events to the Ethics Secretariat (Research.Ethics@uts.edu.au). An adverse event is any event (anticipated or otherwise) that has a negative impact on participants, researchers or the reputation of the University. Adverse events can also include privacy breaches, loss of data and damage to property.

• The Principal Investigator will report to the UTS HREC annually and notify the HREC when the project is completed at all sites. The Principal Investigator will notify the UTS HREC of any plan to extend the duration of the project past the approval period listed above through the progress report.

• The Principal Investigator will obtain any additional approvals or authorisations as required (e.g. from other ethics committees, collaborating institutions, supporting organisations).

• The Principal Investigator will notify the UTS HREC of his or her inability to continue as Principal Investigator including the name of and contact information for a replacement.

I also refer you to the AVCC guidelines relating to the storage of data, which require that data be kept for a minimum of 5 years after publication of research. However, in NSW, longer retention requirements are required for research on human subjects with potential long-term effects, research with long-term environmental effects, or research considered of national or international significance, importance, or controversy. If the data from this research project falls into one of these categories, contact University Records for advice on long-term retention.

You should consider this your official letter of approval. If you require a hardcopy please contact Research.Ethics@uts.edu.au.

If you have any queries about your ethics approval, or require any amendments to your research in the future, please do not hesitate to contact Research.Ethics@uts.edu.au.

Yours sincerely,

A/Prof Beata Bajorek
Chairperson
UTS Human Research Ethics Committee
C/- Research Office
University of Technology Sydney
E: Research.Ethics@uts.edu.au

APPENDIX 3.3: PARTICIPANT INFORMATION SHEET: LEAKY GUT SURVEY

PARTICIPANT INFORMATION SHEET

Health-Seeking Behaviour of Adults with Suspected Increased Intestinal Permeability

UTS HREC APPROVAL NUMBER ETH19-4012

WHO IS DOING THE RESEARCH?

My name is Bradley Leech and I am a PhD student at the University of Technology Sydney. My supervisors are Prof David Sibbritt (David.Sibbritt@uts.edu.au), Dr Amie Steel (Amie.Steel@uts.edu.au) and Dr Erica McIntyre (Erica.McIntyre@uts.edu.au).

WHAT IS THIS RESEARCH ABOUT?

This research aims to investigate the health-seeking behaviour, views and preferences for the assessment and management of adults with suspected or diagnosed leaky gut (intestinal permeability).

INCLUSION/EXCLUSION CRITERIA

Before you decide to participate in this research study, we need to ensure that you are eligible to take part. To be eligible to participate you must think or know you have leaky gut, be 18 years or older and living in Australia.

IF I SAY YES, WHAT WILL IT INVOLVE?

If you decide to participate, we will ask you to complete an online survey that will take approximately 30 minutes to complete. In this survey, you will be asked to provide basic demographic characteristics and information about your views and preferences for the management, diagnosis and treatment of leaky gut.

DO I HAVE TO SAY YES?

Participation in this study is voluntary. It is completely up to you whether or not you decide to take part.

ARE THERE ANY RISKS/INCONVENIENCE?

Yes, there is an inconvenience. This survey will take approximately 30 minutes to complete.

IF I SAY YES, CAN I CHANGE MY MIND LATER?

You can change your mind at any time, and you do not have to say why. We will thank you for your time so far and won't contact you about this research again.

WHAT WILL HAPPEN IF I SAY NO?

Nothing. If you decide not to participate, it will not affect your relationship with the researchers or the University of Technology Sydney. No further contact will be made by the research team regarding this survey.

WHAT WILL HAPPEN TO INFORMATION ABOUT ME?

Your information will be treated confidentially, no personal details that could identify you will be obtained during the survey. We plan to publish the results of this study and use them as the consumer views and preference for the development of a clinical practice guideline for the assessment and management of increased intestinal permeability.

At the end of the survey, you will be given the option to be contacted to be a part of future phases of the clinical practice guidelines project. In all instances, your information will be treated confidentially.

WHAT IF I HAVE CONCERNS OR A COMPLAINT?

If you have concerns about the research that you think I or my supervisor can help you with, please feel free to contact us on Bradley.Leech@uts.edu.au or 02 9514 4172. If you would like to talk to someone who is not connected with the research, you may contact the Research Ethics Officer on 02 9514 9772 or Research.ethics@uts.edu.au and quote this number UTS HREC Approval Number ETH19-4012.

APPENDIX 3.4: INFORMED CONSENT: LEAKY GUT SURVEY

By selecting 'NEXT' you agree to the following:

I agree to participate in the research project *Health-Seeking Behaviour of Adults with Suspected Increased Intestinal Permeability (UTS HREC NO. ETH19-4012)* being conducted by Mr Bradley Leech (Bradley.Leech@uts.edu.au).

I understand that the purpose of this study is to investigate the health-seeking behaviour, views and preferences for the assessment and management of adults with suspected or diagnosed leaky gut (intestinal permeability).

I have read the Participant Information Sheet.

I understand that my participation in this research will involve completing an online survey lasting approximately 30 minutes.

I am aware that I can contact Mr Bradley Leech if I have any concerns about the research. I also understand that I can stop the survey at any time I wish, without consequences, and without giving a reason.

I am aware that if I decide not to participate, this will not affect my relationship with the researchers or the University of Technology Sydney. No further contact will be made by the research team regarding this survey.

I agree that I have had an opportunity to have all of my questions answered fully and clearly.

I agree that the research data gathered from this project may be published in a form that does not identify me in any way.

APPENDIX 3.5: PHASE TWO UTS HREC ETHICS APPROVAL

Dear Applicant,

Re: ETH20-5291 - "Clinical Practice Guideline for the Assessment and Management of Increased Intestinal Permeability"

Your local research office has reviewed your application and agreed that it now meets the requirements of the National Statement on Ethical Conduct in Human Research (2007) and has been approved on that basis. You are therefore authorised to commence activities as outlined in your application, subject to any conditions detailed in this document.

You are reminded that this letter constitutes ethics approval only. This research project must also be undertaken in accordance with all UTS policies and guidelines including the Research Management Policy.

Your approval number is UTS HREC REF NO. ETH20-5291

Approval will be for a period of five (5) years from the date of this correspondence subject to the submission of annual progress reports.

The following standard conditions apply to your approval:

- Your approval number must be included in all participant material and advertisements.
- Any advertisements on Staff Connect without an approval number will be removed.
- The Principal Investigator will immediately report anything that might warrant review of ethical approval of the project to the Ethics Secretariat (Research.Ethics@uts.edu.au).
- The Principal Investigator will notify the UTS HREC of any event that requires a modification to the protocol or other project documents, and submit any required amendments prior to implementation. Instructions on how to submit an amendment application can be found here.
- The Principal Investigator will promptly report adverse events to the Ethics Secretariat. An adverse event is any event (anticipated or otherwise) that has a negative impact on participants, researchers or the reputation of the University. Adverse events can also include privacy breaches, loss of data and damage to property.
- The Principal Investigator will report to the UTS HREC annually and notify the HREC when the project is completed at all sites.
- The Principal Investigator will notify the UTS HREC of any plan to extend the duration of the project past the approval period listed above through the progress report.
- The Principal Investigator will obtain any additional approvals or authorisations as required (e.g. from other ethics committees, collaborating institutions, supporting organisations).
- The Principal Investigator will notify the UTS HREC of his or her inability to continue as Principal Investigator including the name of and contact information for a replacement.

This research must be undertaken in compliance with the Australian Code for the Responsible Conduct of Research and National Statement on Ethical Conduct in Human Research.

You should consider this your official letter of approval.

If you have any queries about this approval, or require any amendments to your approval in future, please do not hesitate to contact your local research office or the Ethics Secretariat.

APPENDIX 3.6: PARTICIPANT INFORMATION SHEET: IP GUIDELINE

PARTICIPANT INFORMATION SHEET

Clinical Practice Guideline for the Management of Increased Intestinal Permeability

UTS HREC APPROVAL NUMBER ETH20-5291

WHO IS DOING THE RESEARCH?

My name is Bradley Leech and I am a PhD student at the University of Technology Sydney. My supervisors are Prof David Sibbritt (David.Sibbritt@uts.edu.au), Dr Amie Steel (Amie.Steel@uts.edu.au) and Dr Erica McIntyre (Erica.McIntyre@uts.edu.au).

WHAT IS THIS RESEARCH ABOUT?

This study aims to improve the management of increased intestinal permeability by clinicians in private practice in Australia by developing a clinical practice guideline for the management of intestinal permeability (IP Guideline).

INCLUSION/EXCLUSION CRITERIA

You have been identified by the Working Group as a key stakeholder related to the IP Guideline.

IF I SAY YES, WHAT WILL IT INVOLVE?

If you decide to participate, you will be asked to actively participate in the Stakeholder Group. Through a survey study design, you will be asked to;

- Participate in Stakeholder Group survey,
- Report all relevant conflicts of interests,
- Evaluate the acceptability and feasibility of each recommendation,
- Provide insight on the extent published evidence reflects outcomes,
- Provide feedback on guideline wording to ensure recommendations are understandable.

DO I HAVE TO SAY YES?

Participation in this study is voluntary. It is completely up to you whether or not you decide to take part.

ARE THERE ANY RISKS/INCONVENIENCE?

Yes, there is an inconvenience. Involvement in the IP Guideline requires approximately 30 minutes of your time.

IF I SAY YES, CAN I CHANGE MY MIND LATER?

You can change your mind at any time, and you do not have to say why. If you are unable to participate you will be required to notify the Mr Bradley Leech at the earliest possible moment.

WHAT WILL HAPPEN IF I SAY NO?

Nothing. If you decide not to participate, it will not affect your relationship with the researchers or the University of Technology Sydney. No further contact will be made by the research team regarding this research project.

WHAT WILL HAPPEN TO INFORMATION ABOUT ME?

As a requirement of the National Health and Medical Research Council (NHMRC) *Guidelines for Guidelines* Handbook to meet the 2016 *NHMRC Standards for Guidelines* all members name, profession or discipline, organisational affiliation and role in the guideline development process is required to be reported. Additionally, all disclosure of interests or each member will be reported. This information you will be published in the IP Guideline.

WHAT IF I HAVE CONCERNS OR A COMPLAINT?

If you have concerns about the research that you think I or my supervisor can help you with, please feel free to contact us on Bradley.Leech@student.uts.edu.au or 02 9514 4172. If you would like to talk to someone who is not connected with the research, you may contact the Research Ethics Officer on 02 9514 9772 or Research.ethics@uts.edu.au and quote this number [UTS HREC **ETH20-5291**]

APPENDIX 3.7: INFORMED CONSENT: STAKEHOLDER SURVEY

INFORMED CONSENT

By signing below, you agree to the following terms and conditions:

1. I agree to participate in the research project *Clinical Practice Guideline for the Management of Increased Intestinal Permeability (UTS HREC* **ETH20-5291***)* being conducted by Mr Bradley Leech (Bradley.Leech@uts.edu.au), Prof David Sibbritt (David.Sibbritt@uts.edu.au), Dr Amie Steel (Amie.Steel@uts.edu.au) and Dr Erica McIntyre (Erica.McIntyre@uts.edu.au).
2. I understand that the purpose of this study is to improve the management of increased intestinal permeability by clinicians in private practice in Australia by developing a clinical practice guideline for the management of intestinal permeability.
3. I have read the Participant Information Sheet.
4. I understand that my participation in this research will require approximately 30 minutes.
5. I am aware that I can contact Mr Bradley Leech if I have any concerns about the research. I also understand that I am free to withdraw my participation from this research project at any time I wish, without consequences, and without giving a reason. If I am unable to participate, I will be required to notify Mr Bradley Leech the at the earliest possible moment.
6. I am aware that if I decide not to participate, this will not affect my relationship with the researchers or the University of Technology Sydney. No further contact will be made by the research team regarding this survey.
7. I agree that I have had an opportunity to have all of my questions answered fully and clearly.
8. I agree that the research data gathered from this project including name, profession or discipline, organisational affiliation and role in the guideline development process will be published. Additionally, all disclosure of interests will be reported. This information will be published in the *Clinical Practice Guideline for the Management of Increased Intestinal Permeability*.

Stakeholder Group Member Name Signature
Date

APPENDIX 3.8: NHMRC EVIDENCE STATEMENT

Key question(s):			**Evidence table**
1. Evidence base *(number of studies, level of evidence and risk of bias in the included studies)*			
	A	**One or more level I studies with a low risk**	
	B	**One or two Level II studies with a low risk of bias or SR/several Level III studies with**	
	C	**One or two Level III studies with a low risk**	
	D	**Level IV studies or Level I to III**	
2. Consistency *(if only one study was available, rank this component as 'not applicable')*			
	A	**All studies consistent**	
	B	**Most studies consistent and**	
	C	**Some inconsistency, reflecting genuine**	
	D	**Evidence is inconsistent**	
	N	**Not applicable (one study only)**	
3. Clinical impact *(Indicate in the space below if the study results varied according to some unknown factor (not simply study quality or sample size) and thus the clinical impact of the intervention*			
	A	**Very large**	
	B	**Substantial**	
	C	**Moderate**	
	D	**Slight/Restricted**	
4. Generalisability *(How well does the body of evidence match the population and clinical*			
	A	**Evidence directly generalisable to target**	
	B	**Evidence directly generalisable to target**	
	C	**Evidence not directly generalisable to the**	
	D	**Evidence not directly generalisable to**	
5. Applicability *(Is the body of evidence relevant to the Australian healthcare context in terms of*			
	A	**Evidence directly applicable to Australian**	
	B	**Evidence applicable to Australian**	
	C	**Evidence probably applicable to**	
	D	**Evidence not applicable to Australian**	

Other factors *(Indicate here any other factors that you took into account when assessing the evidence base (for example, issues that might cause the group to downgrade or upgrade the*

EVIDENCE STATEMENT MATRIX

Please summarise the development group's synthesis of the evidence relating to the key question, taking all the above factors into account.

Component	Rating	Description
1. Evidence base		
2. Consistency		
3. Clinical impact		
4. Generalisability		

5. Applicability		
Indicate any dissenting opinions		
RECOMMENDATION *What recommendation(s) does the guideline development group draw from this evidence? Use action statements where possible.*	**GRADE OF RECOMMENDATION**	
PRACTICE POINT (CONSENSUS-BASED RECOMMENDATION) *If there is no good quality evidence available but there is consensus among Guideline committee members, a consensus-based recommendation (practice point) can be given.*		
UNRESOLVED ISSUES *If needed, keep note of specific issues that arise when each recommendation is formulated and that require follow-up.*		

IMPLEMENTATION OF RECOMMENDATION *Please indicate yes or no to the following questions. Where the answer is yes please provide explanatory information about this. This information will be used to develop the implementation plan for the guidelines.*	
Will this recommendation result in changes in usual care?	**NO/YES**
Are there any resource implications associated with implementing this recommendation?	**NO/YES**
Will the implementation of this recommendation require changes in the way care is currently organised?	**NO/YES**
Are the guideline development group aware of any barriers to the implementation of this recommendation?	**NO/YES**

APPENDIX 3.9: DISCLOSURE OF INTEREST FORM

Disclosure of Interest Form

Before the appointment to the guideline development group, all conflicts of interest need to be identified, transparently reported and appropriately managed to reduce the risk of bias. The Chair will review disclosures and determine whether or not a management plan is required.

Question 1

Over the past 12 months, have you or, as far as you are aware, any immediate family members been **employed by an entity having a commercial or other interest** in intestinal permeability? This includes but limited to any employment where the entity is involved in the development or sale of any nutraceutical or therapy for intestinal permeability or employment where the entity is involved in the assessment (pathology testing) of intestinal permeability.

No (go to question 2)

Yes: Please provide all relevant details below

Question 2

Over the past 12 months, have you or, as far as you are aware, any immediate family members **been given any financial benefits** from any entity which has a commercial interest in intestinal permeability?

No (go to question 3)

Yes: Please provide all relevant details below

Question 3

Over the past 12 months, have you or, as far as you are aware, any immediate family members **received any support, payment or been employed** by any entity who is involved in the assessment (pathology testing) of intestinal permeability?

No (go to question 4)

Yes: Please provide all relevant details below

Question 4

Over the past 12 months, have you or, as far as you are aware, any immediate family members **received any support, payment or been employed** by any entity who is involved in the development or sale of any nutraceutical or therapy for intestinal permeability?

No (go to question 5)

Yes: Please provide all relevant details below

Question 5

Are you **affiliated or associated with any organisations** whose interests are either aligned with or opposed to any subject matter related to the assessment or management of intestinal permeability?

No (go to question 6)

Yes: Please provide all relevant details below

Question 6

Are there any other **relationships or activities not declared above** that could be perceived potentially to influence your contribution?

No

Yes: Please provide all relevant details below

Declaration of Interest

Name

In signing this form, I hereby agree:

- that the information provided was correct on the date entered below
- this information is provided to the Working Group member for their consideration.
- to update this information throughout the development of the IP Guideline.
- allow the publication of any interest I have disclosed in this form, and any interests declared after I complete this form and any management plan in the final guideline.

Signature of potential member

Date

Office use only

Appointment approved without a management plan

Appointment approved with a management plan

Appointment declined due to conflict of interest unable to be managed

Signature of Chair

Print name

Date

APPENDIX 3.10: REGISTER OF DISCLOSURES OF INTEREST

Questions	Stakeholder Group Members							
	Dr Jason Hawrelak	Dr Nirala Jacobi	Dr Michael Osiecki	Dr Christine Houghton	Dr Ronald Goedeke	Benedict Freudenmann	Kirsty Wirth	Vanita Dahia
Over the past 12 months, have you or, as far as you are aware, any immediate family members been **employed by an entity having a commercial or other interest** in intestinal permeability?	No	Yes	Yes	Yes	No	Yes	No	No
Over the past 12 months, have you or, as far as you are aware, any immediate family members **been given any financial benefits** from any entity which has a commercial interest in intestinal permeability?	No	Yes	Yes	Yes	No	Yes	No	No
Over the past 12 months, have you or, as far as you are aware, any immediate family members **received any support, payment or been employed** by any entity who is involved in the assessment (pathology testing) of intestinal permeability?	No	Yes	No	No	No	Yes	No	Yes
Over the past 12 months, have you or, as far as you are aware, any immediate family members **received any support, payment or been employed** by any entity who is involved in the development or sale of any nutraceutical or therapy for intestinal permeability?	No	No	Yes	Yes	No	Yes	No	No
Are you **affiliated or associated with any organisations** whose interests are either aligned with or opposed to any subject matter related to the assessment or management of intestinal permeability?	No	No	Yes	Yes	No	No	No	No
Are there any other **relationships or activities not declared above** that could be perceived potentially to influence your contribution?	No	No	No	No	No	No	No	No
Are there any disclosures requiring a management plan?	**No**	**No**	**No**	**No**	**No**	**No**	**No**	**No**

APPENDIX 3.11: CONFLICT OF INTEREST MANAGEMENT PLAN

All conflicts of interest should be identified, transparently reported and appropriately managed to reduce the risk of bias. The conflict-of-interest management plan is a process for determining if a declared interest represents a conflict of interest, and how this member and their conflict of interest will be managed.

The chair may use one of the following methods to manage any conflict of interest [389]:

- The conflicted member may be excluded from survey items related to the specific area or issue.
- A conflicted member may be excluded from reviewing any recommendations associated with the conflict.
- The conflicted member may contribute in the area of conflict; however, any response or discussions made by a conflicted member will be carefully reviewed by the chair to assess risk of bias.
- In a case where the conflict of interest may cause bias, the conflicted member may not be appointed to the guideline development group.

NHMRC. (2018). Guidelines for Guidelines: Identifying and managing conflicts of interest. Retrieved from https://www.nhmrc.gov.au/guidelinesforguidelines/plan/identifying-and-managing-conflicts-interest

APPENDIX 3.12: TERMS OF REFERENCE FOR STAKEHOLDER GROUP

TERMS OF REFERENCE

Purpose of IP Guideline

The Clinical Practice Guideline for the management of increased intestinal permeability (IP Guideline) aims to improve the treatment of altered intestinal permeability by clinicians in private practice of Australia.

Term

This *Terms of Reference* is effective from 18th of April and continues until the completion of the survey. The survey should take no more than 30 minutes.

Roles and responsibilities of the Stakeholder Group

As a member of the Stakeholder Group you will be involved in providing your expertise and experience for the management of intestinal permeability. You will be expected to;

- Participate in the Stakeholder Group survey,
- Report all relevant conflicts of interests,
- Evaluate the acceptability and feasibility of each recommendation,
- Provide insight on the extent published evidence reflects outcomes,
- Provide feedback on guideline wording to ensure recommendations are understandable.

Communication

As a member of the Stakeholder Group communication will take place over email.

Benefits and reimbursement

As a member of the Stakeholder Group your contribution will be recognised within the published IP Guideline. You will also be provided with a printed and PDF copy of the final IP Guideline. Furthermore, your time will be reimbursed with a $100 visa card.

Member's Details

In order to ensure effective communication, all members of the Guideline Development Group (Working Group and Stakeholder Group) are required to provide their contact details.

Please fill out your details below.

Title:	
Name:	
Email address:	

Phone number:	
Postal Address:	
Qualifications:	
Affiliation:	

By signing below, you agree to the following terms and conditions:

1. I understand my roles and responsibilities as a Stakeholder Group member and will fulfil them to the best of my ability.
2. I have read and understand the *Participant Information Sheet* and any questions have been answered.
3. I have completed the *Member's Details* above.
4. I will notify the Working Group if I am unable to continue my involvement in the IP Guideline.
5. All information provided in the *Terms of Reference* is correct and up to date.

Stakeholder Group Member Name Signature Date

APPENDIX 3.13: STAKEHOLDER SURVEY

Q1 What is your full name?

_____

Q2
There is a total of 38 recommendations. For each recommendation, you will be asked whether you agree with the recommendation, how important and appropriate you think the recommendation is and whether you would change anything about the recommendation. These recommendations were developed for Australian clinicians to use in clinical practice to improve the management of intestinal permeability in Australian adults.

Each recommendation will vary in its categories (Evidence-based recommendations, Consensus-based recommendations and Practice points) and the strength of recommendation (Strong recommendation, Recommendation, Option, Consensus-based recommendation, Practice point and No recommendation). Please download Interpreting the recommendations to aid in the interpretation of the recommendations.
The recommendations are broken down into the following groups: dietary choices, probiotic, prebiotic and synbiotic supplementation, amino acid supplementation, plant-based medicine supplementation, essential fatty acid supplementation and mineral supplementation.

The development of each recommendation is supported by published literature. A summary of the research is found in the following documents. Each document contains the clinical questions that were asked, a summary of the clinical need for the research questions, a summary of the evidence, the risk of bias assessment, NHMRC evidence statement and the justification for the recommendations. If you are unsure about the recommendation while providing your feedback, review the supporting document to better understand the evidence and why the recommendation was developed. Dietary choices supporting information Probiotic, prebiotic and synbiotic supplementation supporting information Amino acid supporting information Plant based medicine supporting information Essential fatty acid supporting information Mineral supporting information

You will be provided with recommendations in the following format at the top of each page. Then you'll be asked 5 question regarding this recommendation.

Category: Evidence-based recommendation
Strength: Recommendation
Recommendation 1.1 People with intestinal permeability should consider consuming no more than 10 standard drinks a week and no more than 4 standard drinks on any one day in accordance with the Australian Dietary Guidelines during the treatment of intestinal permeability.

Q3 The following are dietary based recommendations. A complete summary of the evidence can be downloaded in Dietary choices supporting information

Q4
Category: Evidence-based recommendation
Strength: Recommendation

Recommendation 1.1 *People with intestinal permeability should consider consuming no more than 10 standard drinks a week and no more than 4 standard drinks on any one day in accordance with the Australian Dietary Guidelines during the treatment of intestinal permeability.*

Q5 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q6 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q7 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q8 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q9 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q10 Please describe what you would change about this recommendation

_____

Q11
Category: Consensus-based recommendation
Strength: Consensus-based recommendation

Recommendation 1.2 *People with intestinal permeability may consider limiting or avoiding alcohol consumption during the short-term treatment of intestinal permeability.*

Q12 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q13 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q14 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q15 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q16 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q17 Please describe what you would change about this recommendation

_____

Q18
Category: Evidence-based recommendation
Strength: Recommendation

Recommendation 1.3 *People with intestinal permeability should consider consuming a diet high in dietary fibre from a diverse range of sources.*

Q19 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q20 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q21 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q22 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q23 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q24 Please describe what you would change about this recommendation

_____

Q25
Category: Consensus-based recommendation
Strength: Consensus-based recommendation

Recommendation 1.4 *Clinicians are advised to recommend patients to consume 38g for men and 28g for female of dietary fibre daily while treating patients with intestinal permeability.*

Q26 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q27 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q28 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q29 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q30 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

- ○ Yes (1)
- ○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q31 Please describe what you would change about this recommendation

_____

Q32
Category: Consensus-based recommendation
Strength: Consensus-based recommendation

Recommendation 1.5 *Clinicians are encouraged to recommend gluten-free sources of dietary fibre to patients with confirmed intestinal permeability.*

Q33 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q34 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q35 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the

management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q36 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q37 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q38 Please describe what you would change about this recommendation

_____

Q39
Category: Evidence-based recommendation
Strength: Recommendation

Recommendation 1.6 *People with intestinal permeability should consider consuming the Acceptable Macronutrient Distribution Range of protein (15-25%), fats (20-35%) and carbohydrates (45-65%) in accordance with the Australian Dietary Guidelines.*

Q40 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q41 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q42 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q43 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q44 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q45 Please describe what you would change about this recommendation

_____

Q46
Category: Evidence-based recommendation
Strength: Recommendation

Recommendation 1.7 *People with intestinal permeability should consider NOT consuming a diet high in fat.*

Q47 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q48 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q49 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q50 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q51 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

- ○ Yes (1)
- ○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q52 Please describe what you would change about this recommendation

_____

Q53
Category: Evidence-based recommendation
Strength: Recommendation

Recommendation 1.8 *People with intestinal permeability should consider NOT consuming a diet high in fructose.*

Q54 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q55 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q56 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q57 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q58 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q59 Please describe what you would change about this recommendation

_____

Q60
Category: Evidence-based recommendation
Strength: Option

Recommendation 1.9 *People with intestinal permeability may consider consuming the estimated energy requirements in accordance with the Australian Dietary Guidelines.*

Q61 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q62 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q63 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q64 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q65 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q66 Please describe what you would change about this recommendation

_____

Q67
Category: Evidence-based recommendation
Strength: Option

Recommendation 1.10 *Clinicians may consider using a kilojoule restricted diet in the short-term treatment of people with confirmed intestinal permeability.*

Q68 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q69 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q70 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q71 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q72 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q73 Please describe what you would change about this recommendation

_____

Q74
Category: Evidence-based recommendation
Strength: Strong recommendation

Recommendation 1.11 *Clinicians should only advise a strict gluten-free diet if clinical symptoms or pathology indicate a gluten intolerance, sensitivity or allergy.*

Q75 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q76 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q77 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q78 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q79 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q80 Please describe what you would change about this recommendation

_____

Q81
Category: Evidence-based recommendation
Strength: Strong recommendation

Recommendation 1.12 *Clinicians should only advise a gluten-free diet during the short-term treatment of people with confirmed intestinal permeability that report clinical symptoms in response to the consumption of gluten after the investigation for gluten intolerance, sensitivity or allergy has been carried out.*

Q82 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q83 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q84 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q85 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q86 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q87 Please describe what you would change about this recommendation

_____

Q88
Category: Evidence-based recommendation
Strength: Strong recommendation

Recommendation 1.13 *Clinicians should offer a low gluten diet for the management of people with confirmed intestinal permeability that report no clinical symptoms or pathology indicating a gluten intolerance, sensitivity or allergy.*

Q89 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q90 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q91 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q92 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q93 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

- ○ Yes (1)
- ○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q94 Please describe what you would change about this recommendation

_____

Q95 The following are probiotic, prebiotic and synbiotic recommendations. A complete summary of the evidence can be downloaded in Probiotic, prebiotic and synbiotic supplementation supporting information

Q96
Category: Evidence-based recommendation
Strength: No recommendation

Recommendation 2.1 *There is insufficient evidence to form a recommendation on the use of probiotics as a collective group for the treatment of people with intestinal permeability.*

Q97 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q98 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q99 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q100 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q101 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q102 Please describe what you would change about this recommendation

_____

Q103
Category: Evidence-based recommendation
Strength: Option

Recommendation 2.2 *Clinicians may consider using Saccharomyces boulardii supplementation in the treatment of people with intestinal permeability.*

Q104 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q105 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q106 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q107 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q108 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q109 Please describe what you would change about this recommendation

_____

Q110
Category: Evidence-based recommendation
Strength: Option

Recommendation 2.3 Clinicians may consider the use of effective probiotics for a period of 3 months when treating people with intestinal permeability.

Q111 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q112 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q113 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q114 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q115 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

- ○ Yes (1)
- ○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q116 Please describe what you would change about this recommendation

_____

Q117
Category: Practice point
Strength: Practice point

Recommendation 2.4 Clinicians may consider researching probiotic strains for their effectiveness before using them to treat people with intestinal permeability.

Q118 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q119 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q120 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q121 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q122 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q123 Please describe what you would change about this recommendation

_____

Q124 **Category:** Practice point
Strength: Practice point

Recommendation 2.5 Clinicians may consider the use of probiotics which are supported by pre-clinical research in conjunction with other treatment interventions for the management people with intestinal permeability.

Q125 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q126 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q127 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q128 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q129 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q130 Please describe what you would change about this recommendation

_____

Q131 **Category:** Evidence-based recommendation
Strength: Recommendation

Recommendation 2.6 People with intestinal permeability should consider the consumption of fermented milk products such as kefir.

Q132 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q133 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q134 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q135 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q136 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q137 Please describe what you would change about this recommendation

_____

Q138
Category: Evidence-based recommendations
Strength: Option

Recommendation 2.7 People with intestinal permeability may consider NOT consuming Yakult light®.

Q139 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q140 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q141 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q142 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q143 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q144 Please describe what you would change about this recommendation

_____

Q145
Category: Evidence-based recommendations
Strength: No recommendation

Recommendation 2.8 There is insufficient evidence to form a recommendation on the use of prebiotics as a collective group for the treatment of people with intestinal permeability.

Q146 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q147 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q148 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q149 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q150 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q151 Please describe what you would change about this recommendation

_____

Q152
Category: Practice point
Strength: Practice point

Recommendation 2.9 Clinicians may consider researching prebiotic for their effectiveness before using them in the treat of people with intestinal permeability.

Q153 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q154 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q155 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q156 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q157 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q158 Please describe what you would change about this recommendation

_____

Q159
Category: Practice point
Strength: Practice point

Recommendation 2.10 Clinicians may consider the use of prebiotic which are supported by pre-clinical research in conjunction with other treatment interventions for the management people with intestinal permeability.

Q160 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q161 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q162 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q163 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q164 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

- ○ Yes (1)
- ○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q165 Please describe what you would change about this recommendation

_____

Q166
Category: Consensus-based recommendation
Strength: Consensus-based recommendation

Recommendation 2.11 Clinicians may consider NOT using polydextrose in the treatment of people with intestinal permeability.

Q167 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q168 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q169 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q170 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q171 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q172 Please describe what you would change about this recommendation

_____

Q173
Category: Evidence-based recommendation
Strength: Option

Recommendation 2.12 Clinicians may consider the use of effective synbiotic in the treatment of people with intestinal permeability.

Q174 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q175 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q176 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q177 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q178 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q179 Please describe what you would change about this recommendation

_____

Q180
Category: Evidence-based recommendation
Strength: Option

Recommendation 2.13 Clinicians may consider the use of effective synbiotic for a period of 3 months when treating people with intestinal permeability.

Q181 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q182 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q183 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q184 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q185 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q186 Please describe what you would change about this recommendation

_____

Q187
Category: Evidence-based recommendation
Strength: Option

Recommendation 2.14 Clinicians may consider NOT using polydextrose and Bifidobacterium animalis ssp. lactis 420 in the treatment of people with intestinal permeability.

Q188 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q189 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q190 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q191 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q192 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q193 Please describe what you would change about this recommendation

_____

Q194
Category: Practice point
Strength: Practice point

Recommendation 2.15 Clinicians may consider the use of synbiotic which are supported by pre-clinical research in conjunction with other treatment interventions for the management people with intestinal permeability.

Q195 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q196 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q197 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q198 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q199 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q200 Please describe what you would change about this recommendation

_____

Q201
Category: Evidence-based recommendation
Strength: Recommendation

Recommendation 2.16 Clinicians should consider NOT using probiotics for the treatment of people with nonsteroidal anti-inflammatory drug induced intestinal permeability.

Q202 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q203 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q204 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q205 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q206 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q207 Please describe what you would change about this recommendation

_____

Q208
Category: Evidence-based recommendation
Strength: Recommendation

Recommendation 2.17 Clinicians should consider NOT using prebiotics for the treatment of people with nonsteroidal anti-inflammatory drug induced intestinal permeability.

Q209 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q210 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q211 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q212 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q213 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

- ○ Yes (1)
- ○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q214 Please describe what you would change about this recommendation

_____

Q215
Category: Evidence-based recommendation
Strength: Recommendation

Recommendation 2.18 Clinicians should consider NOT using synbiotics for the treatment of people with nonsteroidal anti-inflammatory drug induced intestinal permeability.

Q216 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q217 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q218 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the

management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q219 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q220 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q221 Please describe what you would change about this recommendation

_____

Q222 The following are amino acid recommendations. A complete summary of the evidence can be downloaded in Amino acid supporting information

Q223
Category: Evidence-based recommendation
Strength: Strong recommendation

Recommendation 3.1 *Clinicians should offer glutamine supplementation for the treatment of people with intestinal permeability.*

Q224 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q225 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q226 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q227 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q228 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q229 Please describe what you would change about this recommendation

_____

Q230
Category: Consensus-based recommendation
Strength: Consensus-based recommendation

Recommendation 3.2 *Clinicians may consider the use of glutamine supplementation in conjunction with other treatment interventions for the management of people with intestinal permeability.*

Q231 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q232 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q233 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q234 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q235 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

- ○ Yes (1)
- ○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q236 Please describe what you would change about this recommendation

_____

Q237
Category: Evidence-based recommendation
Strength: Recommendation

Recommendation 3.3 *Clinicians should consider the use of short-term lactoferrin supplementation for the treatment of people with nonsteroidal anti-inflammatory drug induced intestinal permeability.*

Q238 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q239 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q240 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the

management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q241 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q242 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q243 Please describe what you would change about this recommendation

_____

Q244 The following are plant-based medicine recommendations. A complete summary of the evidence can be downloaded in Plant based medicine supporting information

Q245
Category: Evidence-based recommendation
Strength: No recommendation

Recommendation 4.1 *There is insufficient evidence to form a recommendation on the use of plant-based medicines as a collective group for the treatment of people with intestinal permeability.*

Q246 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q247 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q248 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q249 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q250 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q251 Please describe what you would change about this recommendation

_____

Q252
Category: Practice point
Strength: Practice point

Recommendation 4.2 *Clinicians may consider the use of plant-based medicines which are supported by pre-clinical research in conjunction with other treatment interventions for the management people with intestinal permeability.*

Q253 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q254 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q255 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q256 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q257 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q258 Please describe what you would change about this recommendation

_____

Q259 The following are essential fatty acid recommendations. A complete summary of the evidence can be downloaded in Essential fatty acid supporting information

Q260
Category: Evidence-based recommendation
Strength: No recommendation

Recommendation 5.1 *There is insufficient evidence to form a recommendation on the use of essential fatty acid supplementation for the treatment of people with intestinal permeability.*

Q261 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q262 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q263 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q264 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q265 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

○ Yes (1)

○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q266 Please describe what you would change about this recommendation

_____

Q267 The following are mineral recommendations. A complete summary of the evidence can be downloaded in Mineral supporting information

Q268
Category: Evidence-based recommendation
Strength: Option

Recommendation 6.1 *Clinicians may consider using zinc supplementation in the treatment of people with intestinal permeability.*

Q269 How would you rate your **understanding** of the recommendation?

○ Very poor	○ Poor	○ Neutral	○ Good	○ Very good

Q270 How would you rate your level of **agreement** or **disagreement** with this recommendation?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q271 How would you rate your level of agreement or disagreement with the **appropriateness** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q272 How would you rate your level of agreement or disagreement with the **importance** of this recommendation for clinicians to follow in the management of Australian adults with suspected or diagnosed intestinal permeability?

○ Strongly disagree	○ Disagree	○ Neutral	○ Agree	○ Strongly agree

Q273 In your opinion, would you **change anything** about this recommendation? This may include the wording, the strength of recommendation or any other aspect of the recommendation.

- ○ Yes (1)
- ○ No (2)

Display This Question:

If In your opinion, would you change anything about this recommendation? This may include the wordin... = Yes

Q274 Please describe what you would change about this recommendation

_____

APPENDIX 4.1: MANUSCRIPT OF ORIGINAL RESEARCH

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Original Article

Health-seeking behaviour, views and preferences of adults with suspected increased intestinal permeability: A cross-sectional survey of Australian adults

Bradley Leech*, Erica McIntyre, Amie Steel, David Sibbritt

Australian Research Centre in Complementary and Integrative Medicine, Faculty of Health, University of Technology Sydney, Ultimo, Australia

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ABSTRACT

Background: The public health consequence of increased intestinal permeability (IP) is currently limited by the lack of patient-centred research. This study aims to describe the health-seeking behaviour of Australian adults with suspected IP.

Methods: A cross-sectional survey of 589 Australian adults who have been diagnosed with IP or have suspected (undiagnosed) IP.

Results: The majority (56.2%) of participants with suspected IP reported self-diagnosing their condition, with the majority (56.7%) of these participants preferring to be assessed using an accurate method by a general practitioner or naturopath. On average, Australian adults with suspected IP spent 11.1 (95% CI: 9.5, 12.8) years between first suspecting IP and receiving a formal diagnosis. Over the previous 12 months, participants spent an average of $699 on consultation fees, $2176 on dietary supplements for the treatment of IP, and an average of $287 on the assessment of IP. Furthermore, participants who find it difficult to live on their available household income spent significantly more (mean=$2963) on dietary supplements compared to participants who find it easy to live on their available household income ($1918) (p=0.015).

Conclusion: The investigation of Australian adults with suspected IP found the majority of participants experienced a considerable length of time between first suspecting IP and receiving a diagnosis of IP. The out-of-pocket expenditure associated with the management of IP suggests a financial burden for people with suspected IP. The results of this study provide novel patient-centred considerations that can be used to inform a clinical practice guideline for the management of IP.

1. Introduction

The single layer of epithelial cells that separate the internal and external environment of the small intestine is renewed every four to five days, playing an essential role in maintaining intestinal homeostasis.[1] Increased intestinal permeability (IP) involves the disassembling of tight junction proteins between the cells of the small intestine, resulting in a loss of intestinal barrier integrity.[2] With an estimated prevalence of 10-87% in health conditions with a known association,[3] altered IP has been suggested to play an important role in health and disease in both public and private healthcare.[4]

The clinical relevance and consequence of altered IP remain a controversial topic within conventional medicine.[5] Yet, published literature continues to identify IP as a target for disease prevention and therapeutic intervention.[4] IP has been suggested to precede the onset of a number of chronic health conditions such as Crohn's disease,[6] liver disease,[7] type 1 diabetes,[8–11] coeliac disease,[9] rheumatoid arthritis,[12] gestational diabetes,[13] and diarrhoea-predominant irritable bowel syndrome.[14,15] Altered IP is also associated with autoimmune conditions, metabolic conditions, liver diseases, and gastrointestinal conditions.[3,16] Although IP is a reaction within the small intestine, many of the measurable and clinically relevant risk factors are systemic, suggesting that IP is more than a digestive health issue and a possible feature of disease.[16]

* Corresponding author at: Australian Research Centre in Complementary and Integrative Medicine, Faculty of Health, University of Technology Sydney, 15 Broadway, Ultimo, NSW, 2007, Australia.
E-mail address: Bradgleech@gmail.com (B. Leech).

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Previous research has investigated the assessment and management of IP from the practitioner standpoint, where practitioners acknowledge the involvement of IP in many health conditions found in clinical practice.[17,18] Within clinical practice, the pathology tests available are invasive, require patients to pay out-of-pocket, and involve a substantial amount of time to perform.[18] Practitioners that frequently treat IP in clinical practice are reported to avoid using validated pathology tests due to the financial cost to the patient and prioritise case history to diagnose IP.[18] While the frequency of methods used by patients, including the accuracy of self-diagnosis remains unknown, the self-diagnosis of other chronic illnesses such as diabetes is considered to be somewhat accurate.[19] Furthermore, no research to date has considered patients views and preferences towards the assessment and management of IP, resulting in knowledge gaps for evidence-based practice. Incorporating patients views and preferences in the decision-making process is often overlooked however, a positive impact on the outcome of healthcare is observed when patients views and preferences are considered.[20] As such, this study aims to describe the health-seeking behaviour of Australian adults with suspected IP while also exploring the views and preferences surrounding the assessment and management of IP.

2. Methods

2.1. Study design and setting

A cross-sectional study design using an online self-reported survey was utilised with approval from the Human Research Ethics Committees (HREC) of the University of Technology Sydney (#ETH19-4012).

2.2. Participants and recruitment

Participants were recruited via social media platforms and a purpose-built webpage, with snowball sampling methods also used. The survey was open for two months between September 2019 and November 2019. Eligibility to participate in this study required participants to either suspect or know they have altered IP, be aged 18 years or more, living in Australia and have internet access. Survey responders with incomplete demographic characteristics, accounting for <5% of total data were excluded from analysis. This study was designed to capture people that may have suspected IP or confirmed IP, to best reflect the type of patients that present to clinical practice for the treatment of IP.[18]

2.3. Survey instrument

The developed online survey utilised the questionnaire items which were obtained from published literature and modified to suit Australians with suspected IP.[17,18] To improve the survey's reliability, standardised five-point Likert scales were used for scaling questions. The survey included three main domains: demographic characteristics, diagnosis of IP, and the financial expenditure related to IP. The questionnaire was first pilot tested using lay people to assess the time required to complete the survey and language clarity, with corrections made accordingly.

2.3.1. Demographic characteristics

The participants were asked about their gender, age, height, and weight. Body mass index (BMI) was calculated from height and weight measurements. BMI was then categorised to underweight, healthy weight, overweight, and obese.[21] The participants were also asked their country of birth, the state or territory where they live, and whether they live in an urban, rural or remote location.

2.3.2. Diagnosis of increased intestinal permeability

Participants were asked a number of questions in relation to the assessment of IP including: the year they believed their IP started, the year their IP was diagnosed, the method used to confirm their IP, at what point their IP was assessed, the number of times their IP was assessed, and the qualifications of the practitioner involved in the assessment of their IP. In addition, participants preference for IP testing method characteristics, the preferred method and time point for IP assessment, and the qualifications of their preferred practitioner were all asked. To gauge the preference and importance towards being assessed for IP and the likelihood of treatment adherence if results returned a positive test of altered IP, five-point Likert scales were used. The term 'assessed' and 'assessment' are used throughout this article to describe the action participants used for measuring, evaluating or identifying IP.

2.3.3. Financial expenditure

A number of items participants were asked to report: the out-of-pocket expenditure of treating IP, practitioner consultation fees, and cost of measuring IP. Participant's income manageability was determined by how well they manage their household income, categorised as 'difficult all the time', 'difficult some of the time', 'not too bad', or 'easy'. The amounts are reported in Australian dollars (AUD).

2.4. Data collection

The survey was administered through the online platform *SurveyGizmo*. After data collection, data was exported to a statistical software program STATA® 16 for data checking and statistical analyses.

2.5. Statistical analyses

Responses to questionnaire items were reported as means, standard deviations, 95% confidence intervals (CIs) or frequencies and percentages. Chi-square analysis was used for tests of association between categorical variables and Student's t-tests were used for continuous variables across a binary variable. Ordinal variables such as those on Likert scales were assessed with non-parametric tests, including Mann-Whitney U test and the Wilcoxon signed ranks test, where appropriate. Analysis of variance (ANOVA) was used to measure the difference between a continuous variable across a categorical variable.

3. Results

3.1. Demographic characteristics

A total of 982 people responded to the survey, of which 393 responses did not meet the eligibility criteria or were classified as having too much incomplete data, leaving a total of 589 participants. Most participants were female (n=548, 93%), living within an urban area (n=416, 70.6%) in either New South Wales (n=175, 29.7%) or Queensland (n=161, 27.3%) (Table 1). The mean age of the participants was 45.0 (SD=12.1) with a mean BMI of 27.0 (SD=6.9). The income manageability of participants was described most commonly as 'easy or not too bad' (n=209, 46.5%) and 'difficult some of the time' (n=145, 32.3%). Half the surveyed population reported altered IP as their primary health concern (n=300, 50.9%) with a range of other autoimmune, inflammatory gastrointestinal, and metabolic conditions reported for the other half (Table 1).

3.2. Diagnosis of increased intestinal permeability

The most frequently used methods to diagnose IP were self-diagnoses (n=330, 56.2%) and case history, according to a health-

Table 1
Demographic characteristics of study participants (n=589)

Demographics	Distribution of responses	
	n	%
Gender		
Female	548	93.0
Male	41	7.0
BMI classification		
Underweight	19	3.3
Healthy weight	268	46.1
Overweight	138	23.8
Obese	156	26.9
Country of birth		
Australia	476	81.0
Other	112	19.0
State or territory		
New South Wales	175	29.7
Queensland	161	27.3
Victoria	103	17.5
Western Australia	63	10.7
South Australia	36	6.1
Australian Capital Territory	23	3.9
Tasmania	18	3.1
Northern Territory	10	1.7
Area of residence		
Urban	416	70.6
Rural	161	27.3
Remote	12	2.0
Income manageability		
Easy or not too bad	209	46.5
Difficult some of the time	145	32.3
Difficult all the time	95	21.2
Primary health concern		
Increased intestinal permeability	300	50.9
Other autoimmune diseases	40	6.8
Hashimoto's thyroiditis	28	4.8
Gastrointestinal issues	24	4.1
Chronic fatigue syndrome	21	3.6
Rheumatoid arthritis	18	3.1
Obesity	15	2.6
Mental health	13	2.2
Hormonal issues	10	1.7
Fibromyalgia	9	1.5
Gastrointestinal *Candida albicans*	8	1.4
Psoriatic arthritis	7	1.2
Mould exposure	7	1.2
Irritable bowel syndrome	6	1.0
Ankylosing spondylitis	6	1.0
Asthma	6	1.0
Food intolerances	6	1.0
Cardiovascular disease	6	1.0
Mast cell activation syndrome	6	1.0
Other health conditions	53	9.0
	Mean	**SD (range)**
Age in years	45.0	12.1 (18-82)
Body Mass Index (BMI)	27.0	6.9 (15.4-64.5)

Table 2
Health seeking behaviour for the assessment and management of increased intestinal permeability by Australian adults (n=589)

Health Seeking Behaviour	Distribution of Response	
	n	%
Method of assessment (n=587)		
Self-diagnosed	330	56.2
Case history according to a practitioner	130	22.2
IgG food sensitivity test	29	4.9
Hemaview - live blood analysis	23	3.9
Stool zonulin	22	3.8
Lactulose|mannitol urine test	17	2.9
I don't know	16	2.7
Iridology	12	2.0
Serum zonulin	4	0.7
Kinesiology	4	0.7
Stage that IP was measured (n=134)		
Before treatment	102	17.3
During the treatment phase	24	4.1
After treatment was completed	8	1.4
Number of times measured for IP (n=589)		
0	459	77.9
1	104	17.7
2 +	26	4.4
Time between initial and second assessment (n=26)		
Between 1 and 6 months	2	7.7
Between 6 and 12 months	11	42.3
Between 12 and 24 months	6	23.1
Over 2 years	7	26.9
Year IP was diagnosed (n=237)		
< 3 years	140	59.1
4-6 years	46	19.4
7-9 years	22	9.3
> 10 years	29	12.2
Year participant believe IP started (n=498)		
< 3 years	84	16.9
4-6 years	82	16.5
7-9 years	77	15.5
> 10 years	255	51.2

IP: increased intestinal permeability

care practitioner (n=130, 22.2%) (Table 2). From the participants that were assessed for IP, 17.3% (n=102) were assessed before receiving treatment, 4.1% (n=24) during the treatment phase, and only 1.4% (n=8) after treatment was completed. Of the participants who were diagnosed with IP, 59.1% (n=140) reported being diagnosed within the last three years. However, on average, participants with suspected IP spent 11.1 (95% CI: 9.5, 12.8) years between first suspecting IP and receiving a diagnosis. No statistically significant difference was found between the length of time between when participants first suspected IP to the year they were diagnosed and whether they were diagnosed by a medical practitioner or another healthcare practitioner (p=0.120). The vast majority of participants were not assessed for IP (n=459, 77.9%) with only 17.7% (n=104) assessed once, and 4.4% (n=26) assessed more than twice. For the participants that were assessed two or more times, the second assessment of IP typically took place between 6 and 12 months (n=11, 42.3%). A significant association between the number of times IP was assessed and the person (practitioner or self) who diagnosed IP was found (p<0.001). Specifically, healthcare practitioners and medical practitioners more frequently assessed IP (n=74, 33.9%; n=39, 33.6%, respectively) compared to those who self-diagnosed (n=4, 1.9%).

3.3. Practitioners involved in the diagnosis of increased intestinal permeability

Most participants (n=374, 67.4%) first suspected they had IP, whereas 32.6% (n=181) had a practitioner first suggest IP as a possible diagnosis. Participants were most frequently diagnosed with IP by self-diagnosing (n=274, 47.9%), followed by a naturopath (n=207, 36.2%), integrative medicine practitioner (n=82, 14.3%), nutritionist (n=53, 9.3%), and general practitioner (n=50, 8.7%) (Table 3). Most participants preferred their IP to be assessed by a naturopath (n=363, 63.5%), followed by a general practitioner (n=310, 54.2%), integrative medicine practitioner (n=259, 45.3%), nutritionist (n=225, 39.3%), gastroenterologist (n=221, 38.6%) or a dietitian (n=162, 28.3%). From the participants that self-diagnosed,

Table 3
Practitioners involved in the diagnosis of increased intestinal permeability (n=572)

Who diagnosed increased intestinal permeability	Diagnosis of increased intestinal permeability					
	Initial diagnosis		Total preferred practitioner		Preferred practitioner for self-diagnosed	
	n	%	n	%	n	%
Naturopath	207	36.2	363	63.5	118	56.7
Integrative medicine practitioner	82	14.3	259	45.3	75	36.1
Nutritionist	53	9.3	225	39.3	69	33.2
General practitioner	50	8.7	310	54.2	118	56.7
Herbalist	19	3.3	101	17.7	31	14.9
Kinesiologist	19	3.3	86	15.0	22	10.6
Dietitian	17	3.0	162	28.3	60	28.9
Chinese medicine practitioner	15	2.6	110	19.2	35	16.8
Homeopath	13	2.3	77	13.5	24	11.5
Acupuncturist	11	1.9	78	13.6	26	12.5
Chiropractor	11	1.9	58	10.1	18	8.7
Gastroenterologist	10	1.8	221	38.6	90	43.3
Ayurvedic practitioner	6	1.1	73	12.8	25	12.0
Osteopath	4	0.7	40	7.0	13	6.3
Nurse	3	0.5	53	9.3	19	9.1
Nurse practitioner	3	0.5	52	9.1	18	8.7
Pharmacist	1	0.2	54	9.4	14	6.7
Self-diagnosed	274	47.9	-	-	-	-

their preferred practitioner for the assessment of IP was a general practitioner (n=118, 56.7%) or a naturopath (n=118, 56.7%).

3.4. Expenditures related to the assessment and management of intestinal permeability

On average, participants reported spending $698.78 on consultation fees and $2175.96 on dietary supplements over the previous 12 months (Table 4). There was a statistically significant difference between income manageability and the average amount spent on dietary supplements. Specifically, participants who find it 'difficult all the time' to live on their available household income spend significantly more (mean=$2963.28) on dietary supplements over 12 months compared to participants who described their income manageability as 'easy or not too bad' ($1918.56; p=0.015). No significant differences were found between who diagnosed their IP and the average amount spent on dietary supplements in the previous 12 months (p=0.167). However, participants that were diagnosed by a medical practitioner spent on average $2309.16 on dietary supplements over the previous 12 months, whereas those who were self-diagnosed spent on average of $1793.40. Participants on average spent $286.76 on the assessment of IP with no significant difference found with either income manageability or the source of diagnosis.

There is a statistically significant difference between who diagnosed their IP and the average amount spent on consultation fees in the previous 12 months (p<0.001). Specifically, those who were diagnosed by a medical practitioner, or another kind of healthcare practitioner spent significantly more (mean=$980.63 and $996.29 respectively) on consultation fees compared to participants who self-diagnosed IP ($226.45). No difference was found for the average amount spent on consultation fees between a medical practitioner or healthcare practitioners.

3.5. Views and preferences towards the costs involved with intestinal permeability

Participants reported that the cost involved in testing IP is 'very important' in their decision to be tested (n=260, 58.8%), with many participants (n=218, 48.8%) indicating they are willing to spend between $51.00 and $150.00 on the testing procedure for IP (Table 4). However, the importance of cost in the decision to be tested decreased as income manageability increased (p<0.001). Furthermore, as income manageability increased, so did the amount participants were willing to spend on the testing procedure for IP (p<0.001).

Regardless of income manageability, participants reported a preference towards allocating finances to dietary treatment interventions (n=309, 70.6%) for the management of IP followed by dietary supplements (n=265, 60.9%) and lifestyle treatments (n=240, 55.4%) (Table 4). Although half the participants (n=248, 56.5%) reported the financial allocation for the assessment of IP to be 'very important', increased income manageability was associated with the preference towards allocating finances to the assessment of IP (p=0.018). Irrespective of income manageability, participants reported medication use to be 'not important' for financial allocation (n=296, 71.6%).

3.6. Views and preferences towards the assessment and management of intestinal permeability

The majority of participants (n=527, 89.6%) would prefer to be assessed for IP regardless of income manageability (p=0.054) with 75.0% (n=442) reporting the assessment of IP to be 'very important' (Table 5). Accuracy (n=554, 94.9%), accessibility (n=476, 81.4%), and affordability (n=408, 69.5%) were all commonly reported to be 'very important' characteristics for the assessment of IP; whereas non-invasive methods (n=470, 80.6%) and length of time involved to perform the assessment (n=352, 61.1%) were both commonly reported to be 'not important' characteristics for the assessment of IP. Participants further commonly reported the preference to be assessed for IP using blood pathology (n=459, 78.1%) followed by urine collection (n=354, 60.2%) and a stool test (n=325, 44.3%), with a case history assessment by a practitioner (n=242, 41.2%) to be the least preferred method of IP assessment. The time point that participants commonly prefer to be assessed for IP were; before receiving treatment for IP (n=354, 60.1%), for monitoring disease (n=231, 39.2%), when asked by the patient (n=213, 36.2%), for monitoring IP (n=204, 34.6%), after receiving treatment for IP (n=169, 28.7%), when advised by the practitioner (n=160, 27.2%), and during the treatment of IP (n=117, 19.9%).

The majority of participants (n=549, 93.2%) reported they would be 'very likely' to adhere to a treatment protocol if assessed and diagnosed with altered IP (Table 5). In terms of the preferred

Table 4
Expenditures related to the assessment and management of increased intestinal permeability and association with income manageability (n=447)

Expenditures	Mean expenses per person		Income manageability						
			Difficult all the time		Difficult some of the time		Easy or not too bad		p-value
	n	Mean	n	Mean	n	Mean	n	Mean	
Expenses for the management of IP in the previous 12 months									
Consultation fees	424	$698.78	91	$903.49	136	$745.99	197	$571.62	0.057
Dietary supplements	309	$2175.96	67	$2963.28	96	$2019.36	146	$1918.56	0.015
Expenses for the assessment of IP									
All assessment methods	74	$286.76	13	$238.46	24	$315.21	37	$285.27	0.847
Food sensitives - IgG	19	$515.53	4	$385.00	4	$738.75	11	$481.81	0.746
Stool zonulin	16	$329.38	4	$252.50	4	$500.00	8	$282.50	0.089
Hemaview - live blood analysis	15	$204.33	1	$120.00	8	$243.13	6	$166.66	0.620
Iridology	10	$167.50	3	$110.00	2	$100.00	5	$229.00	0.515
Lactulose/mannitol urine test	9	$115.00	-	-	4	$63.75	5	$156.00	0.030
Kinesiology	4	$77.50	1	$100.00	2	$105.00	1	$0.00	0.484
Serum zonulin	1	$70.00	-	-	-	-	1	$70.00	-
	n	%	n	%	n	%	n	%	p-value
Importance of cost in the decision to be tested									
Very important	260	58.8	72	75.8	94	65.3	94	46.3	<0.001
Important	112	25.3	13	13.7	37	25.7	62	30.5	
Not important	70	15.8	10	10.5	13	9.0	47	23.2	
Amount willing to spend on IP assessment									
$0-$50.00	107	23.9	43	45.3	38	26.2	26	12.6	<0.001
$51.00-$150.00	218	48.8	41	43.2	67	46.2	110	53.1	
$151.00 or over	122	27.3	11	11.6	40	27.6	71	34.3	
Preference towards expense allocation to									
Dietary treatments									
Very important	309	70.6	65	70.7	101	70.1	143	70.8	0.953
Important	101	23.1	20	21.7	33	22.9	48	23.8	
Not important	28	6.4	7	7.6	10	6.9	11	5.5	
Dietary supplements									
Very important	265	60.9	53	60.2	90	63.4	122	59.5	0.968
Important	107	24.6	22	25.0	33	23.2	52	25.4	
Not important	63	14.5	13	14.8	19	13.4	31	15.1	
The assessment of IP									
Very important	248	56.5	46	51.7	88	61.1	114	55.3	0.018
Important	117	26.7	18	20.2	39	27.1	60	29.1	
Not important	74	16.9	25	28.1	17	11.8	32	15.5	
Lifestyle treatments									
Very important	240	55.4	58	64.4	78	55.3	104	51.5	0.349
Important	135	31.2	22	24.4	43	30.5	70	34.6	
Not important	58	13.4	10	11.1	20	14.2	28	13.9	
Medications									
Very important	56	13.6	13	15.3	18	13.7	25	12.8	0.841
Important	61	14.8	11	12.9	17	13.0	33	16.8	
Not important	295	71.6	61	71.8	96	73.3	138	70.4	

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method of treating IP, participants 'strongly prefer' the use of dietary products (n=392, 82.2%), followed by lifestyle habits (n=357, 76.5%), and dietary supplements (n=324, 68.6%). On the contrary, 82.8% (n=351) of participants 'slightly prefer' the use of medications to treat IP, representing the least preferred method of IP treatment.

4. Discussion

This is the first study to describe the health-seeking behaviours and explore the views and preferences of adults with suspected or diagnosed IP. The results of this study suggest inconsistencies between the healthcare provided to Australian adults with suspected IP and the healthcare this patient population would prefer to receive. Most notably, the majority of participants experienced a considerable length of time between first suspecting IP and receiving a diagnosis of IP. They also reported challenges involved in the accurate diagnosis of IP and the out-of-pocket expenditure associated with IP.

4.1. Diagnosis of increased intestinal permeability

Our results indicate that those participants without a formal diagnosis of IP are self-diagnosing; however, have a desire to be assessed using an accurate method by a healthcare practitioner. This discrepancy in the assessment of IP may be contributed in part to the common practices of healthcare practitioners. Practitioners that frequently treat IP in clinical practice avoid measuring IP due to the financial cost to the patient and prioritise case history assessment for diagnosing IP.[18] However, the results of this study suggest that Australian adults with suspected IP are willing to allocate finances to an accurate and accessible method of IP assessment before receiving treatment. The inconsistencies between the healthcare provided to Australian adults with suspected IP and their pre-

Table 5
Views and preferences towards the assessment and management of increased intestinal permeability by Australian adults (n=589)

Views and preferences	Distribution of Responses	
	n	%
Preference to be assessed for IP		
Prefer to be assessed	527	89.6
Prefer not to be assessed	61	10.4
Importance to be assessed for IP		
Very important	442	75.0
Important	78	13.2
Not important	69	11.7
Likelihood of adhering to treatment if assessed and positive for IP		
Very likely	549	93.2
Neutral	23	3.9
Very unlikely	17	2.9
Importance of various assessment characteristics for IP		
Accuracy		
Very important	554	94.9
Important	25	4.3
Not important	5	0.9
Accessibility		
Very important	476	81.4
Important	78	13.3
Not important	31	5.3
Affordability		
Very important	408	69.5
Important	122	20.8
Not important	57	9.7
Time involved		
Very important	113	19.6
Important	111	19.3
Not important	352	61.1
Non-invasive method		
Very important	56	9.6
Important	57	9.8
Not important	470	80.6
Preference of assessment method		
Blood test	459	78.1
Urine collection	354	60.2
Stool test	325	55.3
Case history	242	41.2
Preference for assessment time point		
Before treatment	354	60.1
For monitoring disease	231	39.2
When asked by the patient	213	36.2
For monitoring IP	204	34.6
After treatment	169	28.7
When advised by the practitioner	160	27.2
During treatment	117	19.9
Preference for treatment method		
Dietary products		
Strongly prefer	392	82.2
Prefer	50	10.5
Slightly prefer	35	7.3
Lifestyle habits		
Strongly prefer	357	76.5
Prefer	73	15.6
Slightly prefer	37	7.9
Dietary supplements		
Strongly prefer	324	68.6
Prefer	88	18.6
Slightly prefer	60	12.7
Medication		
Strongly prefer	46	10.9
Prefer	27	6.4
Slightly prefer	351	82.8
Important areas for practitioners to comprehend		
Dietary treatments for IP		
Very important	395	95.2
Important	18	4.3
Not important	2	0.5
Lifestyle treatments for IP		
Very important	389	94.0
Important	23	5.6
Not important	2	0.5

(continued on next page)

Table 5 (continued)

Views and preferences	Distribution of Responses	
	n	%
Signs and symptoms of IP		
Very important	390	93.8
Important	25	6.0
Not important	1	0.2
Biomarkers associated with IP		
Very important	378	91.5
Important	28	6.8
Not important	7	1.7
Risk factors for IP		
Very important	373	90.8
Important	34	8.3
Not important	4	1.0
Methods to accurately assess IP		
Very important	376	90.4
Important	31	7.5
Not important	9	2.2
Conditions associated with IP		
Very important	376	90.2
Important	36	8.6
Not important	5	1.2
Dietary supplements for IP		
Very important	366	89.1
Important	33	8.0
Not important	12	2.9
Time point that IP should be assessed		
Very important	362	87.4
Important	45	10.9
Not important	7	1.7
Individuals that require to be assessed for IP		
Very important	354	85.1
Important	53	12.7
Not important	9	2.2
Medications for IP		
Very important	215	52.4
Important	85	20.7
Not important	110	26.8

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ferred healthcare suggest the preferences of the consumer may not always be considered.

As with other health-related conditions, IP is subject to underdiagnosis, over-diagnosis and misdiagnosis within clinical practice.[22,23] Of particular concern from our findings is the high rate of self-diagnosis of IP. This high self-diagnosis rate may result in a misdiagnosis, causing potential anxiety to the patient, unnecessary treatment burden when not required or result in other more serious health conditions being undiagnosed. The high self-diagnosis rate may also have an overall negative effect on practitioner-patient relationship with the potential utilisation of inaccurate or inappropriate treatments.[24] Our study also revealed that Australian adults with suspected IP would prefer a general practitioner or naturopath to assess them for IP. However, the lack of acknowledgement of IP by medical practitioners[5] may be a driving factor for the large number of Australian adults with suspected IP not receiving a formal diagnosis and a contributing factor as to why it takes 11 years for IP to be diagnosed. Whether the length of time for a formal diagnosis of altered IP is contributed to behaviours of the patient or the practitioner is unknown; however, the shortage of validated testing methods and no gold standard testing method are factors influencing healthcare practitioners not to measure IP and to treat regardless.[18]

A common practice for practitioners is the use of case history in the diagnosis of a number of health conditions, especially functional bowel disorders.[25,26] Even with the extensive algorithms of patients case history, there is still a poor agreement between practitioners and the diagnostic criteria of functional bowel disorders.[27] A concern when applying an algorithmic model of diagnosis to IP is that there is no validated algorithm and the associated case history features of IP remain uncertain, especially as previously perceived symptoms of IP are not associated with diagnostic markers of IP.[16,18] The clinical similarities between gastrointestinal conditions[26] and the under examined clinical features of IP, limits the accuracy of case history as a diagnostic method for IP.

4.2. Financial expenditure of increased intestinal permeability

The out-of-pocket expenditure associated with the assessment and management of IP suggests a financial burden for Australian adults with suspected IP. Although a financial burden calculation is not possible with the data collected in this study, other Australian based studies provide further support for a potential financial burden. For instance, the mean out-of-pocket expenditure for the assessment and management of suspected IP is similar to the amount spent on chronic health conditions in Australia.[28-30] Furthermore, the out-of-pocket expenditure for consultation fees and dietary supplements over a 12 month period is greater than the mean annual expense for Australian adults with gastrointestinal disorders.[31] As Australia has one of the highest out-of-pocket expenditure for medication in the world,[28,32] healthcare practitioners should consider the out-of-pocket expenses related to IP management, especially people with a low income manageability.

The results of this study suggest a significant difference between the income manageability and the average amount spent on dietary supplements, with those who find it 'difficult all the time' to live on their available household income spending significantly more on dietary supplements compared to the 'easy or not

too bad' income groups. Other studies suggest people with poor financial status are more likely to face a financial burden in relation to the out-of-pocket expenditure.[29,33] Whether a person's income manageability is a cause or consequence for the out-of-pocket expenditure on IP remains unknown; however, is a worthy area for further investigation.

4.3. Views and preferences of increased intestinal permeability

The results of this study suggest that Australian adults with suspected IP place little importance or value on medication use for the treatment of IP. The strong aversion towards medication use highlights a potential barrier for future pharmacological treatments under development.[34,35] Whether Australian adults with suspected IP will use such medication remains an area for future research. However, what this study does suggest is dietary products (dietary interventions) are the preferred method for the treatment of IP. Dietary interventions are also the most frequently used type of treatment for IP by practitioners in Australia,[17] highlighting agreement between the preferred treatment method for IP and the care given by healthcare practitioners. Utilising the results of this study, patients' views and preferences can help inform the development of a clinical practice guideline for the assessment and management of IP.

4.4. Limitations

There are a number of potential limitations of our study that need to be considered when interpreting our findings. Our sample has a greater percentage of females than the Australian general population, hence caution is required if generalising findings to the Australian population. Although this study aimed to explore Australian adults with suspected IP, whether or not participants have diagnosed IP is unknown. Therefore, these results are more relevant to those who suspect they have IP rather than those with a confirmed diagnosis. Self-reported data collection has the potential for recall bias. However, as this was the first survey to describe the health-seeking behaviours of Australian adults with suspected IP, this study does provide new and important information, thus advancing the research agenda on this topic.

4.5. Conclusion

The investigation of Australian adults with suspected IP has highlighted major inconsistencies between the healthcare provided and the healthcare this patient population would prefer to receive, especially regarding the diagnosis of IP. Most notably, the majority of participants experienced a considerable length of time between first suspecting IP and receiving a diagnosis of IP. The out-of-pocket expenditure associated with the assessment and management of IP suggests a financial burden for people with suspected IP. The results of this study provide novel patient-centred considerations that can be used to inform a clinical practice guideline for the assessment and management of IP as an important public health initiative.

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Author contributions

Conceptualization: BL, EM, AS, and DS. Methodology: BL, EM, AS, and DS. Investigation: BL, EM, AS, and DS. Formal analysis: BL, EM, AS, and DS. Writing – original draft: BL. Writing – review & editing: BL, EM, AS, and DS.

Conflict of interest

The authors declare that they have no conflict of interests.

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Ethical statement

This study was approved by the Human Research Ethics Committees (HREC) of the University of Technology Sydney (#ETH19-4012).

Data availability

Data associated with this study will be available upon request.

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APPENDIX 5.1: MANUSCRIPT OF ORIGINAL RESEARCH

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The Subjective Well-being and Health-Related Quality of Life of Australian Adults with Increased Intestinal Permeability and Associations with Treatment Interventions

Bradley Leech, BHSc (Hons),[1,i] Erica McIntyre, PhD,[1,2,ii] Amie Steel, PhD,[1,iii] and David Sibbritt, PhD[1]

Abstract

Objective: The integrity and function of the gastrointestinal system is important in disease prevention and management. This study aims to describe the management methods used by Australian adults with suspected increased intestinal permeability (IP) and the association with subjective wellbeing (SWB) and health-related quality of life (HRQoL).

Design and Setting: Cross-sectional survey of Australian adults diagnosed with IP or have suspected (undiagnosed) IP.

Outcome Measures: Questionnaire items investigating demographic characteristics, self-reported outcome of IP and treatment methods used to manage IP. Participants' HRQoL and SWB according to the 20-Item Short Form Health Survey (SF-20) and Personal Wellbeing Index-Adult (PWI-A) scale, respectively.

Results: Participants ($n = 589$) frequently used dietary products (87.9%), dietary supplements (72.9%) and lifestyle therapies (54.6%) for managing IP. Participants had lower (i.e., worse) mean SWB scores for all domains compared to the Australian population ($p < 0.001$). The number of days IP reported to affect daily living was negatively correlated with SWB and HRQoL ($p < 0.001$). Participants that reported an improvement in their IP in the previous 12 months were more likely to be treated by a healthcare practitioner (OR = 2.04, $p = 0.015$), use dietary supplements (OR = 2.66, $p = 0.003$), participate in vigorous exercise (OR = 2.99, $p < 0.001$) and employ vagus nerve stimulation (OR = 3.10, $p = 0.010$). Conversely, they were less likely to consume gluten (OR = 0.35, $p < 0.001$) or use nonsteroidal anti-inflammatory drugs (OR = 0.35, $p = 0.022$). Self-reported improvement of IP ($\beta = 10.70$, $p < 0.001$) and use of dietary products ($\beta = 12.12$, $p = 0.008$) were predictors of a higher level of SWB.

Conclusions: Altered IP may pose a greater health burden than previously thought, with poor SWB and HRQoL reported in Australian adults with self-reported IP. Our results highlight the potential clinical relevance and consequence of altered IP, providing the first indication of a possible relationship between altered IP and both SWB and HRQoL.

Keywords: intestinal permeability, intestinal barrier dysfunction, subjective wellbeing, health-related quality of life

[1]Faculty of Health, Australian Research Centre in Complementary and Integrative Medicine, University of Technology Sydney, Ultimo, Australia.
[2]Institute for Sustainable Futures, University of Technology Sydney, Ultimo, Australia.
[i]ORCID ID (https://orcid.org/0000-0003-4866-9638).
[ii]ORCID ID (https://orcid.org/0000-0002-3970-9979).
[iii]ORCID ID (https://orcid.org/0000-0001-6643-9444).

Introduction

The health of the gastrointestinal system has become a target of interest for disease prevention.[1] One specific gastrointestinal target area is the integrity of the intestinal barrier of the small intestine. During increased intestinal permeability (IP), the tight junction proteins between the cells of the small intestine disassemble in response to the protein zonulin.[2] The single layer of epithelium cells in the small intestine contributes to the biochemical and physical barrier to the array of foreign pathogens, allergens, and other toxins.[3] The prevalence of altered IP has been suggested to be 10%–87% in health conditions with a known association.[4] During a loss of intestinal integrity, a cascade of reactions contributes to systemic symptoms and disease progression with the mitigation of zonulin that is suggested to inhibit or reduce disease onset.[5,6] Although no defined symptoms of IP have been identified,[5,7] a range of risk factors are known to be associated with altered IP.[5] The clinical risk factors associated with IP provide a potential platform for treatment interventions and areas for further investigation.

The management of altered IP may involve the use or avoidance of dietary products (e.g., increasing dietary fiber, avoidance of gluten and alcohol), lifestyle therapies (e.g., stress management, vagus nerve stimulation), dietary supplements (e.g., vitamin A, probiotics, *Curcuma longa*, fish oil), and medication evaluation (e.g., avoidance of nonsteroidal anti-inflammatory drugs [NSAIDs] and antibiotics or the use of larazotide acetate).[6,8–10] These methods are proposed to have multiple direct and indirect modulatory actions that regulate intestinal integrity.[8,9] Many of the treatments used by practitioners for the management of IP have previously been shown to align with preclinical research.[9] Although these treatment methods are frequently used in clinical practice, there still remains limited evidence for the effective management of altered IP. A broad health services research-based study may help identify the potential areas for further clinical trials.[11]

The clinical relevance and consequence of altered IP in clinical practice have recently been questioned,[12] despite identified associations between IP and a wide range of health conditions.[4] Questions regarding the clinical relevance and consequence of altered IP may stem from a low level of awareness and understanding regarding the potential effect of altered IP on individuals, especially their quality of life (QoL) and subjective well-being (SWB). QoL is an important contributor to overall disease burden alongside financial burden, mortality, and morbidity.[13,14] Health-related quality of life (HRQoL) is a multidimensional concept that measures the impact of health status on QoL and includes mental, physical, emotional, and social functioning.[15] In addition to HRQoL, a person's SWB—also referred to as life satisfaction—can be a determinant in quantifying the clinical relevance and consequence of ill health. The SWB is a multidimensional construct comprising cognitive and affective components that reflect an individual's appraisal of their satisfaction with their life.[16,17] Understanding the SWB of individuals with particular health conditions may help identify populations with greater mortality risk[18] and guide the development of targeted supportive interventions.

The impact of altered IP on individuals' HRQoL and SWB, and the treatments used in the management of IP, remains under-examined. As such, this study has two primary aims: to describe the SWB and HRQoL of Australian adults with suspected IP and to explore the treatment methods used by this population group.

Materials and Methods

Study design and setting

A cross-sectional study design using an online self-reported survey was deployed. Approval for the study was obtained from the Human Research Ethics Committees (HREC) of the University of Technology Sydney (ETH19-4012). The health-seeking behavior, views, and preferences of this study cohort have previously been published.[19]

Participants and recruitment

Participants were recruited via social media platforms and a purpose-built webpage, with snowball sampling methods used. The authors shared the survey on their social media and known Facebook groups, such as Leaky Gut and Microbiome Support Group Australia. The survey was open for 2 months between September 2019 and November 2019. Eligibility questions asked participants whether they believe they have IP (self-diagnosed) or have been diagnosed with IP. To participate in the study, participants were also required to be 18 years of age or older, living in Australia, and have Internet access. The target population, although broad, represents an under-examined population group; as such, this study was designed to capture people with suspected IP or confirmed IP. As IP is suggested to be underdiagnosed, including participants who self-diagnose IP best reflects the target population and the patients who present to clinical practice for the treatment of IP.[7] Survey responders with incomplete demographic characteristics, accounting for <5% of total data, were excluded from analysis.

Survey and data collection

The online survey administered through the online platform *SurveyGizmo* utilized questionnaire items previously developed to investigate IP in Australia.[7,9] The survey was pilot tested by four lay individuals to assess language clarity, with the required corrections made. The survey included four main domains: demographic characteristics, treatment methods for altered IP, SWB, and HRQoL.

Demographic characteristics

The participants were asked about their gender, age, height, and weight from which body mass index (BMI) was calculated and categorized to underweight, healthy weight, overweight, and obese.[20] The participants were further asked about their country of birth, the state or territory where they reside, and whether this was in an urban, rural, or remote location. The participant's income manageability was determined by how well they manage their household income, categorized as "difficult all the time," "difficult some of the time," "not too bad," or "easy."

Self-reported outcome of increased IP

Two questions were asked to explore the potential severity of IP. First, participants were asked whether they

believed their IP has become "better," "worse," or "no change" over the previous 12 months. Participants were then asked how many days a week does their IP affect their daily living with the option of 0–7 days.

Treatment of increased IP

A selection of survey items involving dietary products, lifestyle therapies, dietary supplements, and medications that may either improve or exacerbate IP along with open-ended questions were used to document how frequently these methods were used. The frequency of use for dietary products, lifestyle therapies, dietary supplements, and medications were measured by using a six-point scale ("never," "less than once a month," "1–3 times a month," "once a week," "2–6 times a week," "every day"). These treatment methods were further explored in relation to the person who prescribed the treatment, mainly the qualification of the practitioner or whether the treatment was self-prescribed.

Subjective well-being

Participants' SWB was measured by using the Personal Well-being Index—Adult (PWI-A) scale—an instrument validated in Australian population samples.[21] The PWI scale consisted of seven domains of satisfaction: *standard of living*, *personal health*, *achieving in life*, *personal relationships*, *personal safety*, *community-connectedness*, and *future security*.[21] The PWI scoring system of each domain is reported on a 0–10 scale, with 0 representing no satisfaction at all and 10 being completely satisfied.

Quality of life

The 20-Item Short Form Health Survey (SF-20) was used to measure participants' HRQoL.[22] The SF-20 assesses six health domains: physical functioning (six questions), role functioning (two questions), social functioning (one question), mental health (five questions), current health perceptions (five questions), and bodily pain (one question).

Statistical analyses

Data were exported to STATA® 16 for statistical analyses. Variables were reported as means, standard deviations (SDs), 95% confidence intervals (CIs), or frequencies and percentages, where appropriate. Chi-square analysis was used to examine the association between two categorical variables, with Student's *t*-tests used for continuous variables across a binary variable. Analysis of variance was used to measure the difference between a continuous variable across a categorical variable. Spearman's correlation analysis was used to measure the correlation between the number of days that IP affects daily living, SWB, and HRQoL. Logistic and linear regression models were used when considering multiple factors. Variables associated with SWB, HRQoL, or the number of days that IP affects daily living—with a bivariate p-value <0.25[23]—were entered into the respective multivariate logistic or linear regression models, to adjust for potential confounders. A stepwise backward elimination process was then used to identify the most important independent predictors.

For analysis, participants' use of dietary products, lifestyle therapies, dietary supplements, and medications were grouped as frequently ("once a week," "2–6 times a week," and "every day") and infrequently ("less than once a month," "1–3 times a month," and "never"). Although participants were able to select either "exacerbation," "improvement," or "no change" for the self-reported outcome of IP in the previous 12 months, only data from exacerbation and improvement were used during analysis. Practitioners were categorized as "medical practitioners" (integrative medicine practitioners, general practitioners, and gastroenterologists) and "healthcare practitioners" (all practitioners).

Analysis and interpretation of the data collected from the PWI-A scale were undertaken according to a previously published work.[24] Participants who answered consistently 0/10 or 10/10 across all PWI domains were excluded due to a risk of response bias.[24] For analysis, the raw scores were transformed to a 0–100 scale. The combined mean score from the seven domains represents the participants' overall SWB. A two-sample *t*-test was used to compare the normative mean of the surveyed sample and the Australian population.[25]

The analysis and interpretation of the SF-20 were undertaken according to a previously published work.[22] For analysis, the SF-20 item scores were transformed to a scale of 0 to 100, with 0 representing the worst perceived health-related outcome. Item scores for each domain were combined and averaged to produce the final domain score (0–100). Higher scores reflect better perceived health-related outcomes, except for bodily pain where a higher score indicates more bodily pain.

Results

Demographic characteristics

There were 982 responses to the survey, of which 393 responses were excluded as the initial eligibility questions were not answered and thereby classified as not meeting the eligibility criteria; this left a total of 589 participants. Most participants were female (93%), with a mean age of 45.0 years (SD = 12.1; range 18–82) and a mean BMI of 27.0 (SD = 6.9). Participants' BMI were classified as healthy weight (46.1%), obese (26.9%), overweight (23.8%), and underweight (3.3%). Most participants were born in Australia (81.0%) and resided in New South Wales (29.7%), Queensland (27.3%), Victoria (17.5%), or Western Australia (10.7%), in an urban (70.6%), rural (27.3%), or remote area (2.0%). Most participants described their income manageability as "easy or not too bad" (46.5%), followed by "difficult some of the time" (32.3%) or "difficult all the time" (21.2%). The major health concerns reported by participants were IP ($n = 300$, 50.9%), autoimmune conditions ($n = 40$, 6.8%), Hashimoto's thyroiditis ($n = 28$, 4.8%), gastrointestinal issues ($n = 24$, 4.1%), chronic fatigue syndrome ($n = 21$, 3.6%), and rheumatoid arthritis ($n = 18$, 3.1%).

Practitioners consulted with, and treatments used, for managing increased IP

Participants most frequently reported using dietary products (87.9%), dietary supplements (72.9%), and lifestyle therapies (54.6%) for the management of IP. Medications were infrequently used by participants for the

TABLE 1. FREQUENCY OF TREATMENT METHODS USED FOR INCREASED INTESTINAL PERMEABILITY ($N=483$)

	Treatment methods used for increased IP[a]									
	Total		*Dietary products*		*Lifestyle therapies*		*Dietary supplements*		*Medications*	
Who prescribed treatment	n	%	n	%	n	%	n	%	n	%
Self-prescribed	288	59.6	256	53.0	163	33.8	156	32.3	11	2.3
Naturopath	208	43.1	175	36.2	104	21.5	180	37.3	0	0.0
Integrative medicine practitioner	93	19.3	77	15.9	47	9.7	84	17.4	11	2.3
General practitioner	81	16.8	60	12.4	30	6.2	34	7.0	21	4.4
Nutritionist	60	12.4	57	11.8	28	5.8	47	9.7	0	0.0
Dietitian	37	7.7	34	7.0	12	2.5	15	3.1	0	0.0
Chinese medicine practitioner	28	5.8	17	3.5	12	2.5	24	5.0	0	0.0
Chiropractor	28	5.8	16	3.3	22	4.6	15	3.1	0	0.0
Acupuncturist	24	5.0	12	2.5	11	2.3	18	3.7	0	0.0
Herbalist	24	5.0	15	3.1	14	2.9	22	4.6	0	0.0
Gastroenterologist	20	4.1	13	2.7	5	1.0	8	1.7	3	0.6
Kinesiologist	20	4.1	14	2.9	12	2.5	12	2.5	0	0.0
Ayurvedic practitioner	12	2.5	10	2.1	5	1.0	9	1.9	0	0.0
Homeopath	12	2.5	8	1.7	7	1.5	9	1.9	0	0.0
Osteopath	7	1.5	6	1.2	5	1.0	5	1.0	0	0.0
Pharmacist	5	1.0	1	0.2	1	0.2	5	1.0	1	0.2
Nurse	4	0.8	4	0.8	2	0.4	2	0.4	2	0.4
Nurse practitioner	2	0.4	2	0.4	2	0.4	1	0.2	1	0.2

[a]Participants were able to select multiple treatment methods.
IP, intestinal permeability.

treatment of IP (8.5%). Self-prescribing of treatment methods for the management of IP was most frequently reported (59.6%), followed by prescription from a naturopath (43.1%), integrative medicine practitioner (19.3%), general practitioner (16.8%), and nutritionist (12.4%) (Table 1). Both dietary products (53.0%) and lifestyle therapies (33.8%) were frequently self-prescribed. However, dietary supplements and medications were most frequently prescribed by a naturopath (37.3%) and general practitioner (4.4%), respectively.

Self-reported outcome of increased IP

In the previous 12 months, more participants reported that their IP had improved (55.8%). Half of the participants (50.0%) reported that IP affected their daily living 7 days a week. Further, participants who described an improvement in their IP during the previous 12 months reported that IP affected their daily life 4.0 days a week (95% CI: 3.6–4.4); however, participants who described exacerbation of their IP in the previous 12 months reported that IP affected their daily life 6.0 days a week (95% CI: 5.7–6.3; $p<0.001$).

A self-reported improvement in IP was associated with participants who were treated by a practitioner compared with those who were not treated by a practitioner (76.1% vs. 23.9%; $p<0.001$). Participants who reported that their IP had worsened in the previous 12 months had a significantly higher mean BMI compared with those who reported an improvement in their IP in the past 12 months (28.4 vs. 25.5; $p<0.001$). Multivariate logistic regression analysis found that the use of NSAIDs ($\beta=1.08$; 95% CI: 0.17–1.98; $p=0.021$), lifestyle therapies ($\beta=1.08$; 95% CI: 0.46–1.70; $p=0.001$), and *Saccharomyces boulardii* ($\beta=1.56$; 95% CI: 0.46–2.67; $p=0.006$) were predictors of a greater number of days each week that IP was reported to affect daily living. However, reporting an improvement of their IP in the previous 12 months ($\beta=-1.78$; 95% CI: −2.39 to −1.17; $p<0.001$), and infrequently ($\beta=-0.90$; 95% CI: −1.64 to −0.16; $p=0.017$) or frequently ($\beta=-0.82$; 95% CI: −1.49 to −0.16; $p=0.016$) practicing yoga were found to be predictors for a fewer number of days affecting daily living each week.

TABLE 2. TREATMENT-RELATED CHARACTERISTICS AND THE IMPROVEMENT OF INCREASED INTESTINAL PERMEABILITY IN THE PREVIOUS 12 MONTHS ($N=287$)

Characteristics	*Odds ratio (95% CI)*	p
Treating person		
Self	1.00	
Health care practitioner	2.04 (1.15–3.61)	0.015
Gluten		
Never	1.00	
Frequently	0.35 (0.20–0.61)	<0.001
Vigorous exercise		
Never	1.00	
Frequently	2.99 (1.61–5.53)	<0.001
Vagus nerve stimulation		
Never	1.00	
Frequently	3.10 (1.31–7.31)	0.010
NSAIDs		
Never	1.00	
Infrequently	0.48 (0.26–0.86)	0.014
Frequently	0.35 (0.15–0.86)	0.022
Using dietary supplements		
No	1.00	
Yes	2.66 (1.40–5.05)	0.003

NSAIDs, nonsteroidal anti-inflammatory drugs.

Table 3. Associations Between Common Dietary Products, Lifestyle Therapies, Medications, and the Self-Reported Outcome of Intestinal Permeability in the Previous 12 Months ($N=483$)

			Self-reported outcome of increased IP in the previous 12 months		
	Total		Exacerbation	Improvement	
	n	%	%	%	p
Dietary products					
Red meat					
Frequently	357	74.2	44.6	55.4	0.909
Infrequently	124	25.8	43.9	56.1	
Organic foods					
Frequently	331	69.4	37.6	62.4	<0.001
Infrequently	146	30.6	58.8	41.2	
Dairy					
Frequently	279	57.9	50.6	49.4	0.012
Infrequently	203	42.1	36.4	63.6	
Refined sugar					
Frequently	239	49.8	59.6	40.4	<0.001
Infrequently	241	50.2	30.8	69.2	
Fermented foods					
Frequently	220	46.2	36.1	63.9	0.004
Infrequently	256	53.8	52.3	47.7	
Gluten					
Frequently	213	44.4	64.1	35.9	<0.001
Infrequently	267	55.6	30.3	69.7	
Apple cider vinegar					
Frequently	179	37.3	36.1	63.9	0.026
Infrequently	301	62.7	49.0	51.0	
Bone broth					
Frequently	173	36.1	32.5	67.5	0.001
Infrequently	306	63.9	51.3	48.7	
Collagen					
Frequently	166	35.0	31.1	68.9	<0.001
Infrequently	308	65.0	51.3	48.7	
Alcohol					
Frequently	148	30.9	47.4	52.6	0.472
Infrequently	331	69.1	43.1	56.9	
Lifestyle therapies					
Breathing exercises					
Frequently	212	45.5	34.4	65.6	<0.001
Infrequently	254	54.5	55.3	44.7	
Stress management					
Frequently	210	45.2	34.0	66.0	<0.001
Infrequently	255	54.8	57.1	42.9	
Meditation					
Frequently	191	40.8	38.7	61.3	0.037
Infrequently	277	59.2	50.6	49.4	
Vigorous exercise					
Frequently	146	30.9	28.4	71.6	<0.001
Infrequently	327	69.1	51.4	48.6	
Yoga					
Frequently	133	28.5	31.3	68.8	0.001
Infrequently	333	71.5	50.7	49.3	
Vagus nerve stimulation					
Frequently	61	13.3	20.5	79.6	<0.001
Infrequently	399	86.7	48.8	51.2	
Medications					
NSAIDs					
Frequently	63	13.4	69.2	30.8	0.001
Infrequently	407	86.6	40.2	59.8	
Prednisone					
Frequently	16	3.5	41.7	58.3	0.827
Infrequently	447	96.5	44.9	55.1	
Methotrexate					
Frequently	11	2.4	57.1	42.9	0.704
Infrequently	448	97.6	43.9	56.1	
Antibiotics					
Frequently	6	1.3	60.0	40.0	0.657
Infrequently	460	98.7	43.6	56.4	

Treatment-related characteristics of increased IP improvement

Participants who reported an improvement in their IP in the previous 12 months were more likely to be treated by a health care practitioner (OR = 2.04, $p=0.015$), use dietary supplements (OR = 2.66, $p=0.003$), participate in vigorous exercise (OR = 2.99, $p<0.001$), and employ vagus nerve stimulation (OR = 3.10, $p=0.010$) (Table 2). Further, participants who reported an improvement in their IP during the previous 12 months were also less likely to consume gluten (OR = 0.35, $p<0.001$) or use NSAIDs (OR = 0.35, $p=0.022$).

Associations between common dietary products, lifestyle therapies, medications, and the self-reported outcome of increased IP

Participants who reported frequently consuming organic foods ($p<0.001$), fermented foods ($p=0.004$), bone broth ($p=0.001$), collagen ($p<0.001$), or apple cider vinegar ($p=0.026$) described an improvement in their IP in the previous 12 months compared with those who infrequently consumed these dietary products (Table 3). Further, participants who reported infrequently consuming dairy products ($p=0.012$), refined sugar ($p<0.001$), or gluten-containing products ($p<0.001$) described an improvement in their IP in the previous 12 months compared with participants who reported frequently consuming these dietary products. Participants who reported frequently practicing breathing exercises ($p<0.001$), stress management ($p<0.001$), meditation ($p=0.037$), vigorous exercise ($p<0.001$), yoga ($p=0.001$), or vagus nerve stimulation ($p<0.001$) more commonly described an improvement in their IP in the previous 12 months compared with participants who infrequently reported practicing these lifestyle therapies. Participants who infrequently used NSAIDs ($p=0.001$) more commonly described an improvement in their IP in the previous 12 months compared with participants who frequently used NSAIDs.

Frequency of dietary supplements use for the treatment of increased IP

The most frequently used dietary supplements for the management of IP were probiotics (36.1%), herbal mixtures (26.6%), prebiotics (21.7%), zinc (21.7%), glutamine (19.4%), magnesium (19.1%), and vitamin D (15.6%) (Table 4). Dietary supplements were most frequently used by participants who described an improvement in their IP during the previous 12 months compared with those who described exacerbation of their IP (63.3%–86.8% vs. 13.2%–36.7%). Participants frequently reported using dietary supplements as prescribed by a practitioner rather than self-prescribed (66.7%–87.8% vs. 12.2%–33.3%) (Table 4). There was a statistically significant association between the use of dietary supplements and the self-reported outcome of IP. Specifically, participants who used zinc ($p=0.05$), glutamine ($p=0.02$), magnesium ($p=0.006$), vitamin C ($p=0.03$), or vitamin B complex ($p=0.001$) described an improvement in their IP during the previous 12 months.

Table 4. Associations Between Dietary Supplements and Self-reported Outcome of Increased Intestinal Permeability in the Previous 12 Months and Percentage for Person Prescribing Each Treatment ($N = 346$)

	Self-reported outcome of IP in the previous 12 months					*Person who prescribed treatment*	
	Total		*Exacerbation*	*Improvement*		*Self-prescribed*	*Practitioner prescribed*
Dietary supplements	n	%	%	%	p	%	%
Probiotic	125	36.1	33.3	66.7	0.483	27.0	73.0
Herbal mixtures	92	26.6	28.4	71.6	0.111	26.4	73.6
Prebiotic	75	21.7	27.6	72.4	0.113	23.0	77.0
Zinc	75	21.7	25.4	74.6	0.05	20.0	80.0
Glutamine	67	19.4	22.5	77.6	0.02	25.8	74.2
Magnesium	66	19.1	19.2	80.9	0.006	29.2	70.8
Vitamin D	54	15.6	35.9	64.1	0.956	30.2	69.8
Vitamin C	50	14.5	18.8	81.3	0.03	20.4	79.6
Vitamin B complex	49	14.2	13.2	86.8	0.001	12.2	87.8
Omega 3	48	13.9	33.3	66.7	0.689	33.3	66.7
Curcuma longa	42	12.1	23.3	76.7	0.114	30.9	69.1
Slippery elm	41	11.9	28.6	71.4	0.366	17.1	82.9
Aloe vera	39	11.3	24.1	75.9	0.146	23.7	76.3
Digestive enzyme	37	10.7	36.7	63.3	0.963	13.5	86.5
Multivitamin	37	10.7	20.7	79.3	0.062	24.3	75.7
Amino acid mix	31	9.0	32.0	68.0	0.637	29.0	71.0
Saccharomyces boulardii	21	6.1	18.8	81.3	0.131	14.3	85.7
Vitamin A	19	5.5	33.3	66.7	0.806	26.3	73.7

Subjective well-being and HRQoL

There was a statistically significant difference in overall SWB and each domain of SWB between Australian adults with suspected IP and the Australian population ($p < 0.001$). Specifically, Australian adults with suspected IP had lower (i.e., worse) average scores for all domains compared with the Australian population. A *t*-test showed that participants who described exacerbation of their IP had a worse ($M = 54.7$, SD = 20.3) SWB than those reporting an improvement ($M = 66.1$, SD = 19.6) in their IP ($p < 0.001$). Spearman's correlation analysis revealed that the number of days that IP affects daily life had a negative correlation with SWB and HRQoL ($p < 0.001$). Results for correlation analyses are summarized in Table 5.

Subjective well-being and common dietary products, lifestyle therapies, and medications

Pairwise comparison found a statistically significant difference between the overall SWB of participants, and the frequency of common dietary products, lifestyle therapies, and medication use. Participants who used any form of

Table 5. Spearman's Correlation Between Quality of Life and Subjective Well-being with the Number of Days Increased Intestinal Permeability Affects Daily Life Each Week (0–7 Days)

	n	*Mean*	*SD*	*Correlation coefficient*	p
Subjective well-being					
Personal well-being index	422	60.3	20.3	−0.402	<0.001
Standard of living	422	65.0	25.5	−0.313	<0.001
Health	422	43.4	24.6	−0.453	<0.001
Achieving in life	422	56.1	25.6	−0.377	<0.001
Personal relationship	422	64.2	26.3	−0.261	<0.001
Personal safety	422	75.3	24.3	−0.193	<0.001
Community connectedness	422	59.3	27.2	−0.277	<0.001
Future security	422	58.8	27.9	−0.273	<0.001
Quality of life					
Physical functioning	423	61.9	33.8	−0.275	<0.001
Role functioning	423	57.3	42.5	−0.335	<0.001
Social functioning	423	60.5	32.3	−0.388	<0.001
Mental health	423	55.0	21.6	−0.294	<0.001
Health perception	423	37.2	28.5	−0.474	<0.001
Bodily pain	423	50.4	25.1	0.316	<0.001

Score ranges from 0 to 100. A high score indicates better health except for pain, where a high score indicates more pain.
SD, standard deviation.

TABLE 6. MULTIPLE REGRESSION PREDICTING SUBJECTIVE WELL-BEING AND HEALTH-RELATED QUALITY OF LIFE

		Health-related quality of life						
	Subjective wellbeing (n = 301), β (95% CI), p value	*Physical functioning (n = 417), β (95% CI), p value*	*Role functioning (n = 306), β (95% CI), p value*	*Social functioning (n = 306), β (95% CI), p value*	*Mental health (n = 304), β (95% CI), p value*	*Health perception (n = 302), β (95% CI), p value*	*Bodily pain (n = 304), β (95% CI), p value*
Improvement of IP in previous 12 months	10.70 (6.01 to 15.39), <0.001		21.06 (11.60 to 30.51), <0.001	18.83 (11.72 to 25.94), <0.001	10.57 (5.56 to 15.58), <0.001	21.88 (15.76 to 27.99), <0.001	−11.74 (−17.53 to −5.94), <0.001
Using dietary products	12.12 (3.21 to 21.03), 0.008				15.79 (6.24 to 25.33), 0.001		
Using lifestyle therapies	−6.30 (−11.05 to −1.54), 0.010		−14.97 (−24.59 to −5.35), 0.002	−9.30 (−16.53 to −2.07), 0.012	−7.30 (−12.36 to −2.23), 0.005	−7.45 (−13.58 to −1.33), 0.017	5.86 (0.07 to 11.64), 0.047
BMI							
Normal weight	1.00	1.00			1.00	1.00	1.00
Obese	−5.70 (−10.99 to −0.41), 0.035	−15.51 (−22.59 to −8.43), <0.001			−5.91 (−11.58 to −0.24), 0.041	−12.89 (−19.88 to −5.91), <0.001	12.69 (6.05 to 19.33), <0.001
Treating person							
Self	1.00	1.00				1.00	
Medical practitioner	−6.35 (−11.52 to −1.18), 0.016	−13.06 (−20.46 to −5.66), 0.001				−9.76 (−16.57 to −2.95), 0.005	

BMI, body mass index; CI, confidence interval.

dietary products ($M = 61.0$, SD = 20.4) for the treatment of IP were found to have better SWB compared with those who never used dietary products ($M = 54.6$, SD = 18.7) ($p = 0.023$). Further, participants who never consumed gluten-containing foods ($M = 65.2$, SD = 21.5) were found to have better SWB compared with participants who frequently consumed gluten ($M = 59.1$, SD = 19.8) ($p = 0.037$). However, participants who frequently consumed alcohol ($M = 64.9$, SD = 18.7) were found to have better SWB compared with those who never consumed alcohol ($M = 54.0$, SD = 22.4) ($p < 0.001$). Further, participants who frequently practiced breathing exercises ($M = 63.0$, SD = 19.4; $M = 56.2$, SD = 21.5) ($p = 0.014$), stress management ($M = 62.4$, SD = 19.0; $M = 56.1$, SD = 23.7) ($p = 0.036$), vigorous exercise ($M = 66.2$, SD = 18.4; $M = 55.3$, SD = 20.9) ($p < 0.001$), or yoga ($M = 68.0$, SD = 17.0; $M = 56.0$, SD = 20.9) ($p < 0.001$) were found to have better SWB compared with participants who never participated in these lifestyle therapies. Lastly, participants who never used NSAIDs ($M = 62.5$, SD = 21.5) were found to have better SWB compared with those who frequently used them ($M = 54.3$, SD = 19.5) ($p = 0.026$).

Multiple regression predicting SWB and HRQoL

Seven regression models predicting overall SWB, and each HRQoL domain were undertaken. The results of these regression models found that the outcome of IP in the previous 12 months, BMI, the treating person, and the use of dietary products and lifestyle therapies were all statistically significant predictors of overall SWB and each HRQoL domain (Table 6). Specifically, improvement of IP ($\beta = 10.70$, $p < 0.001$) and using dietary products ($\beta = 12.12$, $p = 0.008$) were predictors of better SWB whereas being obese ($\beta = -5.70$, $p = 0.035$), treated by a medical practitioner ($\beta = -6.35$, $p = 0.016$), and using lifestyle therapies ($\beta = -6.30$, $p = 0.010$) were predictors of worse SWB. Regarding HRQoL, all domains except physical functioning saw improvement in IP as a statistically significant predictor for higher HRQoL (Table 6).

Discussion

This study is the first to explore the HRQoL and SWB of Australian adults with suspected IP. Our results suggest that altered IP may pose a greater health burden than previously thought, providing the first indication that Australian adults with altered IP are susceptible to poor SWB and HRQoL. Further, several participant characteristics were found to be associated with the improvement or exacerbation of IP (Fig. 1).

Increased IP and SWB and HRQoL

Our results suggest that Australian adults with suspected IP have a lower SWB compared with the Australian population. Further, improvement in IP is suggested to be a significant predictor of SWB and HRQoL. These results provide the first indication that a relationship between both SWB and HRQoL and altered IP exists in a diverse range of health conditions. In support of this relationship, Australian adults with gastrointestinal disorders (many of which are associated with altered IP)[4] have been found to have a lower HRQoL compared with Australian adults without gastrointestinal disorders.[26] Further, a lower QoL has been reported in diarrhea-predominant irritable bowel syndrome patients with IP compared with those with a normal intestinal integrity.[27] The association between altered IP and both SWB and HRQoL contributes to a much needed clinical understanding of altered IP, especially as the consequence and clinical relevance of altered IP in clinical practice have recently been questioned.[12] Further, the correlation found between both SWB and HRQoL and the number of days that IP affects daily living suggests that the previously reported symptoms[7] and biomarkers[5] of altered IP are not the only clinical consequence of altered IP, with both SWB and HRQoL now suggested to be involved.

Health care and increased IP

The care provided by health care practitioners compared with self-care differs not only in the treatments used but also in the reported outcomes. First, this study identified a high prevalence of self-prescription of treatment interventions, primarily dietary products, and lifestyle therapies, for the management of IP. Dietary supplements and medication were most frequently prescribed by a health care practitioner. These findings coincide with existing research that suggests that complementary and integrative medicine practitioners frequently use dietary supplements while also using a multimodal and personalized approach for the management of altered IP.[9] Working alongside a health care practitioner has also been suggested to provide greater health outcomes compared with no clinic-based support.[28–30] This may explain why in this study Australian adults who report an improvement in their IP are two times more likely to be treated by a health care practitioner. Second, our study found that only 24% of self-treated participants reported an improvement in their IP compared with 76% of practitioner-treated participants. These findings suggest that the care provided by health care practitioners to Australian adults with suspected IP may have beneficial effects on the outcomes of altered IP. Further, health care practitioners, especially those with limited experience in the management of altered IP, may draw on the findings of this study to gain a deeper understanding as to the treatment methods used by Australian adults with IP.

Features associated with increased IP improvement

Participants who reported an improvement in their IP were 35% less likely to consume gluten or use NSAIDs. Our results also found that participants who indicated that they avoided consuming gluten-containing foods and never used NSAIDs were associated with a better SWB. These results concur with clinical studies that show that the consumption of gluten-containing products and the use of NSAIDs induce IP.[31,32] Practitioners who treat IP also advocate for their patients with IP to avoid gluten and NSAIDs.[9]

The finding that vitamin C and vigorous exercise is associated with improvement of IP conflicts with existing research. First, preliminary research suggests that 500 mg of vitamin C (ascorbic acid) may induce a rearrangement of the actin cytoskeleton and thereby an exacerbation of IP.[31,33]

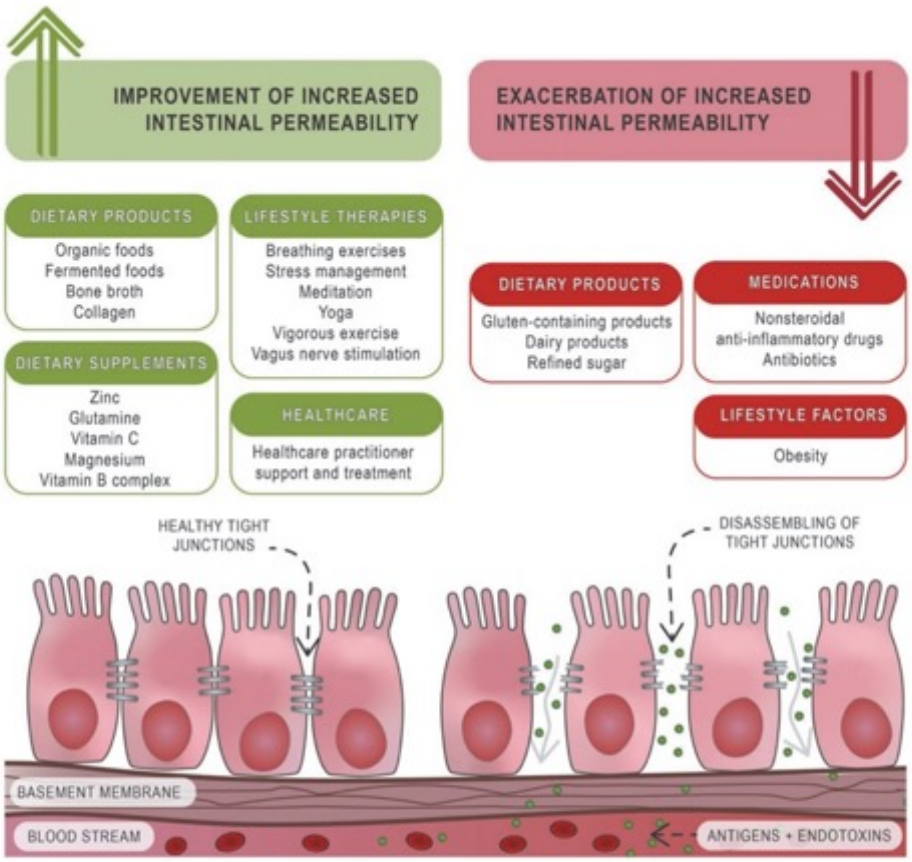

FIG. 1. Participant's characteristics found to be associated with the improvement and exacerbation of increased intestinal permeability.

Potentially, the association between vitamin C intake and improvement of IP may be the result of the frequent use in dietary supplements, especially as participants who reported an improvement in their IP were 2.7 times more likely to use dietary supplements. Research has demonstrated a causative link between vigorous exercise and altered IP.[34] As a result of redistribution of blood flow and splanchnic hypoperfusion during vigorous exercise, damage to mucosal and epithelial cells may occur, thereby paving the way for exacerbation of IP.[34] The improvement associated with vigorous exercise in our study may be the result of improved health; for example, as health and well-being improve so does the ability to participate in exercise.[35] Further large-scale trials and epidemiological research is needed to confirm both of these hypotheses.

Research agenda

Our study provides useful information where further research can draw on the findings to inform clinical trials and clinical practice guidelines. The identified characteristics found to be associated with the improvement and exacerbation of IP warrant further investigation (Fig. 1). Many of these associated features are yet to be investigated for their effect on IP, with clinical research focusing primarily on dietary supplements and dietary products for the treatment of IP. However, there has been limited investigation exploring the effectiveness of lifestyle therapies in the management of IP.[9] Nevertheless, many of these lifestyle therapies are reported to have beneficial health outcomes in health conditions with a known association with altered IP.[36,37]

These results provide a foundation for future clinical trials where a study exclusively conducted in primary care ensuring a homogenous study population and standardized diagnostic criteria may confirm the results of this study.

The findings from this study may also help to inform the development of a clinical practice guideline for the management of altered IP. By understanding the treatment methods used, the development of recommendations can incorporate the views and preferences of Australian adults with suspected IP to enable relevant and appropriate recommendations for this patient group.

Limitations

Although this study involved participants with self-reported suspected IP, whether there was a confirmed diagnosis of IP is unknown. However, previous research has shown that people with self-reported irritable bowel syndrome have similar health care utilization and QoL as those with diagnosed IBS.[38] Many of the health conditions that participants report experiencing are known to be more prevalent in females and are suggested to be associated with IP, which may explain why 93% of participants were female.[4] Therefore, these results are considered relevant to females who suspect they have IP rather than Australian adults with a confirmed diagnosis of altered IP. The self-reported outcome of IP has the potential for recall bias and may not reflect improvement or exacerbation of IP. Therefore, to confirm the relationship between both SWB and HRQoL and altered IP, a clinical study that measures IP and evaluates both SWB and HRQoL is required. However, this study provides important and novel information, advancing the research agenda on the clinical consequence of altered IP, and suggests potential treatment strategies that are worth investigating.

Conclusion

The integrity of the small intestine may pose a greater health burden than previously thought, with susceptibility to poor SWB and HRQoL reported in Australian adults with altered IP. Our results strengthen the clinical relevance and consequence of altered IP, providing the first indication that a relationship between both SWB and HRQoL and altered IP exists. Clinical trials may use these findings to further explore the potential use of the treatment interventions used by Australian adults with suspected IP.

Authors' Contributions

B.L. led the development of the study, conducted the study, and drafted the article. D.S., A.S., and E.M. provided expertise on all stages of the study and revised the article.

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Address correspondence to:
Bradley Leech, BHSc (Hons)
Faculty of Health
Australian Research Centre in Complementary and Integrative Medicine
University of Technology Sydney
15 Broadway
Ultimo
NSW 2007
Australia

E-mail: bradgleech@gmail.com

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