Mapping and predicting benthic habitats in estuaries using towed underwater video

Thesis submitted by

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Certificate

I certify that the work in this thesis has not previously been submitted for a degree nor has it been submitted as part of requirements for a degree.

I also certify that the thesis has been written by me and that any help that I have received in the research or preparation of the thesis, and all sources used, have been appropriately acknowledged.

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Abstract

With the global carbon crisis a matter of worldwide concern, efforts to preserve natural habitats that sequester carbon are of utmost importance. However, the processes which enable aquatic plants to survive and thrive are poorly known, as is the extent of their distribution and how they change over multiple scales. The aim of this research was to develop methodologies to help define the relationships between key benthic habitats and bio-physical variables and spatially predict their distribution and abundance within south-west Australian estuaries through towed underwater video. This thesis identified multiple non-destructive methods along with their strengths and limitations, to characterise benthic cover from underwater video, and highlighted optimal methods based on equipment, end goals, time and funding available. Additionally, I emphasize that no one method used in isolation was suitable for the analysis of underwater video from the shallow and turbid habitats from my study sites, but that a combination of methods was required for optimal characterisation.

This research is one of the first to model and spatially predict fine-resolution (5% intervals) percent cover of benthic habitats within estuaries from post-processed underwater video using biological and physical datasets with a state-of-the art machine learning method called 'Random Forests'. This method is often used within terrestrial landscape ecology, but rarely within estuarine systems. Random Forests performed well with 79-90% variation explained by the models for each key benthic habitat and partial plots illustrated strong relationships between physical variables and biotic habitats. The most influential parameters driving biotic habitat distributions were longitude (19%), depth (13%), and latitude (11%), although this relationship varied between estuaries and on the degree of estuary connectivity to the sea (permanently-opened, artificially-opened and normally-closed). Predictive performance of key benthic habitat models was moderate to excellent and associated uncertainty maps of standard deviation of each model was highly variable in areas of habitat fragmentation.

Broad-resolution distributions of biotic habitats were found to be important in understanding local-scale physical processes. Seagrasses were the most common biotic habitat in five estuaries, although higher numbers of seagrass species occurred in the permanently-opened Leschenault Estuary (e.g. *Ruppia megacarpa, Halophila ovalis* and *Heterozostera tasmanica)*, while seasonally-opened (Wilson Inlet) and normally-closed

(Wellstead, Stokes and Beaufort) estuaries supported monospecific meadows of *R. megacarpa*. Red and green macroalgae had inverse latitudinal distributions, with red alga occurring in northern estuaries with higher amounts of seawater incursion and freshwater input. Green alga, especially green film alga were more prominent in the more stagnant, and normally-closed waters of the southern estuaries. Motile commercial fishery species such as crabs (*Portunus pelagicus*) were common in northern estuaries where access to marine influence was essential for their survival. Encrusting benthic polychaete worms such as *Ficopomatus enigmaticus* and the black mussel *Mytilus edulis* were found shallow sections of southern estuaries, which were able to tolerate extreme changes in water quality due to estuary bar closure, and often encrusting the hard substratum of submerged trees and rocks. This study demonstrated advances in modelling techniques of species abundances and distribution from underwater video and highlighted the importance of bio-physical relationships on spatial patterns of different seagrass species and other biotic habitats such as algae beds, polychaete mounds and mussel clumps in estuaries.

Estuarine habitats are at the forefront of climate change effects and experience rapid changes (within weeks to months) in their spatial distribution and abundance. I developed a real-time, rapid and accurate method to capture broad-resolution semi-quantitative (barren, low, moderate and high percent cover) changes in benthic habitats using underwater video, as traditional remote sensing methods such as aerial photography and satellite imagery can often take up to weeks and months to post-process for spatial habitat distribution. I tested the accuracy of two benthic habitat assessment protocols: the broad-resolution real-time classification protocol (called the "Rapid Benthic Assessment") against the fine-resolution post-processed habitat classification. I also tested the validation of the broad-resolution percent cover categories of seagrass from the RBA method using in situ samples of *R. megacarpa* and *H. ovalis*. The high correlation between the RBA and the fine-resolution method indicated that a high degree of detail and accuracy was retained by the RBA method. The visualisation of benthic habitats almost impossible to map through traditional remote sensing means was made possible through rapid data acquisition and visualisation from underwater video. This study demonstrated that real-time delineation of estuarine habitats allowed for rapid data analysis and representation within hours of data collection.

This research will enable resource management authorities to make informed decisions on monitoring benthic habitats which have global significance within estuarine systems from baseline habitat maps, supplement existing maps and understand how bio-physical attributes shape benthic habitat distributions.

Chapter 1: *General Introduction*

Chapter 1: General Introduction

Biological habitats are important for coastal and marine processes, especially habitats such as seagrass and algal beds which act as ecosystem engineers, and sequester approximately 10% of the world's carbon sequestration, a value which surpasses terrestrial forests, and are important contributors to managing the global carbon crisis (Kennedy et al., 2010; Fourqurean et al., 2012). Mapping and predicting species spatial distribution and abundance within these shallow habitats and the processes that affect them are essential in effectively understanding, managing and conserving coastal and marine environments. The global carbon crisis is the excessive consumption of carbon-based energy sources, which releases carbon dioxide into the atmosphere at levels which outpace natural absorption and thus, exacerbating the effects of climate change through global warming (Canadell et al., 2007). This has direct implications to future generations of humanity and at multiple scales which have been identified for seagrasses as altered spatial distributions from sea level rise, growth rates and sexual reproduction by increased temperature and changing salinity (Short and Neckles, 1999). Shallow underwater habitats like estuaries, coral reefs and seagrass beds cover approximately 7% of the world and abut one of the most densely populated parts of our planet (Costanza et al., 1997). Additionally, they provide enormous ecosystem services (Costanza et al., 1997) and are extremely vulnerable to anthropogenic disturbance through agriculture, catchment clearing, aquaculture and climate change. Therefore, maintaining healthy estuarine and marine habitats which sequester carbon and reduce the anthropogenic carbon footprint left on the earth is of paramount importance to humanity.

1.1 Importance of estuaries

Estuaries are essential transitional environments that lie between terrestrial and marine conditions, and are heavily influenced by multiple physical gradients and processes (Brearley, 2005; Prandle, 2009). They experience changes in tidal regimes and salinity from marine sources, as well as freshwater input and deposits of terrestrial sediments and nutrients through connecting river systems (Cloern, 1987; Mann and Lazier, 2006). Conversely, marine connections can deposit sandy, marine-derived sediments at or near the estuary bar. Estuaries are vulnerable to particular climatic regimes which are dependent on their spatial location; often pertaining to water temperature, amount of rainfall, and degree of tidal, wave or wind mixing of the water column. These physical gradients and processes affect ecosystem structure, functioning and the biotic habitats that they can support (Mann and Lazier, 2006). Estuaries are also a source of 'blue carbon', in that the aquatic vegetation often found within them sequester carbon through their root systems at amounts that rival and even surpass tropical rainforests (Kennedy et al., 2010; Mcleod et al., 2011). Current studies prove that biotic-habitats such as seagrasses, mangroves and salt marshes sequester as much as 50% of organic carbon per hectare within sediments, and act as one of the most cost-effective global forms of natural carbon capture and storage (Kennedy et al., 2010).

Estuaries are also one of the most productive ecosystems in the world. They provide valuable food sources and habitats for a number of animals, including fish that complete their lifecycles within the estuary (Lenanton and Potter, 1987), birds by providing habitats in which to survive and breed (Goss-Custard et al., 1995), mammals by providing areas of foraging and breeding, and for humans that utilize estuaries for recreation and commercial fisheries. The presence of vegetative plants within estuaries and along their shorelines can also act to trap sediments and act as natural barriers to dissipate wave energy, and prevent shoreline erosion. Vegetation such as seagrasses are known to be nursery grounds for many invertebrate and fish species, including commercially important species such as mullet (*Mugil cephalus*), black bream (*Acanthopagrus butcheri*), blue swimmer crabs (*Portunus pelagicus*), and Western king prawn (*Penaeus latisulcatus*) (Brearley, 2005).

However, estuaries are also in worldwide decline (Kennish, 2002; Lotze et al., 2006) and are one of the most degraded temperate marine ecosystems (Jackson et al., 2001). At the interface of the marine and freshwater environment, estuaries are affected by rainfall, freshwater inflow, sediment supply through their rivers and adjoining catchment areas. They are also affected by anthropogenic factors such as catchment clearing, nutrient input via agricultural practices into connecting waterways and urban development from population growth adjacent to estuaries and coastlines. This has major impacts on the structure and function of the existing ecosystems through 'dead zones' from eutrophication, harmful algal blooms, salt intrusions through ground-water from clearing practices, habitat loss, potential phase shifts in the surviving ecological communities, and cumulative chain of events often having disastrous impacts on the communities depending on the estuary for survival (Yamamuro et al., 2006; Dolbeth et al., 2007; Diaz and Rosenberg, 2008; Pillay et al., 2010). As our demands upon estuaries increase, so too does our need to understand the ecological and physical processes affecting the distribution and abundance of the benthic habitats within them.

Habitat-formers within estuarine and marine environment often provides hard substratum for colonisation, canopy areas, and viable surfaces for epiphytic algal growth (e.g. seagrass beds, polychaete tubes, algal and kelp forests) (Franschetti et al., 2006). Moreover, these environments often provide structurally complex areas for both benthic and pelagic organisms to live, thereby increasing local biodiversity (Hughes et al., 2009; Buhl-Mortensen et al., 2010; Anderson et al., 2011). Habitat-formers also function to stabilize sediments and reduce sedimentation and turbidity in estuaries (Fonseca and Fisher, 1986). The composition, extent and complexity of biotic habitats can also influence the species that use these biogenic habitats (Levin et al., 2001). This is seen in studies by Connell and Jones (1991), where areas of high complexity supported greater proportions of adult blennioid *(Forsterygion varium)* fish in New Zealand, and higher mortality rates of juveniles in low complexity sites. Similarly, crustacean abundance and species richness increased with the occurrence of *Halimeda sp.* in seagrass meadows (Stoner and Lewis, 1985; Humphries et al., 1992). Also, marine invertebrates had greater abundances in structurally complex habitats of algal mats (Dean and Connell, 1987), and within seagrass beds of *Zostera marina* (Boström and Bonsdorff, 1997).

Estuarine variability is likely to affect multiple ecosystems and the habitats within them through indirect (e.g. influx of nutrients fuelling algal growth at the surface) and direct (e.g. shading effects on seagrasses and reducing photosynthesis) processes. Changes in ecosystem functioning are more likely to arise from a chain of cumulative events, rather than a single factor or process. For example, seagrass loss in Australia has been attributed to adjacent industrial expansion and consequent increases in wastewater and effluent from the addition of treatment plants and processing and refining plants (Cambridge and McComb, 1984; Kendrick et al., 2002). This in turn, increased the amount of productivity within the system, fuelling algal blooms and epiphytic algae overshading seagrass habitats, which then reduced effective photosynthesis of the chloroplasts in seagrass leaves. While this example is limited to Australia, other studies in Florida Bay (Madden et al., 2009), Chesapeake Bay (Kemp et al., 2005), and Mondego estuary (Cardoso et al., 2004) have reported similar cascading effects of poor water quality and eutrophication on their associated habitats. Additionally, algal habitats support a different range of species communities, thereby driving species phase shifts from seagrass to algal communities (Hauxwell et al., 2003). Moreover, the shift from a seagrass habitat that has a complex 3D structure provides more available habitat than to a phyto-plankton dominated 2D structure with considerably less complexity (Schmidt et al., 2012). However, some algal assemblages often play fundamental roles in habitat structure and function and, similar to seagrass habitats, they provide nurseries, shelter and food for juvenile species of fish, crab, prawn and shrimp (Humphries et al., 1992; Neira and Potter, 1992; Bulleri et al., 2002). Maintaining ecosystem function and biodiversity is one of the key building blocks of freshwater and marine ecosystems, so the ability to monitor estuarine variability within the system is of high importance to neighbouring townships and management, which depend on the estuary for survival.

The ability to accurately assess different habitat types present in estuarine and marine environments is important in management and conservation planning. The most efficient form of displaying patterns within benthic marine systems is through illustrations and maps (Dennison et al., 2007). These maps are often used to convey status, trends and interactions, either through anthropogenic or natural causes (Kostylev et al., 2001; Dennison et al., 2007). While they may be simple in form, the visual elements are crucial for effective communication to various stakeholders (Dennison et al., 2007). Maps attempt to describe and predict the benthic habitat by utilizing a range of physical and remote sensing techniques, identify knowledge gaps and highlight spatial patterns (Dennison et al., 2007). In our current state of environment, with increasing effects of climate change, we need effective mapping methods to guide resource management decisions and prioritize monitoring needs to protect the resources we have in order ensure success of habitat preservation and restoration for future generations.

Ecological theory states that environmental variables, in part, contribute to species distribution and abundance, and approximate predictions of these biological factors from indirect variables such as latitude and longitude, can be reasonably estimated (Austin, 2007). Additionally, biological indicators such as seagrass species distribution, seagrass condition and associated community assemblages and canopy cover have often been used to assess estuarine health because they rapidly respond to changes in highly variable nutrient input, salinity, water flow and reduced sediment redox conditions (Madden et al., 2009; Parnum and Gavrilov, 2009; Smale et al., 2011). Perturbations in estuaries occur as a chain of events, and often with the early warning symptoms of change reflected in the biology. The spatial variability of seagrass beds changes annually and seasonally, and the importance of mapping the distribution of seagrass beds and associated physical water quality and geographic parameters over time can be indicative of natural or anthropogenic issues affecting growth, species composition, spatial distribution of seagrass beds or point sources of pollution. Similarly, other biological indicators such as phytoplankton (Paerl et al., 2003), fish (Hughes et al., 1998; Dulvy et al., 2008) and macro-invertebrates (Rosenberg et al., 2004; Leonardsson et al., 2009) have the ability to detect disturbances within water bodies. However, recent studies by Ellis and Bell (2013) have indicated that some fish, due to their 'generalist' nature, can be poor environmental indicators of estuarine quality. In order to adequately protect the marine environment and biodiversity, a thorough understanding of underlying spatial patterns is paramount (Andrew and Mapstone, 1987). Management of these habitat-formers is therefore of great importance to the agencies responsible in maintaining healthy environmental conditions promoting their long-term growth and spatial distribution.

1.2 Estuaries in south-west Australia

There are multiple definitions within the literature for what constitutes an estuary which are often based on a northern hemisphere perspective and vary between disciplines, so I have adopted the definition by Potter et al. (2010), which encompasses the main characteristics of estuaries in Australia:

"An estuary is a partially enclosed coastal body of water that is either permanently or periodically open to the sea and which receives at least periodic discharge from a river(s), and thus, while its salinity is typically less than that of natural sea water and

varies temporally and along its length, it can become hypersaline in regions when evaporative water loss is high and freshwater and tidal inputs are negligible".

While there are many schools of thought in regards to nomenclature of estuary closure between Australian states (e.g. Intermittently Closed and Open Lakes and Lagoons (ICOLLs) in New South Wales, and Intermittently Open Estuaries in Victoria), I have addressed the estuaries in this study by hydrodynamic regime as found in Brearley (2005):

1) Permanently opened
2) Seasonally open-closed
3) Normally closed
4) Permanently closed

These classifications indicate the degree of opening and closure to marine influence, salinity regime, river flow and rainfall, all of which affect the ecosystems and species present in the estuaries. In south-west Australia, there are 8 permanently opened, 20 seasonally opened, 18 normally closed and 5 permanently closed estuaries. Permanently closed estuaries do not have any degree of marine intrusion, and are governed by freshwater input associated with connecting riverine systems. In addition, normally closed and permanently closed estuaries are often found along the southern edge of Western Australia, while seasonally opened and permanently opened estuaries are more common along the south-west coastline of Australia (Potter et al., 1990). Estuaries in south-west Australia have a Mediterranean climate, that is, cool and wet during winter, and long dry summers, with annually variable amounts of freshwater influx to estuaries from winter rains (Brearley, 2005). The larger estuaries on the western coast of Australia appear to have longer riverine systems, and are often permanently to seasonally open. However, the estuaries on the southern coast are often normally closed, following a gradual decrease in rainfall, but open in exceptionally high rainfall seasons (Brearley, 2005).

In the past, traditional remote sensing techniques to detect the limits of seagrass habitats within south-west Australian estuaries have been hampered by tannin-stained and turbid waters, and the shallowness of the study locations (Bastyan et al., 1995). Estuaries not only lie at the interface between terrestrial and marine systems, they also lie between two forms of mapping capabilities: the nature of estuaries makes them too turbid for aerial or satellite imagery to penetrate to the seafloor, and too shallow to effectively run the large vessels required for multibeam mapping (Churst et al., 2010). This thesis attempts to overcome these innate issues by using underwater video as a primary tool to map and characterise the estuarine landscape. In addition, indirect environmental parameters such as latitude and longitude, distance from the coast, freshwater influx and marine influence were used to determine if there were any effects on the distribution of benthic estuarine habitats.

1.3 Study area

The five estuaries within this study lie on the south-west coast of Australia, between Perth and Esperance (Leschenault Estuary, Wilson Inlet, Beaufort Inlet, Wellstead Estuary and Stokes Inlet). These were chosen on the basis of logistical convenience and a pre-existing survey program between Geoscience Australia and Department of Water, Western Australia. These estuaries are highly turbid through sediment resuspension and tannins, and are relatively shallow. These estuaries vary in their opening regimes, catchment sizes, community assemblages and climate exposure (Brearley, 2005). Leschenault Estuary has been permanently opened to the ocean since 1952 by a modified man-made channel called “the Cut”. Wilson Inlet is classed as seasonally open-closed, mostly closed to the Southern Ocean by a sandbar during most of the year, but has been artificially breached every winter dating back to approximately 103 years ago, when rising water levels from catchment runoff during periods of high rainfall threaten to inundate the township and adjacent agricultural areas (Brearly, 2005). Wellstead Estuary, Beaufort and Stokes Inlet are normally closed to the ocean. Beaufort Inlet differs physically from Stokes and Wellstead as it has an unusually high sand bar blocking the estuary to the ocean (3 m). This means that the length of time the estuary is open for lasts only a few weeks compared to Stokes Inlet, which once opened, stays open for many months and has been known to stay open for years.

As a consequence of sporadic opening regimes, the estuaries are subject to seasonally-variable fluctuations in temperature, salinity, freshwater input via rainfall, nutrient enrichment and eutrophication that can degrade the water quality, increase macroalgal and phytoplankton blooms, and can substantially change benthic communities, particularly as a result of hypersaline conditions (Hodgkin & Lenanton, 1981; Lavery et al., 1991; Brearly, 2005; Carruthers et al., 2007; Chuwen et al., 2009; Potter et al., 2010). Furthermore, pelagic species which migrate to (catadromous) and from (andadromous) marine to freshwater settings utilize estuaries to spawn and must be able to withstand these changing conditions (Potter et al., 1999). A few species occur in all estuaries and in enough numbers for local commercial fishery importance, namely: *Mugil cephalus, Aldrichetta fosteri,* while *Pomatomus saltarix* make up an important recreational effort (Potter et al., 1990).

Estuaries of south-west Australia typically support a variety of aquatic vegetation including seagrasses (e.g. *Ruppia megacarpa, Halophila ovalis, Zostera muelleri, Heterozostera tasmanica* and *Posidonia australis*) and mixed macroalgal assemblages (e.g. *Gracilaria sp., Caulerpa sp., Lamprothanium papulosa, Chaetomorpha sp., Polyphysa peniculus*) (Congdon and McComb, 1981; Hodgkin & Clark, 1988a,b; Hillman et al., 2000; Carruthers et al., 2007). Seagrass meadows and macroalgal assemblages characteristically rim the edges of estuaries and are often intermixed in shallow water where they compete for space, light and nutrients (Wurm, 2000); provide substantial above ground complexity; and are known to be important nursery grounds and adult refugia for a wide range of marine taxa, including commercial species of fish (King George whiting *Sillaginodes punctata*, whitebait *Hyperlophus vittatus*), and crustacea (Blue swimmer crabs *Portunus pelagicus*; Western King Prawn *Penaeus latisulcatus*, shrimp *Palaemonetes australis*) (Humphries et al., 1992; Brearly 2005). Additionally, due to the relatively shallow nature of the estuaries, *R. megacarpa* often grows to the water surface. Conversely, the inner regions of deeper estuaries are unvegetated and often bioturbated with burrows and feeding tracks left by a variety of infaunal and epifaunal animals, including worms (*Ficopamatus enigmaticus*), blue swimmer crabs (*Portunus pelagicus*) and burrowing fishes (Semeniuk, 2000). For the purposes of my thesis, although *R. megacarpa* has been known as submerged aquatic vegetation in the literature, it is referred hereinafter as seagrass.

1.4 Aims and objectives

This study examines the relationship between the physical and geographical aspects of estuarine habitats and biological structure and composition within these estuaries. A wide range of methods have been used to collect and analyse underwater imagery for both ecological and habitat mapping purposes. In **Chapter 2**, I review and evaluate five methodologies applicable to quantifying benthic habitats, outlining their strengths and weaknesses, and their application to shallow, turbid estuarine environments. The spatial structure and configuration of habitat-forming biota can have major implications on the physical stability and structure of estuaries and population and community structure of associated species. To characterise and map biotic habitats, in **Chapter 3**, key biotic habitats were quantified from underwater video footage in five turbid south-west Australian estuaries, ranging from permanently opened to normally closed estuarine systems. Fine-resolution percent cover (5% intervals) was recorded for key benthic habitat-forming species, such as seagrasses (*R. megacarpa, H. ovalis and H. tasmanica*), macroalgae (branching, filamentous and film algae for red and green algae), mussels (*Mytilus edulis)*, and polychaete worms (*F. enigmaticus*), along with indicators of seagrass health (epiphyte load), across each estuary from video transects and drop camera stations, while associated physical and water quality parameters were collected from stations across each estuary. These bio-physical variables were then modelled using a state-of-the-art machine-learning method (Random Forests) to predict the distribution and abundance of key habitat-forming biota in unsampled areas. I then evaluate these relationships to create maps to describe and predict major benthic habitat types using Random Forests.

Estuaries are dynamic and often highly turbid ecosystems where the presence and health of biotic habitats, such as seagrasses and macroalgae, can change dramatically in space and time. To map dynamic biotic habitats, in **Chapter 4**, a real-time characterisation method (Rapid Benthic Assessment) was developed which characterised broad-resolution semi-quantitative covers of dominant benthic habitats from underwater video in the five turbid south-west Australian estuaries. Here, broad-resolution percent cover categories (low, medium, high) of seagrass species, seagrass length (short, medium, long), epiphyte presence, and semi-quantitative categories of key biotic habitats (green macroalgae, red macroalgae, polychaetes worms, mussels) were recorded *in situ* in real-time from video transects and drop camera stations across each estuary. Seagrass percent cover and length were also recorded directly using benthic cores. To determine the accuracy and precision of the RBA approach, real-time characterisations were compared against the direct measures of seagrass percent cover and length collected from benthic cores, and the fine-resolution post-processing approach of Chapter 3. Finally, **Chapter 5** discusses biological patterns between the five hydrodynamically-classified estuaries, conclusions based on the feasibility and application of using underwater video to map benthic habitats in estuarine systems, and recommendations for future work in mapping benthic habitats. Chapters 3 and 4 have been written as manuscripts for review and may therefore have some information repeated throughout the text. In addition, 'I' is consistently used throughout this thesis, but 'we' is used in corresponding manuscripts.

Chapter 2: *A review of video methods to characterise seafloor systems, with application to south-west Australian estuaries*

Chapter 2: A review of video methods to characterise seafloor systems, with application to south-west Australian estuaries

Abstract

Many technological advances have improved the collection method and resolution of underwater video and still image capture to identify and monitor biotic underwater habitats. To assess the effectiveness and suitability of these methodologies for estuarine habitats within south-west Australia, this review identified and categorised current methods used to estimate abundance and/or percent cover of benthic habitats from underwater video and images. Examples of each methodology are provided, along with their strengths and limitations, and emphasized that no single observational method viewed in isolation was suitable for the analysis of benthic habitats from the shallow and turbid estuaries of south-west Australia, but that a combination of methods was appropriate to ensure effective allocation of limited resources.

2.1 Introduction

The ability to identify and determine ecological and spatial change within a benthic community is central to conservation management (Fabricius and De'ath, 2004), and accurate and precise spatial and temporal information through the development of baseline habitat maps plays a crucial role in being able to make informed decisions (Burrough & McDonnell, 2005). It is estimated that as little as 5-10% of seafloor habitats are mapped at the same resolution as terrestrial habitats (Wright & Heyman, 2008). Mapping underwater habitats can be challenging in estuaries and coastal systems for a number of reasons: tannin-stained water from the presence of organic compounds (Wrigley et al., 1988; Brearley, 2005) and turbidity due to suspended sediment input from rivers (Cloern, 1987), can affect visibility in the water column. Currents and tides affect sample collection; high relief bedforms can affect data capture, and shallow water depths preclude the use of high-resolution multi- or single-beam sonar systems. Mapping spatial habitat patterns of distribution and abundance relies on the ability to accurately and precisely estimate biotic percent cover. Precise methods are important to ensure repeatability of measurements of percent cover between and within observers (Andrew and Mapstone, 1987). The development of accurate baseline habitat maps allows us to effectively compare across time scales and potentially infer natural or anthropogenic processes causing change in biotic habitats.

Currently, mapping systems integrate data within ecological modelling from a wide variety of fields: biological, geomorphological, geochemical and physical to detect relevant species-relationships and their interactions within and among habitats (Anderson et al., 2009b; Pittman and Brown, 2011; Buhl-Mortensen et al., 2012). Effective modelling allows us to visualise spatial patterns (Mellin et al., 2010), and identify natural or anthropogenic processes and environmental variables governing species distribution and abundance (Anderson et al., 2009b; Pittman and Brown, 2011). This enables us to efficiently manage estuarine and marine systems through reliable maps by monitoring changes in their spatial patterns of distribution and abundance over time (Fourqurean et al., 2001; Brown et al., 2011). Within estuaries, water quality variables such as nutrient input, temperature, salinity and dissolved oxygen govern the availability of suitable areas for colonisation of biotic habitats, and subsequently, the animals that rely on these habitats to survive. Additionally, geographical variables such as latitude, longitude, and coastal, river and bar distances may determine the spatial distribution and abundance of biotic habitats (Leathwick et al., 2006; Austin, 2007). To date, these factors have been analysed in isolation and have not been combined to predict spatial patterns in benthic habitats for maps of estuarine systems in south-west Australia using machine learning methods such as Random Forests. Ultimately, the magnitude and extent of spatial mapping of marine habitats is a matter of end goals, equipment, study area, time and cost.

Underwater videography has been used for many applications in benthic habitat mapping, and has several advantages that are complimentary to other remote sensing survey techniques. It is a very effective fine-scale *in situ* tool to map and ground-truth aquatic benthic habitats by; visually corroborating habitats which are mapped through large-scale remotely sensed hydroacoustic equipment (e.g. side scan sonar, multibeam sonar), aerial photography (Compact Airborne Spectrographic Imager (CASI), CASI 2), and satellite photography (e.g. Landsat Enhanced Thematic Mapper Plus (ETM+), Quickbird, Advanced Land Observing Satellite (ALOS) Advanced Visible and Near

Infrared Radiometer (AVNIR)) (Anstee et al., 2009; Murphy and Jenkins, 2010). Underwater video has also been used to visualise habitats often surveyed by SCUBA as it provides a permanent record for further post-processing by multiple users, multiple daily transects can be completed within short time frames, and is non-destructive to the surrounding environment. Underwater video can survey multiple habitat types at deeper depths compared to SCUBA diving. It has the potential for rapid on-board data analysis (Anderson et al., 2008) to ensure environmental managers can visualise spatial habitat patterns of the seabed within hours of data collection, make informed decisions regarding survey plans, and identify targeted locations for sample collection within a time-frame suitable to their budgetary requirements. Additionally, it has the advantages of unrestricted bottom time and can survey larger expanses of seafloor compared to SCUBA.

Multiple methods exist to post-process and analyse underwater video for ecological data and depend on the end goals of the study (Table 2.1). Choosing an appropriate method to analyse video depends on the purpose of the study which can include; measures of percent cover (Buhl-Mortensen et al., 2009), measures of abundance of a species within the video frame (Harvey et al., 2002); species presence (Haghi et al., 2012); ground-truthing substratum and associated biology (Anderson et al., 2008); behaviour (Willis et al., 2000); or fishing and trawling effects (Grizzle et al., 2009). In addition, the aquatic system studied influences the method used to characterise the seabed, whether identifying habitat gradients through transects, capturing broad habitat types in a large scale environment through drop camera stations, or focusing on fine-scale changes of the seafloor through intensive sampling. This decision is also dependent on understanding the tradeoffs (advantages and disadvantages) between video methods to ensure effective allocation of limited resources, the system studied and the video camera system used.

Consideration of the method used also depends on a number of factors; the orientation of the video camera system and the quality of the video obtained: is the camera perpendicular to the benthos (i.e. downward-looking video camera) or at a forward-facing oblique angle? A perpendicular view of the seabed allows an observer to objectively assess quantitative parameters such as area and percent cover of benthic substratum, provided there is a scale present (Kohler and Gill, 2006), while an oblique-angled camera encompasses a larger area than a perpendicular view of the benthos, has an element of perspective, where height can be a measurable quantity, and taxa are more recognisable by their shape (Wakefield and Genin, 1987; Dolan et al., 2008). The quality of the video has also been known to affect habitat classification where poor quality images often hinders progress and results in incorrect classification.

Not only is it important to attain comparable results between multiple methods (standardisation), the need for accuracy in re-creating spatial distributions of estuarine and marine benthic communities is vital for effective resource management, restoration efforts and determining ecological resilience (Dethier et al., 1993; Suding, 2011). The comparison of multiple methods to analyse underwater video for biological percent cover has been studied previously (Dethier et al., 1993; Lam et al., 2006). However, this research addresses important gaps in the existing knowledge of seafloor habitat mapping by providing a broad spectrum of methods, application to current technologies using underwater video and providing in-depth comparisons of the end goals, benefits and constraints of each method. It is timely given the increasing focus on habitat mapping both in near-shore and offshore marine environments, and the growing impact of anthropogenic and climatic changes on these systems. This study compares five of the most commonly used sampling methodologies to obtain estimates of abundance and percent cover of benthic habitats from underwater video and still images and identifies the most suitable method for monitoring south-west Australian estuaries.

Table 2.1: A review of the methods available for estimating abundance and percent cover from underwater video and lists their goals, advantages and disadvantages. Please refer to strength and weaknesses of each method for an in-depth description of the method.

Method	Goals	Advantages	Disadvantages	Literature cited
Point counts	• Percent cover • Species assemblage • Patch and area	• Objective • Random points can be superimposed on image • Repeatable by trained observers • Length and area measurements (if scale present)	• Logistically time consuming • Downward facing images required • Tall objects are overestimated • Requires high quality images with clearly defined benthic habitat categories	Ryan, 2004; Kohler and Gill, 2006; Carleton and Done, 1995; Dumas et al., 2009
Digital image segmentation	• Benthic cover estimation	• Accurate area of percent cover	• Logistically time consuming • Difficult to use with oblique-angled images • High reflectance reduces efficiency of detecting habitats • Downward facing images required	Whorff and Griffing, 1992; Tkachenko, 2005; Wakeford et al., 2008
Automated classification	• Percent cover of benthic habitat • Patch and area	• Automated classification of habitat types • Reduced time spent processing data • Good for fine-scale detail monitoring of repeated sites	• High complexity of image requires long processing time • Requires high image quality • Not always accurate as it is based on user-defined classes • Downward facing images required	Williams et al., 2012; Teixidó et al., 2011; Seiler et al., 2012

Method	Goals	Advantages	Disadvantages	Literature cited
Visual estimation	• Semi-quantitative categories for percent cover • Benthic assemblage	• Rapid data collection and analysis while in the field • Broad coverage • Semi-quantitative/ranked data • Comparative measures of length and size • Robust • Highly repeatable • Can use forward or downward-facing camera system	• Broad biota categories • Not many 'finer' scale studies • Can be subjective	Coles et al., 2009; De'ath and Fabricius, 2000; DeVantier et al., 1998; Fabricius and De'ath, 2004; Mellors, 1991; Stein et al., 1992; Fourqurean et al., 2002
Categorical classification	• Presence/absence to ground-truth remote sensing information	• Larger coverage over areas • Reduced amount of time spent processing data • Can use forward or downward-facing camera system	• Loss of detail (presence or absence data only)	Roob et al., 1998; Blake et al., 2000; Blake and Ball, 2001; Dolan et al., 2008; Nelson et al., 2011
Baited Remote Underwater Video Stations*	• Count of motile species • Method is more focused on species assemblages, but has been used to characterise benthos	• Objective • Comparative measures of length and size • Forward-facing camera	• Fish assemblages can be dependent on bait type	Cappo et al., 2004; Cappo et al., 2011

* This method is outside the scope of this review as this method is mainly used to describe fish assemblages and relative abundance.

2.2 Strengths and weakness of each approach to estimate biological abundance and cover

2.2.1 Point counts

Traditionally, percent cover estimates of biota and benthic substratum were estimated using points distributed within quadrats. Intertidal ecologists are the most common users of point count methods to count biota, as well as coral reef ecologists, who regularly sample percent cover of benthic biota such as sponges, coral, bryozoans, algae or substratum (Paine, 1977; Underwood, 1981; Dethier et al., 1993; Carleton and Done, 1995; Ryan, 2004; Kohler and Gill, 2006). This method is also increasingly used by coastal and near-shore ecologists for assessing change in benthic habitats using Autonomous Underwater Vehicles (AUVs), snorkel and diver tows, Remotely Operated Vehicles (ROVs) (Lam et al., 2006; Dumas et al., 2009; Barrett et al., 2010; Leaper et al., 2011; Smale et al., 2012). The method uses an image, or screenshot that is perpendicular to the sample area, often outlined by a quadrat. A number of points are randomly or systematically overlaid on the image, and the biota or substratum type under each point visually identified and entered by manually transcribing the data within a database program, or using a specialised program such as the commonly used Coral Point Count with Excel extensions (CPCe Kohler and Gill, 2006). The program allows a user to allocate points within the image as stratified, uniform, or equally spaced grids. The user can select a number of sampling points to estimate abundance or percent cover, where percent cover is calculated by the total proportion of points for particular species and multiplied by 100 (Kohler and Gill, 2006).

This method is also used by the Australian Institute of Marine Science in the Long Term Monitoring Program to analyse change in reef composition, cover and, health of the Great Barrier Reef. The Long Term Monitoring Program by AIMS uses permanent transects in 47 reefs along the GBR, and collects 50 still images at 1 m intervals, which are then overlaid with 200 points to estimate benthic cover (Jonker et al., 2008). The point count method has been used with data from Autonomous Underwater Vehicles (AUVs) in various studies to monitor the status of the spatial distribution of marine benthic habitats and species diversity in temperate Australia (Barrett et al., 2010; Smale et al., 2012), often with the integration of broad-scale high-resolution multi-beam bathymetry data. Additionally, the outputs from the AUV surveys can produce 3-D reconstructions of the seafloor from stereo images and a number of co-located physical variable measurements to visualise spatial patterns and features at fine-scales and detect change in distribution of key benthic habitats (Williams et al., 2012).

The strengths of this method is that it is repeatable by other trained observers and can be used for length and area measurements if scale bars or laser projections are present within the still photograph (Harvey et al., 2002). It generates a permanent record of the benthic habitat at the time of image capture, and if performed correctly, is an objective, unbiased method. Point counts are used with images or video that is directly perpendicular to the seafloor, often with actual quadrats present in the image. Point counts can also be used to estimate presence/absence data, count data, and point frequency data. In a study examining the number of points (sampling effort) required to estimate accurate and cost-effective sampling strategies, Dumas et al. (2009) identified that a sample size of 9 points per 1 m^2 was a reliable and cost-effective sampling strategy with no significant differences between sampling intensities of 25 vs. 50 vs. 100 within a 1 m^2 quadrat.

The number of points allocated per quadrat still varies widely among numerous studies (Ryan, 2004; Lam et al., 2006; Alquezar and Boyd, 2007; Jonker et al., 2008), as does quadrat size (Foster et al., 1991; Tkachenko, 2005). This approach requires images to have a consistent resolution, colour and contrast quality to provide unbiased estimates of percent cover and comparable estimates between images. This is often done by running a standard colour/contrast filter over all of the images (Kohler and Gill, 2006). A major problem with the point count method on oblique video and stills is that the percent covers of tall taxa are over-estimated due to perspective issues (i.e. parallax error). For example, tall taxa appear to be larger and cover more area than other benthic taxa, when in reality organisms may actually occupy the same size and area. Point counts trialled on the video data from the Commonwealth Environment Research Facilities Marine Biodiversity Hub survey of the Carnarvon shelf overestimated sponges (many of which were tall) by up to 30% (Tran, unpublished data) (Brooke et al., 2009). Consequently, while point counts are a robust and repeatable method for down-looking images, they are likely to provide biased estimates when applied to forward-facing images, especially where organisms are tall. The point count method can also be time consuming if there are large amounts of video and stills (McDonald et al., 2006). Secondary to this problem is the requirement of physical storage for video images and stills, since greater quality stills require larger storage devices. There is also the potential for the method to focus on more abundant species and subsequently miss rarer species.

2.2.2 Digital segmentation of habitats within still images

Digitising each organism in an image and calculating the area occupied by each species is a convenient way to accurately measure the percent cover of benthic organisms' at large scales (Tkachenko, 2005). This method calculates the surface area of the biological substratum by using a computer program to outline and digitise benthic species (e.g. some software programs used include DISTANCE v6.0, IMAGE J, and ArcMAP). Species can have a unique range of colours within photographic still images which can be used to delineate between multiple species (Tkachenko, 2005). Researchers have used digitised images of percent cover to measure temporal changes in live coral cover to determine rates of growth and competitive interactions between corals, and to estimate differences in condition following disturbances such as storms and cyclones (Connell et al., 2004). Similarly, still imagery was digitised to measure coral percent cover in shallow coral reef communities on Lizard Island, Australia, over 23 years to quantify acute environmental disturbance events by monitoring differences in spatial, temporal and taxonomic change (Wakeford et al., 2008).

One of the major advantages of digital segmentation of still images is due to the methods' applicability to monitor competition between different sessile species over time and space by comparing changes in percent cover. Digitising stills has been proven to calculate a more accurate area of percent cover compared to the point count method of estimating percent cover (Tkachenko, 2005). A comparison of visual methods (visual estimation, even, random and stratified point counts, and digitisation) was used to estimate percent cover of intertidal benthic species by Meese and Tomich (1992). They found that digitising photographs was the most precise method between observers and recommended that a combination of visual techniques with digitisations was required to increase accuracy and precision. Images taken in the field provide permanent records and can be further analysed at a later date. Digital segmentation can also be used when analysing images from an oblique camera angle.

Undefined species with indistinguishable outlines or unfocussed images pose an issue to analysis, and post-processing can be more time consuming than the point count method, further highlighting the requirement for high quality images that aren't hampered by environmental (turbidity or salinity gradients) or technical issues. Species which don't have distinguishable outlines are also difficult to digitise in programs that identify different species by colours or traceable outlines. Digitising an area with different perspectives can introduce issues with scale as mentioned previously. Another issue is the amount of time it takes to calculate digitised areas in software programs. The time taken to digitise an image within a study by Tkachenko (2005) took an average of 15 min, but could reach up to several hours depending on habitat complexity. Significant problems with this method are that it depends on the altitude at which the image is taken; colours between images must be similar for each species; lighting must be identical within all images (i.e. having the exposure in each image exactly the same); and classification is dependent on the taxonomic skill of the observer.

2.2.3 Automatic classification

Automatic classification of benthic habitats from underwater still images is a relatively new and novel technique that is used to monitor the health and status of benthic species, as well as change in distribution and abundance over time (Bewley et al., 2012). This method often uses still or stereo imagery to characterise habitats in 3D, designed to reduce manual interpretation of images and hence, cost and time (Seiler et al., 2012). The process uses a series of algorithms that automatically collect benthic data (rugosity, colour, texture and patchiness) from still images and assigns the image to a predefined habitat class using machine learning methods (Seiler et al., 2012). This method is relatively cost-effective, generates permanent records and has a relatively high classification rate (Seiler et al., 2012). Teixidó et al. (2011) used a similar semi-automatic method to demonstrate its effectiveness over a series of benthic communities from still images over four different habitat types; coral reefs, coralligenous habitats, deep-water corals, and Antarctic habitats. The method uses hierarchical segmentation algorithms, and post-processing time is reduced by up to 40% compared to point-count methods of estimating percent cover. Automated classification has been developed to automatically detect specific species from AUV data, and involves using supervised learning to accurately delineate differences in species present.

Automatic classification has many benefits, most of which revolve around the collection of high resolution still images, and the analysis and output of unique information (e.g. 3D visualizations). Additionally, the study of benthic habitats can be conducted at fine (individual corals within single still images) or coarse (rock habitat over multiple transects) scale, using ROV or SCUBA collected images (Teixidó et al., 2011). Time is significantly reduced analysing percent cover once user-defined habitat classes are specified compared to traditional point count methods (Teixidó et al., 2011). Automated and semi-automated techniques can also mitigate observer bias by eliminating human error from image analysis.

However, automatic classification depends on users to identify species from a proportion of still images for the algorithm to learn from, and is therefore restricted by the need for experienced observers and taxonomist input into the program. It can also be time-dependent with increasing habitat complexity and quality of the image, where underexposed or overexposed images are often misclassified (Seiler et al., 2012). Collection of underwater still images can be difficult to correct for colour as the water column absorbs the red colour spectrum first. Consequently, depth and illumination are often factors which need to be considered (Bewley et al., 2012). Additionally, using AUV to collect still images produces relatively large datasets from high resolution stills, which requires large amounts of storage and computing power.

2.2.4 Visual estimation

The main goals of using visual estimation techniques is to rapidly classify marine benthic habitats in the field while shortening the time spent post-processing data. This method has been shown to be effective in delineating cover categories to a high degree of accuracy (Mellors, 1991). Semi-quantitative categories can include ranked data using numerical, alphabetical terms or categories, and have been used quite effectively over the past few years (Done, 1982; Maragos and Cook, 1995; DeVantier et al., 1998; Fabricius and De'ath, 2004). In a study by Stein et al. (1992), habitat patches were delineated by different bottom types from continuous underwater video using a two-tiered approach of primary and secondary substratum (i.e. sand, mud, gravel, cobble etc). Primary substratum covered at least 50% of the image, and secondary was classified by more than 20%. For example, if substratum was classified as sand-mud, it would mean sand covered >50% and mud >20%. The use of semi-quantitative values to classify video data in the field can significantly reduce time spent post-processing the data when habitat patches are distinct.

Mellors (1991) used a ranked visual technique involving five categories to successfully estimate above-ground seagrass biomass within 0.25 m^2 visual quadrats which is currently used today (DeVantier et al., 1998; De'ath and Fabricius, 2000; Fabricius and De'ath, 2004; Coles et al., 2009). In particular, Fabricius and De'ath (2004) use "Rapid Ecological Assessment Surveys" to rank coral cover of main benthic taxonomic groups in categories from 0-5 in 20% intervals for rapid estimates of percent cover over 100-600 m. Broad-scale categories were found to be insensitive to observer bias (Miller and De'ath, 1996). While this may reduce taxonomic resolution and miss out on fine-scale spatial habitat patterns, an overall view of the habitat within a broader scale of the study area provides environmental managers the broad-scale detail needed in short amounts of time.

The real-time Characterisation of the Benthos and Ecological Diversity (C-BED) method by Anderson et al. (2008) provides an overview of primary and secondary substratum types, as well as bedform relief (i.e. sand ripples, waves, flat etc) and biota that are present. This method is a combination of the semi-quantitative classification scheme of benthic substratum type by Roob (1998) and Stein et al. (2001), whilst incorporating large distances that can be surveyed within the time frame available. The C-BED method estimates percent cover of substratum from underwater towed video over a 15 second window of the seafloor. This means that the characterisation is not just a snap-shot (i.e. still photo) of the seafloor, but encompasses a broader picture of the habitat setting within the video. Real-time characterisations of the seabed as well as biota occurrence were also made by Buhl-Mortensen et al. (2012), which are similar to Anderson et al. (2008), albeit their sequences were 30 seconds long and characterised substratum cover to 5 percent cover intervals (i.e. 5, 10, 15 etc.).

The strengths of this methodology is that it captures the 'broader-scale' view of the structure and composition of benthic habitat, measures spatial variability within that time-frame, is able to use oblique or down-ward facing underwater cameras, and is able to be undertaken rapidly during the field survey (i.e. 'real-time' *in situ* assessment). This method is repeatable by trained observers both for field assessment and post-processing in the lab (Anderson et al., 2008). The advantage of the C-BED method is that it allows for rapid analysis of *in situ* sampling and has the added advantage of providing information to survey planners on where specific habitats are located. Most importantly, these methodologies enable large amounts of seafloor (habitat and biota) to be characterised and their distributions mapped. This methodology has the advantage of reducing survey costs due to rapid data collection and processing while in the field. Additionally, one main advantage of this method is the speed that percent cover estimates can be obtained compared to point quadrat methods, which can take up to double the time (Dethier et al., 1993, McDonald et al., 2006). This methodology also takes into account the entire benthic system present in the image, including rare species, which point counts may miss. Direct observations of real-time video compared to video stills also allows observers to make a more informed observation about the environment of the substratum and biological factors influencing the habitat (i.e. there may be fish in the habitat, but observing one frame from 15 seconds may not capture this information, whereas a characterisation encompassing a larger time-range will).

The disadvantages of visually estimating percent cover of benthic substratum are: it is subjective, a higher number of replicate samples may not result in higher accuracy, and it requires concentrating on specific distributions of taxa and estimating percent cover of within the area, rather than counting and recording biota underneath a certain point (Dethier et al., 1993). New observers must be trained by experienced observers as inexperience has been shown to affect variability between observers. Another disadvantage of this method is that it cannot be used for quantitative analysis unless some quantifiable aspects have been included and/or a representative scale of percent cover has been identified beforehand (Braun-Blanquet, 1932). Additionally, the method may not capture small changes in cover (McAuliffe, 1990). This method is more useful for broader-scale (tens of meters) benthic habitats, rather than the finer-scale (meters) habitats.

2.2.5 Categorical classification

Categorical classification mainly involves classifying biota into presence or absence from underwater video data and corroborates this with remotely sensed data such as bathymetric data obtained from multibeam sonar or aerial photography (Roob et al., 1998, Rattray et al., 2009, Foster et al., 2009, Hasan et al., 2012). This method has been used in many Victorian mapping surveys of estuaries and coastal environments to identify and assess marine benthic changes through historic aerial photography and ground-truthed video data to classify vegetated data identified in aerial photographs (Blake et al., 2000). These classified categories are based on sediment types and presence or absence of algae and seagrasses (Ierodiaconou et al., 2007, Blake and Ball, 2001, Roob et al., 1998). In a study by Ierodiaconou et al. (2007), automatic decision tree classifiers were used to predict the spatial distribution of ecological groups from meaningful combinations of geophysical data and ground-truthed video data. Further to this, Hasan et al. (2012) used broad-scale categories of biota presence combined with acoustic data to create habitat maps of spatial distribution. Additionally, within logistically limited surveys, video designs that are focussed in their goals and outputs and only require underwater video for the confirmation of specific benthic fauna, often benefit from the speed of this methodology (Wilson et al., 2007).

The benefit of this methodology, is that it covers large amounts of area within short periods of time and subsequently, produces high amounts of data, provides presence and absence of taxa quickly from post-processed data or in real-time. This can be quite effective in environments which experience rapid changes to their ecological habitats from environmental or anthropogenic impacts, where results are used to ground-truth other remote sensing methods, and enable research scientists and management authorities to make informed decisions regarding future survey site selection. Additionally, many ecological species distribution based models are based on presence and absence data which focus on predicting the spatial distribution of taxa in regards to the physical environment (Elith and Leathwick, 2009).

On the other hand, there is the potential for loss of taxonomic resolution in highly complex habitats, and this method may require skilled observers for species identification as well as the potential misclassification of species in dense habitats with mixed species assemblages. This method is unlikely to provide enough information to explain specific spatial distributions and abundances that are important in determining species-relationships (e.g. species competition). Additionally, no biological abundances or percent covers of benthic habitats and key taxa are recorded with this method.

2.3 Discussion

It is critical to select the most appropriate methodology to assess habitats within estuaries, and in this review, I show that while no one method is ideal, visual methods are best suited for the turbid and shallow estuaries of this study. Of the methods I review above, three (random point count, automated classification, digital analysis) are restricted to downward-facing images of the seafloor, assuming no further post-processing of the video is performed (e.g. the use of the Canadian perspective grid proposed by Wakefield and Genin, 1987). Conversely, categorical classification and visual estimation techniques allow for the analysis of oblique-angled images. Some advantages of using an oblique-angled underwater camera are that it covers a larger area in a single frame compared to vertical stills, and it also provides an estimate of height. Each method has its benefits and disadvantages, all which need to be taken into consideration before deciding and embarking on a survey plan.

The video pertaining to this thesis was collected using a forward-facing camera at an oblique-angle. To consider using quadrat methods such as *point counts* or *digital image analysis*, a perspective grid would have needed to be overlain over each image to provide an accurate estimate of area (Wakefield and Genin, 1987). Paused frames within video transects also highlights the need to ensure that frames analysed in the video do not overlap with consecutive frames. The video also estimates a distorted view of the sea floor and may be subject to user bias if there was no attempt to make any corrections to either limit the frame of view (e.g. using 1/3 of the video screen instead of the full screen), or limit the perspective within the frame to determine the area of analysis (e.g. using the Canadian Perspective grid by Wakefield and Genin, 1987).

Given the ecosystem studied, the oblique-angled video system, and the time available for this research, the C-BED method was chosen to be the most applicable method to rapidly characterise seafloor habitats and biota as it was easily incorporated into the survey setup, with a rapidly adaptable system which allowed for quantitative and semi-quantitative measurements of biotic habitats. Transient environments such as estuaries rapidly change within short time periods, so rapid methods such as the C-BED method and semi-quantitative methods similar to Fabricius and De'ath (2004) which are able to characterise benthic habitats in real-time, are suitable for broad-scale habitat mapping. A combination of visual estimation methodologies for percent cover of substratum and biotic categories were also estimated by using slightly modified semi-quantitative categories similar to Fabricius and De'ath (2004). Additionally, fine scale studies can be completed with the archived video if required.

Consideration must be applied when choosing the most suitable method for survey, and this decision is dependent on end goals, funding, equipment availability, and habitat. Despite the logistical constraints in mapping habitats from underwater video, interesting and important patterns are often found using underwater video as a sampling method, as demonstrated by discoveries of marine benthic polychaete communities that are reliant on chemosynthetic-based interactions from cold-seep areas between 400 – 6000 m (Sibuet and Olu, 1998) and cold-water coral communities of *Lophelia pertusa* which function as important fish habitats in deep-water ecosystems (Costello et al., 2005). By using the most appropriate methods to map and characterise the spatial distribution and abundance of benthic habitats in estuaries, we can better understand the processes, drivers and interactions that lie between the physical and biological aspects of the system.

Chapter 3: *Quantifying and predicting the distribution and abundance of key habitat-forming biota across five estuaries in south-west Australia*

Chapter 3: Quantifying and predicting the distribution and abundance of key habitat-forming biota across five estuaries in south-west Australia

Abstract

The spatial structure and configuration of habitat-forming biota can greatly impact the physical stability and structure of estuaries, along with the population and community structure of associated species. I characterised biotic habitats from fine-scale post-processed underwater video and modelled their distribution and abundance with interpolated physical, geochemical and geographical parameters to predict their spatial distribution and abundance using the Random Forests ensemble classifier for five turbid estuaries in south-west Australia. Predictive models of key habitat types (seagrasses *Ruppia megacarpa*, *Halophila ovalis* and *Heterozostera tasmanica*, green and red macroalgae, polychaetes worm mounds formed by *Ficopamatus enigmaticus,* and mussel clumps formed by the black mussel, *Mytilus edulis*) along with their associated uncertainty maps were developed from fine-scale percent cover and illustrated the spatial distribution of habitats within each estuary. In addition, partial plots described strong relationships of the four most influential abiotic predictors for each habitat within each estuary. The primary parameters driving Random Forests models were found to be longitude (19% ranked importance), depth (13%), and latitude (11%). Model predictability varied between estuaries, dependent on opening regimes and increased with latitude. The results of this study can be used to supplement existing baseline maps of estuarine habitats and add to the understanding of the bio-physical interactions and spatial distributions of vulnerable biogenic habitats (e.g. seagrasses) and invertebrate species in estuaries.

3.1 Introduction

Estuaries are some of the most highly productive ecosystems in the world and are subject to climate change effects and anthropogenic pressure (Duarte, 2002; Hemminga & Duarte, 2000). Estuaries receive and capture runoff and nutrients from the terrestrial environment, recycle nutrients (Cloern, 1987; Radke et al., 2004), and support diverse plant and animal life by providing shelter, breeding grounds and migratory pathways for coastal and oceanic infaunal and epifaunal species, mammals and commercially important fisheries species (Loneragan & Potter, 1990; Humphries et al., 1992; Beck et al., 2001; Larkum et al., 2006). In addition, estuaries are often areas of high primary productivity, due to primary producers, such as bacteria, providing the basis of local food webs to aquatic plants (Flores-Verdugo et al., 1990; Maher, 2011). Recent studies have demonstrated that seagrasses, mangroves and salt marshes, by trapping sediments, sequester carbon 100 times faster than tropical rainforests and act as some of the most global cost-effective solutions to natural carbon capture and storage (Kennedy et al., 2010; Mcleod et al., 2011). Estuaries and coastal watersheds are also a focus for human settlement as they provide many sources of revenue through tourism, recreational and commercial activities, goods and services (Tagliani et al., 2003, Barletta & Costa, 2009). In Australia, more than 81% of the population live within 50 km of the coast and estuaries for livelihood or the general aesthetic of living adjacent to water features (Hugo, 2011). As a result, these coastal and estuarine systems are prone to change and often incur high levels of pollution from human and industrial effluent, sedimentation from agriculture, overfishing, storm water run-off (Jeng et al., 2005), and disturbance caused by urban development such as the creation of piers, excavation of channels, and land reclamation.

Estuaries are also highly vulnerable to climate change and sea-level rise, as they are the interfaces between marine and freshwater processes (Hugo, 2011). Other potential impacting pressures include enhanced coastal erosion due to higher incidences of storms, weathering (IPCC, 2001; Scavia et al., 2002; Orviku et al., 2003), and current and tidal changes from changing oceanographic regimes (Harley et al., 2006). Inundation due to sea level rise (IPCC, 2001) is also a problem which will be exacerbated by global warming. These climate changes are likely to drive regime shifts in biological habitats and species diversity, alter physical shape and bio-physical functioning of these land-ocean interfaces, with far reaching consequences on human inhabitation of coastal zones, commercial fisheries, tourism, and the general health and aesthetics of these ecosystems. Clearly, understanding the bio-physical processes within estuaries and the climate and anthropogenic changes that affect them is important for their preservation.

Effective management of estuarine areas and their bio-physical habitats requires many forms of information. In particular, a detailed understanding of the baseline spatial distribution of these habitats is important in order to make informed decisions about the ecosystem structure and function (Underwood, 1981; Andrew & Mapstone, 1987; Roy et al., 2001). The health and status of estuarine systems can be monitored by mapping the water quality and the corresponding spatial extent of bio-physical habitats (Robbins, 1997; Smale et al., 2011). These maps can help managers understand the ecosystem processes and species-environment relationships and their importance to the often highly interconnected marine and terrestrial ecosystems that they abut by providing information on fragmented benthic habitats.

Mapping estuaries is a challenging task due to inherent difficulties in location accessibility, sampling equipment and limitations of mapping habitats underwater. Physical and biological data can be time consuming and costly to collect, given the large distances between estuaries along the coastlines of Australia. Unlike terrestrial areas where optical remote sensing technologies such as LiDAR (Light Detection And Ranging) and Landsat can provide detailed broad-scale images of the landscape, these technologies are unable to penetrate water to great depths and even then only in very clear coastal waters (Goldfinger, 2009). Additionally, LiDAR can be ineffective in the delineation of multiple underwater habita&ts with dense cover (Gilvear et al., 2004; Knight et al., 2009). Further offshore, technological advancements in acoustic sonar systems now enable the collection of high resolution seafloor maps from coastal and deep offshore marine environments (Brown et al., 2011). High resolution swath sonar systems are mounted to the hulls or pole-mounted to the side of marine vessels, but these techniques are limited to depths greater than 10 m due to the associated transducer draft below the vessel (Rooper & Zimmermann, 2007). Estuaries not only lie at the interface between terrestrial and marine systems, they also lie between these two forms of mapping capabilities: the nature of estuaries makes them too turbid for LiDAR or Landsat to penetrate to the seafloor, and too shallow to effectively run the large vessels required for multibeam mapping (Churst et al., 2010).

Because seagrasses and other habitat-formers are likely to be strong indicators of the health of an ecosystem, with the largest effects on the species that rely on these habitats for their survival (Fourqurean & Robblee, 1999; Madden et al., 2009), there is a critical need to find effective technologies to map these highly important and vulnerable ecosystems. Although aerial photography and underwater video footage have often been used in combination to successfully map coastal and estuarine systems (Blake & Ball, 2001; Rooper & Zimmermann, 2007; Roelfsema et al., 2009), many coastal estuaries, especially semi-closed ones, are too turbid to map benthic habitats using aerial imagery (Bastyan et al., 1995). Some estuaries in south-west Australia have poor visibility due to turbidity; resuspension of fine sediments by wind-generated waves, and tannin-stained or highly coloured water from humic substances present in the surrounding soil and vegetation of estuaries (Wrigley et al., 1988; Brearley, 2005). Consequently, previous attempts to map estuaries in south-west Australia using aerial photography failed to detect coverage of seagrasses (Bastyan et al., 1995).

Underwater video has not only shown to be a valuable tool to ground-truth habitat maps from aerial photography (Brown et al., 2011), but also to successfully characterise the benthic habitats within estuaries and marine habitats (Becker et al., 2010), identify unique and sensitive habitats (Carleton & Done, 1995; Becker et al., 2010), distinguish habitat change due to natural and anthropogenic impacts (McDonald et al., 2006), quantify ecological indicators to determine estuarine health (Ellis & Bell, 2013) and to examine the adequacy of Marine Protected Areas (MPAs) and other management strategies (Stevens & Connolly, 2004; McDonald et al., 2006). Video imagery has been used to map the distribution and characteristics of physical (e.g. substratum type, structure (relief) and bedform) and biological habitats (e.g. seagrasses, macroalgal assemblages, mussel beds, polychaete worm mounds) (Holmes et al., 2007; Anderson et al., 2008; Creese et al., 2009; Anderson et al., 2011), even in water too turbid for aerial photography (Norris et al., 1997). Towed-video can provide clear images of the benthic habitat and allows for large areas to be covered quickly without damaging the environment (Miller & De'ath, 1996; Anderson, 2005), thus providing a potential way of quantifying bio-physical habitats within turbid estuarine ecosystems. Recent advances in geographic information systems and technology means that data from video can be accurately georeferenced and recorded in high quality. This ability to geo-locate sampling points enables researchers and managers to accurately re-survey the same locations through time to determine the spatio-temporal dynamics and long-term changes in these systems. However, extensive amounts of video are required to map entire landscapes, so techniques involving interpolation are valuable as they have the ability to create continuous coverage from point sources (Burrough & McDonnell, 2005).

Analytical modelling advances in machine learning methods such as Random Forests (RF) have enabled researchers to view bio-physical relationships of ecological habitats within a spatial context (Breiman, 2001). Random Forests modelling can be used to identify the major parameters driving benthic habitat distribution and to predict occurrences in un-sampled areas (Breiman, 2001). This technique is also able to handle multiple variable types, account for non-linear relationships, and tolerate missing values. This is important, as ecological datasets are at times incomplete and often variable, and ecological patterns are frequently non-linear and complex (De'ath & Fabricius, 2000; Maher, 2011). Understanding and mapping benthic habitats using remote sensing techniques and modelling methods can increase our understanding of the functioning of these environments and ensure that with proper management, they will survive through time.

In this study, we collected a variety of biological and physical data through the use of underwater video, sediment and water quality sampling to characterise the benthic habitats of five estuaries of south-west Australia. The sampling in this study provides a 'snapshot in time' of the bio-physical structure and composition of the benthic habitats in these estuaries. The objectives of this study were to: (1) quantify percent cover and map the distribution of benthic habitats within five south-west Australian estuaries using underwater video, (2) map the spatial distribution of associated physical and water quality parameters using interpolation methods from field collected data, (3) model the relationships between the spatial distributions of major benthic habitat-formers in relation to associated physical variables and evaluate the use of these relationships to create predictive maps to describe and predict major benthic habitat types using Random Forests.

3.2 Methodology

3.2.1 Study areas

Five estuaries along the south-west coast of Western Australia were examined in this study (Leschenault Estuary, Wilson Inlet, Beaufort Inlet, Wellstead Estuary and Stokes Inlet) spanning approximately 850 km from Leschenault Estuary in the north (latitude 115.70145 E, longitude 33.26898 S) to Stokes Inlet on the south coast (latitude 121.16389 E, longitude- 33.81865 S) (Fig. 3.1). These estuaries are wave-dominated microtidal systems, but vary to some degree in their morphology and physiographic setting (Heap et al., 2001) (Table 3.1). Estuaries differed in their size (mean 17.7 ± 8.0 km^2, range: 4.5 km^2 (Beaufort inlet) to 48 km^2 (Wilson Inlet)), and shape, from oval (Leschenault Estuary) to elongate (Stokes Inlet), with the width of each estuary affecting the wind fetch, water column mixing by wind, and distribution of surficial sediments (Chuwen et al., 2009). The sediment composition between each estuary was relatively similar with fine-grained muds in the deeper basins of both permanently opened and artificially opened estuaries while coarser-grained sandy sediments occur along the shallower shorelines (Hodgkin & Clark, 1988a). All five estuaries are fed by relatively large catchments and are located in a subtropical to temperate climate with relatively low rainfall (Table 3.1). The estuaries in this study also vary in opening regimes, and some are often subject to isolation from the sea by a sand bar. Stokes Inlet, Beaufort Inlet and Wellstead Estuary are normally closed to the ocean and are located in areas of low rainfall, but they open briefly during exceptionally high river flow caused by high rainfall. Wilson Inlet is artificially breached annually during winter when high catchment flow threatens to inundate the township, and Leschenault Estuary has been artificially opened via “the Cut” since 1952 (Hodgkin & Lenanton, 1981; Hodgkin & Hesp, 1998; Chuwen et al., 2009).

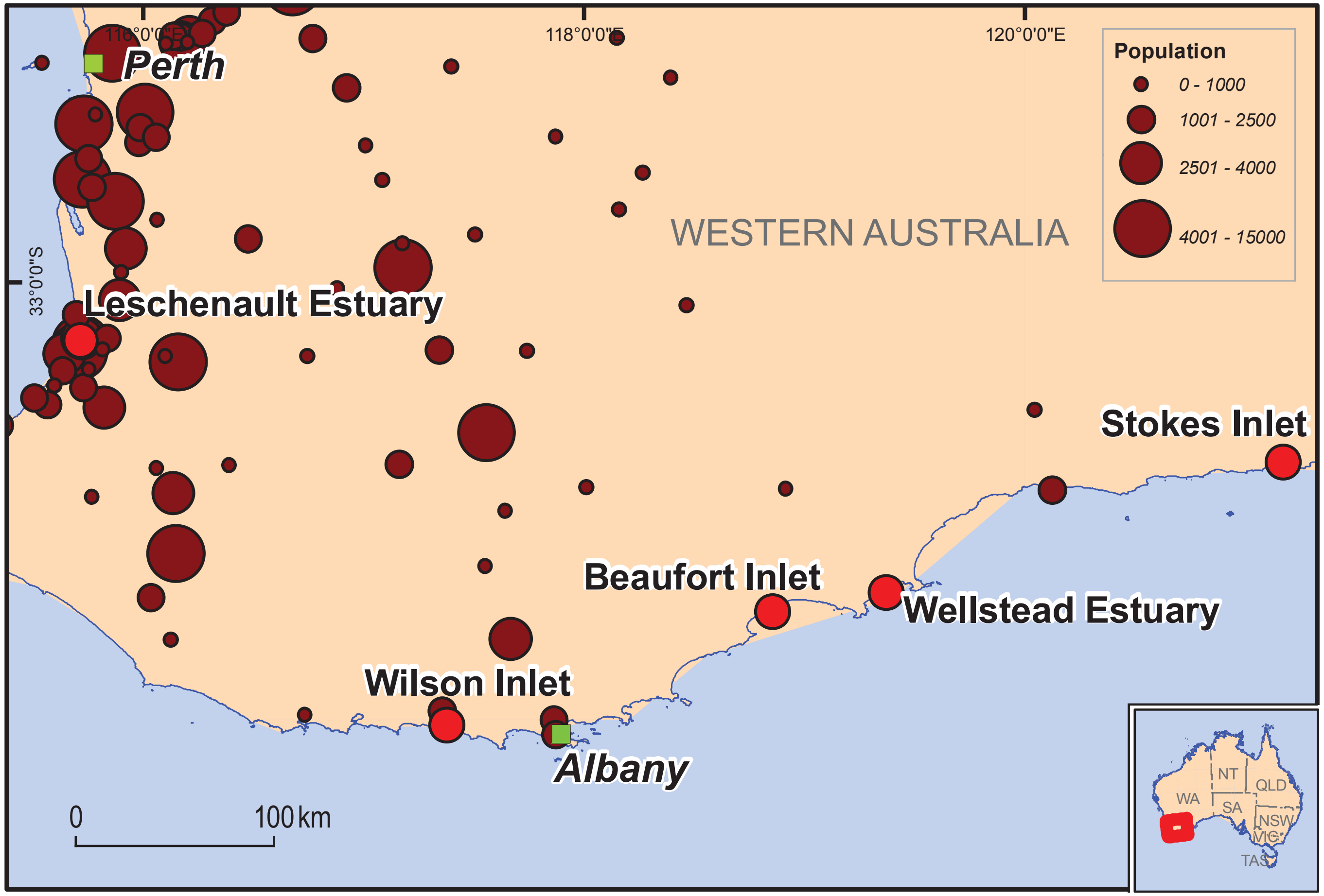


Figure 3.1: The location of the five estuaries from this study in Western Australia. Red circles indicate the five estuaries surveyed, green squares represents cities and proportional dark red circles represent population of townships.

Table 3.1: The physiographic settings of the five estuaries surveyed in Western Australia

Estuary	Estuary size	Estuary shape & dimensions^	Depth range	Connection to the sea	Associated catchment	Catchment cleared	Mean annual discharge	Mean Annual Rainfall	Refs*
Wellstead Estuary	3.20 km^2	Elongate 2 x 6 km (widest)	0-1.5 m	Normally closed	1,481 km^2	>80 %	14.0	494 (239-741)	3,4,5,6
Beaufort Estuary	4.81 km^2	Oval 1 x 3 km	0- 1.5 m	Normally closed	6,260 km^2	>84 %	3.6	600	3,4,6
Stokes Inlet	14 km^2	Elongate 2 x 10 km (when full)	0–7 m	Normally closed	6,348 km^2	65-70 %	11.9	520 (296-769)	3,4,5,6
Leschenault Estuary	26.16 km^2	Oval 3 x 14 km(when full)	0-2 m	Permanently open (artificially since 1951)	4,552 km^2	48 %	570.0	964	3,4,6
Wilson Inlet	45.23 km^2	Elongate/Oval 14 x 4 km	0-5 m	Open in Winter (~70-100 days)	2,911 km^2	60 %	161.4	765 (577-975)	1,2,3,4,5,6

^ Refer to Figure 3.4 for details on estuary shape
*References: [1]Ranasinghe & Pattiaratchi, 1999; [2]Hodgkin & Clark, 1988a, [3] OzCoasts website, [4]Hodgkin & Hesp, 1998, [5]Commonwealth Bureau of Meteorology, unpublished data, [6]Pen, 1999.

3.2.2 Survey design

Physical data acquisition

Two surveys were undertaken to collect physical and biological data for five estuaries. Four estuaries (Leschenault Estuary, Stokes Inlet, Wellstead Estuary and Beaufort Inlet) were sampled early in 2009 (17th March – 6th April), while Wilson Inlet was sampled late in 2008 (21st – 26th October) due to survey scheduling availabilities between Geoscience Australia and the Department of Water, Western Australia. To characterise the bathymetry of each estuary, existing depth contour data were obtained from the Department of Water, Western Australia. Depths for each estuary were then plotted in ArcGIS (ESRI version 9.2) and used to generate spatially continuous layers of depth and slope (changes in depth and measured by degrees) for each estuary (Table 3.2). Using ArcGIS, estuarine shapefiles along with depth and slope layers were used to allocate sampling effort across the full extent and depth range of each of the five estuaries. Sampling sites were allocated within a grid formation across each estuary and in depths > 0.5 m. This method ensured that samples were collected across the spatial extent and depth range of the five estuaries. Depths < 0.5 m (e.g. inaccessible shallow mudflats) were not sampled due to the difficulties associated with towing a video camera and sampling shallow sites from a small boat. All sampling was undertaken during daylight hours (between 8 am and 2 pm) to ensure enough light was available to accurately characterise the benthic habitat using underwater video and a range of water quality parameters. Additionally, the camera system was fitted with 2 x 250 watt lights to ensure visibility in deeper habitats. Water quality parameters were measured at each site (secchi depth, water temperature, salinity and dissolved oxygen) and collected sediment samples, with a total of 103 sites sampled during the two surveys (Table 3.2).

At each sampling site, a secchi disk was deployed to: 1) examine spatial variability in water clarity within and between estuaries relative to physical and hydrodynamic features, and 2) ensure water clarity was sufficient (> 2 m) to undertake subsequent towed-underwater observations. To do this, a secchi disk (20 cm diam. disk with opposing black and white squares) connected to rope with incremental depths of 10 cm, was deployed vertically in the water at each site until it could not be seen from the surface. The depth at which the disc could not be seen anymore was recorded as the secchi depth (Hill & Wilkinson, 2004). Next, water quality parameters (water temperature, salinity and dissolved oxygen) were measured at the surface and bottom of the water column, by placing a Yellow Springs International Sonde (YSI© 600 XLM) in the water to 10 cm below the water surface, and then lowering it to the bottom of seabed and raising it by 10 cm (by marked gradations on the cord) above the seabed when the boat was stationary. The YSI Sonde was attached to a linked user interface onboard and measurements were recorded of the surface and bottom of the water column after it had been submerged. Next, sediment samples were collected at all 103 sites to measure total inorganic carbon (TCO_2), sediment grain size (percent mud and sand), porosity, total organic carbon, total nitrogen - and their respective stable isotopic compositions ($\delta^{13}C$ and $\delta^{15}N$). Each core was collected via a pole attached to a ball valve and a Perspex corer (9 cm diameter) and sampled for total inorganic carbon (TCO_2) following Radke et al. (2004). The remaining sediments were then sampled for grain size analysis by placing the rest of the core in separate FALCON vials (95-130 g).

Table 3.2: Summary information on physical samples (water quality, pore water, sediment composition) collected from five south-west estuaries.

Estuary	**No. sites sampled**	**No. water quality samples collected (surface /bottom)**	**No. sediment samples**	**TCO2 (pore water samples) mmol m-2***	**δ15 N***	**δ13C***	**Sand-mud-gravel composition (%)***
Wellstead Estuary	13	13/13	13	0	13	13	13
Beaufort Estuary	10	10/9	10	0	10	10	10
Stokes Inlet	20	20/20	20	20	17	20	20
Leschenault Estuary	29	23/23	29	29	29	29	29
Wilson Inlet	31	0/0	31	31	27	31	31
Total	**103**	66	103	80	96	103	103

* Please refer to Section 3.2.2 for unit descriptions and physical sample collection methods

Sediment analyses

To calculate the amount of organic reactivity within surficial sediments at each site, pore water samples were collected at two different times; once as soon as the sample was collected and then 8 hours later, as described by Papadimitriou et al. (2002) and Radke et al. (2004). These samples were analysed using an Apollo SciTech AS-C3 Dissolved Inorganic Carbon analyser (Murray et al., 2007). Grain size distributions of mud (< 63 μm) and sand (63μm – 2,000 μm) for each sub-sample were then measured using a Malvern Mastersizer 2000 laser-particle size analyser (Woolfe & Michibayashi, 1995). Grain size analysis was completed using the method of moments with GRADISTAT v.4.0. (Blott & Pye, 2001). Percent volume laser data was used to calculate grain size statistics of sorting, mean grain size, skewness and kurtosis, which were expressed in metric units. Freeze-dried sediments were analysed for porosity (the measure of void space between grains) by measuring water loss and calculating the difference and stable isotopes, total organic carbon and total nitrogen were measured using a Thermo Finnigan Flash elemental analyser (Murray et al., 2007). Additionally, carbon samples were measured for the separation of N_2 and CO_2 gas in gas chromatography columns (Murray et al., 2007).

3.2.3 Towed video and drop camera survey

Towed video and drop camera sampling sites were chosen to overlap with the sampling design of the physical data collection sites, so that most sites recorded both physical data and biological towed-video/drop camera observations. However, not all sites had both sampling types due to logistical constraints. Underwater video transects were allocated equally across the extent of each estuary to traverse over depth-associated biological habitat transitions. In contrast, drop camera stations were allocated throughout the basin of each estuary such that the number of sample sites depended on the size of each estuary and time availability. Underwater towed-video and drop camera observations (Frame of View; FoV) were used to map and characterise the substratum type (rock, boulders, shell, soft sediments (sand and mud)), biological (seagrass/algae), bedform structure (e.g. flat, low, moderate or high relief sediments, sediment ripples or waves and bioturbated bedforms), and the associated epibenthos (percent cover of key taxa, and presence of all epibenthos) of each estuary. A total of 41 towed-video transects and 99 drop cameras were collected from the five estuaries, sampling a total of 39.40 linear kms in depths of 0.4 to 6 m (Table 3.3).

Table 3.3: Summary information on the number of towed video-transects and drop camera samples collected from the five south-west estuaries.

Estuary	**No. Towed Video Transects**	**No. Drop camera Sites**	**No. FoV^ characterisations total (transects + drop camera)**	**Linear km* surveyed**	**Depths sampled (m)**
Wellstead Estuary	9	13	446 (224 + 222)	5.50	0.4 – 1.4
Beaufort Estuary	7	10	336 (165 + 171)	4.14	0.5 – 3.1
Stokes Inlet	8	21	666 (309 + 357)	8.21	0.5 – 7.2
Leschenault Estuary	8	25	721 (324 + 397)	8.89	0.5 – 1.3
Wilson Inlet	9	30	1,028 (533 + 495)	12.67	0.7 – 4.5
Total	**41**	**99**	**3197**	**39.40**	

* Linear kms estimated using average speed (1 knot) and p/30 s x number of observations.

^ Frame of View as defined in Section 3.2.3

To characterise alongshore and offshore patterns in benthic habitat structure, towed-video transects were sampled perpendicular to the shore for approximately 300 m (between 120 – 1,000 m), depending on estuary width, with transects separated by approximately 200 m around each estuary. However, the distance between transects varied up to 3 km due to time restrictions allocated for each estuary (Fig. 3.2). To sample the central regions of each estuary, drop camera observations were allocated within a grid formation throughout the basins of each estuary. Drop camera sites characterised substratum type and key taxa and were shorter in distance than transects, but were adequate enough for the person on board the boat to record the benthic environment to a minimum of four FoV characterisations per site (4-7; min. of 2 mins). At each site, the camera was lowered to approximately 1 m above the seabed and allowed to drift with the current for approximately ~1-5 mins (50-100 m). Each site comprised a central point, with four additional sites collected approximately 200 m apart and positioned north, east, south, and west from the central point. Wilson Inlet did not have a central drop camera site and consisted of four site points allocated in a square formation due to modifications made to consequent surveys. As mentioned above, physical samples in the subsequent 2009 survey were collected from a central drop camera site which made it easier to determine drop sites sampled for physical variables. As time was limited, this combination of methods was used to ensure that habitat composition and transitions across depth and offshore gradients were adequately quantified.

Benthic habitats within each estuary were characterised along each transect and at each drop camera site using a small forward-facing (30 x 50 cm) Raytech towed-video camera system (see Roob et al., 1998; Anderson et al., 2008; Nichol et al., 2009) (Fig. 3.3). During each transect, the camera system was deployed from the back of a small punt (2 x 5 m) and towed at a constant altitude of approximately 1 m above the seabed. A real-time video feed to a ship-board computer enabled researchers to observe (Fig. 3.4) and avoid collision with the seabed or underwater objects by altering the amount of cable supplied to the camera. All video footage was recorded to mini-DV tapes using a portable Sony HD Mini-DV player (Model: GV-HD700, see Anderson et al., 2009a). The spatial locations of each data entry were recorded while in the field using GNAV Real-time GIS Tracker' program (© Gerry Hatcher, 2002), which was attached to a hand-held Garmin 76S differential GPS. Every FoV characterisation was linked to GPS positions as each point contained UTC date and time, both of which were found on the video time stamp of each video transect and drop camera site.

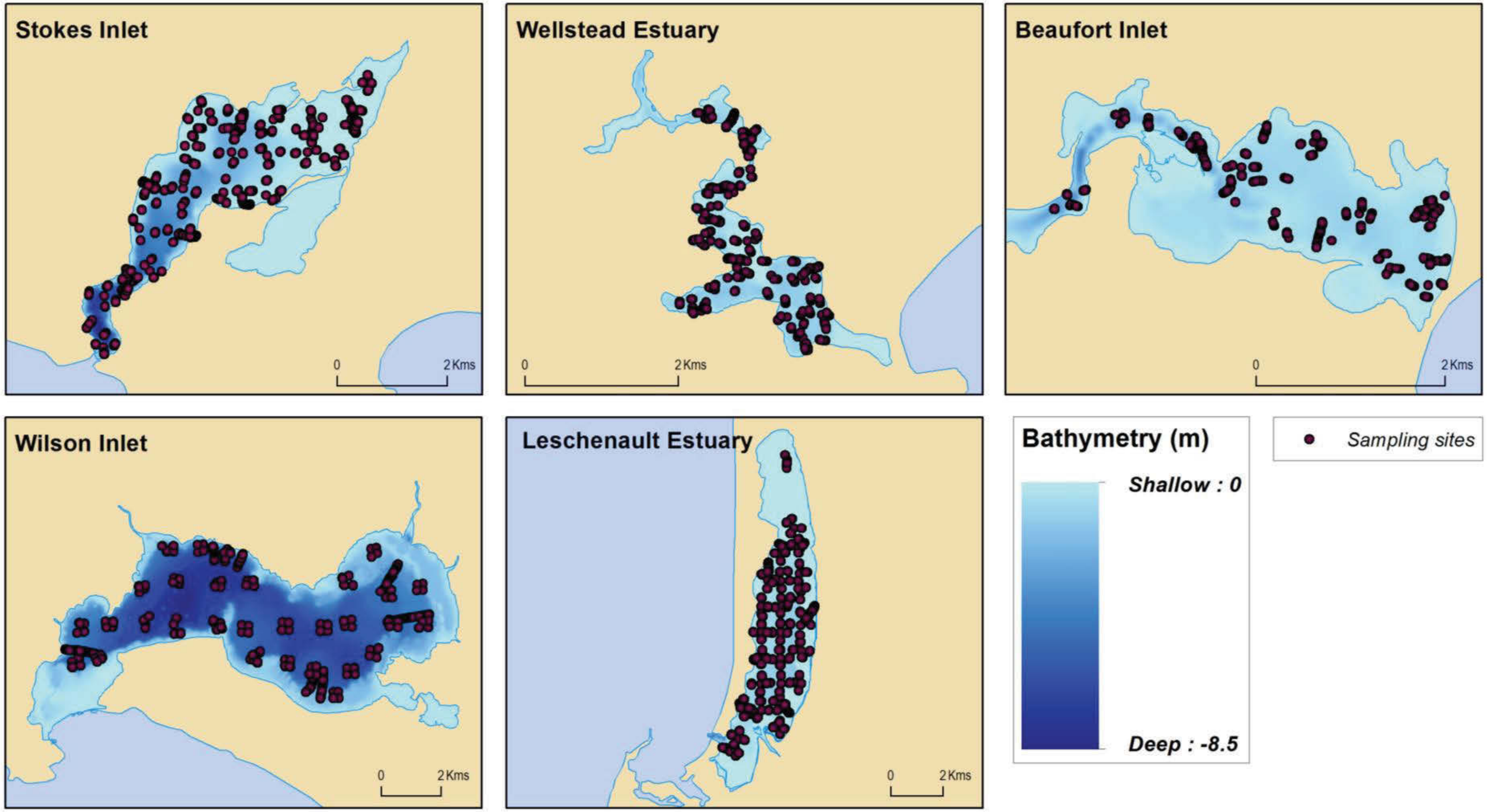


Figure 3.2: The location of video sampling sites in each estuary sampled in Western Australia, with underlying bathymetry

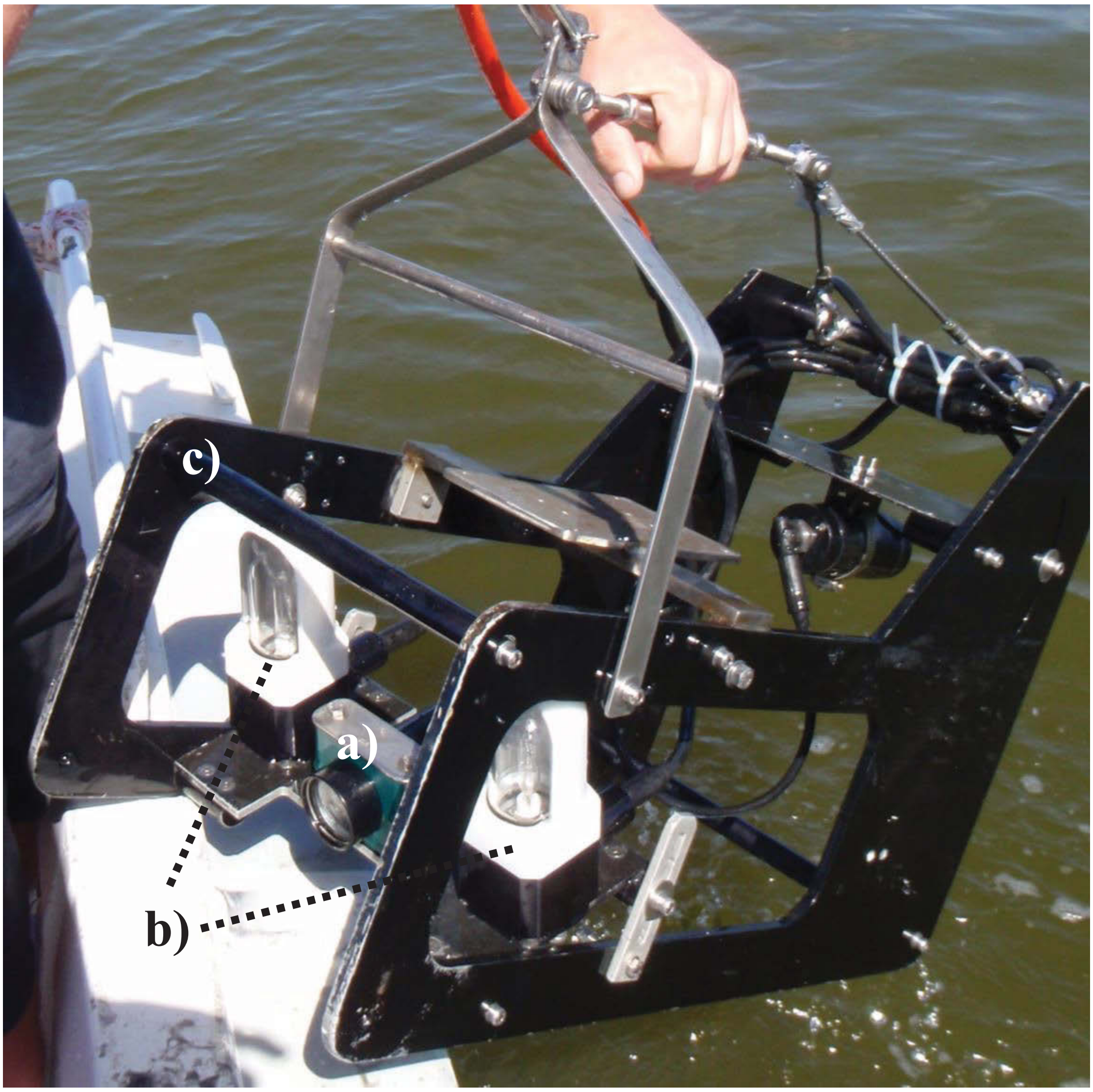


Figure 3.3: Small hand-deployable Raytech high-resolution towed-video system. System includes: (a) high-resolution video camera; (b) two 250watt lights; (c) steel frame with dimensions 30 x 50 cm.

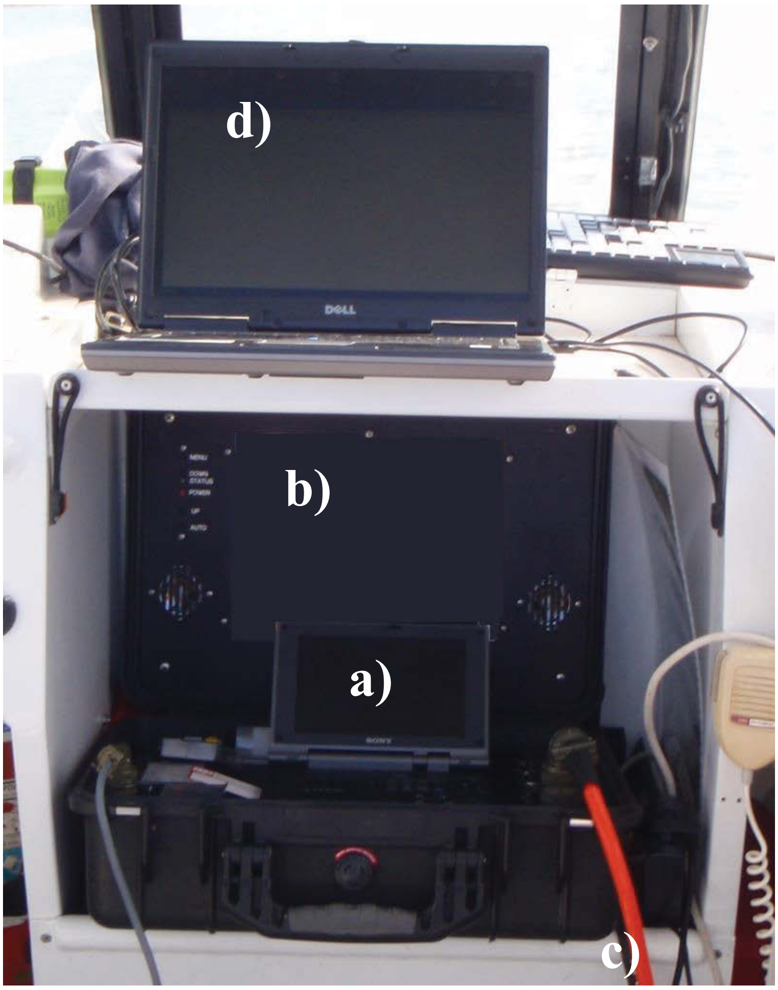


Figure 3.4: Image of the top-side unit containing: (a) Sony mini-DV cassette recorder; (b) video monitor; (c) coaxial cable connection to camera; (d) laptop computer is connected to GPS and programmable Cherry©Keyboard.

Underwater video footage was post-processed where substratum type, bedform structure, habitat forming taxa, and all macrobenthos were recorded for a 15-sec FoV (approx. 15 m) every 30 seconds of video footage. For each FoV substratum composition (rock, boulders (<25.5 cm), shell hash, mud, sand – defined by Greene et al., 1999; Greene et al., 2007) was visually quantified to 5% intervals. However, due to the difficulty in consistently distinguishing muddy sands from sandy mud, mud and sand were subsequently combined into a single 'soft sediment' category. Sediment geomorphology (bedform and relief) was characterised for each 15-sec FoV following the protocol of Anderson et al. (2008) and Anderson et al. (2009a) where bedform (hummocky, ripples) and relief (flat (0 m), low (<1 m), moderate (1-3 m)) was recorded. The percent cover of all habitat-forming species (e.g. species of seagrass, macroalgae, and invertebrates) was then visually assigned within each 15-sec FoV to 5% intervals, similar to studies by Mortensen & Buhl-Mortensen (2004) and Cappo et al. (2011). Habitat-forming taxa included three seagrass species (e.g. *Ruppia megacarpa, Halophila ovalis* and *Heterozostera tasmanica*), two red macroalgal forms (e.g. filamentous, branching), four green macroalgal forms ((branching *Lamprothamnium papulosum*, branching *Polyphysa peniculus*), green-slime and unknown)), and two invertebrate taxa (mussel clumps formed by *Mytilus edulis* and polychaete mounds created by *Ficopamatus enigmaticus*). As seagrasses are often a dominant component of estuaries and may vary in their physical structure and condition, Blade length was recorded and epiphyte load estimated for each seagrass species per FoV (Table 3.4). Seagrass length was visually estimated to three categories of short (<10 cm), medium (10 – 40 cm) and long (>40 cm). Seagrass condition was classified by the amount of estimated epiphytic algae (epiphyte load) smothering seagrass leaves to 5% cover precision. Epiphyte load can be an important indicator of eutrophication, where nutrient enrichment from catchment clearing, agriculture, and urban waste (introducing mainly phosphorous and nitrogen) can result in prolific algal growth (Ierodiocounou & Laurensen 2002; Krause-jenson et al., 2007). We acknowledge that estimating epiphytic cover from video is likely to result in a high source of error, but aimed to compare these data with the empirical data from Chapter 4. All other epibenthos/taxa observed within each 15-sec FoV were identified to lowest taxonomic category discernable and recorded as presence/absence (e.g. shrimp, crabs, and fish: full list of taxa provided in Table A1 of Appendix). All FoV video data were entered into a Microsoft Access Database (Microsoft ® Office Access 2007).

As part of a pilot study to compare video sampling methods for estimating percent cover of benthic habitats, percent cover was also estimated using the Coral Point Count extension (CPCe) point-count method (Kohler & Gill, 2006). This was done to ensure that both methods were comparatively similar in percent cover estimations of key benthic habitats from underwater video. Here, 200 FoV video characterisations (600 separate frames) were randomly selected to include a range of habitats within each of the five estuaries. For each 15-sec FoV, 20 CPCe points from three randomly separated frames that did not overlap (separated by ~5 sec intervals) were used to estimate total percent cover. The values were then compared with FoV % cover method to determine whether there was a difference between the two approaches. The two methods were highly positively correlated and indicated little difference between the methods (r^2=0.88 Pearson's correlation coefficient, $p<0.0001$). The point-count method provides an accurate and repeatable method of analysing seafloor images but is highly time consuming (up to 7 times longer than my FoV method), as it involves acquiring the still images for the 15-sec FoV, overlaying points over a still image of the seafloor, counting the number of randomly allocated points for each benthic habitat and recording this data within a database. Consequently, percent cover estimates to 5% intervals using the FoV percent cover method was deemed suitable for the study.

3.2.4 Data analysis

To determine the importance of physical configuration on benthic habitat configuration, several physical and spatial variables (e.g. depth and slope) were calculated in ArcGIS. Spatial variables (distance to the shore, distance to estuary bar opening, and distance to nearest river entrance) were measured using the point distance tool in ArcGIS. Raster layers were then created for each distance variable (distance to coast, river and sand bar) using the Euclidian distance tool in Spatial Analyst, and latitude and longitude with Map Algebra in ArcGIS. Bathymetric contour data sourced from the Department of Water, Western Australia, at a resolution of 0.5 m were interpolated to create a spatially continuous bathymetric layer using the TopoToRaster Tool (Wahba & Wendelberger, 1980). The bathymetric contour data was then used to derive slope and topographic complexity (maximum rate of change in elevation of adjacent cells) values across the seafloor of each estuary.

Sediment samples collected within each estuary were then interpolated using the Inverse Distance Squared (IDS) method to create spatially continuous sediment layers for all physical and sedimentary data (e.g. Percent mud, percent sand, porosity etc.). Due to the relatively low numbers of samples per estuary (Table 3.2; 10 – 31 samples), IDS was chosen as this method is simplistic and robust enough to handle smaller sample sizes (Li & Heap, 2008). Two measurements, Relative Mean Absolute Error (RMAE% relative error in predictions) and Relative Root Mean Square Error (RRMSE%) were used to determine the interpolation and model performance for each physical variable in each estuary using the predicted and measured variables cross validation plot in Geospatial Analyst, ArcGIS (Table A2) (Li et al., 2011).

The formulas for calculating performance of interpolation and models are:

$$RMAE = \frac{1}{n}\sum_{i=1}^{n} |(p_i - o_i)/o_i|$$

and,

$$RRMSE = [\frac{1}{n}\sum_{i=1}^{n}([p_i - o_i]/o_i)^2]^{1/2}$$

Where n=number of observations, o=observed value, p=predicted value, o_i=standardized observed value (Li & Heap, 2008).

Means and standard errors for each substratum type were then calculated for each estuary and are graphically presented. Percent occurrence of epifaunal presence was calculated by the number of characterisations present in proportion to the total number of FoV observations per estuary. The relationship between each estuary from physical and derived variables was also assessed using linear models with estuaries as the single factor, and a Tukey's HSD post-hoc test to determine which estuaries significantly differed from each other (Table 3.4). Homogeneity of variances and normality assumptions were examined by residual plots.

Predictive models: Relationship between physical sediments and biological data

Ensemble regression trees (called 'Random Forests' (RFs)) were used to visualise the spatial distribution and abundance of benthic habitat-formers (Breiman, 2001). These models used physical, biological and spatial data to explain and predict environmental patterns within the five estuaries. RFs are an ensemble method which consists of many classification trees, where the output of each tree is averaged. Each habitat-former (e.g. *R. megacarpa* etc.) was modelled using the 'randomForest' package in R (R Development Core Team, 2011) (Liaw & Wiener, 2002) with standard deviation modelled using the 'ModelMap' package (Freeman & Frescino, 2009). Variable importance plots were used to determine the main variables contributing to the model (Stroble et al., 2009). The model randomly used 75% of the data to create a model, while the remaining 25% of the data was used for validation. The same measures of

RMAE and RRMSE were used to evaluate how well the model performed on the remaining 25% of the data.

Standard deviation maps were used to illustrate the degree of uncertainty from each prediction (Freeman & Frescino, 2009). The interpolated physical layers (e.g. space, depth, slope and sediments) were then included as input variables into the most optimal RF model (highest variance explained). Continuous prediction layers of percent cover of habitat-formers (e.g. *R. megacarpa*) were then created for each estuary in R using the 'raster' package by Hijmans & Etten (2012). We calculated the RMAE on the withheld portion of the data (25% of the test data). Partial plots were used to display the relationship between predicted percent cover of each habitat type and physical variables (Liaw & Wiener, 2002). Due to the slight attenuation of the predicted surface of each benthic habitat (i.e. either over-estimated low values and under-estimated high values) when all distance-derived variables were included in the RF model, each model was run on three different combinations of the data: 1) all predictor variables, 2) all predictor variables except for distance variables, and 3) all predictor variables except for 'distance to estuary bar opening' and 'distance to nearest river entrance' variables. This was based on preliminary analyses of how well each model performed in Wilson Inlet using *R. megacarpa* coverage. We selected the best performing model to represent realistic prediction ranges of each benthic habitat in each estuary using the RMAE of the model, comparison of the output ranges of the prediction surface, and visual comparison between the underwater video points of measured percent cover from each benthic habitat and the predicted surface. For modelling purposes, red and green algae morphologies were each amalgamated and predicted.

3.3 Results

3.3.1 Physical and spatial setting

3.3.1.1 Permanently and artificially opened vs. normally closed estuaries

Estuaries in the north and south-west were larger, received higher annual rainfall, and artificially or permanently opened, whereas estuaries along the south were normally closed, smaller in size, received less annual rainfall, and had a greater proportion of their catchment cleared (Table 3.1 & 3.4). In the north, hydrodynamics of estuaries were driven by tidal influences, while in the south, rainfall catchment runoff (reflected in the median rainfall distribution) were key drivers (Table 3.1). Dissolved oxygen content was significantly different between large and small estuaries (e.g. Leschenault and Stokes to Beaufort and Wellstead Estuary (Table 3.4, $F_{4, 98}$, p<0.0001)). Stokes and Leschenault Estuary had significantly higher dissolved oxygen content in surficial waters, compared to Beaufort and Wellstead Estuary. Also, Leschenault had higher dissolved oxygen content in bottom waters than Beaufort Inlet (Table 3.4, $F_{4, 98}$, p<0.0001) indicating permanently opened estuaries were well-mixed throughout the water column. Clear depth-related differences between surface and bottom dissolved oxygen content were also evident in the Stokes and Wellstead Estuaries, indicating that stratification of the water column in both estuaries was present at the time of survey. Similarly, salinities were also significantly different between estuaries, with some localised differences within estuaries (Table 3.4). Smaller estuaries, with high amounts of their catchments cleared (e.g. Beaufort Inlet and Wellstead Estuary), had greater variability in their salinity ranges between surface and bottom waters (Table 3.1 & 3.4). Stokes and Wellstead Estuaries had different surface and bottom water temperatures compared to Leschenault Estuary (Table 3.4). Slope and distance measures between river entrances, estuary bar entrances and the shoreline also varied significantly between estuaries (Table 3.4). Normally closed estuaries contained higher ranges of slope compared to artificially and permanently opened estuaries ($F_{4, 98}$, p<0.0001) illustrating the different spatial configurations of these estuaries and the degree of closure from the coast (Table 3.4).

Table 3.4: Summary of physical variables collected for each estuary collected in this study. Means and ± Standard Error with minimum and maximum in brackets. ANOVAs were completed on each variable and associated P-values with 4, 98 df. Tukey's HSD post-hoc test indicated as connecting lines with no significance between estuaries (subsequent symbols pertaining to each estuary). Water quality variables were not available for Wilson Inlet.

Physical Variables [label]*	**Leschenault** Mean ± SE (range)	**Wilson** Mean ± SE (range)	**Beaufort** Mean ± SE (range)	**Wellstead** Mean ± SE (range)	**Stokes** Mean ± SE (range)	**Tukey's**	**P**
Depth (m) [depth]	1.1 ± 0.1 (0.4-2.1)	3.0 ± 0.2 (1-4.5)	1.2 ± 0.2 (0.5-2.7)	1.1 ± 0.1 (0.5-1.7)	2.5 ± 0.5 (0.6-8.0)	Wi S L B We	<.0001
Slope [slope]	0.1 ± 0.0 (0.0-0.8)	0.2 ± 0.0 (0.0-0.8)	0.4 ±0.1 (0.0-1.5)	0.3 ± 0.1 (0.0-0.9)	0.5 ± 0.2 (0.0-3.2)	S B We Wi L	<.0001
Distance to coast (m) [coast]	541.9 ± 41.9 (80.3-1075.3)	789 ± 73.3 (155.1-1638.8)	212.6 ± 28.4 (91.7-364.9)	110.4 ± 17.7 (32.3-220.9)	275.7 ± 43.2 (41.3-772.1)	Wi L S B We	<.0001
Distance to river (m) [river]	4830.3 ± 557.2 (729.8-10876.9)	3144.6 ± 257.7 (729-5684)	1408 ± 255.3 (328.6-2612.4)	1990.2 ± 254.7 (335.4-3264.7)	2682.9 ± 380.5 (524.3-6174.5)	L Wi S We B	<.0001
Distance to estuary bar (m) [bar]	5460.9 ± 548.8 (626.4-11309.3)	7716.5 ± 580.3 (1717.4-12525)	2002 ± 426.3 (262.7-4065.2)	2233.9 ± 251.5 (909.8-3826.2)	3971.3 ± 417.5 (190-7014.1)	We B S L Wi	<.0001
Sediment parameters							
Mud [mud]	60.9 ± 4.3 (5.3-91.5)	55.5 ± 7.7 (0-100)	79.6 ± 4.8 (40.2-92.7)	70.4 ± 7.5 (16.8-97.0)	35.6 ± 9.2 (1.6-96.6)	B We L Wi S	.0050
Sand [sand]	39.1 ± 4.3 (8.5-94.7)	44.5 ± 7.7 (0-100)	20.4 ± 4.8 (7.3-59.8)	29.6 ± 7.5 (3.0-83.2)	64.4 ± 9.2 (3.4-98.4)	S Wi L We B	.0080
Mean grain size [mean]	58.7 ± 12.2 (14.7-298.1)	116.5 ± 27.3 (4.8-461.5)	23 ± 8.8 (8.6-101.4)	36.8 ± 10.6 (8.7-116.8)	167 ± 29.3 (7.3-367.7)	S Wi L We B	.0164
Porosity [por]	74.6 ± 1.9 (47.0-89.8)	70.6 ± 3.5 (34.0-88.3)	82.4 ± 2.8 (59.3-91.3)	85.4 ± 2.5 (64.6-92.5)	62.1 ± 4.7 (41.8-90.7)	We B L Wi S	.0005

Physical Variables [label]*	**Leschenault** Mean ± SE (range)	**Wilson** Mean ± SE (range)	**Beaufort** Mean ± SE (range)	**Wellstead** Mean ± SE (range)	**Stokes** Mean ± SE (range)	**Tukey's**	**P**
Sorting [sort]	5.1 ± 0.2 (2.6-7.9)	3.4 ± 0.3 (1.3-7.1)	5.2 ± 0.5 (3.6-9.3)	4.1 ± 0.2 (2.9-6.2)	3.2 ± 0.2 (1.9-4.8)	B L We Wi S	<.0001
Skewness [skew]	-0.5 ± 0.1 (-3.2-0.2)	-0.5 ± 0.2 (-3.2-0.4)	0.2 ± 0.1 (-0.6-0.4)	-0.3 ± 0.2 (-2-0.1)	-2.1 ± 0.4 (-4.6-0.3)	B We L S Wi	.0411
Kurtosis [kurt]	3.2 ± 0.4 (1.9-14.8)	4 ± 0.6 (1.9-19.3)	2.6 ± 0.1 (1.8-3.2)	3.1 ± 0.3 (2.3-33)	11.9 ± 2.1 (2.3-33)	Si Wi L We B	<.0001
%N [n]	0.3 ± 0 (0-1.0)	0.6 ± 0.1 (0-1.1)	0.4 ± 0 (0.1-0.7)	0.8 ± 0.1 (0.1-1.3)	0.2 ± 0.1 (0-0.6)	We Wi B L S	<.0001
%C [c]	2.3 ± 0.3 (0.1-6.8)	4.6 ± 0.7 (0-9.4)	3.9 ± 0.4 (0.8-5.4)	6.6 ± 0.9 (1.1-11.0)	1.6 ± 0.5 (0-5.0)	We Wi B L S	<.0001
$\delta^{13}C$ [d13C]	-17.1 ± 0.2 (-19.4 - -14.5)	-21.4 ± 0.1 (-22.5 - -19.9)	-24.8 ± 0.3 (-26.1 - -22.6)	-19.9 ± 0.5 (-22.3 - -16.8)	-22.4 ± 0.4 (-24.5 - -18.7)	L We Wi S B	<0.001
$\delta^{15}N$ [d15N]	2.2 ± 0.2 (-0.02-3.8)	4.5 ± 0.1 (2.3-5.6)	3.7 ± 0.1 (3.1-4.3)	0.6 ± 0.2 (0.5-3.0)	3.9 ± 0.4 (0.5-6.8)	We L B S Wi	<0.001
Water quality parameters							
Dissolved Oxygen (Surface) [dos]	7.1 ± 0.1 (6.2-8.7)	-	6.1 ± 0.2 (5.0-7.2)	6.0 ± 0.1 (5.11-6.6)	6.9 ± 0.1 (6.2-7.6)	L S B We	<.0001
Dissolved Oxygen (Bottom) [dob]	7.1 ± 0.2 (5.3-10.1)	-	5.2 ± 0.5 (1.7-6.7)	6.1 ± 0.2 (5.1-7.7)	6.1 ± 0.2 (0.6-7.6)	L S We B	.0038
Salinity (Surface) [sals]	39.1 ± 0.5 (35.8-43.4)	-	45.1 ± 0.8 (40.7-48.3)	39.2 ± 0.5 (35.9-41.8)	35.1 ± 0.2 (33.3-36.4)	B We L S	<.0000
Salinity (Bottom) [salb]	39.5 ± 0.5 (36.6-43.4)	-	46.5 ± 0.3 (45.3-48.1)	39.5 ± 0.4 (36.7-41.3)	38.0 ± 1.6 (33.7-55.1)	B L We S	.0002
Temperature (Surface) [tems]	19.4 ± 0.3 (16.6-21.4)	-	20.7 ± 0.5 (18.0-23.4)	21.2 ± 0.4 (18.6-23.8)	21.2 ± 0.3 (18.9-23.7)	We S B L	.0001

Physical Variables [label]*	**Leschenault** Mean ± SE (range)	**Wilson** Mean ± SE (range)	**Beaufort** Mean ± SE (range)	**Wellstead** Mean ± SE (range)	**Stokes** Mean ± SE (range)	**Tukey's**	**P**
Temperature (Bottom) [temb]	18.8 ± 0.6 (6.3-20.8)	-	20.5 ± 0.4 (18.2-22.1)	20.7 ± 0.3 (19.1-22.2)	21.2 ± 0.2 (19.4-23.1)	S We B L	.0007
Secchi Depth (m) [secchi]	1.1 ± 0.1 (0.4-1.9)	-	0.7 ± 0.0 (0.5-0.8)	0.7 ± 0.0 (0.5-0.9)	0.8 ± 0.0 (0.5-1.1)	L S B We	.0146

*Abbreviated Labels within physical variable column for Figures 3.9-3.13

3.3.1.2 Shallow vs. deep estuary environments

Wilson and Stokes Inlets were the two deepest estuaries and had similar sediment characteristics (high mean grain size, low sorting), compared to the other three estuaries (Table 3.4, $F_{4,\ 98}$ p<0.05). Although Stokes was the deepest, Wilson Inlet was deeper over a much larger basin region than all other estuaries (Table 3.1) while Stokes Inlet had a shallower overall basin (0 - 4 m) but supported a significantly deeper channel (~ 7 m) through the south facing bar entrance (Table 3.1). Additionally, the deep channel within Stokes Inlet revealed stratification in salinity, temperature and dissolved oxygen content. Secchi depths in Leschenault Inlet differed significantly between Beaufort and Wellstead Estuary, indicating greater water column visibility within Leschenault Estuary (Table 3.4, p<0.015). Sediment sorting was significantly different between Beaufort and Wilson Inlet ($F_{4,\ 98,}$ p<0.05). Well-sorted sediments (typically similar sized sandy sediments) were located in relatively shallow sections of all five estuaries, while poorly-sorted sediments (wide range of sediment sizes dominated by muds) were found in deeper sections of estuaries. Additionally, in Stokes and Wilson Inlets, well-sorted sandy sediments were also found in areas of rippled sediments which were adjacent to the bar and river entrances, as seen from the video. Conversely, kurtosis (i.e. the height and width of the sediment distribution curve) ranged from mesokurtic (normal distribution) within some central sections of estuaries, to leptokurtic (more concentrated or 'peaked' distribution) adjacent to bar and river entrances (Table 3.4), indicating greater physical transport processes affecting the sediment at different spatial locations. Surficial sediments (e.g. mud content, porosity, sorting, kurtosis, skewness and mean grain size) varied significantly between estuaries (Table 3.4). Grain size distributions showed that most estuaries accumulated progressively higher mud content and poorly sorted sediments at depth a, particularly across the deep inner basins (Table 3.4). However, this spatial relationship was not apparent in the permanently opened Leschenault Estuary, where sediments were relatively sandy within the marine opening of the "Cut" and often muddy within the basin of the estuary. Inversely, sand content was higher around the edges and in the shallow sections of other estuaries. Porosity also differed significantly between normally closed and artificially- and permanently-opened estuaries, with higher porosity values in Wellstead and Beaufort estuaries compared to Leschenault and Wilson Inlets, with the exception of Stokes Inlet, which had the lowest porosity values (Table 4; $F_{4,98,}$ p<0.0005) (Table 3.4)). Skewness (i.e.

the degree and direction of departure from a normal distribution) was significantly different between all estuaries and followed similar patterns to sorting, with negatively skewed sediments adjacent to river entrances and estuary bar areas, indicating potential higher energy systems within these areas. Negatively skewed sediments indicate larger particles were deposited first, while finer sediments are transported either to the basin of the estuary or out to sea. In contrast, the inner sections of each estuary (i.e. basin areas) supported more normally distributed sediment grain sizes ($F_{4, 98}$, p<0.04). Isotopic compositions of carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$) were both significantly different ($F_{4,98}$, p<0.0001 between Leschenault Estuary and Beaufort Inlet, where $\delta^{13}C$ was lower at deeper depths and within and adjacent to rivers, which indicates higher terrigenous input in Beaufort Inlet (Table 3.4). Conversely, $\delta^{15}N$ was lower in sheltered, shallower areas of Beaufort Inlet.

3.3.2 Benthic habitat composition from video analysis

3.3.2.1 Physical habitat composition from underwater video

Underwater towed video (total of 39.40 linear km and 3,197 Frame of View (FoV) data points) was collected from shallow rim and deep basin zones across all estuaries (Table 3.3). Estuaries were characterised as soft-sediment environments with 64% mean cover ± 0.6 (S.E) and >87% occurrence of all sample points comprised of either homogeneous soft-sediments (18% occurrence) or soft mixed (69% occurrence) habitats. In total, soft-sediments comprised 65% cover ± 0.6 (S.E) (range: 0 – 100%), with mostly sandy sediments found in the shallow rim zone (46% occurrence), and muddy sediments from the deeper basin zones (32% occurrence). Coarser shell hash was also present in all five estuaries (10% of all observations), but only in discrete locations (1.41 mean % cover ± 0.1 (S.E), range: 0 – 70%), either along the shoreline, adjacent to estuary bar entrances, and within Parry River in Beaufort Inlet. In contrast to soft-sediments, hard substratum was rarely recorded (<0.4% of all observations; rocks (0.03%) or boulders (0.4%)), restricted to the shallow rim zones of Wilson and Beaufort Inlet, respectively. Seabed relief varied both between estuaries and spatially within estuaries. Irregular or hummocky bedforms were common in the shallow rim zones of these estuaries (mean: -1 m ± 0.02), while flat relief was commonly found in all estuaries within the deeper basin areas (mean depth: -2.6 m ± 0.03 S.E). Rippled sediments were commonly found adjacent to higher energy environments such as river entrances and estuary bars in Leschenault, Stokes and Wilson Inlet (mean depth: -1.7 m ± 0.02 (S.E)) and often contained well-sorted sediments. Stokes Inlet had greater slope and depth gradients and consequently, displayed a greater variety in relief, where low and moderate relief bedforms were present in areas with high slope (mean slope: 1.4 ± 0.02 (S.E) and 1.5 ± 0.02 (S.E), respectively). In comparison, Beaufort and Wellstead had relatively shallow basins and contained similar spatial distributions of hummocky sediments throughout the estuary, with no discernible relationship between relief and depth (Table 3.1). Leschenault Estuary was relatively shallow throughout the basin, whereas Wilson Inlet had a clearly defined basin with depths up to 4 m.

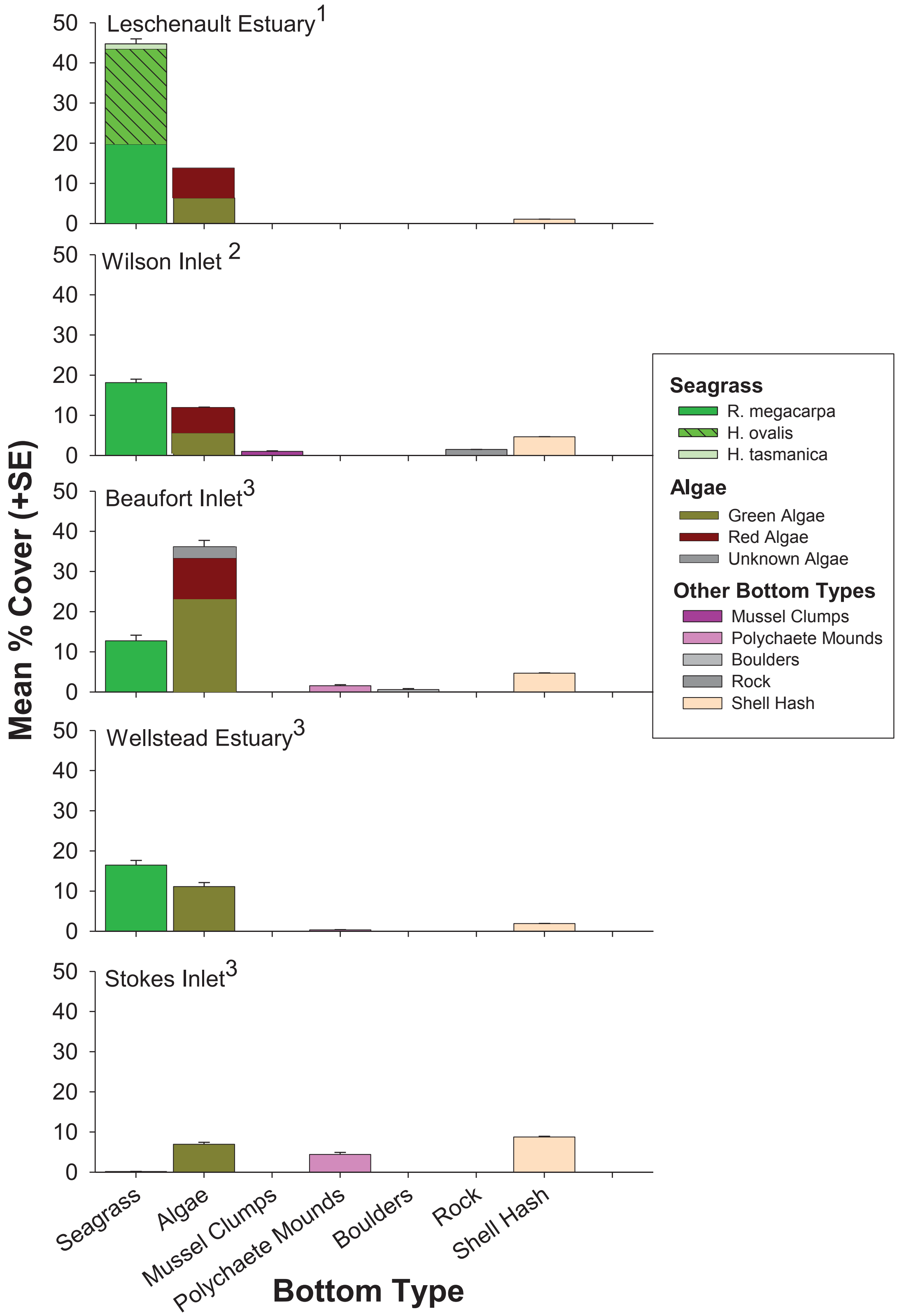


Figure 3.5: Relative composition of substratum and habitat-forming biota within and between the five estuaries, recorded from towed underwater video. Superscript indicates opening regimes: [1] = Permanently opened, [2] = artificially opened and [3] = normally closed. Error bars represent standard error of the mean. Please refer to Table 3.3 for the number of characterisations per estuary.

3.3.2.2 Biological habitat composition from underwater video

Three categories (seagrass, macroalgae and invertebrates) of biological habitats were recorded within this study (Fig. 3.5), and combined they covered 34% of the surveyed seabed (occurring at 77% of all locations with at least one biota type mentioned above). Marine plants, comprised of seagrasses and macroalgal species, were the most common habitat-forming group recorded, covering 32% of the seabed surveyed (occurring at 74% of all locations). Invertebrate habitat-formers comprised less than 2% of the total benthic cover (occurring at 9% of all locations). However, the occurrence and extent of these biological habitats varied between estuaries (Fig. 3.5). Total biotic habitat occurrence was highest in Leschenault, Wilson, Beaufort, and Wellstead Estuaries (range: 72 – 91% occurrence), whereas Stokes Inlet only had 50% occurrence of biotic habitats present.

Seagrass was the most abundant biotic cover recorded (overall mean % cover 20 ± 0.5 S.E., range: 0 – 100), occurring in all five estuaries and was the dominant biotic cover in Leschenault, Wellstead and Wilson Inlets. Three species of seagrass (*R. megacarpa, H. ovalis* and *H. tasmanica*) were recorded from the video, but the presence and spatial distribution of these species varied between estuaries. *R. megacarpa* was common in all five estuaries (35% total occurrence, mean % cover 13.7 ± 0.1 SE; Fig. 3.5 & 3.6) and in relatively shallow depths. However, *R. megacarpa* was only found in very low cover in Stokes Inlet (0.02% total occurrence, mean % cover 0.16% ± 0.05 SE). *H. ovalis* and *H. tasmanica* were only recorded in Leschenault Estuary (13.4% ± 1.0 and 3.2% ± 0.2 total occurrence, respectively) – the most northern of all five estuaries and the only estuary permanently opened to the sea. *H. ovalis* was distributed throughout Leschenault Estuary, with the greatest abundance found in the shallow northern and southern sections of the estuary, but absent from the basin. *H. tasmanica* co-occurred with *H. ovalis* in the southern sections of the estuary, where it was restricted to the sandy sediments of the ‘Cut’ – a channel open to marine influence in the Leschenault Estuary (Fig. 3.7).

Macroalgae was also a common habitat-former in all five estuaries (total of 58% occurrence, mean % cover 12.4 ± 1.6 (S.E)), both alone (29% occurrence 11.4% ± 0.34 SE) and intermixed within seagrass meadows (29% occurrence; 13.6% ± 0.33 SE). A total of six major macroalgal forms were present, but their presence and spatial distribution varied within and between estuaries. Red macroalgal forms (filamentous red, branching-red, unknown/other) were restricted to the three northern estuaries and accounted for a mean cover of 15.4%. In contrast, green algal forms (unknown, green slime, and branching (*Lamprothamnium papulosum, Polyphysa peniculus and Chaetomorpha sp.*)) were found in very low abundances from the northern-most Leschenault Estuary but were more abundant in the other, normally closed estuaries (Fig. 3.5). However, due to the low resolution of the video, some green branching forms were not easily distinguishable, so were consolidated into a broad category of 'unknown green algae'. *P. peniculus* and green slime were common in normally closed estuaries. Green slime forms were present in Leschenault Estuary (35% total occurrence, mean cover of 0.01% ± 0.01 (S.E)) but were more abundant in artificially opened (e.g. Wilson Inlet mean cover of 5.5% ± 0.2 (S.E) and normally closed estuaries (e.g. Wellstead Estuary (mean cover 11.2% ± 0.4 (S.E), Beaufort (mean cover 24.2% ± 0.4 (S.E)) and Stokes Inlet (mean cover 7.2% ± 0.2 (S.E))). Different species (growth forms) varied in their distributions, with green branching forms (*L. papulosum* and *P. peniculus*) common in relatively shallow areas, whereas green slime forms occupied deeper depth ranges within estuarine basins in all estuaries except for Leschenault Estuary.

Branching red algae was limited to Leschenault, Wilson and Wellstead Estuary; throughout Leschenault Estuary (mean cover of 5.7% ± 0.5), to localised distributions within deeper basin zones and along the northern section of Wilson Inlet (mean cover of 0.3% ± 0.04), and with very low occurrences in Wellstead Estuary (mean cover of 0.004% ± 0.004). However, filamentous red algae forms were present in all estuaries except for Stokes Inlet (mean cover of 4.4% ± 0.2). Filamentous red forms were distributed throughout Beaufort Inlet (mean cover of 10.6% ± 1.1) and in greatest abundances on the eastern and southern sections of Wilson Inlet at up to 4.3 m (mean cover of 6.4% ± 0.4), as well as adjacent to the 'Cut' and up to 1.1 m in Leschenault Estuary (mean cover of 0.4% ± 0.1). Additionally, filamentous red algae forms were more commonly found in conjunction with seagrass (83% of all filamentous red algal form occurrences; mean cover of 6.2% with seagrass and 0.9% mean cover without seagrass).

In contrast to marine plants, invertebrate habitat-formers were much less common (7.5% total occurrence) and limited to two species: *Ficopamatus enigmaticus,* a polychaete tubeworm (Family Serpulidae), which creates large ball-like features (up to 7 m in diameter) that were present in small to large clusters (up to 1 m in diameter) in 1-7 m water depth in all three of the normally closed estuaries (Stokes, Wellstead, and Beaufort) (6% total occurrence; mean cover of 1.2% ± 0.1 SE); and the black mussel, *Mytilus edulis* that was present as clumps, in small localised areas on the northern and western side of the estuary adjacent to Young's Lake and Cuppup Creek, within Wilson Inlet (1.5% total occurrence; mean cover of 0.3% ± 0.1 SE) (Figs. 3.5-3.6). Although abundances were often locally high for both species, the distribution of both species were highly patchy. Polychaete mounds were found in the sheltered central basin sections of Wellstead Estuary and Beaufort Inlet and along the deep channel slope in Stokes Inlet (7% occurrence of all locations). Although, polychaete mounds were often found in moderate densities (up to 50% cover), these patches were very localised with low overall % cover (mean cover 2.6% ± 0.3) in each of the three closed estuaries (Fig. 3.6). Similarly, mussel clumps were also present in moderate densities (up to 60% cover) in discrete areas of Wilson Inlet (2.4% occurrences of all locations), with low overall % cover (mean cover 1.1% ± 0.01) (Figs. 3.5-3.6).

Although 87% of all seagrasses had epiphytic algae, the seagrasses were generally in good health, with few plants completely covered in algae. The mean epiphytic load on seagrasses was highest in normally closed estuaries, with the exception of Stokes inlet, where there was very low seagrass coverage. Beaufort Inlet and Wellstead Estuary had mean epiphytic loads of 76% ± 2.3 SE and 43% ± 1.5 SE, respectively. In comparison, artificially opened estuaries and permanently opened estuaries had lower mean epiphytic loads (26% ± 1.1 SE, 38% ± 1.0 SE, respectively). High epiphyte loads often occurred in relatively sheltered areas of Leschenault Estuary, Beaufort and Wilson Inlet, smothering seagrass leaves, with singular leaves weighed down from the amount of epiphytic cover. In normally closed estuaries, epiphytic load was often associated with seagrass growth, whereas artificially opened and permanently opened estuaries had distinct areas of heavy epiphytic cover. Seagrass length was longest along the edges of each estuary and associated with moderately high percent cover of seagrass in all estuaries except for Stokes Inlet. Seagrass length was positively correlated with seagrass cover ($r=0.67$, $n=3195$, $p<0.0001$), but was not strongly correlated with epiphyte load ($r<0.45$, $n=3195$, $p<0.0001$). In comparison, short seagrasses were distributed adjacent to the shoreline of Stokes Inlet.

3.3.3 Random Forests predictions for key species of seagrass and invertebrates

Several habitat-formers (seagrasses *R. megacarpa*, *H. ovalis* and *H. tasmanica*, red and green macroalgae, and invertebrate clumps) were abundant enough to model bio-physical relationships, and these relationships were used to predict estuary-scale distributions (Figs. 3.6-3.8). A variety of variables were found to be important for the prediction of different habitat-formers in South-west Australian estuaries (Figs. 3.9-3.13). Random Forests models performed well and explained 79-90% of the overall seagrass distributions in all estuaries except for Stokes Inlet, possibly due to the low occurrences of *R. megacarpa* (Table 3.5). Five factors (depth, spatial location (latitude and longitude), sediment type and grain size) were important in predicting the distributions of seagrass across each estuary (Fig. 3.9). However, for Stokes Inlet, geochemical parameters (salinity (4.44% relative influence), temperature (3.59% relative influence)) and latitude (3.75% relative influence) were the most important predictors, possibly due to the low occurrences of *R. megacarpa* in this particular estuary (Table 3.5). Partial response plots for *R. megacarpa* indicated that this species preferred water depths <2 m to flourish, although this depth was dependent on the spatial configuration of each estuary (Fig. 3.9). Additionally, within normally closed estuaries, *R. megacarpa* preferred a mean grainsize of 20 µm and the eastern sections of permanently and artificially opened estuaries (Fig. 3.9). These relationships were often non-linear and differed between estuaries, indicating that different factors influenced seagrass distribution at an estuary level. Predicted percent cover of *R. megacarpa* was highest around the shallower regions of each estuary, particularly in water depths of between 0.4 m (shallowest depths surveyed) and 1 m, with relatively low percent covers found in the deeper sections (≥3 m) of each estuary (Fig. 3.6). Wellstead Estuary and Beaufort Inlet both have relatively shallow basins (≤ 1.5 m max. depth) and as a result supported considerably greater predicted percent covers of *R. megacarpa* within the inner basin of each estuary with no apparent depth trends (Fig. 3.6).

Table 3.5: Summary of model performance using Random Forests for biological habitat-formers. RRMSE and RMAE were determined using 25% of the withheld data to create the models.

Model /Estuary	**Distance Variables excluded^**	**RRMSE%**	**RMAE%**	***% Variation Explained***
R. megacarpa				
Stokes Inlet	River, estuary bar	435.78	40.58	-3.37*
Wellstead Estuary	River, estuary bar	59.82	1.76	80.84
Beaufort Inlet	River, estuary bar	95.58	7.39	79.38
Wilson Inlet	River, estuary bar, coast	123.68	43.32	82.84
Leschenault Estuary	River, estuary bar	24.45	0.28	89.53
H. ovalis				
Leschenault Estuary	River, estuary bar	33.98	1.13	84.75
H. tasmanica				
Leschenault Estuary	River, estuary bar	172.95	6.00	56.35
Green Algae				
Stokes Inlet	River, estuary bar, coast	99.05	5.90	54.91
Wellstead Estuary	River, estuary bar	119.11	12.20	55.36
Beaufort Inlet	River, estuary bar	53.98	3.54	69.94
Wilson Inlet	River, estuary bar	36.99	1.57	88.02
Red Algae				
Wellstead Estuary	None	245.56	7.24	80.79
Beaufort Inlet	None	92.08	10.99	61.12
Wilson Inlet	River, estuary bar	105.10	1.79	76.67
Leschenault Estuary	River, estuary bar	86.18	3.48	58.54
Polychaete Mounds				
Stokes Inlet	None	155.64	14.31	55.77
Wellstead Estuary	River, estuary bar	244.55	2.53	-18.14*
Beaufort Inlet	River, estuary bar	190.38	38.32	12.52
Mussel Clumps				
Wilson Inlet	River, estuary bar	148.81	1.09	40.92

^ Please refer to Section 3.2.5 as to why these variables were excluded.
* Indicates extremely poor modelling performance for these benthic habitats

The combination of *R. megacarpa, H. ovalis* and *H. tasmanica* only occurred in Leschenault Estuary, the only estuary that is permanently open to the ocean. The spatial distribution of all seagrass species was largely dependent on latitude, longitude and slope (Figs. 3.9-3.10) suggesting spatial and topographic variables were influential in modelling these species. *R. megacarpa* was commonly found in high distributions along the eastern shore of Leschenault Estuary (mean % cover of 23.6% (Fig. 3.6)). Model performance of *H. ovalis* was high and explained 85% variation. *H. ovalis* was commonly found in the northern and southern sections of the estuary, with low percent cover predicted in the central section of the estuary (mean % cover of 24% ± 1.0, range: 0 – 90%). In contrast, *H. tasmanica* was distributed within the southern section of the estuary, in conjunction with *H. ovalis*. However, overall distribution and abundance of the species was rather low (mean % cover of 1.3 ± 0.2, range: 0 – 65%). Consequently, the model did not perform well, with only 59% of its distribution explained by the model (Table 3.5; Fig. 3.7). Importantly, within Leschenault Estuary, all three seagrass species occupied separate and well defined areas of the estuary (Fig. 3.7), although there was some overlap in the distributions of *R. megacarpa* and *H. ovalis* within the northern area of Leschenault Estuary as well as overlap of *H. ovalis* and *H. tasmanica* within the southern section of the estuary and the “Cut” (Fig. 3.7).

Modelled green macroalgae distributions were explained by spatial location (e.g. latitude and longitude), bathymetry (>1 m), and water quality parameters (e.g. secchi depth and dissolved oxygen content) (Fig. 3.11). RF models explained between 54.9 – 88.0% variations in each estuary, with the model for Wilson Inlet performing well while the model for Stokes Inlet performed rather poorly (Table 3.5). Green algae predictions were modelled for all estuaries except for Leschenault Estuary, where it was absent (Fig. 3.6). Wellstead Estuary had high predicted coverage of green algae within the middle reaches, while Beaufort Inlet had relatively higher abundances within Parry River (Fig. 3.6). In contrast, Wilson Inlet had higher green algae coverage adjacent to the estuary bar entrance (Fig. 3.6), occupying similar spatial distributions to rippled and hummocky areas as seen from the video. Red macroalgae occurred in all estuaries except Stokes Inlet, with distributions explained by a combination of spatial location, $\delta^{13}C$ and $\delta^{15}N$ isotopic compositions, and sediment grainsize. RF models for red algae performed relatively well in Wellstead Estuary and ranged between 58.5 – 80.8% in the amount of variation explained (Table 3.5; Fig. 3.6). However these predictors were inconsistent between estuaries (Fig. 3.12). Red macroalgae was mainly explained by a combination of spatial location and isotopes $\delta^{13}C$ in all estuaries except for Beaufort Inlet. In Beaufort Inlet, red algae distribution was driven by water quality characteristics (e.g. dissolved oxygen content and temperature), highlighting that there were no strong relationships between red algae cover and any other measured physical characteristics. Red algae in Beaufort Inlet were predicted to occur in high percent cover within the middle reaches of the Inlet. Wilson Inlet and Leschenault Estuary occupied similar distributions in that they both had high distributions of red algae in areas furthest away from marine influence (e.g. the estuary bar). Wellstead Estuary had low predicted coverage (0 – 4%) throughout the estuary and performed relatively well (80.8% variation explained; Table 3.5).

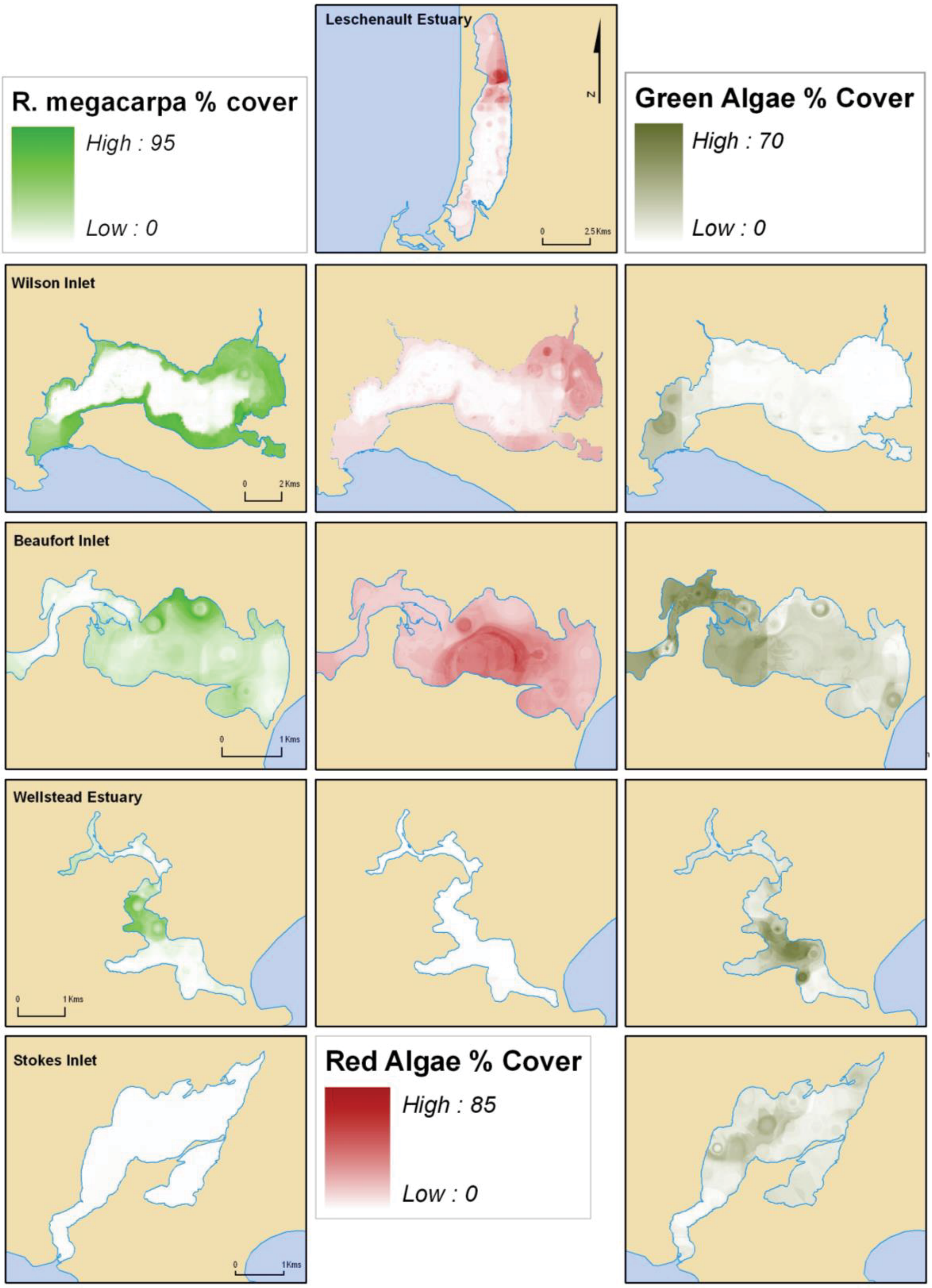


Figure 3.6: Maps of the predicted percent covers of *R. megacarpa*, red and green algal morphologies within five estuaries of south-west Australia

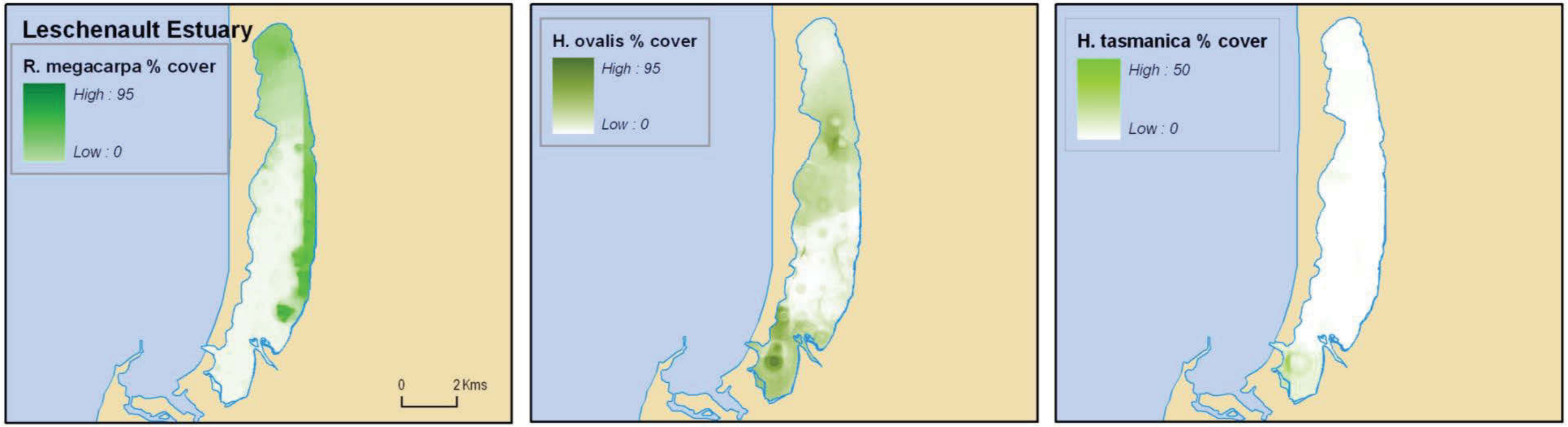


Figure 3.7: Maps of the predicted percent cover of *R. megacarpa*, *H. ovalis* and *H. tasmanica* in Leschenault Estuary.

Polychaete mounds occurred in normally closed estuaries, with distributions explained by longitude and sediment characteristics (porosity and % sand) (Fig. 3.13). However, this relationship was not present in all estuaries, with different predictors explaining polychaete mound distribution (Fig. 3.13). For example, the spatial coverage of polychaete mounds in Stokes Inlet coincided with areas of the highest slope transition in addition to moderate dissolved oxygen on the bottom (Fig. 3.8). Predicted models performed relatively poorly and only explained between 13 – 56% of variation for three estuaries. The predicted distribution for polychaete mounds within Wellstead Estuary highlighted small, localised patches adjacent to the shore and within sheltered arms of the estuary (Fig. 3.8). In Beaufort Inlet, predicted coverage of polychaete mounds was restricted to the northern and eastern sections of the estuary and within relatively low coverage of *R. megacarpa*. RF predictions for polychaete mounds in Wellstead Estuary performed relatively poorly compared to other key biological habitats (Table 3.5). This meant that the particular suite of variables used to predict polychaete mounds in Wellstead Estuary did not produce the best predictive maps, indicating the negligible effect of predictors in the model for this benthic habitat. In addition, the poor performance of the models was also due to the low number of occurrences of polychaete mounds in each estuary.

Mussel clump cover in Wilson Inlet was best explained by sedimentological (e.g. porosity and percent sand) and geochemical parameters (e.g. total carbon content) (Fig. 3.8). The RF model performed poorly and explained only 40.9% of the variation, due to the relatively low occurrence of mussel clumps within Wilson Inlet. Mussel clump percent cover was localised to northern and eastern sections of Wilson Inlet (Fig. 3.8), but was found more frequently in high cover of *R. megacarpa*.

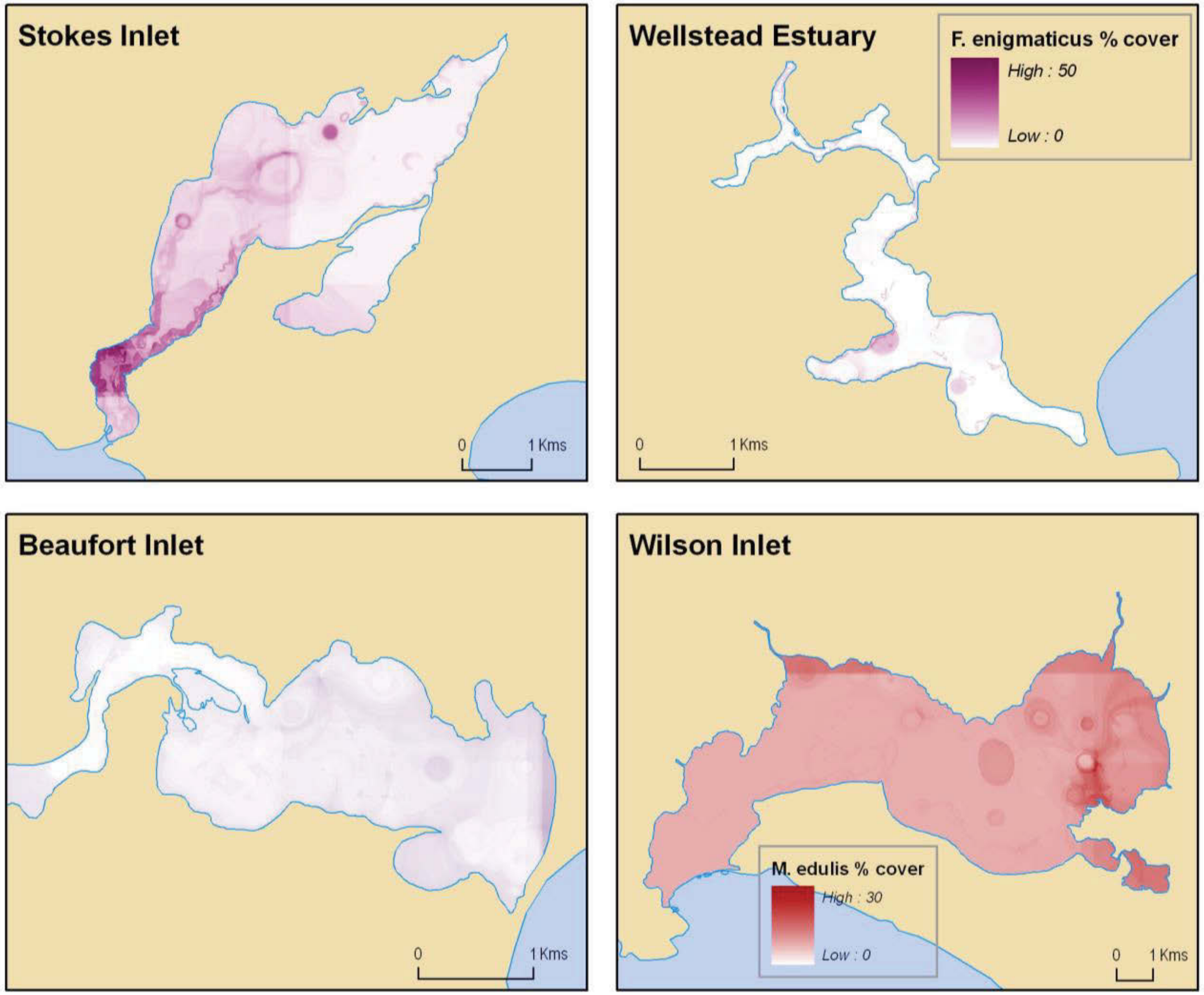


Figure 3.8: Maps of the predicted percent cover of *Ficopomatus enigmaticus* in three estuaries of south-west Australia and predicted coverage of *Mytilus edulis* in Wilson Inlet.

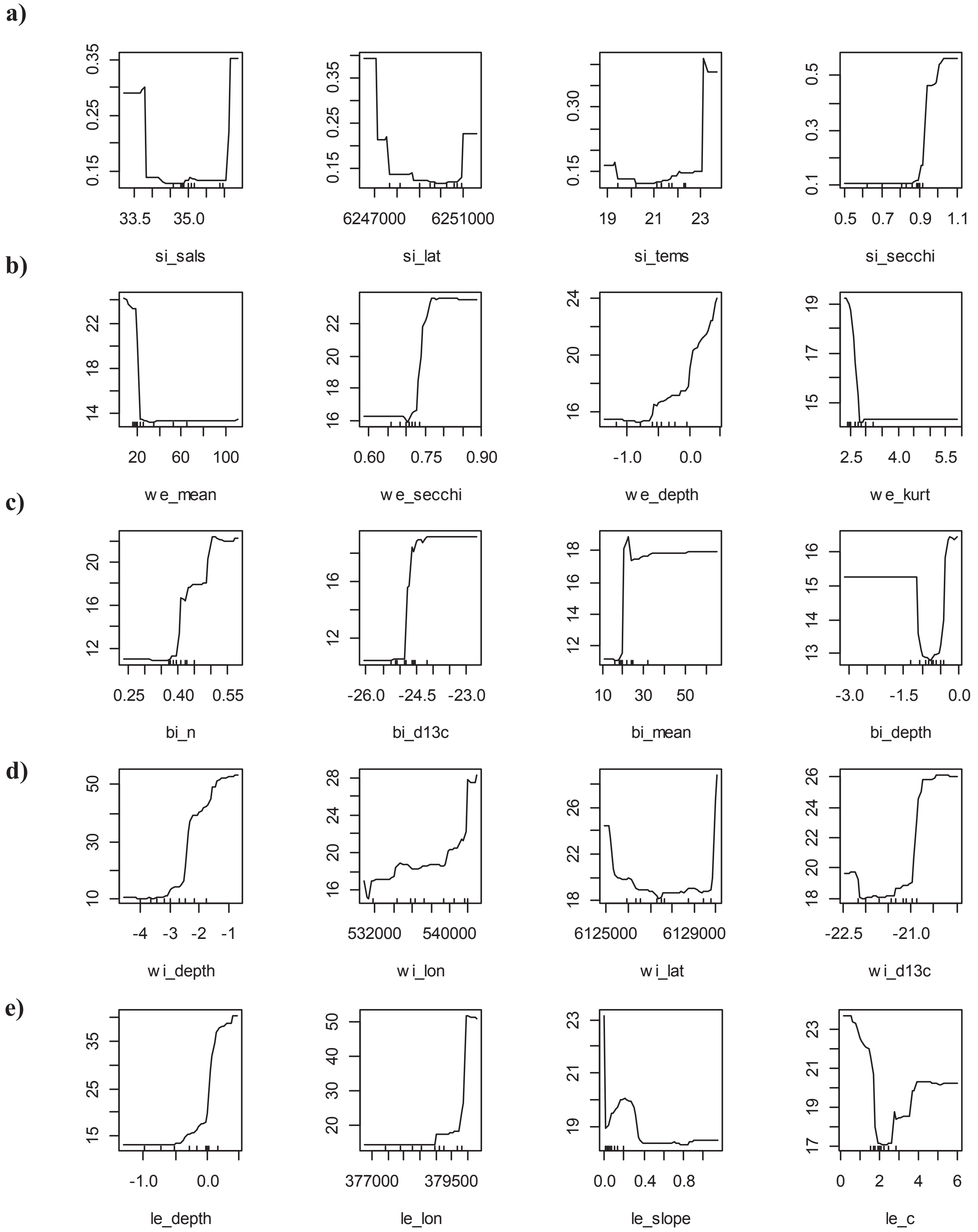


Figure 3.9: Partial plots for *R. megacarpa* for the four most influential response variables in: (a) Stokes Inlet; (b) Wellstead Estuary; (c) Beaufort Inlet; (d) Wilson Inlet; (e) Leschenault Estuary. The first two letters on x-axis indicate estuary, followed by physical variable (see Table 3.4 for reference). All y-axis labels are 'fitted function'.

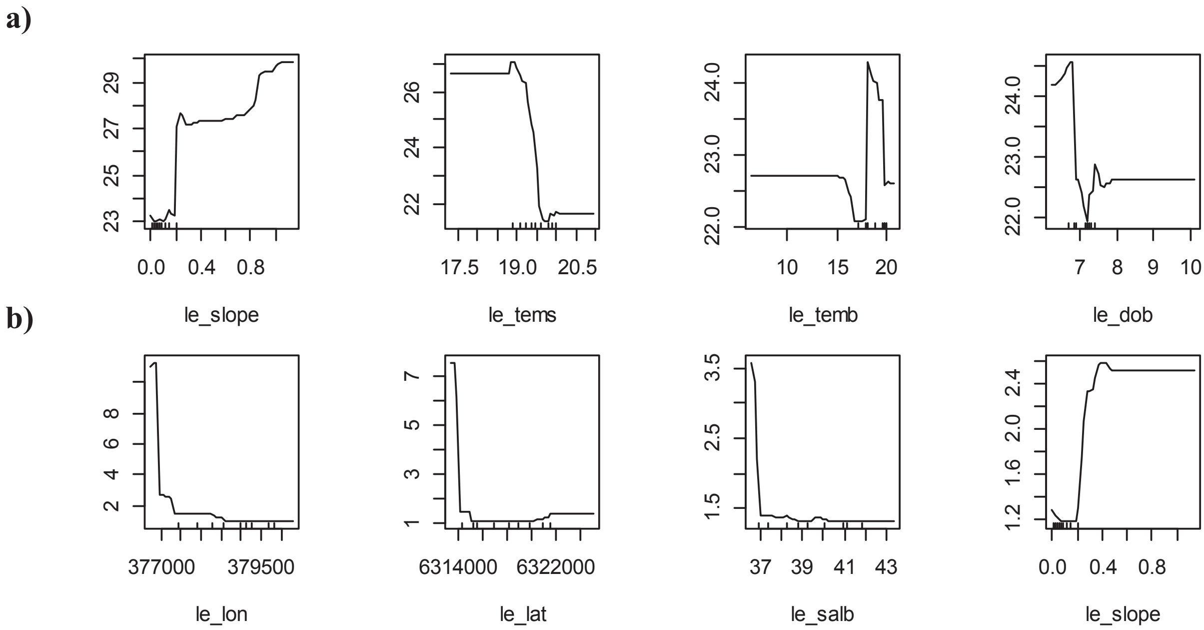


Figure 3.10: Partial plots for the four most influential response variables for: (a) *H. ovalis*; (b) *H. tasmanica* in Leschenault Estuary. The first two letters on x-axis indicate estuary, followed by physical variable (see Table 3.4 for reference). All y-axis labels are 'fitted function'.

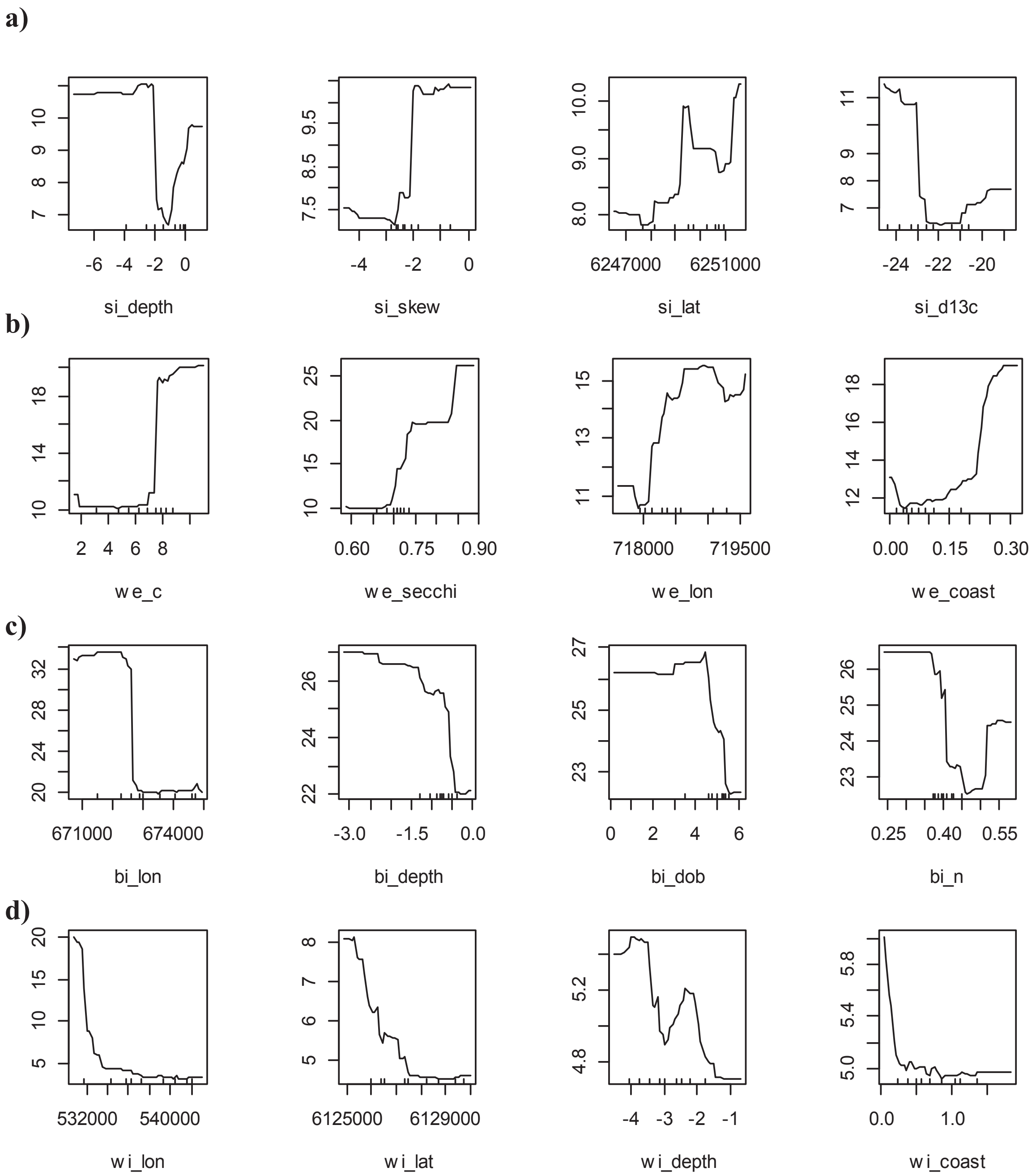


Figure 3.11: Partial plots for the four most influential predictors for Green algae in: (a) Stokes Inlet; (b) Wellstead Estuary; (c) Beaufort Inlet; (d) Wilson Inlet. The first two letters on x-axis indicate estuary, followed by physical variable (see Table 3.4 for reference). All y-axis labels are 'fitted function'.

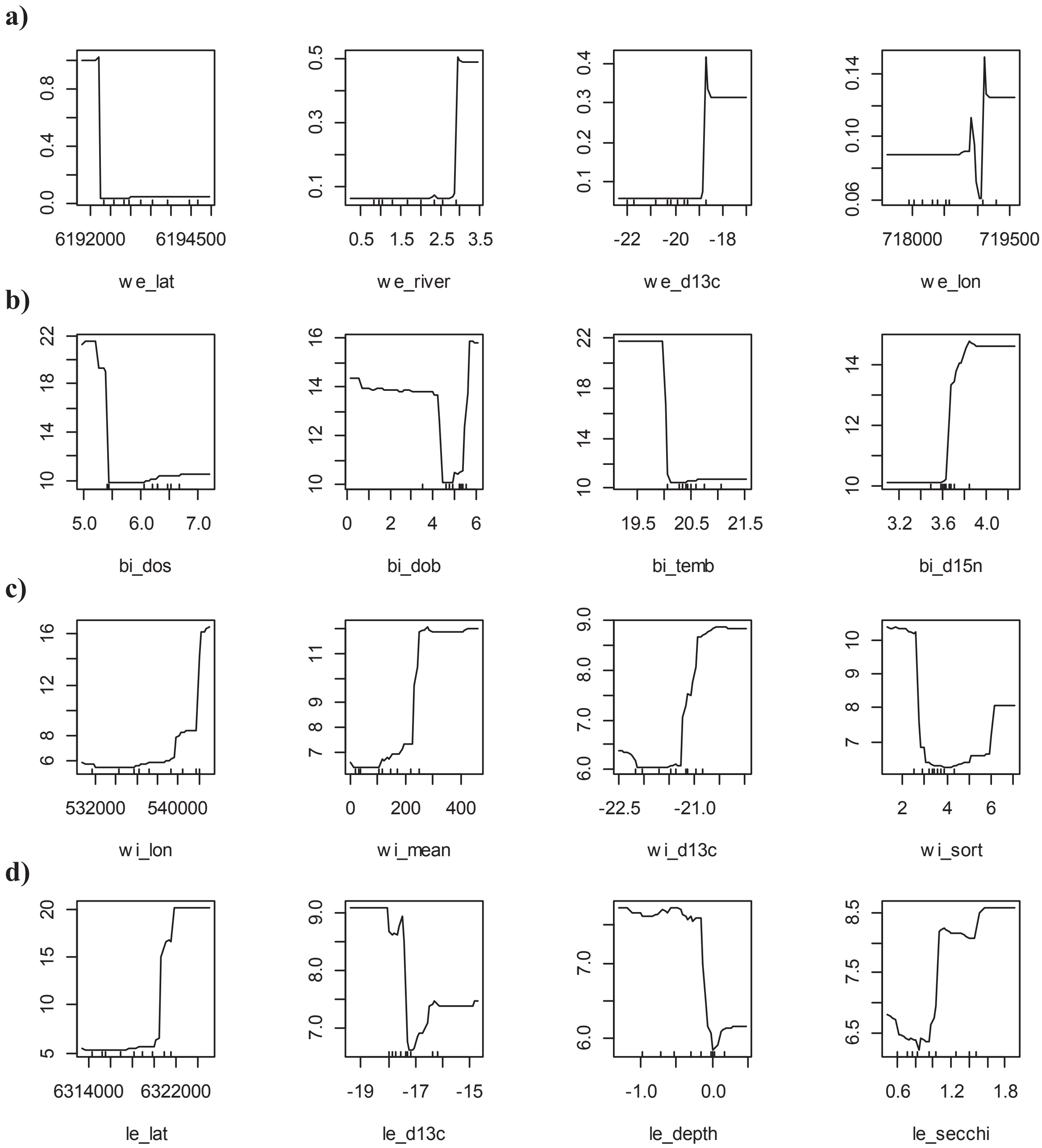


Figure 3.12: Partial plots for the four most influential predictors for Red algae in: (a) Wellstead Estuary; (b) Beaufort Inlet; (c) Wilson Inlet; (d) Leschenault Estuary. The first two letters on x-axis indicate estuary, followed by physical variable (see Table 3.4 for reference). All y-axis labels are 'fitted function'.

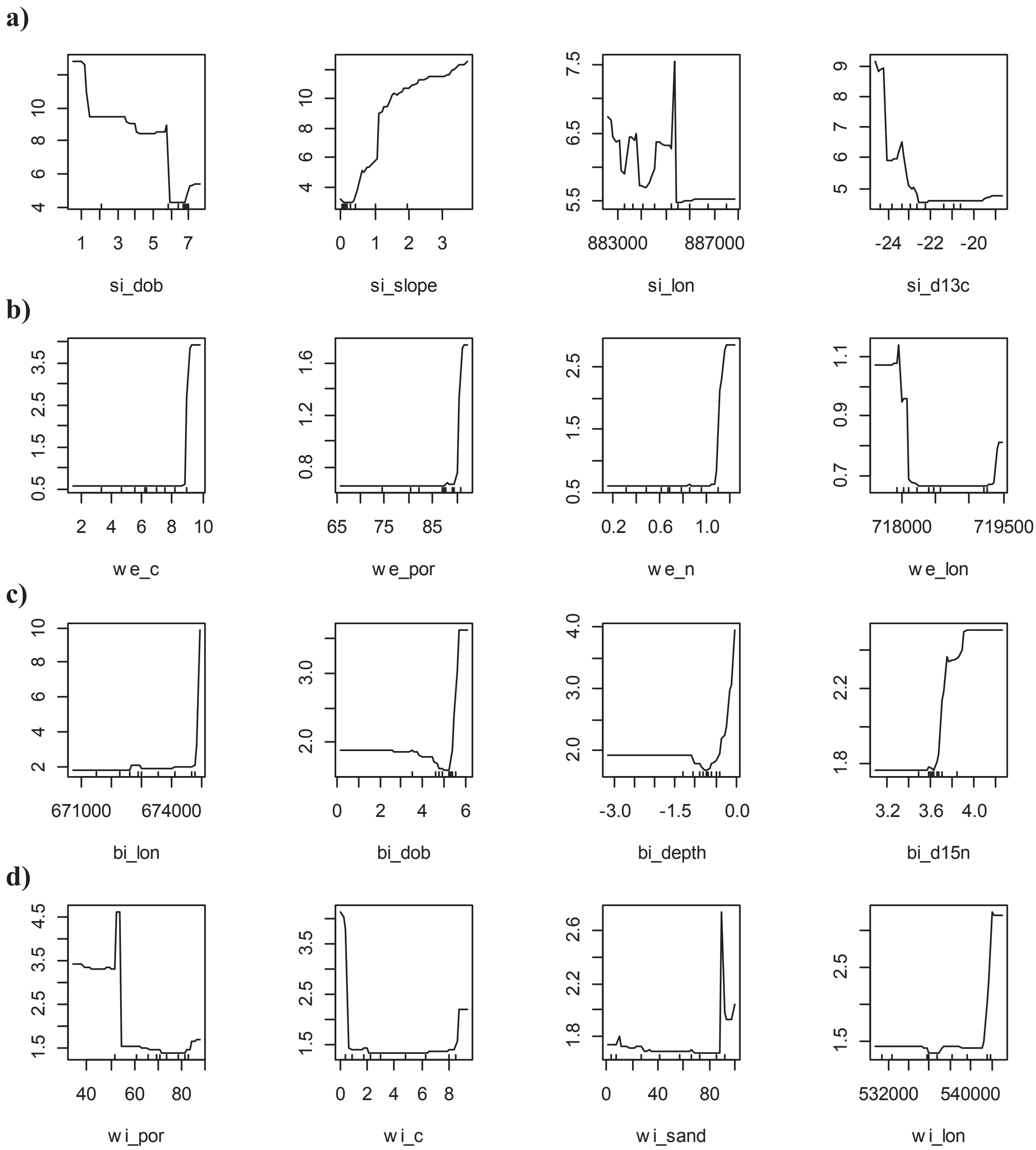


Figure 3.13: Partial plots for the four most influential predictors in: (a) Stokes Inlet; (b) Wellstead Estuary; (c) Beaufort Inlet for Polychaete mounds; (d) Wilson Inlet for Mussel clumps. The first two letters on x-axis indicate estuary, followed by physical variable (see Table 3.4 for reference). All y-axis labels are 'fitted function'.

3.3.4 Model Performance vs. validation dataset

Predictive performance was measured using two indicators: RRMSE and RMAE. Additionally, the standard deviation calculated for each RF model was also mapped for each benthic habitat (Figs. 3.14-3.16). The predicted distributions for each key habitat type were based on regression models using Random Forests and were highly variable in their predictive performance as well as sampling density (Table 3.3). Most models performed relatively well and ranged between a RMAE of 1-14% with the best models developed in Leschenault Estuary for *R. megacarpa* and *H. ovalis*, and in Wilson Inlet for red and green macroalgae (Table 3.5). The poor performance of predictions for mussel clumps and polychaete mounds in Wilson Inlet and Wellstead Estuary, respectively, was to be expected as the model was based on relatively low biotic occurrences, which has been shown to affect model performance.

Uncertainty in the modelled habitat-formers from the standard deviation maps for each estuary varied between 0 – 44% RMAE, indicating a high variability in percent cover of each benthic habitat (Table 3.5). Standard deviation was commonly associated with change in habitat percent cover (i.e. high SDs was associated with high percent cover). This is due to the video traversing over rapidly changing percent covers of substratum, for example, seagrass patches, and is reflected in the standard deviation maps (Figs. 3.14-3.16). As seen with the predicted spatial coverage of each habitat, the standard deviation predictions had horizontal or vertical banding as well as localised 'bulls-eye' effects (Figs. 3.14-3.16), indicating that the model found predictors such as latitude and longitude to explain the most variance within the models, and also showed the spatial clustering effect of the Inverse Distance Weighted interpolation method used on the underlying physical predictors. In other words, the RF model found no better variables to best explain the distribution of the benthic habitats in each estuary.

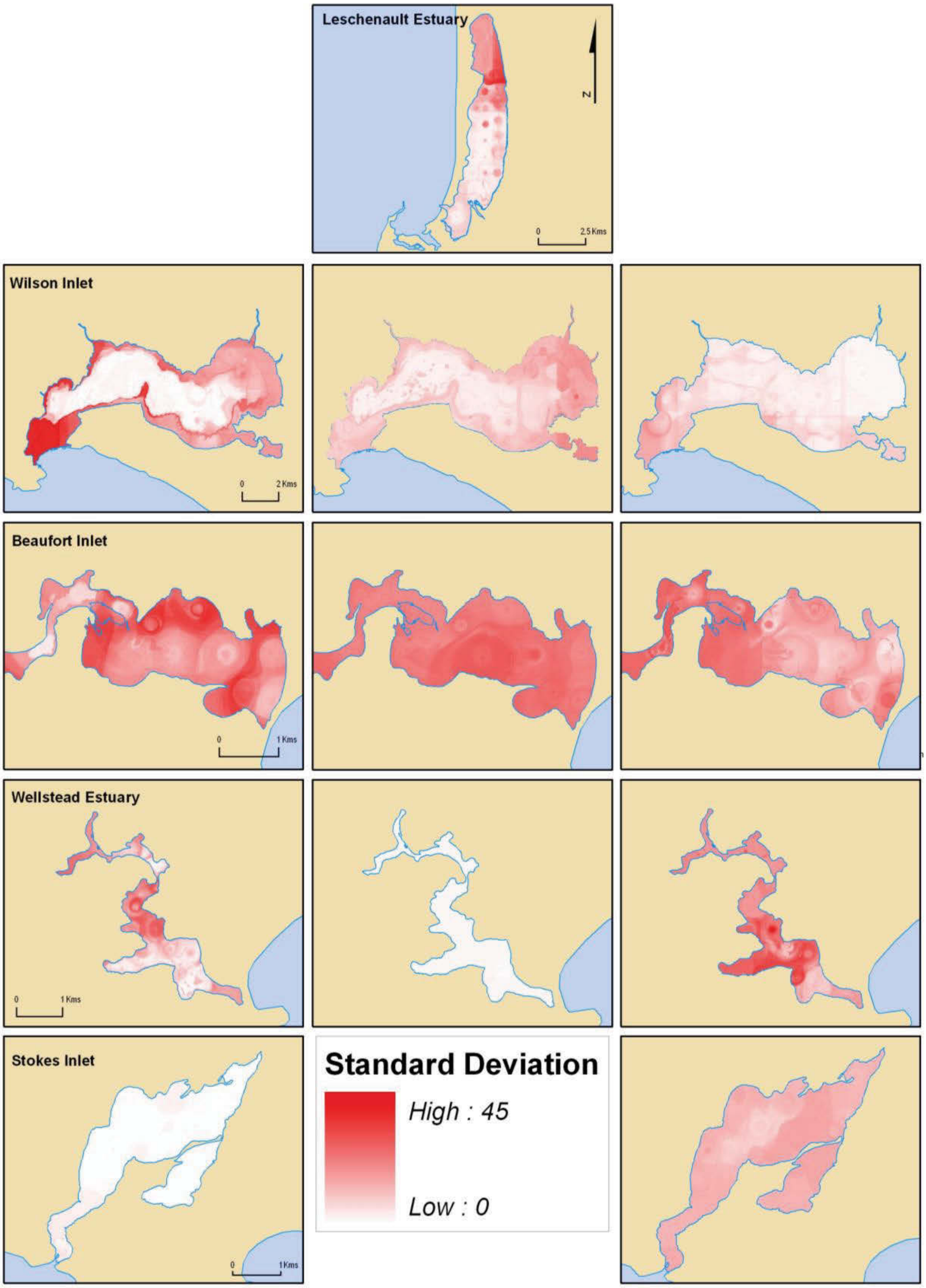


Figure 3.14: Maps of the standard deviation associated with predicted percent cover of *R. megacarpa*, red and green algal morphologies within five estuaries of south-west Australia.

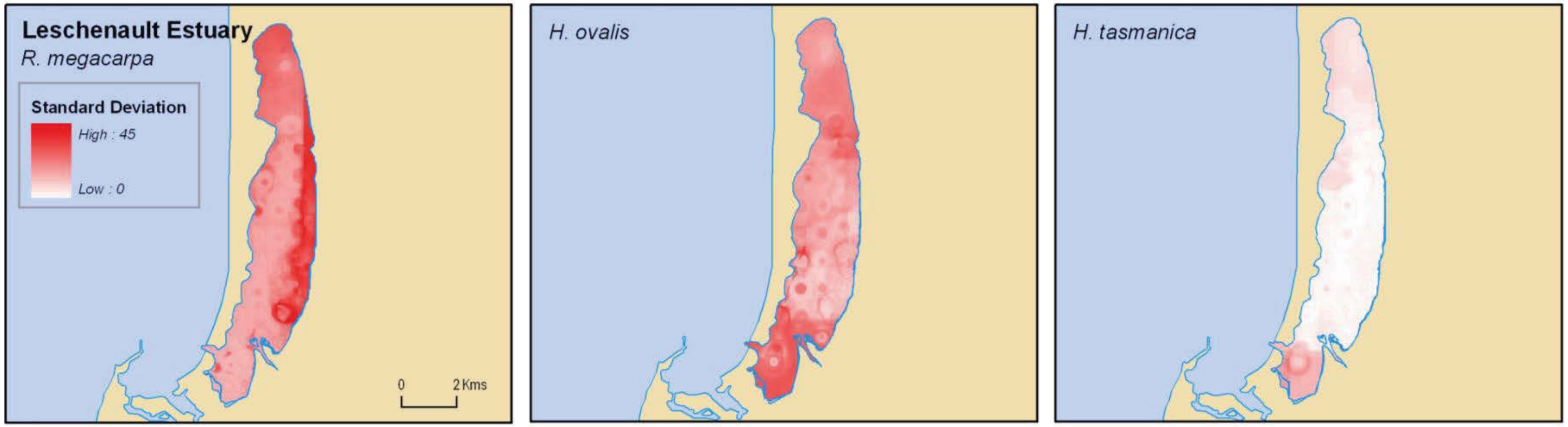


Figure 3.15: Maps of the standard deviation associated with predicted percent cover of *R. megacarpa*, *H. ovalis* and *H. tasmanica* in Leschenault Estuary.

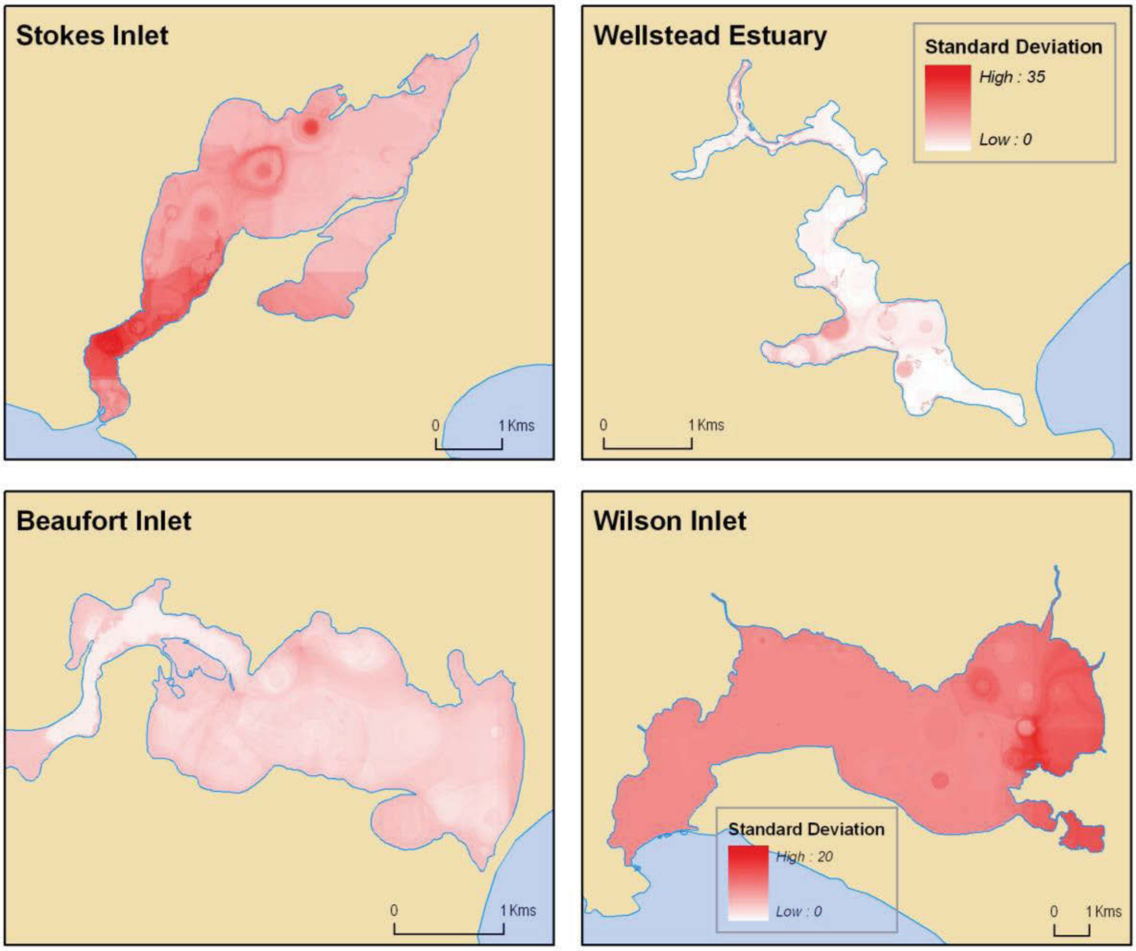


Figure 3.16: Maps of the standard deviations associated with the predicted percent cover of polychaete mounds in three normally closed estuaries and standard deviation associated with the predicted coverage of mussel clumps in Wilson Inlet.

3.4 Discussion

We used underwater video to characterise the benthic habitats within five south-west Australian estuaries and found that species occurrence of seagrass and algae aligned with previously established hydrodynamic regimes (permanently opened, artificially opened and normally closed) (Brearly, 2005). Within estuaries, sediments and water quality differed between shallow and deep habitats, defined as relative to the estuaries in this study, as well as areas influenced by high or low energy regimes. Random Forests models performed well and effectively predicted continuous spatial distributions of seagrass and invertebrate habitats among south-west Australian estuaries from underwater video, physical samples, and spatial variables. While the Field of View (FoV) characterisations provided us with the ability to describe broad-scale habitat patterns, they are limited to point locations. Consequently, RF modelling has allowed us to envisage these patterns within unsampled areas, in addition to modelling the relationship between the physical factors for each habitat and incorporating these patterns into highly effective visual tools in the form of maps.

3.4.1 Patterns of key benthic habitats in estuaries

The number and mean cover of seagrasses and red algal morphologies declined from north to south-west and from permanently opened to normally closed estuaries, while green algal morphologies were common in normally closed estuaries. Seagrass meadows characteristically rimmed the edges of deeper estuaries such as Wilson Inlet and Leschenault, with large areas of bare sediments located within the inner basins (Fig. 3.6). As some estuaries in Western Australia can be closed for months or even years at a time, the seagrass and algal species that occur in these estuaries must be tolerant of extreme changes in water quality, such as salinity, temperature, and levels of oxygen and nutrients in the water column (e.g. Beaufort, Stokes, Wilson and Wellstead Estuary) (Carruthers et al., 2007; Duarte, 2009). However, in permanently opened estuaries along the south-west coast of Australia, seagrasses with marine associations such as *Halophila ovalis, Zostera muelleri* and *Posidonia australis* and mixed macroalgal assemblages were present (e.g. rhodophyta; *Gracilaria* sp., *Chondria* sp., *Laurencia* sp., *Spyridia filamentosa., Ceramium* sp., Phaeophyta: *Hormophysa triquetra, Dictyota* *paniculata*; Chlorophyta: *Caulerpa* sp. *Cladophora* sp., *Enteromorpha* sp.) (Hillman et al., 2000; Carruthers et al., 2007). Leschenault Estuary contained marine seagrasses such as *H. ovalis* and *H. tasmanica. R. megacarpa* is often a dominant component of south-west Australian estuaries, but unlike most seagrasses, its growth can vary dramatically over seasons and years (Brearley, 2005). Within Wilson Inlet, *R. megacarpa* has been shown to favour coarse sediment with a rapid negative response to low light levels (Carruthers et al., 1999). *R. megacarpa* growth is dependent on the amount of light that can penetrate the water column (Carruthers et al., 1999). Consequently, in areas of turbidity caused by suspended sediments or tannins, reduced light penetration through the water column or extreme climate events such as flooding can result in a lack of photosynthetic plant growth (Campbell & McKenzie, 2006). Algae was found in shallow intertidal areas intermixed with seagrass meadows and can often compete with seagrass species for space and nutrient availability as they are both the main primary producers in the system (Wurm, 2000). We found areas of red and green algae of different morphologies within and between areas colonised by seagrass. However, in relatively shallow estuaries with no clearly defined basin areas, such as Beaufort Inlet and Wellstead Estuary, seagrass and algae extended well into the central sections of these two estuaries.

Various green algae are temporally abundant in shallow waters and riverine areas of Stokes (Hodgkin & Clark, 1988b; Hodgkin & Clark, 1989). In the shallow basin of Wellstead Estuary, *Lamprothamnium papulosum* and *Polyphysa peniculus* is dominant. However, the underwater video mainly identified *Polyphysa peniculus* as more common than *Lamprothamnium papulosum* and was attributed to lack of taxonomic resolution from the video footage. Algal assemblages in Wilson Inlet are most abundant along the western end of the estuary. Algal assemblages in Wilson Inlet also comprised red algae; *Polysiphonia* sp. and *Chondria* sp. We found mixed red algal assemblages commonly interspersed with both *R. megacarpa* and *H. ovalis* throughout Leschenault Estuary. Green, red and brown algal assemblages can occur in shallow water along the north, east and southern platforms (Semeniuk et al., 2000). Although many algal species are found in these estuaries, the video was only able to classify larger morphologies of green and red algae, which were consequently reduced to morphologies of branching and filamentous forms. However, amalgamated morphology maps of algae are still valuable as they depict spatial habitats types at a broader scale as well as determining estuary productivity, seagrass decline, and sources of potential pollution. As algae can often outcompete seagrass growth (Hauxwell et al., 2003), being able to monitor this change is of paramount importance if we are to avoid ecosystem collapses that can affect aquatic and human health.

Polychaete mounds were made up of a serpulid worm (*Ficopomatus enigmaticus*) and were commonly found in normally closed estuaries. They form large (>1 m) boulders of intertwined calcareous tubes that extend outwards as they age. They can be viewed as beneficial by increasing oxygen and nutrient levels, and creating large habitat-forming biogenic features up to 7 m in diameter that change the surrounding geomorphology (Hove & Weerdenburg, 1978). They can also flourish in relatively closed systems with little wave energy, reduce particulate loads and are resistant to pollution. However, they are sometimes regarded as pests, and they face little competition with the relatively small numbers of euryhaline species which occur in these estuaries. Our study provided a method of distinguishing localised areas of polychaete mounds within three closed estuaries, and showed that the spatial distribution of polychaete mounds were directly linked with slope, longitude, depth and dissolved oxygen content. Additionally, it has also been found that these worms are more common in normally closed estuaries which are often more saline and experience less annual rain flow, as well as in areas where the estuary bar is not commonly breached (Brearley, 2005). These species form on hard substratum, so are more commonly found on trees and rocks that line the river banks of estuaries; this is reflected in our predicted distributions of polychaete mounds (Brearley, 2005). Mapping the distribution and abundance of these species and how they may relate to physical variables can be beneficial for conservation management as the mounds can be monitored as potential ecological indicators for water quality in areas where these species are found.

3.4.2 Physical setting

Well-sorted sediments were commonly found in shallow areas, reflecting higher wave and wind energy sources. Conversely, deeper sections of estuaries contained poorly-sorted sediments, indicative of more sheltered depositional environments. Additionally, sediment grainsize skewness was often negative within estuary bar and river entrances of Leschenault Estuary and Stokes Inlet, indicating that these areas were subject to high amounts of reworking and deposition. Sediments that were often symmetrical in grainsize skewness were found in areas of deposition. This is a common occurrence and shows similarity in sediment grainsize statistics and estuary distributions which are consistent with the studies of Sheldon (1968) and Kranck (1975). Mean sediment size is influenced by the energy and transport conditions and indicates the average kinetic velocity of the depositing agent, which makes it an ideal indicator for the different energy regimes experienced in various areas of an estuary (Allen, 1971). The estuary bar openings are narrow and subject to sand build up, and in the case of Wilson Inlet, result from onshore sediment transport due to persistent ocean swells during typical dry summer conditions and a lack rainfall during summer months (Ranasinghe & Pattiaratchi, 1999). This supports our findings of higher sand content in sediments adjacent to the estuary bar opening in Wilson Inlet and Leschenault Estuary. In addition, coarse sediment locations found in our study are consistent with previous studies in the same estuaries (Hodgkin & Clark, 1987). Also, the process of catchment clearing increases the amount of sedimentation experienced within the tributaries of each estuary which will have deleterious effects on biotic habitats. Although significant differences existed between stable isotope concentrations of carbon and nitrogen for estuaries, it is quite difficult to unambiguously discern the cause of these differences (Table 3.4; Cloern et al., 2002). Our results suggest that surface sediments are derived from estuarine and marine influence, with estuarine primary productivity driving organic matter reactivity (Cloern et al., 2002).

Salinities were often markedly different between the relatively marine Leschenault and Wellstead Estuaries and hypersaline Beaufort Inlet. This may be a result of the estuary bar state at the time of survey, saltwater intrusion, or the lack of freshwater influx via rainfall from the associated catchments. Additionally, deeper areas of both Stokes and Beaufort Inlet displayed marked temperature, dissolved oxygen and salinity gradients, indicating stratification and a lack of mixing present in these estuaries when closed to the ocean. Moreover, this stratification layer was clearly seen in video characterisations for Stokes Inlet in the areas mentioned above. Stokes Inlet is known to be closed for many years, with highly variable salinity and water levels from fresh and marine influence, and consequently, restricted flora and fauna (Brearley, 2005). This is similar to many other normally closed estuaries in south-west

Australia, which are prone to drying out due to low rainfall and shallow depths (Brearley, 2005). Although water quality variables were collected from one point in time, these variables were among the four most influential predictors for key benthic habitats in the study area (Figs. 3.9-3.13). The interaction between water quality variables and growth of seagrass and algal distribution are likely to affect assemblage structure over time, with often negative impacts associated with high and sustained nutrient levels, extreme weather events and decreased light availability (McKenzie et al., 2012).

3.4.3 Machine learning methods for prediction of key benthic habitats

Machine learning methods such as boosted regression trees and random forests are often advocated for flexibility in modelling, accepting a range of categorical or continuous variables, and the ability to model non-linear relationships which are often seen in ecological data (Leathwick et al., 2006; Elith et al., 2008). Using the Random Forests method in alignment with previous studies on physical parameters affecting the distribution of seagrass, we found the strongest predictor for seagrass abundance was depth, which is inherently linked to amount of irradiance and the ability of the plant to photosynthesize accordingly (Carruthers & Walker, 1999; Larkum et al., 2006). This has also been shown to be the main driver for the distribution and abundance of seagrass growth in other south-west Australian estuaries (Brock, 1982; Carruthers & Walker, 1999; Carruthers et al., 1999). Geographic variables of latitude and longitude were also strong predictors of benthic habitats, and show broad patterns of key habitat distributions, in particular, seagrass species occupying distinct areas in Leschenault Estuary. This also suggests that unmeasured variables may play a role in predicting the spatial distribution, because extremely horizontal or vertical banding patterns of habitats are unlikely to occur naturally, and indicate artefacts in the modelling process. Distance variables from the estuary bar and the rivers in the estuary also influenced the distribution of seagrasses in Leschenault Estuary. However, the inclusion of some distance variables within the RF model caused severe attenuation of the predicted output surface. As the RF model can at times over-predict low values and under-predict high values, we excluded the use of some distance variables in predicting key benthic habitats based on this premise. Using RF to model the predicted distribution of key benthic habitats provides a novel way to analyse data in conjunction with other variables and within this study, offers valuable insight into unsampled areas and allows the spatial representation and distribution of benthic habitats in estuaries.

Of the 14 physical variables, the top five predictors which performed consistently well for the distribution of habitat-formers in south-west Australian estuaries using RF models were: 1) longitude, 2) depth, 3) latitude, 4) sediment carbon content, and 5) benthic dissolved oxygen content. Given that the majority of predictions made for this study were of seagrass beds, it is only logical that carbon content would be one of the top 5 predictors. In addition, algal and shell material (polychaete mounds and mussel clumps) are also common sources of carbon – supporting the fact that estuaries can be of high importance as natural hotspots for organic carbon sequestration (Kennedy et al., 2010).

3.4.4 Strengths, limitations and recommendations

Due to the nature of our survey design, this study provides a 'snapshot' of what the benthic environments were comprised of at the time of data collection. It may therefore not reflect present day coverage but does provide a baseline in which to compare future change. We also highlight that the point count method is more often used with downward-facing cameras, and does not work as efficiently with an oblique view of the seafloor, as larger organisms are often more prominent and account for more percent area of images. In fact, benthic habitats or substratum may only occupy < 50% of the field of view if captured with a downward facing camera, but the reader is advised to consult Wakefield & Genin (1987) and Stevens & Connolly (2003) for the application of a perspective grid on forward facing cameras. Additionally, we recognise that there are inherent limitations with the interpolation methods used in this study, but as we were limited in samples available, we determined that the Inverse Distance Weighted Method was the most appropriate method to use due to the relatively robustness of the method with smaller sample sizes. However, as is common with IDS, samples with higher content often create 'bulls eye' effects due to closer samples having higher similarities than distant samples. Consequently, we have attempted to provide measures of uncertainty for each interpolated variable map per estuary using RMAE and RRMSE for an estimate of prediction error (Table A2 in Appendix). Some of the parameters we used as predictors for key benthic habitats in this study may have performed poorly for particular species, but with the collection of more data, we will be better equipped at understanding ecological processes governing the spatial distribution of habitats in estuaries. Increased data variability was found in the resultant standard deviation maps of each habitat for each estuary (Figs. 3.14-3.16). As high standard deviations were associated with rapidly changing habitat cover (seagrass and algal patches) this suggests that sampling densities may not have been high enough to capture the spatial variation of these small-scale habitats (Li et al., 2011). Consequently, a higher sampling density is recommended in heavily fragmented habitats which can be easily done with archived video. However, the sampling scheme provided in this study was considered sufficient for estuary-wide spatial scales and allowed us to visualise broad spatial patterns. Random Forests models for green and red algae resulted in latitude and longitude banding patterns and indicated that other variables outside of the scope of this study may be better suited in spatial prediction. Possible unmeasured physical variables such as anthropogenic influence through nutrient and sediment input may be important when predicting habitat distribution (Fourqurean et al., 2003). Additionally, there may be issues with spatial discrepancies trying to combine precisely georeferenced samples with interpolated spatial layers, but this lies outside the scope of this study

Predicting the percent cover of benthic habitats using meaningful models within maps is a useful and cost-effective tool for management and monitoring of water quality in Australian estuaries that are often subject to change by natural (e.g. rainfall and climate) and anthropogenic (e.g. catchment clearing, nutrient input) influences. Our data show that a number of variables can explain the distribution and abundance of benthic habitats, which vary vastly between each estuary. The conservation of marine habitats that contribute to the livelihood of adjacent communities and environments that sequester as 50% of organic carbon that natural rainforests do is imperative in the face of climate change and urban development, where sustainable and long-term solutions are advocated.

Chapter 4: *A Rapid Benthic Assessment approach for quantifying broad-scale spatial changes in estuarine habitats*

Chapter 4: A Rapid Benthic Assessment approach for quantifying broad-resolution spatial changes in estuarine habitats

Abstract

Estuaries in south-west Australia are often highly turbid resulting from tannin-stained waters, making the benthic habitats within them difficult to map with conventional remote sensing techniques. Seagrasses contained within estuaries are one of the first habitats to be adversely affected by declining water quality. In this study, we evaluate the accuracy and precision of a real-time method (The Rapid Benthic Assessment) to rapidly quantify benthic habitats across large spatial scales using underwater video. Dominant benthic habitats were estimated using a semi-quantitative categorical approach for seagrass percent cover (absent, low, moderate and high) and length (short, medium, long) in real-time from video transects and drop camera stations. The following methods were compared: 1) broad-resolution real-time classification using underwater video, 2) fine-resolution post-processed classification using video, and 3) validation using *in-situ* samples of *Ruppia megacarpa* sampled from seagrass meadows in three estuaries (Leschenault Estuary, Beaufort Inlet and Wellstead Estuary). Categorical estimates were then compared with fine-resolution percent cover estimates (5%) of *R. megacarpa* recorded from post-processed video footage (presented in Chapter 3). The Rapid Benthic Assessment (RBA) provided a quick and easy method to categorise dominant habitat-forming biota over large spatial scales (1-10's kms). This semi-quantitative approach accurately represented significant differences in above-ground seagrass biomass and density (*R. megacarpa* and *Halophila ovalis*) - highlighting the effectiveness of the method over multiple seagrass species. A high degree of precision was found in biotic substratum cover by comparing the RBA method with fine-resolution post-processed percent cover data (presented in Chapter 3), highlighting the amount of detail retained by the RBA method. However, fine-resolution changes in substratum or biotic cover that were <30% were not adequately captured. Importantly, data were able to be processed and mapped within hours of data collection making them immediately available to scientists and marine managers.

4.1 Introduction

Estuaries support important ecological habitats such as seagrass and algal meadows, which in turn provide refuge and food for a number of invertebrates, marine and freshwater fish species (Edgar & Robertson, 1992; Humphries et al., 1992; Jenkins & Sutherland, 1997; Beck et al., 2001). However, estuaries are often affected by anthropogenic factors such as sediment runoff from agriculture, catchment clearing of native vegetation, untreated sewage, recreational and commercial fishing and aquaculture (Hopkinson & Vallino, 1995; Ladson et al., 2006; Grech et al., 2011). Estuarine habitats and their inhabitants are therefore often affected by changes in water chemistry, changing hydrology regimes, sedimentation, and high nutrient enrichment causing algal blooms of planktonic cyanobacteria (Lukatelich et al., 1987; Roy et al., 2001). Cumulatively, these impacts can lead to habitat degradation, increased turbidity levels, and low oxygen content within the water column and result in seagrass loss and fish kills (Townsend et al., 1992). As estuaries provide a number of amenities to humans as well as sources of revenue, it is of paramount importance to keep estuaries in healthy and functional conditions.

Seagrass habitats can change dramatically in space and time, and are often one of the first indicators of deterioration in the presence of environmental stress (Hackney & Durako, 2005). Seagrass health can decline over large spatial scales very quickly; therefore it is imperative to be able to evaluate seagrass change over large areas rapidly. In many coastal environments, remote sensing methods have been successfully used to map broad-scale distribution of physical and biotic habitats (Ierodiaconou et al., 2011; Klemas, 2011). However, in turbid estuaries, remote-sensing techniques such as aerial photography are restricted in their ability to penetrate the water column, and therefore having limited use in these environments (Bastyan et al., 1995). Similarly, the shallowness of these estuaries, especially in those that are normally closed precludes the use of acoustic mapping methods such as multibeam sonar (Rooper & Zimmermann, 2007). Where turbidity levels are not excessive, towed-video or visual assessments provides a valuable option to quantify the distribution and abundance of physical and biotic habitats, and to infer broad-scale coverage based on point locations (Coles et al., 2009).

Underwater video assessments have been undertaken on a wide range of shallow and deeper water ecosystems (e.g. Anderson et al., 2011; Ierodiaconou et al., 2011) and can provide a range of quantitative information on benthic habitats and communities over broad spatial scales (Haywood et al., 2008; Moore et al., 2009; Cappo et al,. 2011). However, traditional video methods (e.g. point counts per frame) are logistically time consuming, with post-processing often taking months to years to complete (see review in Chapter 2), by which time these habitat may have undergone drastic changes. Rapid real-time characterisation of benthic habitat percent cover and substratum type can provide valuable information to managers by providing preliminary baseline data of habitat extent and help guide conservation management and future marine survey plans (Anderson et al., 2008). Using broad cover categories to classify seagrass biomass while in the field is non-destructive and can considerably reduce time spent post-processing the data (Mellors, 1991). Compared to the other remote sensing methods available for survey, underwater video provides the means by which to characterise and map benthic habitats in real-time, resulting in rapid turn-around of data processing (Anderson et al., 2008). Similar methods are used to rapidly quantify the composition and health of tropical coral reefs (Carleton & Done, 1995; Maragos & Cook, 1995; DeVantier et al., 1998; Hill & Wilkinson, 2004; Maragos et al., 2004; De'ath, 2007), seagrass habitats (Mellors, 1991; Norris et al., 1994; Schultz, 2008) and temperate estuarine systems (Roob et al., 1998; Blake & Ball, 2001).

Estuaries within south-west Australia often contain extensive, highly variable Submerged Aquatic Vegetation (SAV) meadows and differing 'exposure' regimes, variable nutrients and high water level fluctuations (Mellors, 1991; Brearley, 2005; Carruthers et al., 2007). Additionally, these estuaries are known to be eutrophic and often exposed to anthropogenic pressures (e.g. agriculture/farming practices, urbanisation, catchment clearing, and salinisation) which may result in mass fish mortalities, severe deoxygenation by water column stratification, and rapid sedimentation (WRC, 2004). As seagrasses are dynamic habitats which are sensitive to stressors, monitoring them continuously allows us to accurately represent changes in their spatial distribution over similar time scales of growth and death. We need a method that is rapid enough to keep up with this rate of flux and disturbance. Being able to capture that change effectively is important in understanding the way the habitat functions and is structured. Therefore, we need the technology and tools to effectively discriminate between different habitats and whether these habitats are in decline in a cost and time-efficient manner (Anderson et al., 2008). By combining multiple sampling technologies to visualise spatial patterns of the seabed, we can begin to understand how parameters work in concert with one another. Conservation management requires accurate information on the distribution of benthic habitats within estuaries to make informed decisions on the preservation and restoration of benthic habitats, and to monitor changes within the estuaries which may be linked with anthropogenic factors. The degradation of these habitats can have major impacts not only from an economic standpoint (e.g. on adjacent townships and communities which depend on the estuarine productivity for survival for commercial and recreational fishing activities), but also from an environmental standpoint (e.g. species which rely on estuaries and the habitats within them as nursery habitats, migratory pathways and areas of refuge from predators).

In this study, we develop and evaluate a Rapid Benthic Assessment (RBA) method to quantify dominant estuarine habitats (e.g. seagrasses, macroalgae, habitat-forming invertebrates) in real-time from towed underwater footage collected across five turbid south-west Australian estuaries (Leschenault Estuary, Wilson Inlet, Beaufort Inlet, Wellstead Estuary and Stokes Inlet). This RBA method is based on the C-BED method (Characterisation of the Benthos and Ecological Diversity) by Anderson et al. (2008) and modified to include broad-scale semi-quantitative categories of seagrass percent cover (low, medium, high), seagrass length (short, medium, long) and indicators of seagrass health (e.g. epiphyte load). Rapid techniques to determine coverage of benthic habitats are needed in estuaries that are: 1) difficult to survey due to their remoteness and spatial extensiveness; 2) turbid due to sediment resuspension and tannins from dissolved organic matter, and 3) often experience rapid changes in their hydrology regimes. To evaluate the accuracy and precision of the RBA approach, real-time characterisations were compared against; 1) direct measures of seagrass density and length collected from benthic cores sampled from three estuaries (Leschenault Estuary, Beaufort Inlet and Wellstead Estuary), and 2) the fine-resolution percent cover (~5%) post-processing approach of Chapter 3 (see section 3.2.3).

4.2. Methods

4.2.1 Study areas

Rapid Benthic Assessment (RBA) surveys were undertaken in five estuaries (Leschenault Estuary, Wilson Inlet, Beaufort Inlet, Wellstead Estuary and Stokes Inlet) (Hodgkin & Clark, 1987, 1988a,b; Brearley, 2005). All five estuaries are located along the south-west coast of Western Australia (WA), between Perth and Esperance (Fig. 3.1). These estuaries are wave-dominated and microtidal, each with differences in morphology, physiographic settings and their degree of temporal connectivity to the ocean (Table 3.1). They are similar to other estuaries worldwide, with intermittent closures to the ocean via a sand bar (Silva & Davies, 1986; Hodgkin & Hesp, 1998; Emmett et al., 2000; Potter et al., 2010). Stokes Inlet, Beaufort Inlet and Wellstead Estuary are classed as normally closed, located in areas of relatively low rainfall, and reflect the final stage of estuarine evolution, with sediments often infilling the estuary (Brearley, 2005). However, these normally closed estuaries open to the ocean through the estuary bar during seasons of exceptionally high rainfall. At the other end of the spectrum, Leschenault Estuary, located in the north, is permanently opened and has been exposed to marine influence since 1952 (Hodgkin & Lenanton, 1981; Hodgkin & Hesp, 1998; Chuwen et al., 2009). Wilson Inlet is defined as an artificially-opened estuary and is manually opened every winter when rising water levels from high rainfall threaten to flood the adjacent townships (Brearley, 2005). Sediment composition is composed of fine-grained muds in the deeper basins of Leschenault Estuary, Wilson Inlet and Stokes Inlet, while sandy sediments occurred along shallower shoreline features (Hodgkin & Clark, 1988a,b, 1989). The aquatic vegetation found in the estuaries of this study includes seagrasses (e.g. *Ruppia megacarpa, Halophila ovalis, and Heterozostera tasmanica*) and red and green macroalgal assemblages (*Lamprothanium papulosa, Chaetomorpha, Polyphysa peniculus*) (Hodgkin & Clark, 1988a,b; Hillman et al., 1995). Common fish species within these estuaries include cobbler (*Cnidoglanis macrocephalus*), black bream (*Acanthopagrus butcheri*) and mullet (*Aldrichetta forsteri*), while invertebrate species include blue swimmer crabs (*Portunus pelagicus*), Western king prawn (*Penaeus latisulcatus*), worms (*Ficopomatus enigmaticus*) and mussels (*Mytilus edulis*) (Brearly, 2005).

4.2.2 Benthic habitat sampling technique

Benthic habitats were visually characterised from underwater video collected in each estuary by a Rapid Benthic Assessment (RBA) method to provide semi-quantitative information on spatial habitat variation. A combination of transect lines were allocated along the shoreline, and short drop camera stations were positioned throughout the central basin of each estuary to maximise the sampling effort according to time allocated to each estuary (see section 3.2.3 for details on sampling design and sample locations). Benthic habitat composition and structure was classified in real-time every 30 seconds using the 3-tiered RBA method that is based on the C-BED (Characterisation of the Benthos and Ecological Diversity) method, developed to characterise and quantify the physical and biological composition and structure of benthos in offshore and deep-sea environments (Anderson et al., 2008; Nichol et al., 2009). At each 30-sec interval, a 15-second window of seafloor was observed and characterised by substratum type (primary (>50%) and secondary (>20%) cover) comprising of rock, boulders, shell, mussel beds, polychaete tube mounds, algae, seagrass and soft sediments (muddy-sand or sandy-mud), vertical relief (flat (0 m), low (<1 m), moderate relief (1-3 m)) or soft sediment bedforms (sediment ripples, waves or hummocks) and all associated epibenthos (flora and fauna) (Table A1) (Stein et al., 1992; Anderson et al., 2008; Nichol et al., 2009). For example, substratum comprising >50% mud and >20% sand, was classified as mud-sand (MS), and if sand comprised >70%, was classified as sand-sand (SS).

Semi-quantitative categories of dominant seagrass percent cover (absent, low (1-33%), moderate (34-66%) and high (67-100%)) and length (short (<10 cm), medium (10-40 cm) and long (>40 cm)) were estimated in real-time. Due to the broad-scale nature of semi-quantitative percent cover categories; observer bias is at a minimum (Miller & Müller, 1999). The RBA method was completed quickly (e.g. < 30 seconds for observation of seafloor and data entry per characterisation) in real-time and broad semi-quantitative categories of seagrass and seabed characteristics were clearly and consistently distinguishable (Miller & De'ath, 1996; Adams et al., 2004; Fabricius & De'ath, 2004). For each 30-sec interval, the percent cover of dominant seagrass species (e.g. *R. megacarpa, H. ovalis* and *Heterozostera tasmanica*) were categorized as absent, low, moderate or high (Fig. 4.1). Broad-scale percent cover estimates of *R. megacarpa* were determined by visual assessment and were made with the agreement of two observers within the field. Three blade length categories (short, medium and long) were recorded for *R. megacarpa* as these were readily distinguishable in different meadow states, and were simple to categorise in real-time. *H. ovalis* grows along the seabed with little vertical growth (i.e. <10 cm), so length was not recorded for this species. Finally, the occurrence of all other epibenthos (i.e. all organisms visible to the naked eye) and bioturbation marks (e.g. pits, mounds and craters – refer to appendix A for a detailed list of bioturbation marks) were recorded as presence/absence to the lowest taxonomic category discernable (e.g. fish, crab, shrimp etc).

RBA data were entered in real-time using a Cherry Programmable keyboard (© Cherry, 2008). This required a two person team to enter RBA data into GNAV (i.e. a single observer and a key-board recorder). Using a programmable keyboard enabled pre-defined habitats and taxa to be encoded prior to the field survey, allowing for rapid seabed characterisations during the survey. Additional habitats and taxa were also easily and quickly (3-10 seconds) encoded onto the keypad during the survey. The spatial position of each data entry were captured automatically in real-time using the GNav Real-time GIS Tracker' (© Gerry Hatcher, 2002) attached to a hand-held Garmin 76S differential GPS. GNav also enabled positional data to be captured every 1-2 seconds along each transect.

At the completion of the two surveys, RBA video data were processed by parsing data strings of semi-colon delimitated text from files created from GNav using Statistic Analysis Systems (SAS Institute Inc, 2001) and exported into a Microsoft Access database (Microsoft ®Office Access 2003). RBA data were then exported to ArcGIS to be viewed spatially (©1999-2008 ESRI Inc, v.9.3) along with other spatial data layers such as depth contours, estuary configuration (shape), the locations of rivers and bar entrances, and any man-made features (e.g. the "Cut" in Leschenault Estuary).

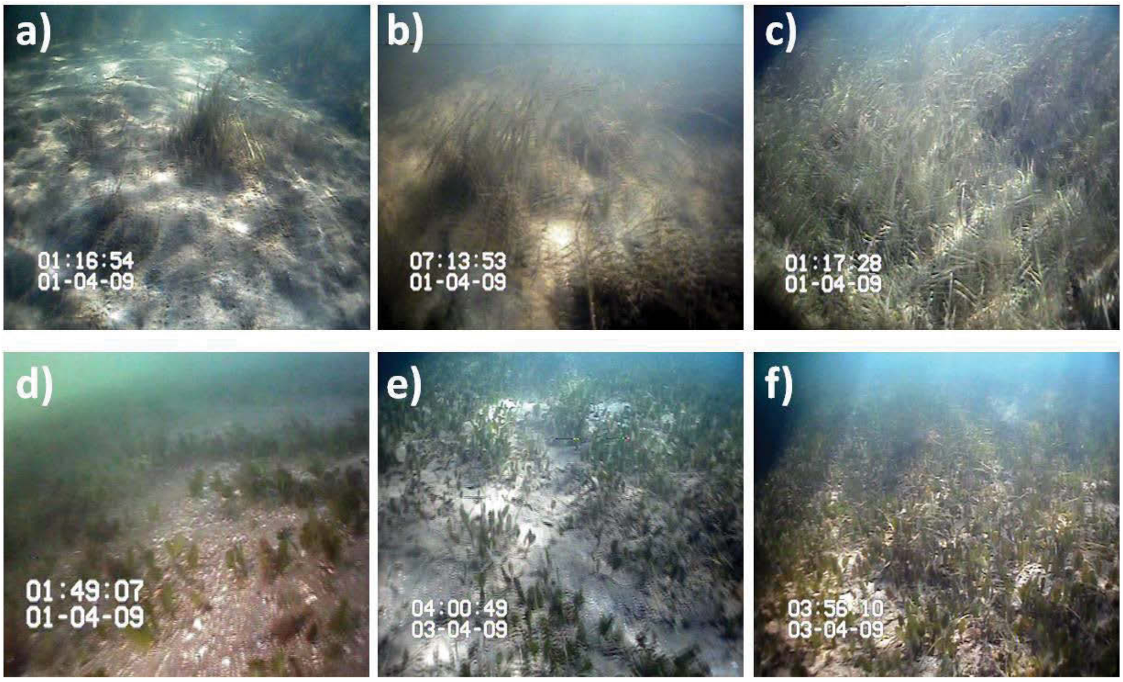


Figure 4.1: Screen grabs from underwater video of the broad RBA percentage covers of seagrass: (a-c) Low, moderate and high percent cover of *R. megacarpa*; (d-f) Low, moderate and high percent cover of *H. ovalis*.

4.2.3 **In situ** *seagrass sampling*

To determine if video observations of percent cover and seagrass blade length were adequately quantified using the *in situ* RBA method, above-ground samples of plant material were collected from a range of seabed densities and locations within the three estuaries (Beaufort, Wellstead and Leschenault Estuary), using *in situ* hand-held push cores (Table 4.1, Fig. 4.2). Sites were pre-selected haphazardly from RBA characterisations in each estuary, ensuring that samples were collected from all three percent cover categories (e.g. low, moderate and high categories). At each site, three replicate cores separated by a radius of ~ 5 m were collected by a single person on snorkel. Snorkelling was deemed suitable as seagrass occurrence within these estuaries occurred in water depths shallower than 2 m. Two different core types were used depending on the type of seagrass species present. Where *R. megacarpa* was the dominant taxa, a 20 cm diameter x 60 cm long BROMAR core (described in (O'Neill et al., 2007) was used to ensure the core did not cut the long delicate blades (Fig. 4.2). However, where *H. ovalis* was dominant, a smaller and shorter perspex corer (14.5 cm diam. x 10 cm) was employed, as it was easier to thread the relatively short (~10cm) and wide blades of *H. ovalis* through the Perspex core (e.g. Leschenault Estuary). We used an ANalysis Of VAriance (ANOVA) to examine if there is an effect of using two different biomass collection devices on one seagrass species (*R. megacarpa)*. As two methods of seagrass collection were used, biomass variables cannot be rigidly standardised. However, the ANOVA did not detect a difference between the two methods used to collect *R. megacarpa* so we combined *in situ* results.

Figure 4.2: Seagrass collection methods showing the two different coring devices: (a) The 20 cm diameter. BROMAR core used to sample in *R. megacarpa*; (b) the 14.5 cm diam. perspex core used to sample *H. ovalis*; (c) biomass collection process while in the field.

Table 4.1: Summary information on the number of *in situ* samples collected for *R. megacarpa* and *H. ovalis* from three south-west Australian estuaries

Estuary	Size of estuary	No. of *in situ* samples for *R. megacarpa*	No. of *in situ* samples for *H. ovalis*^	No. RBA characterisations total (transects + drop camera)	Depths sampled
Wellstead Estuary	3.20 km²	9	n/a	446 (224 + 222)	0.4 – 1.4
Beaufort Estuary	4.81 km²	9	n/a	336 (165 + 171)	0.5 – 3.1
Stokes Inlet	14 km²	n/a*	n/a	666 (309 + 357)	0.5 – 7.2
Leschenault Estuary	26.16 km²	9	9	721 (324 + 397)	0.5 – 1.3
Wilson Inlet	45.23 km²	0	n/a	1,028 (533 + 495)	0.7 – 4.5
Total	**93.4 km²**	**27**	**9**	**3197**	

^ *H. ovalis* was only present in Leschenault Estuary as it is the only estuary in this study to be permanently opened to marine influence.
* Stokes Inlet was not sampled for *in situ* seagrass samples as it only had 0.02% occurrences of *R. megacarpa* within the Low cover category.

During coring, care was taken to ensure adjacent seagrass areas were left undisturbed. Above ground plant material was carefully threaded into the corer and then firmly pushed into the sediment. This procedure ensured that all seagrass leaves within the core area were harvested intact; this was critical as measurements of blade lengths were also required. Once the core was firmly embedded in the sediment, all above-ground seagrass within the core was harvested, while underwater, carefully rinsed from attached sediment and then placed into a labelled zip-lock plastic bags at the surface. The person on board then placed each sample bag on ice within a portable esky, which were later frozen back in the laboratory within 2 - 3 hours of sample collection. After the survey was completed, all biomass samples were transported from Western Australia to the School of the Environment at the University of Technology, Sydney (UTS) using a refrigerated transport company to ensure samples remained frozen under controlled temperatures (slightly below or at 0°C) until processing. Plant material from each core was thawed in the laboratory at UTS (School of the Environment), and subsequently rinsed through a 250 µm sieve in fresh water to remove all sediments and retain seagrass and algal plants. For each core, the number of algae and seagrass genera (and species where possible) were recorded. Seagrass and algal species (where available) were then carefully separated. Epiphytic algae, where present, was carefully removed from each plant using a microscope slide to gently scrape the epiphytic algae off individual blades into a separate container. The total number of seagrass blades were then counted for each seagrass taxa (density) following the protocol of Humphries et al. (1992), whereby a sub-set of 30 blades per sample were measured to the nearest mm (blade length). All seagrass material for each taxa was then folded carefully within aluminium drying envelopes. Each envelope was carefully labelled with waterproof paper tags, placed on a metal tray, and left for 24 hours with one end slightly open to allow moisture to escape during the drying process (Robbins & Boese, 2002). Algae were placed into separate heat-resistant plastic containers, labelled and placed on another drying tray. Trays of seagrass and algal samples were then placed in a 50-60°C (LABEC Fan-Forced Convection oven) oven and dried for 24 hours until constant weight was achieved. Once cooled, the contents of each envelop was carefully weighed and recorded to an accuracy of 0.01 g (g dry wt m^{-2}).

4.2.4 Data Analysis

Means and standard errors for substratum type and biota were calculated per estuary and are represented graphically. Percent occurrence of epifaunal presence and *lebensspurren* ('life traces': bioturbation of soft sediments by macro and meiofauna) after Przeslawski et al. (2012) were calculated by the number of characterisations present in proportion to the total number of estuary characterisations to standardize the data. This was done as there were unequal numbers of observations per estuary and we used a similar 'occurrence index percent' to Maragos et al. (2004). The data were tested for normality using Shapiro-Wilks test and homogeneity of variance by Levene's test in addition to residual plots. Data were transformed using log (above-ground biomass of *R. megacarpa*, epiphytic algae) or square root (shoot density and shoot length of *R. megacarpa*) transformations where necessary. To determine if the broad-scale video-estimated percent cover categories adequately reflected the *in situ* measures of above-ground dry weight, seagrass density, and epiphytic algae, a nested ANOVA was run, with estuary defined as a random factor and nested within cover categories. Tukey's post-hoc test was used to determine where differences between cover categories existed.

To compare the relative proportions of cover between primary and secondary bottom types from the RBA method to the fine-resolution post-processed percent cover method, each primary substratum category present was converted to 50%, and each secondary substratum was converted to 20% to align with the proportions stated in Section 4.3. These were then transformed to represent 100% and summed to each respective bottom type (e.g. algae, soft sediments, boulders etc). Means and standard errors were then calculated per substratum type in Microsoft Access. Measurements of precision between the methods were calculated for each substratum and biotic habitat type on the transformed values of percent cover (Coefficient of Variation (CV) = SD/ $\bar{x}$, where SD = standard deviation, and $\bar{x}$ = mean).

The effectiveness and accuracy of the RBA method from semi-quantitative cover categories for seagrass was compared to the post-processed fine-resolution percent cover intervals from Chapter 3. To establish if there was any correlation between the rapid broad-scale percent cover categories of *R. megacarpa* to the fine-resolution post-processed percent cover of the second chapter, all broad-scale percent cover categories were re-coded to linear values (0, 1, 2, 3) for barren, low, moderate and high cover categories, respectively. Due to the large number of zeros in the dataset, Spearman rank-order correlation coefficients and box plots were used on the re-coded values and the broad-scale percent cover categories. Two-tailed paired t-tests were used to determine if there was a difference in mean estimated percent covers of benthic habitats between the broad-scale RBA method to the fine-resolution post-processing method of Chapter 3. Additionally, to compare the variability of seagrass cover within each video transect for the broad-scale RBA method with the fine-resolution post-processing method from section 3.2.3, we calculated the number of percent cover changes between absent, low, moderate and high, and between each interval of percent cover change, respectively. This meant that some transects were excluded (9 out of 42 transects) as they did not traverse over seagrass habitats and consequently, did not vary between seagrass cover categories.

4.3 Results

4.3.1 Comparisons between the RBA and fine-resolution post-processed methods

Using the RBA method, we collected a total of 3,197 characterisations over two survey periods from five estuaries in south-west Australia, from a total of 99 drop camera stations and 41 transects. These RBA characterisations spanned a total linear distance of 39.4 km^2 and traversed a multitude of benthic habitats. Similar to the fine-resolution post-processed percent cover method in Chapter 3, the RBA method accurately delineated benthic habitats of soft-sediments (mud and sand), seagrass, polychaete mounds and mussel clumps, as well as bedforms of flat, hummocky, low and moderate relief in real-time (Fig. 4.3). Importantly, the time taken to convert the RBA data into formats usable for spatial programs such as ArcMap was considerably shorter than the time taken to post-process the fine-resolution percent cover data (the RBA method took 8 hours to produce results compared to 280 hours to post-process the video to the fine-resolution percent cover detailed in Chapter 3). Surprisingly, after the conversion of primary and secondary substratum categories from the RBA method to mean percent cover, the relative mean substratum proportions between the two methods were comparatively similar (Table 4.2). This indicates that the accuracy of the RBA method was high, with the maximum mean variance of 3% for seagrass cover (Table 4.2), despite the difference in the time taken to post-process the data into fine-resolution percent cover intervals. Additionally, the relative precision for percent cover of substratum between both methods were similar (Table 4.2), even more so for biotic habitats, where the coefficient of variation (CV) measurements for the RBA and fine-resolution method were very precise for algae (CV= 2.34 and 1.53%, respectively), and slightly less for polychaete mounds (CV= 8.4 and 5.57%, respectively). However, percent cover estimates were less than precise for substratum types of the RBA and fine-resolution method of boulders (CV=22.60 and 19.34%, respectively), rock (CV=56.54% for both) (Table 4.2). This was due to the sensitivity of the CV to mean values close to zero.

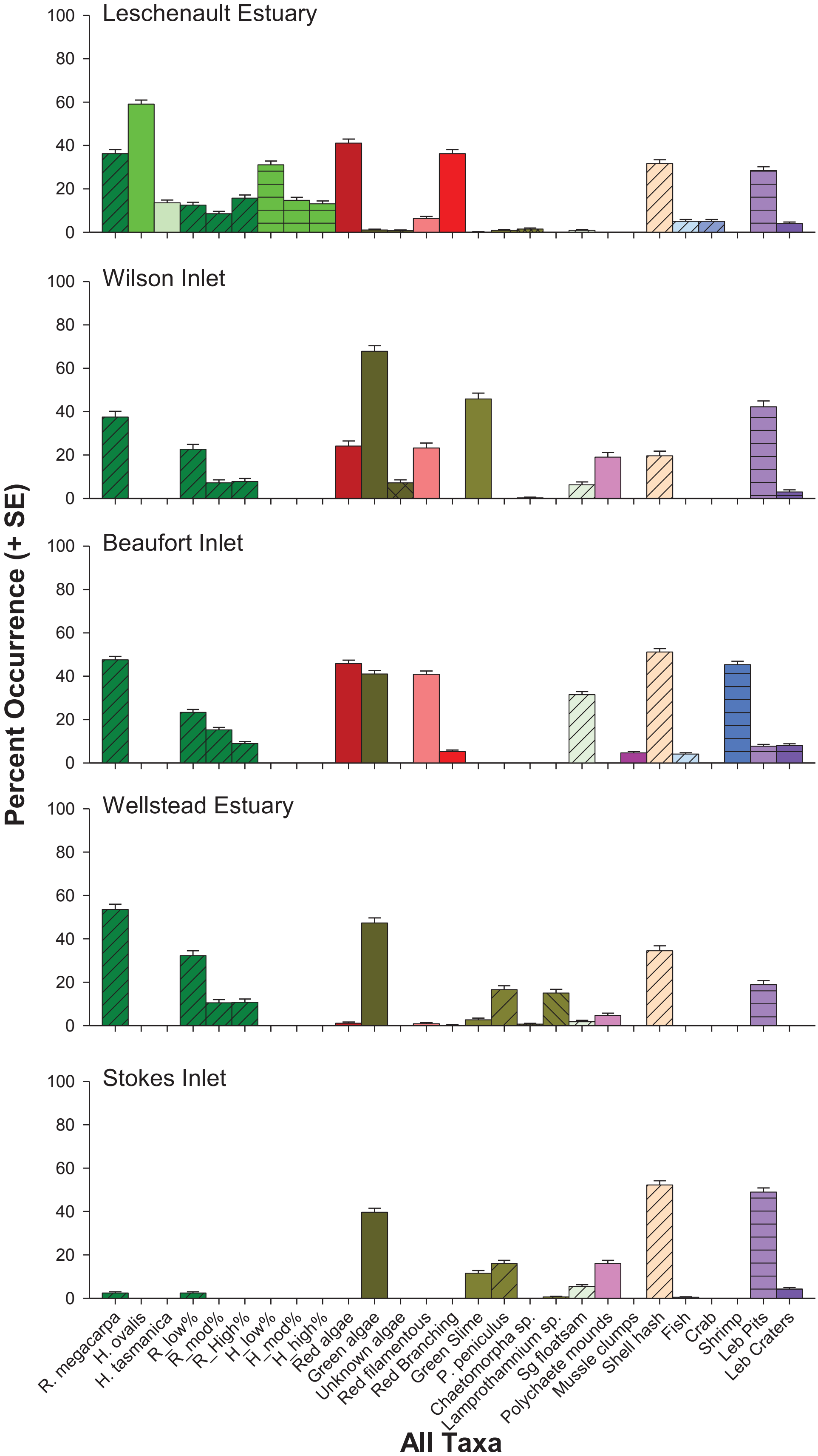


Figure 4.3: Percent occurrence of all taxa RBA percent occurrence of seagrass, morphologies of red and green algae and all other taxa within five estuaries of south-west Australia.

Table 4.2: Comparison of mean substratum percent cover between the RBA broad-resolution (30% categories) method and the fine-resolution (5% categories) method of post-processed percent cover of all estuaries.

Bottom Type*	RBA broad percent cover (mean ± S.E)	Precision (Coefficient of Variation%)	Fine-resolution percent cover (mean ± S.E)	Precision (Coefficient of Variation%)
Algae	10.04 ± 0.4	2.34	12.39 ± 0.33	1.53
Boulders	0.08 ± 0.03	22.60	0.06 ± 0.02	19.35
Mussel clumps	0.27 ± 0.06	13.19	0.34 ± 0.05	8.43
Polychaete mounds	0.92 ± 0.13	8.41	1.17 ± 0.12	5.57
Rock	0.02 ± 0.02	56.54	0.02 ± 0.02	56.54
Seagrass	22.39 ± 0.64	1.64	19.87 ± 0.52	1.47
Shell	1.27 ± 0.12	5.73	1.41 ± 0.1	3.94
Soft sediments (mud and sand)	64.99 ± 1.39	1.12	64.73 ± 0.59	0.51

*Colour key symbolises biotic habitats in Figure 4.3.

Comparisons between the RBA method and the finer-resolution post-processing method identified that very little information was lost with respect to the spatial distribution of abiotic and biotic habitat types (Fig. 4.4). Mean percent covers of mixed soft-sediments were not significantly different from the RBA method to the fine-resolution post-processed method in Chapter 3 and differed overall by 0.5% (Table 4.2; paired t(4)=0.7, p=0.5). Similarly, boulders (paired t(4)=1, p=0.4), rock (paired t(4)=1, p=0.4), and shell hash (paired t(4)=1.9, p=0.1) revealed no significant differences between methods used to estimate percent cover. The RBA method detected near identical (Table 4.2; p=0.2, paired t-test) spatial patterns of seagrass types (broad-scale percent cover and length patterns) along video transects and drop camera stations within each estuary (Fig. 4.2), albeit in less detail compared to the fine-scale post-processed method, where the RBA method failed to capture areas of heterogeneous seagrass habitats along video transects (Fig. 4.5). However, the RBA method revealed that only percent cover changes less than 30% were able to be detected, with fine-resolution changes within patchy seagrass habitats often not recorded (Figs. 4.4-4.6). No significant differences in mean percent cover of alga were detected between methods (paired t(4)=2.3, p=0.08). There was no significant loss of information in spatial distributions between the RBA method and the fine-resolution post-processing method in detecting overall mean percent cover of polychaete mounds (*Ficopomatus enigmaticus*) (Table 4.2; paired t-test (4)=1.46, p=0.2). Similarly, clumps of *Mytilus edulis* in Wilson Inlet indicated similar spatial distribution and did not detect a significant difference in method collection (Table 4.2; paired t(4)=1, p=0.4). Rather, these may be inaccurately represented within the data at a percent coverage much higher than they actually are (e.g. actual percent cover of 5%, but within the low RBA category of 1-33%). Additionally, the RBA method failed to capture more than two habitat combinations, while the fine-scale post-processed method was able to capture multiple habitat combinations.

Bioturbated sediments were identified and comprised *Lebensspuren* pits (26% occurrence), craters (5% occurrence), mounds (1% occurrence) and burrows (<1% occurrence) and were often located in estuaries with deep basins (Fig. 3.3), indicating that these areas contained higher amounts of nutrients within the surficial sediments. Motile species such as crabs and shrimp were more abundant in artificially and permanently opened estuaries (Fig. 4.3). Both the RBA method and the fine-resolution method accurately captured this information.

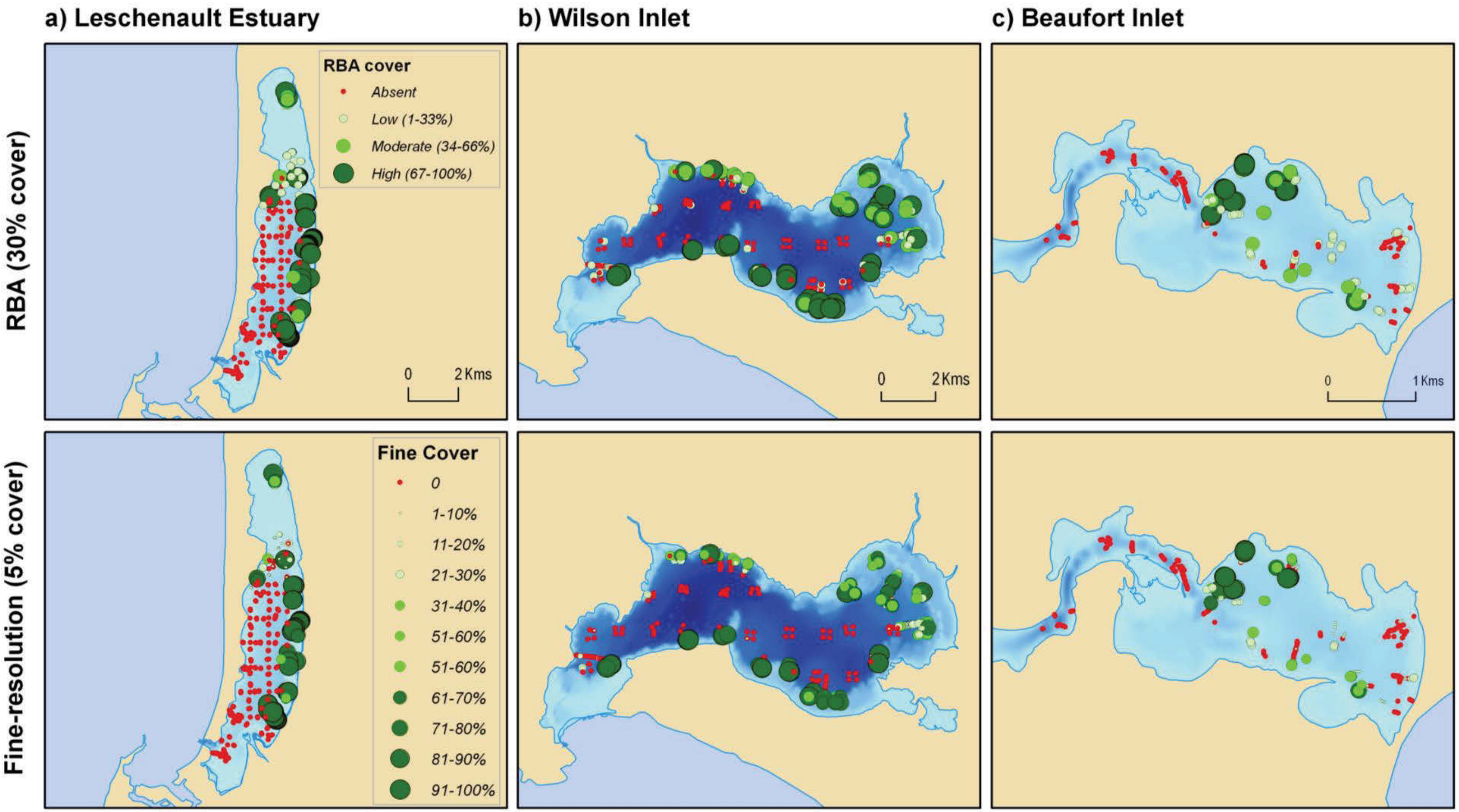


Figure 4.4: Comparison between broad cover categories from the RBA method and the fine-resolution post-processing method in Chapter 3: (a) Leschenault Estuary; (b) Wilson Inlet; (c) Beaufort Inlet.

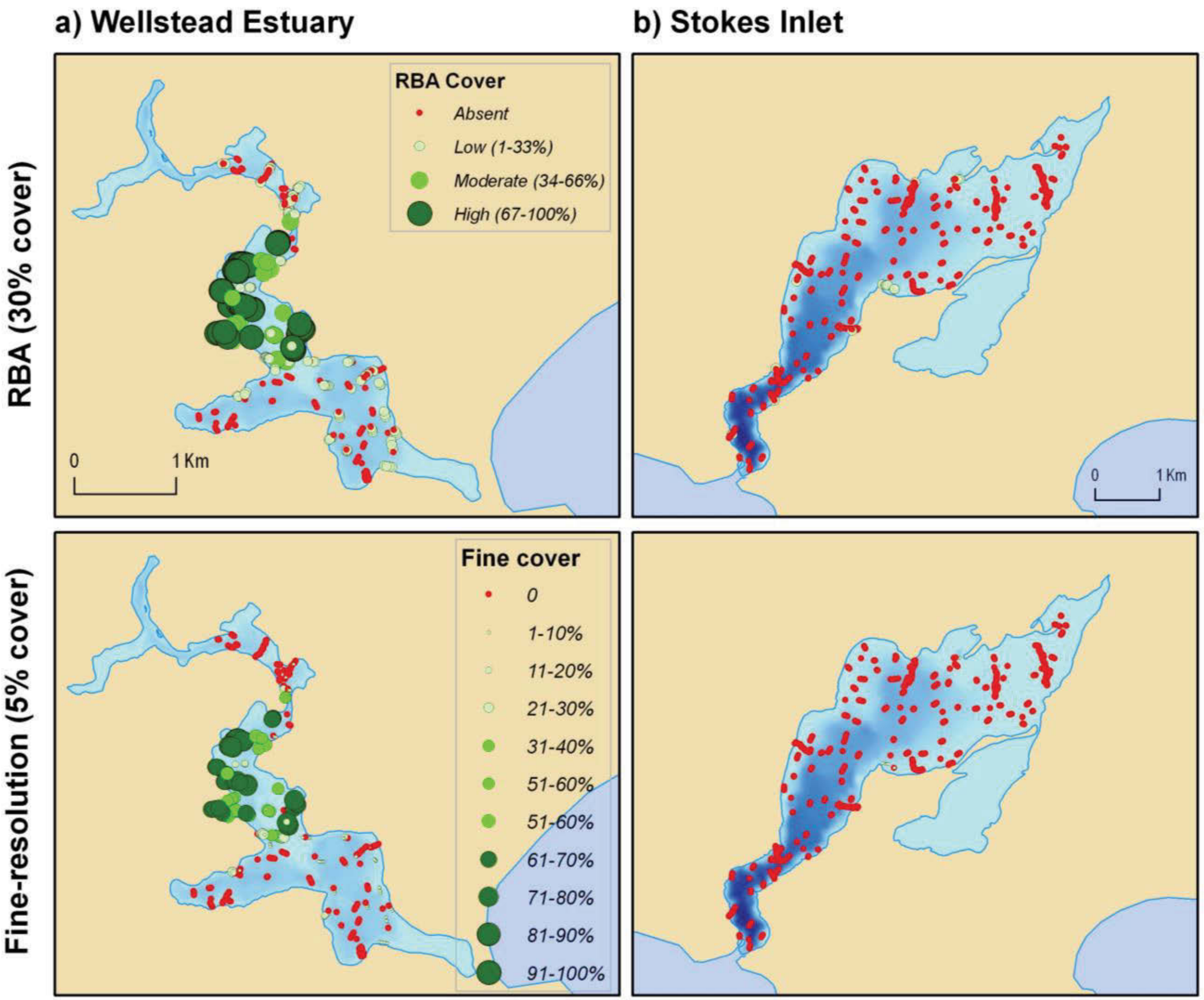


Figure 4.5: Comparison between broad cover categories from the RBA method and the fine-resolution post-processing method in Chapter 3: (a) Wellstead Estuary; (b) Stokes Inlet.

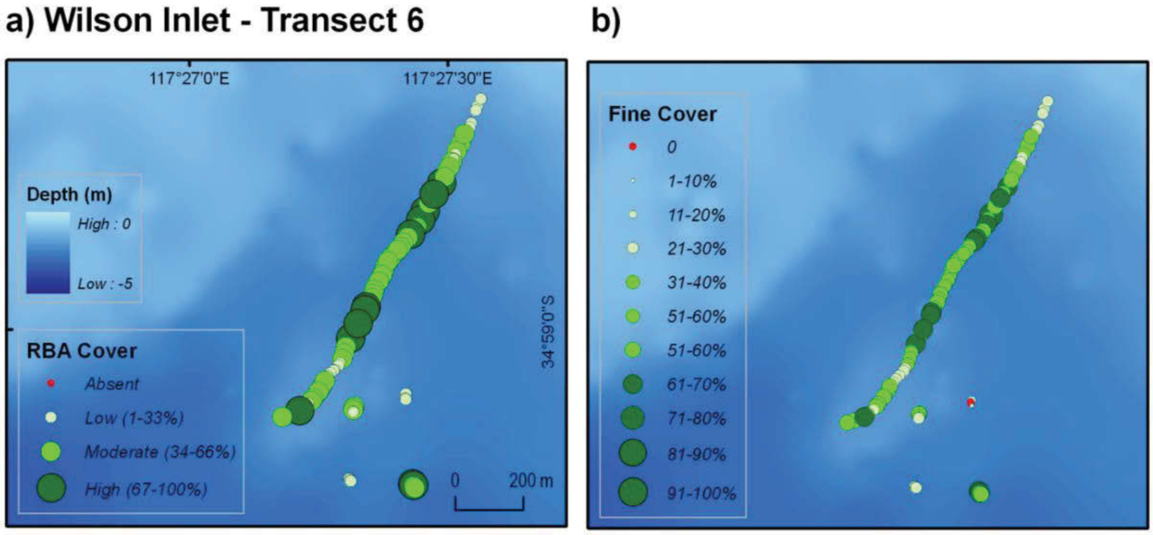


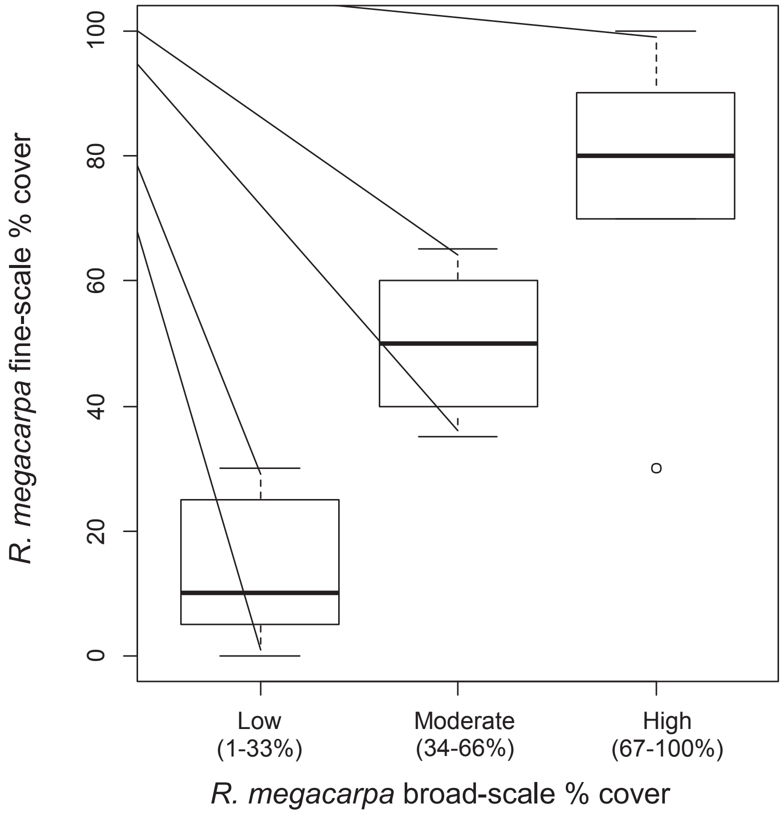
Figure 4.6: Comparison of *R. megacarpa* percent cover from Wilson Inlet along Transect 6: (a) Broad RBA percent cover; (b) fine-resolution percent cover.

4.3.2 Comparisons of seagrass cover between RBA and fine-resolution post-processing method

Broad-scale percent cover categories of seagrass were highly correlated with the fine-resolution (5%) percent cover (Spearman's rank test: p<0.05, correlation coefficient of 0.9) (Fig. 4.7). This identifies that the RBA method recorded similar spatial structures in the seagrass meadows within each of the estuaries, but required much less time to analyse (8 hours instead of 280 hours). Additionally, a highly significant correlation (r^2=0.9, p<0.05) was found between the fine-resolution post-processed data and the real-time broad-scale cover categories of seagrass. This is also supported by high precision calculations for seagrass (CV=1.64 and 1.47 %, respectively) (Table 4.2).

The RBA detected a total of 364 sequential percent cover changes of seagrass for all video transects, which was 56% less than the total number of changes in seagrass cover detected by the fine-resolution post-processed data from Chapter 3. This indicated that the RBA method may not capture the same degree of change for seagrass cover as the fine-resolution post-processed method. However, despite the high number of changes in percent cover of seagrass for the fine-resolution method (651 sequential percent cover changes), most variation was associated with relatively continuous seagrass beds. These changes in percent cover varied between 5-20%, most of which would not be detected by the RBA method as the semi-quantitative categories were too broad. The implications of this are that the RBA method could effectively capture a spatio-temporal change in percent cover greater than 33%, but would fail to detect smaller amounts of change (Fig. 4.7b). From the RBA method, the low percent cover category of seagrass was often short to medium lengths (16% occurrence), moderate percent covers were medium to long lengths (9% occurrence) and high percent covers were often longer in length (6% occurrence). A similar pattern was also detected using the fine-resolution method, highlighting the ability of the RBA method to capture biologically relevant differences within seagrass meadows. Patchy seagrass habitats (21% occurrence) and homogenous seagrass habitats (13% occurrence) were relatively common.

a)



b)

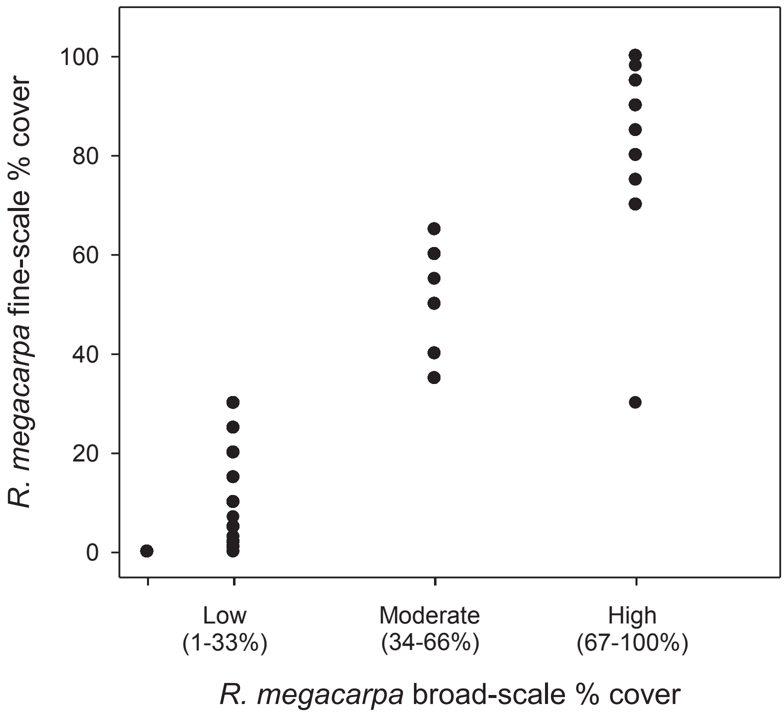


Figure 4.7: Comparison between the RBA method of broad percent cover categories and the fine-resolution percent cover from Chapter 3: (a) Box plot; (b) scatter plot.

4.3.3 **In situ** *seagrass sample comparison*

The percent cover categories of *R. megacarpa* used in the RBA method accurately distinguished areas of low, moderate, and high percent cover measured from *in situ* benthic core samples (Table 4.3; Fig. 4.8; $F_{2,29,}$ $p<0.05$). Both above-ground dry weight and seagrass shoot density significantly differed between categories (Table 4.3; Fig. 4.8a,b; $F_{2,29,}$ $p<0.05$ and $F_{2,98,}$ $p<0.04$, respectively). No significant effect was determined for collection type (Bromar corer vs. perspex corer) ($F_{1,\ 36,}$ $p=0.5$). However, although these broad-scale percent cover categories were significantly different between each category (Tukey's HSD test), the range of above-ground biomass weights and densities did not follow linear relationships (Fig. 4.8a,b). This showed that *in-situ* samples of seagrass were easily distinguishable between the broad-scale categories from above-ground dry weight and seagrass density (Fig. 4.8). Seagrass length categories reliably distinguished short and long seagrass lengths from video but were not significantly different (Table 4.3; $F_{2,29,}$ $p>0.05$), where Tukey's HSD test showed that short and medium lengths were not significantly different (Table 4.3; Fig. 4.8c). Additionally, epiphytic algal biomass was found to be significantly different between all broad-scale percent cover categories (Table 4.3; $F_{2,29,}$ $p<0.05$).

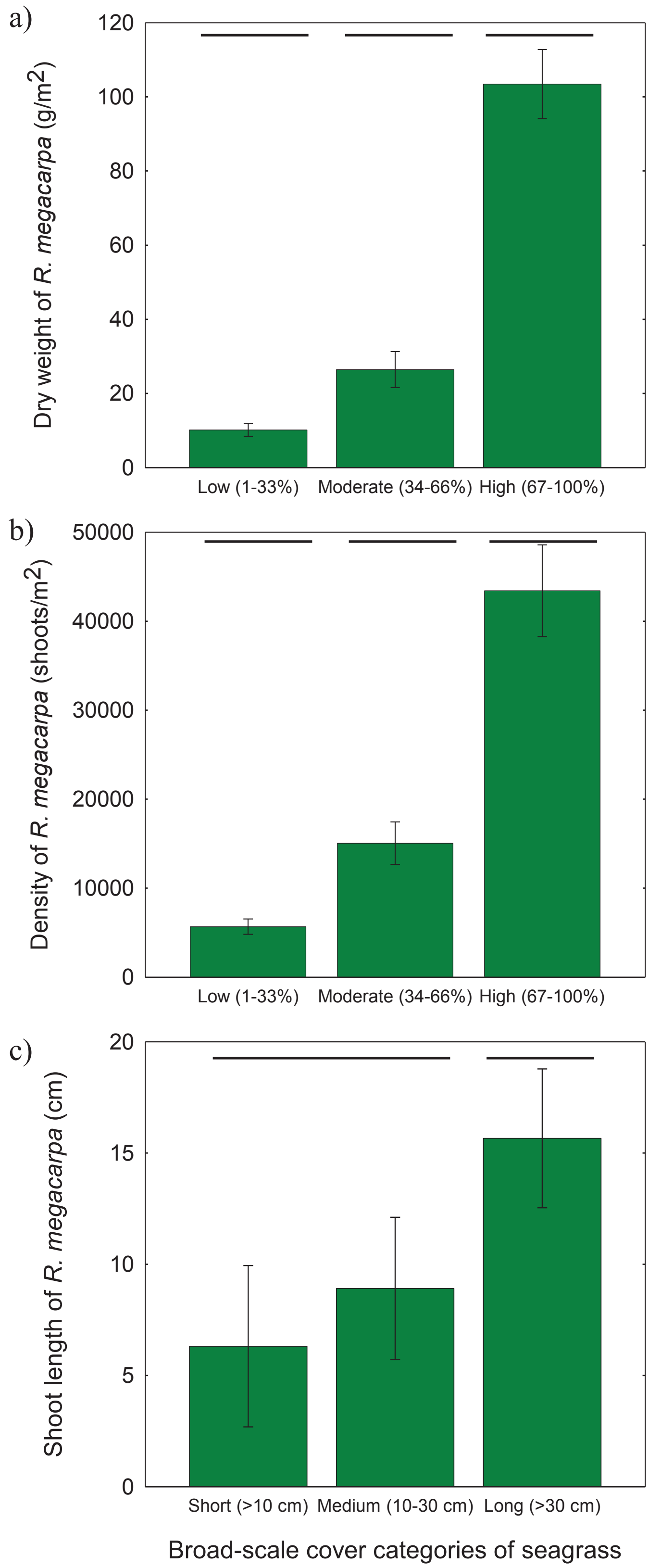


Figure 4.8: Comparisons of broad scale estimates of *R. megacarpa* cover for low (n=9), moderate (n=9) and high (n=9) towed-video categories compared to *in situ* measures from benthic cores of: (a) *R. megacarpa* above-ground biomass (mean dry weight); (b) *R. megacarpa* density (mean number of shoots p/m); (c) *R. megacarpa* shoot length (mean dry weight). Significant differences between categories are indicated using Tukey's HSD test outlined above each cover category.

Table 4.3: Summary of nested analysis of variance of: (a) above-ground dry-weight; (b) epiphytic algae; (c) density and shoot length of *R. megacarpa*.

Source	Df	Sum Sq	Mean Sq	F value	Pr(>F)	Significance
***a)** Dry weight of R.megacarpa*						
%category	2	7.79	3.89	45.25	1.21E-09	***
%category:Location	6	0.56	0.09	1.09	0.39	
Residuals	29	2.50	0.09			
***b)** Dry weight of epiphytic algae*						
%category	2	7.01	3.50	28.27	1.55E-07	***
%category:Location	6	3.93	0.66	5.29	0.0009	***
Residuals	29	3.60	0.12			
***c)** Shoot density of R.megacarpa*						
%category	2	118140	59070	65.33	1.81E-11	***
%category:Location	6	15290	2550	2.82	0.028	*
Residuals	29	26220	900			
***d)** Shoot length of R.megacarpa*						
%category	2	30.21	15.10	2.95	0.07	*
%category:Location	6	27.33	4.56	0.89	0.52	
Residuals	29	148.64	5.13			

4.4 Discussion

Seagrasses are often extensive habitats within estuaries and can be one of the first habitats within a marine-coastal interface to show signs of deterioration from eutrophication, increased nutrients and decreased water quality. As a consequence of this potential rapid rate of change and the major consequences to ecosystem functioning, there is an increasing need to be able to detect rapid changes in seagrass structure and spatial distribution over broad spatial scales (Duarte et al., 2008). Traditional methods are often too slow in processing these data to capture these habitat changes, consequently, these habitats become hard to manage when the change is too rapid to monitor. Therefore, there is a need for a rapid and repeatable method that can be used for monitoring and evaluating habitat change. In this study, abiotic and biotic habitats were quantified in real-time using the Rapid Benthic Assessment (RBA) method. The RBA method provided a quick and easy method to non-destructively quantify benthic habitats within large estuarine areas. Broad-resolution cover categories were accurate and allowed for easy differentiation and recorded in real-time, indicating that percent cover changes in seagrass beds can be accurately captured over broad scales with extremely shorter post-processing times. Broad-resolution percent cover categories of seagrass from the video were supported by in situ samples and accurately categorized within five normally turbid estuaries in south-west Australia.

The amount of seagrass cover change between the RBA method and the fine-resolution post-processing method was dependent on how heterogeneous the seagrass bed was. Over homogeneous seagrass habitats, the RBA method captured comparable amounts of habitat changes, but over heterogeneous transects (e.g. patchy seagrass cover), the fine-resolution post-processing method captured approximately 50% more habitat changes. This is best explained by the intentional design of the RBA method, which incorporated broad semi-quantitative percent cover categories to minimize user bias with the use of a subjective video technique. This is supported by (Dethier et al., 1993), where visual estimates of broad percent covers of seagrass were found to be more accurate and robust compared to point-count methods. Similarly, estimating percent cover of benthic habitats is now becoming more widely used within deeper marine habitats (Buhl-Mortensen et al., 2009; Cappo et al., 2011). In comparison, the fine-resolution post-processing method was more beneficial in capturing more combinations of multiple habitat types, whereas the RBA method captured only primary and secondary substratum types. However, the RBA method was further supported by similar mean percent covers of substratum, highlighting that the RBA method was a very time-efficient and useful tool to analyse broad-scale habitat changes in the field.

Previous attempts to map estuaries within south-west Australia using traditional methods such as aerial photograph resulted in poor distinction between different habitats (Bastyan et al., 1995), whereas in this study, underwater video provided a unique visual advantage to turbidity-limited estuaries. Importantly, the RBA method was time-efficient, simple, and allowed for a visualisation of benthic habitat patterns along transect gradients and within basin areas, with differences in seagrass composition between permanently, artificially opened and normally closed estuaries. Marine species of seagrass were common to Leschenault Estuary (e.g. *H. ovalis and H. tasmanica*) whereas *R. megacarpa* was common as monospecific strands in Wilson, Beaufort, Wellstead and Stokes Inlet (Brearley, 2005). Between all estuaries in this study, seagrass was restricted to depths < 4 m. Seagrass density was generally highest closest to shore, but dependent upon depth throughout each estuary. For example, seagrass occurred along the shallow northern reaches but were absent from the southern edges of the estuary in Wellstead estuary as it was shallow throughout the entire estuary. Additionally, seagrass distribution was also dependent upon a number of factors such as geographic location (latitude and longitude), sediment type and grainsize.

A number of indicators are used in determining the health and status of estuarine areas based on seagrasses. Measures of cover, density, biomass, extent, species abundance, and epiphytic algae are commonly used to assess health and restoration success (Madden et al., 2009). However, fine percent cover estimates can take considerable time to post-process (Mumby et al., 1999), and seagrass habitats may have significantly declined before effective management has been established. The restoration of seagrass habitats has had mixed results in the past and is predominantly dependent upon prevailing water quality conditions. If seagrass degradation was previously caused by anthropogenic disturbances of poor water quality and high turbidity, then restoration and reproductive effort of seagrass in the same conditions may be unsuccessful (Cabaço & Santos, 2012). However, if seagrass degradation was caused by natural events such as storms, and the site showed recruitment of seagrass, seagrass restoration efforts in this area may be successful. While these indicators are visual responses to change, these measurements allow us to determine the distribution and abundance of seagrasses. Seagrass length was highly variable within each estuary, and in Wilson Inlet, is expected to show large seasonal differences (Carruthers et al., 1999). Furthermore, while exact spatial extents of seagrass beds were not determined, maps of the data show likely fragmentation within the beds at large spatial scales (~15 m).

4.4.1 Strengths, limitations and recommendations

The main focus of this chapter was to develop and present a rapid assessment method to delineate between rapid semi-quantitative broad-resolution percent cover of seagrass habitats in five estuaries of south-western Australia. While this method may be useful for rapid change assessment, we highlight that monitoring these study sites over time were outside the scope of this study. Given the estuaries were too turbid to map benthic habitats with large-scale conventional optical remote sensing methods such as aerial photography, we approached benthic habitat mapping of each estuary at a finer-scale with underwater video and supplemented this technique with *in situ* seagrass samples and water quality samples. The high standard errors between the broad scale video classes for the *in situ* seagrass samples potentially indicate that a greater number of samples would have been beneficial to this study. Additionally, we found that the RBA method could not capture fine-scale habitat patterns which occurred at <30%. While the inclusion of other remote sensing technologies and additional *in situ* sampling would have greatly improved the delineation of benthic habitats and broad-scale percent cover categories of seagrass, we were limited by funds and available time for the survey. The RBA method allowed us to concurrently characterise geological bedforms, substratum types and biological habitats and species in real-time, and facilitated the reproduction of broad-scale patterns within each estuary. Underwater video provides a permanent record of data, the ability for further post-processing by multiple observers, and is non-destructive. As such, it allows for repeated monitoring of the same areas without altering the environment. Although the RBA method suppresses the level of ecological detail attained with post-processing, it has the advantages of providing fast and reliable data acquisition, preliminary maps within hours of collection, and providing ability to identify areas of interest in which to focus future effort, with video footage available for validation and further finer detailed examination.

Mapping the spatial extent and occurrence of key taxa allows the visualisation of estuarine habitats and identification of areas of potential impact with respect to seagrass health. This means that authorities responsible for monitoring estuarine systems have the ability to respond quickly to changes that may negatively impact the ecosystem. Habitat change and potential processes governing that change can be detected by collecting data across the same estuaries over time. Furthermore, documented change from underwater video is a useful communication tool for building public awareness of seagrass importance (Orth et al., 2006). It provides a simple, yet effective method of highlighting the importance of capturing change in seagrasses and within estuarine habitats. The video footage from this study was provided to the Department of Water in Western Australia for educational and management purposes.

In summary, a rapid assessment method for delineating between broad-resolution percent cover of seagrass habitats was established and supplemented with *in-situ* seagrass samples with promising results. This has broad implications for conservation and management agencies in mapping broad-scale patterns in shallow and turbid underwater environments within short time-frames (while on survey), and mapping the extent and distribution of habitats that are often prone to rapid changes due to anthropogenic and climatic affects and identify habitats of interest for further surveying. This is particularly important for areas which have no spatial baseline habitat information available.

Chapter 5: *General Discussion*

Chapter 5: General discussion

5.1 Discussion

In this thesis, I focus on using underwater video as a primary method to map and characterise benthic habitats in south-west Australian estuaries. I build upon existing methods and develop a new method to characterise concurrent biological and physical habitats and estimate fine-scale percent cover of benthic habitats within estuarine habitats. I apply a state-of-the-art modelling technique called Random Forests to model the relationship between physical and biological factors and map and predict their spatial distribution in unsampled areas with models performing favourably. Following this, I also compare the fine-scale method with a rapid, real-time broad-resolution method to estimate seagrass percent cover as these habitats often experience swift decline in association with declining water quality. The results of this thesis highlight that underwater video can be analysed in multiple ways to map and characterise benthic habitats depending on time, cost, end goals, resources and study area.

Benthic habitats within estuaries play important environmental and economic roles in productivity, food sources and shelter for a number of vertebrate and invertebrate species, sustain commercial fisheries, and recreational activities in an ever expanding population. Habitat-formers are often significant and valuable to ecosystem functionality by providing complexity and increasing species biodiversity. In addition, biotic habitats such as seagrasses and macro-invertebrates often serve as indicators of estuarine health through their rapid response to changes in their surrounding environment; either from hydrodynamics, sedimentation, erosion or eutrophication (Orth et al., 2006). Physical and environmental parameters which have direct (e.g. pH, temperature, salinity) and indirect (e.g. latitude and longitude correlated with rainfall and climate regime) gradient effects on individual species are often used to explain their associated ecological processes, distribution, and abundance (Austin, 2002). This thesis uses similar aspects of this approach to predict the spatial distribution and abundance of benthic habitats using an ensemble classifier called Random Forests. Determining the spatial distribution of benthic indicators in estuarine and marine systems is a significant component to effective conservation management (Austin, 2002). By identifying and monitoring ecological and anthropogenic changes to estuarine environments, we are better equipped at implementing relevant measures to ensure healthy habitats persist through time.

The results of this research will enable resource management authorities to view broad spatial maps of multiple benthic habitats within estuaries of south-west Australia using a combination of underwater video, physical samples and environmentally-derived parameters. Underwater video is often used to ground-truth remote sensing techniques such as aerial photography, satellite imagery, and multibeam sonar. A variety of methods currently exist for the analysis of underwater video, often with different end goals, strengths and weaknesses associated with each analysis (Chapter 2). Chapter 2 identifies the most suitable method for video analysis with respect to the estuaries and their associated logistical issues in this study. While each method may vary in objectivity and subjectivity, the results from Chapter 3 show that the similarities between objective and subjective approaches are highly comparable. Clearly, this thesis shows that environmental parameters were vital in modelling and predicting the spatial distribution of biotic and invertebrate species within estuarine habitats from fine-scale post-processed video data (Chapter 3). I show that the use of Random Forests as a modelling tool allowed us to visualise spatial distributions and abundances from point samples of underwater video, and examine the relationship between bio-physical interactions of biogenic and invertebrate distributions with overall high performance rates (Chapter 3). The real-time Rapid Benthic Assessment (RBA) developed in Chapter 4 had high precision, accuracy and comparable mean percent covers between the broad-scale cover classes of benthic habitats and the fine-scale post-processed data from Chapter 3. While the RBA method was able to capture a similar number of habitat changes within homogeneous seagrass beds, it was unable to capture similar numbers of habitat changes in heterogeneous beds (Chapter 4). However, the RBA method provides a real-time semi-quantitative assessment of benthic habitats with rapid turn-around of data, with the archived video available for post-processing. The ability to visualise broad-scale habitat patterns in real-time allows us to target important areas for conservation and monitoring in future surveys (Kelly et al., 2001, Dennison et al., 2007). Although this research includes data over a limited temporal window, and therefore restricts the examination of spatial distribution over temporal scales, the outputs of these baseline predictions can be used to compare how future species distributions differ.

5.1.1 Habitat patterns between permanently opened, seasonally opened and normally closed estuaries

Effective science communication begins with clear ideas conveyed to a broad range of stakeholders (Dennison et al., 2007). Conceptual models and maps are effective tools in communicating the ecological status, change, and process of estuarine and marine habitats. The synthesis of data into map and graph form allows us to convey information of habitats and processes efficiently (Dennison et al., 2007). Estuarine conceptual modelling is a useful tool that illustrates the ecological and geochemical processes that occur within estuaries. The conceptual model presented in this chapter (Fig. 5.1) was reproduced from Carruthers et al., (2007) and is used here to illustrate generalised hydrological processes and biotic assemblages of the estuarine systems along the south-west coast of Australia. This means that there are implicit assumptions of the conceptual model which may not be applicable to all estuaries. This conceptual model summarises two hydrodynamic regimes and illustrates similarities in their highly dynamic habitats, processes, and species diversity (Carruthers et al., 2007). Their varying catchment size, annual rainfall, and anthropogenic nutrient inputs affect the biotic assemblages of estuaries, and closure regimes, all of which have influenced the results found in Chapter 3 (Fig. 5.1). Similar processes have been identified in other estuaries around the world and are not limited to Australia, highlighting the importance of generalising broad-scale conceptual patterns that are indicative of worldwide patterns. Other seasonally or artificially closed estuaries are present in South Africa (Whitfield, 1992), the west coast of North America (Emmett et al., 2000), the central coast of Vietnam (Tran, 2011), the south-west coast of Sri Lanka (Silva & Davies, 1986), and the south-east coast of Brazil (De Souza et al., 1986). Conceptual models are effective in rapidly conveying patterns and processes of ecological systems to multiple stakeholders with similar worldwide patterns.

Similar to Carruthers et al. (2007), the results from this thesis highlight that dynamic, ephemeral, mono-specific seagrasses (*R. megacarpa*) and biotic assemblages (algae) were found in normally closed and seasonally opened estuaries. In permanently opened estuaries, in addition to *R. megacarpa*, seagrass species such as *H. ovalis* and *H. tasmanica* dominate the system, each occupying distinct areas of distribution. However, as depth was significantly different between all estuaries, seagrass habitats displayed different spatial patterns since estuaries did not always contain deep, unvegetated basin areas or shallow seagrass habitats adjacent to the shoreline. Instead, some estuaries were shallow throughout, therefore vegetated throughout the riverine areas (e.g. Wellstead Estuary). As seagrasses can be thought of as 'biological sentinels' of the estuarine and marine environment, its decline is often prelude to a much larger problem, heralding the effects of natural and anthropogenic-induced stress (Short et al., 2011). It is therefore increasingly important to monitor their distribution and abundance over spatial and temporal scales.

Additionally, estuarine health was measured in terms of epiphytic algal coverage on seagrass leaves in Chapter 3. Epiphytic algal growth on seagrass leaves is associated with increased nutrient enrichment, can reduce photosynthesis of seagrass leaves (Bulthuis & Woelkerling, 1983; Silberstein et al., 1986; Ralph et al., 2007), and can outcompete seagrass growth (Hauxwell et al., 2003; Valiela et al., 1997). Conversely, epiphytic growth can increase the recruitment and survival of other native species (Rohr et al., 2011). The results in Chapter 3 enabled us to estimate estuarine health by percent cover of epiphytic algal cover on seagrass leaves, identifying areas of excessive epiphyte fouling within each estuary, and potentially locating point sources of pollution or high influx in nutrient enrichment. Given the importance of seagrass as an ecological service provider not only to the adjacent community, but the habitats and ecosystems that survive within them, the need for their efficient management becomes a subject for management, government agencies, and the local community.

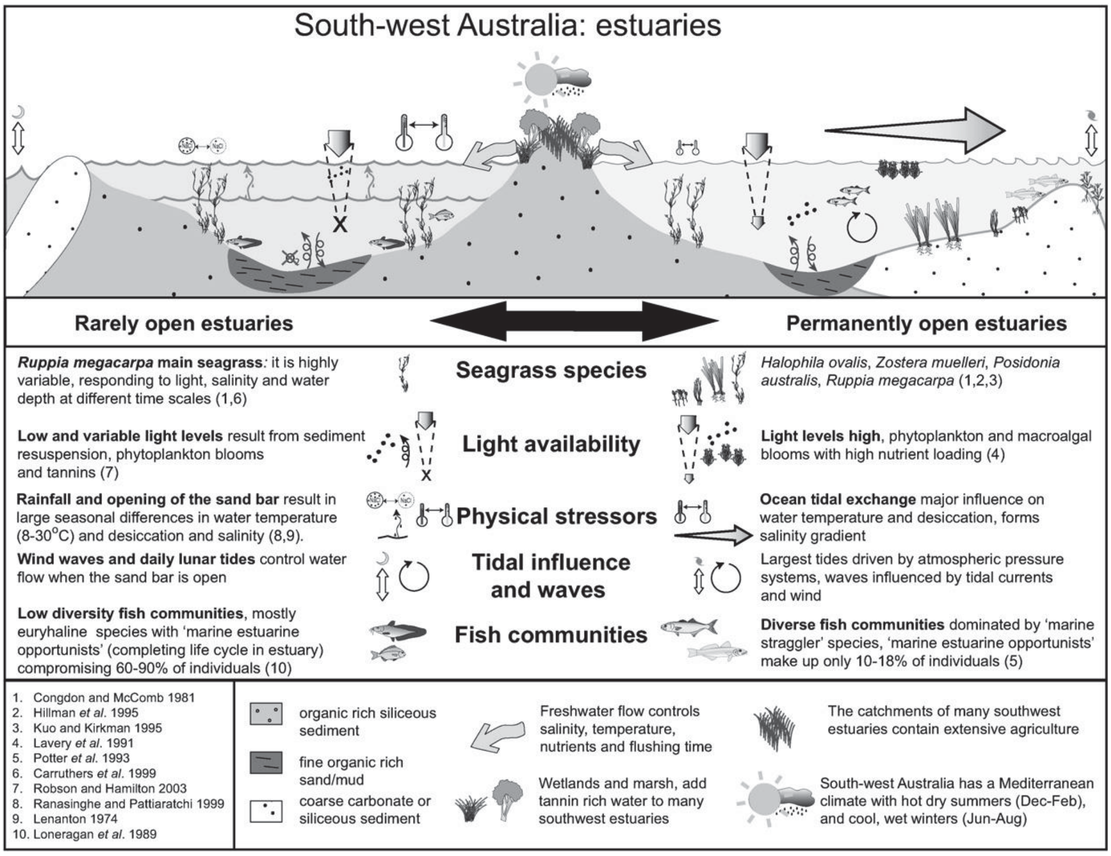


Figure 5.1: Conceptual model of hydrodynamic regimes and habitats present in south-west Australian estuaries (Carruthers et al., 2007) (Reprinted from Journal of Experimental Marine Biology and Ecology, 350, Seagrasses of south-west Australia: a conceptual synthesis of the world's most diverse and extensive seagrass meadows, 21-45, 2006, with permission from Elsevier)

Predicting the spatial distribution and abundance of green and red algae is also important in determining estuarine productivity. Overlapped with seagrass maps, such predictions can identify algal-dominated habitats, areas of species competition, and habitat fragmentation. Algal assemblages were determined from underwater video with the number of red algal morphologies declining from north to south-west and from permanently opened to normally closed estuaries, while green algal morphologies were more common in normally closed estuaries. Although other studies have identified a high number of algal species for some estuaries in this study, the underwater video used in this study was not of a high enough resolution to efficiently determine algal assemblages to species level (Wurm & Semeniuk, 2000). Consequently, it is recommended that a video system should incorporate high resolution still images, as well as the collection of *in situ* samples to allow for species identification. Additionally, polychaete mounds and mussel clumps were modelled and spatially predicted with varying degrees of success in Chapter 3. Although the variance explained by the models were relatively low in comparison to other modelled benthic habitats, the predicted distributions of invertebrates aligned with similar spatial distributions often found in normally closed estuaries along banks and areas of hard substratum (Brearley, 2005). This highlights the usefulness and importance of spatial prediction using Random Forests, which can represent accurate spatial patterns in nature. The synthesis of hydrodynamic and physical processes of estuarine environments is valuable in effective science communication to stakeholders and resource management and can facilitate programs to monitor the health and status of estuarine and marine systems.

The future of key coastal ecosystems such as seagrass and algae beds is somewhat unknown. As population inevitably increases, so too will anthropogenic effects of man-made structures within coastal zones, nutrient runoff from agricultural practices (Orth et al., 2006). While key iconic coastal ecosystems such as coral reefs, mangrove beds and salt marshes are widely known for their charismatic ecosystem inhabitants, seagrasses have only slowly been receiving the attention they deserve (Carruthers et al., 2007; Orth et al., 2006). Hopefully the present time is not too late a time to be investing in their survival and longevity.

5.1.2 Using Random Forests to model species distribution and abundance in estuaries

Species often respond to ecological gradients in a non-linear and asymmetric manner (De'ath & Fabricius, 2000). Traditionally, statistically independent samples are often collected in terrestrial systems (Elith & Leathwick, 2009). However, in their estuarine and marine counterparts, continuous tow data is collected, resulting in a loss of statistical independence (Elith & Leathwick, 2009). Statistical models, such as machine learning methods (e.g. Random Forests), allow us to capture this non-linearity, accept categorical and non-categorical data, and are flexible modelling techniques for ecological data (Liaw & Wiener, 2002; Elith et al., 2008; Leathwick et al., 2006). Ecological relationships and patterns often reveal that certain physical or environmental parameters can influence the spatial distribution of a species (Austin, 2002). Similarly, species distribution models use geographical and environmental gradients to explain biotic presence, absence or abundance.

Within this thesis, RF models performed well in predicting the biotic habitats in estuaries, explaining 79-90% variation of *R. megacarpa* models. They clearly reflected similar patterns driven by depth, latitude and longitude, supporting previous studies of seagrass growth and distribution (Carruthers et al., 1999; Brock, 1982; Carruthers & Walker, 1999). While other seagrass models (*Halophila ovalis* and *Heterozostera tasmanica*) did not perform as well as *R. megacarpa* (85 and 56%, respectively), the models highlighted clear and distinct areas of distribution, which is supported by Wurm & Semeniuk (2000). Unfortunately, the interpolation method of physical parameters was evident in the models, as round 'bulls-eye' distributions are clear artefacts in the spatial predictions of some benthic habitats. Modelled green algal distributions varied between estuaries, and explained between 55-80% variance, again driven by latitude, longitude and depth, but varied between estuaries. Similarly, modelled red algal distributions were explained best by latitude, longitude and isotopes $\delta^{13}C$, but these variables were not consistent between estuaries. However, real spatial patterns in biotic environments are unlikely to occur as bands demonstrated by modelled green algae distributions, which highlights that unmeasured environmental variables may be hindering the model to adequately represent spatial distributions (Dormann et al., 2007). Consequently, it is recommended that future studies incorporate other physical variables known to influence benthic habitat distribution (e.g. light flux, chlorophyll *a* measurements), and more study sites to better explain the distribution and abundance of benthic habitats within estuaries to prevent the interpolation effects affecting the final modelled distributions. This can only improve the performance of the model to predict the spatial distribution of habitats. Additionally, while it may be useful to predict future spatial distributions of biotic habitats with previously known data, challenges lie with estimating the amount of change associated with biological processes and interactions (Elith & Leathwick, 2009).

Using machine learning methods such as RF on underwater video data from estuaries is a novel modelling method to determine the spatial distribution and abundance of biotic habitats. The results in Chapter 3 identified that fine-scale post-processed geo-located underwater video characterisations combined with derived variables of distance, and interpolated physical variables allowed for the spatially continuous prediction of benthic habitats within south-west Australian estuaries. However, incorporating historical events that may be attributed to species distribution is fraught with problems and can often result in models that are ineffective at explaining species distribution (Austin, 2002). Hence, previously reported events such as fish kills which could be attributed to poor water quality and may dictate low species occurrence in estuaries, weren't incorporated in our study even though similar events have occurred in Beaufort and Stokes Inlet. The results from this thesis demonstrate the applicability of using machine learning methods to predict species distributions with high model performance over spatial scales in multiple estuaries.

5.1.3 Rapid characterisations of the seafloor

The RBA method of Chapter 4 demonstrated that real-time characterisations made while in the field were comparable to the post-processed fine-scale percent cover from Chapter 3. This allowed for the visualisation of spatial patterns and provided semi-quantitative data of seagrass density and biomass. Additionally, a high degree of accuracy was found in the broad-scale cover categories of seagrass between *in situ* samples and the underwater video categories. This has broad implications for monitoring a large number of estuaries with short periods of time available for survey as the method allows for rapid data collection and analysis within hours of survey and high estimates of accuracy. Rapid delineation of benthic habitats of the seafloor provides valuable ground-truthed information to broad-scale (tens of kms) remote sensing techniques (Holmes et al., 2007; Becker et al., 2010; Rooper & Zimmermann, 2007). The RBA method also provides a permanent archive for further post-processing and is particularly useful for mapping real-time and rapid delineation of spatial extent and distribution of rapidly changing environments under threat (Wilkinson & Salvat, 2012). However, the broad-scale nature of the RBA method was unable to capture fine-scale changes within seagrass habitats at a scale of 12 m between characterisations (<30% change in cover). In addition, observers must be trained before surveys to ensure consistency in correctly characterising the seafloor (Miller & Müller, 1999). In contrast, the FoV method in Chapter 3 provided quantitative data at the expense of time. While the two methods are able to detect change of benthic habitats to a certain degree, the decision to monitor benthic habitats is often based on scale and dependent on time and funding available to resource management.

Spatial patterns in nature are often inextricably linked with scale and restricted by logistics (Levin, 1992). This means that a balance must be made between the amount of detail required for mapping and characterising benthic habitats and the time and funding available. It is also important to understand how much detail is lost using the methods outlined in this thesis and whether or not the scale of the RBA method is too coarse for the estuarine or marine species studied. The underwater video has the advantage of being easily post-processed to any scale needed. Additionally, the annual cycles of seagrass species vary amongst multiple locations within an estuary (Carruthers & Walker, 1999) so long term monitoring involving multiple transects is advocated. *Ruppia megacarpa* has been shown to rapidly decline within weeks in response to light reduction, either by turbidity or shading by epiphytic algae (Carruthers et al., 1999). Studies by Carruthers et al. (1999) show that a reduction in irradiance for existing *R. megacarpa* beds for more than 3 months has detrimental effects, but less than this period resulted in rapid recovery. Hence, the number of characterisations was deemed to be suitable for real-time characterisations as the space between characterisations was ~ 12 m, with the video available for finer scale post-processing if required.

One key issue in analyses in this thesis is that of data spatial autocorrelation. Spatial autocorrelation is the degree of dependency of video characterisations within each estuary (Legendre, 1993). Positive spatial autocorrelation is the concept where nearby observations are more similar then observations further apart, which causes issues with the lack of independence of samples, and violates statistical tests. Spatial autocorrelation was implicitly accounted for in Chapter 3 by incorporating spatial parameters of latitude and longitude as predictor variables for each RF model. Additionally, leaving 15 seconds between characterisation windows also enabled us to have some control on spatial autocorrelation within the video analysis. However, as mentioned above, there are inherent difficulties in incorporating spatial autocorrelation into models as ecological processes such as competition, larval dispersal, or unidentified environmental variable or process may be governing species abundance and distribution (Austin, 2002). It is recommended that future tests incorporate some form of testing for spatial autocorrelation, such as partial mantel test, or randomisation tests, to ensure that video observations are statistically independent.

5.1.4 Implications for estuarine management

One of the main issues facing resource management of estuaries in Australia is the remoteness of the study sites, the lack of consistency in standards of monitoring between agencies and states, and the increasing anthropogenic pressures on adjacent water systems due to population growth, urban development, agriculture and unsustainable harvest practices. Unfortunately, the funding to monitor and survey estuaries is dependent on state government funds and funding agencies which are prone to annual change, unfortunately not on the time-scales of habitat change. Equally important is the need for long-term solutions, integration between and within local and state governments, resource management, and engagement with commercial and recreational stakeholders, and local communities (Wilkinson & Salvat, 2012). However, the ideal to return to a past baseline of ecosystem health can also be as unrealistic, as over time changing climatic systems, land use and change in biodiversity may have drastically transformed the ecosystem processes and functions (Suding, 2011). Instead, a balance must be made between previously known baselines and spatial and temporal change for the long-term monitoring and survival of the ecosystem.

The results in this thesis have broad ecological implications to resource management with respect to rapidly changing benthic habitats in estuarine systems of Australia by providing models and predicted maps of benthic habitat abundance and distribution and to guide management in making informed environmental decisions (Chapter 3). Additionally, there is a clear validation from the high accuracy attained in using the broad-scale Rapid Benthic Assessment to characterise rapidly changing, turbid and shallow benthic habitats with data analysis possible within hours of data collection (Chapter 4). Although these methods focus on estuarine habitats, they are also applicable to a broad range of marine habitats, as evidenced by the C-BED method which has been used extensively in marine environments of variable depths (Anderson et al., 2008, 2011). With the ever-increasing move to monitor ecosystem health and sustainable habitats under natural and anthropogenic events, effective management of estuaries and the habitats within them are crucial in ensuring the continuation and maintenance of species biodiversity. Key benthic habitats such as seagrasses play monumental roles in organic carbon storage (Fourqurean et al., 2012, Mcleod et al., 2011). Formulation of restoration plans involving catchment management should be based on temporal studies of benthic habitats and is advocated in a wide range of applications from rivers to reefs. Unfortunately, this study does not capture the temporal aspect of benthic habitats in the five estuaries as data were collected over single points in time. However, the use of spatially predictive mapping techniques for visual delineation of benthic habitats is a widely used and effective tool to inform the general public of processes and spatial distributions in estuaries and marine habitats, and combined with temporal datasets it will facilitate detection of changes to these habitats over time.

The estuaries in this study are a small part of the number of estuaries that line the south-west coast of Australia. While the estuaries in this research span a broad range of opening regimes, surveying more estuaries with similar hydrodynamic regimes and similar methods outlined in Chapter 3 and 4 will serve to validate the results found in this study, allowing for broad generalisations to be made of estuarine benthic habitats with differing hydrodynamic regimes. Incorporation of additional environmental and physical parameters which are relevant to the distribution and abundance of benthic habitats within estuaries will be important in increasing the predictive power of modelling techniques and allow for effective monitoring temporal and spatial change in benthic habitats.

This research is one of the first to use underwater video combined with spatial and physical variables to predict biotic habitats in shallow and turbid estuaries of south-west Australia. This highlights the effectiveness of RF modelling within multiple ecosystems as it has been previously used in terrestrial, freshwater and marine environments. Additionally, depending on the scale and time allocated for research, the RBA model is a powerful tool with fast and accurate results and is advocated for broad-scale studies where baseline habitats are required.

Appendix

Table A1: Variable descriptions and types used in regression analyses

Variable	Description	Type
Slope	Slope of estuary benthos	Continuous
Depth	Depth in metres	Continuous
Distance to coast	Distance of sampling points to coast	Continuous
Distance to river	Distance of sampling point to closest river	Continuous
Distance to estuary bar	Distance of sampling points to estuary bar	Continuous
Easting	Easting coordinates for each estuary	Continuous
Northing	Northing co-ordinates for each estuary	Continuous
Camera-scale predictors % cover seagrass		
Ruppia megacarpa	% cover to intervals of 5	Continuous
Halophila ovalis	% cover to intervals of 5	Continuous
Heterozostera tasmanica	% cover to intervals of 5	Continuous
% cover Algae		
Red (branching)	% cover to intervals of 5	Continuous
Red (filamentous)	% cover to intervals of 5	Continuous
Green (branching)	% cover to intervals of 5	Continuous
Green (slime)	% cover to intervals of 5	Continuous
Green (unknown)	% cover to intervals of 5	Continuous
% cover Polychaete mounds		
Ficopamatus enigmaticus	% cover to intervals of 5	Continuous
% cover Mussel clumps		
Mytilus edulis	% cover to intervals of 5	Continuous
% epiphytic algae	% cover of epiphytic algae on seagrass	Continuous
Biota	sessile biota recorded to closest functional taxonomic groups	
Crab	Crustacean – *Portunus pelagicus*	presence/absence
Shrimp	Crustacean – *Caridea sp.*	presence/absence
Fish	Fish species present in estuaries (e.g. King George whiting, whitebait, black bream etc).	presence/absence
Lebensspurren	tracks, trails and pits left by infaunal animals on the	

Variable	Description	Type
	seafloor	
Burrows	Bioturbated burrows with entry hole	presence/absence
Mounds	mound of sediment	presence/absence
Pits	Small depression <25 cm	presence/absence
Craters	Large depression >25 cm	presence/absence
Water quality predictors		
Temperature	Temperature of surface and bottom water (°C)	Continuous
Salinity	Salinity of surface and bottom water	Continuous
Secchi Depth	Depth at which secchi disk can no longer be seen (m)	Continuous
Dissolved Oxygen	Amount of dissolved oxygen in water at surface and bottom of water column (mg/L)	Continuous
Sediment quality predictors		
%Mud	Amount of sediment < 63µm present in sample	Continuous
%Sand	Amount of sand (63µm – 2 mm) present in sample	Continuous
Porosity	Porosity of sample	Continuous
% Carbon	% Carbon content of sample	Continuous
% Nitrogen	% Nitrogen content of sample	Continuous
Skewness	Symmetry or asymmetry of grainsizes to the average	Continuous
Kurtosis	Amount of concentration of grainsizes to the average	Continuous
Sorting	Amount of spread around mean grainsize	Continuous
Mean	Mean grainsize of sediment samples	Continuous

Table A2: Performance of interpolation methods and associated variables for optimal performance for each physical variable in all estuaries.

Estuary	Variable	Optimal Power	Regression Function	Mean	Relative Mean Square Error	Relative Root Mean Square Error %	Mean Absolute Error	Relative Mean Absolute Error %	Coefficient of Variation%
Stokes	Sand	1	0.07* x+58.6	-1.5	41.0	63.7	1.5	2.4	27.9
	Mud	1	0.07* x+34.8	1.5	41.0	115.2	1.5	4.3	47.1
	Porosity	1	0.01* x+61.5	0.3	21.7	34.9	0.3	0.5	12.9
	Secchi	2.11	0.05* x + 0.8	0.02	0.2	18.7	0.02	2.3	6.6
	TCO2 Flux	1	-0.1* x+8.1	0.8	24.1	372.1	0.8	12.5	64.8
	Sorting	1	-0.1* x+3.5	-0.02	0.9	29.7	0.02	0.6	5.2
	Kurtosis	1	-0.09 * x + 12.9	-0.2	10.0	83.6	0.2	0.7	18.5
	Skewness	1	-0.03 * x + -2.1	0.03	1.9	90.1	0.03	1.4	29.9
	Mean	1	-0.04 * x + 173.2	0.1	139.8	83.7	0.1	0.1	27.2
	N	1	0.1 * x + 0.2	0.009	0.2	124.8	0.01	4.9	56.0
	C	1	0.06 * x + 1.6	0.09	2.1	125.9	0.09	5.7	50.5
	Temp (Surface)	7.13	0.2 * x + 17.8	0.1	1.1	5.3	0.1	0.74	2.6
	Temp (Bottom)	6.82	0.1 * x + 18.7	0.1	0.9	4.4	0.1	0.5	2.2
	DO (Surface)	4.44	0.1 * x + 6.0	-0.01	0.2	3.6	0.01	0.1	2.0
	DO (Bottom)	2.20	0.6 * x + 2.3	-0.1	1.3	20.8	0.1	2.0	25.7
	Salinity	7.26	0.2 * x + 28.1	-0.07	0.7	1.9	0.1	0.2	0.9

Estuary	**Variable**	**Optimal Power**	**Regression Function**	**Mean**	**Relative Mean Square Error**	**Relative Root Mean Square Error %**	**Mean Absolute Error**	**Relative Mean Absolute Error %**	**Coefficient of Variation%**
	(Surface)								
	Salinity (Bottom)	2.88	0.8 * x + 8.2	0.8	4.4	11.6	0.8	2.2	16.9
	$\delta^{13}C$	4.16	0.3 * x + -14.9	-0.2	1.4	6.5	0.2	0.9	5.1
	$\delta^{15}N$	2.06	0.1 * x + 3.3	-0.1	1.3	32.6	0.1	3.4	16.6
Beaufort	Sand	1	-0.1 * x + 23.8	0.9	16.4	80.4	0.9	4.2	13.2
	Mud	1	-0.1 * x + 88.7	-0.9	16.4	20.6	0.9	1.1	3.6
	Porosity	1	-0.1 * x + 92.1	-0.7	9.5	11.5	0.7	0.8	1.9
	Secchi	1	-0.1 * x + 0.6	5.31125e-05	0.001	0.2	5.31125e-05	0.01	16.9
	TCO2 Flux	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
	Sorting	1	-0.1 * x + 5.9	0.1	1.7	32.02	0.1	2.1	5.1
	Kurtosis	1	-0.1 * x + 2.9	-0.03	0.5	17.5	0.03	1.2	3.7
	Skewness	1	-0.1 * x + 0.2	-0.009	0.3	1514	0.01	4.3	25.9
	Mean	1	-0.1 * x + 25.9	1.2	30.0	130.6	1.2	5.2	20.2
	N	1	-0.2 * x + 0.5	-0.005	0.2	43.01	0.01	1.3	7.9
	C	1	-0.2 * x + 4.5	-0.05	1.4	36.2	0.05	1.3	6.3
	Temp (Surface)	8.82	0.2 * x + 17.2	-0.4	1.4	6.7	0.4	2.1	3.2
	Temp (Bottom)	1	-0.1 * x + 20.5	-0.001	0.01	0.06	0.001	0.01	5.6
	DO (Surface)	6.1	0.3 * x + 4.1	-0.1	0.7	11.02	0.1	2.4	8.4
	DO (Bottom)	1	-0.1 * x + 5.5	0.0008	0.01	0.3	0.001	0.02	28.2
	Salinity (Surface)	1	-0.1 * x + 50.8	0.04	2.7	5.9	0.04	0.1	1.2

Estuary	Variable	Optimal Power	Regression Function	Mean	Relative Mean Square Error	Relative Root Mean Square Error %	Mean Absolute Error	Relative Mean Absolute Error %	Coefficient of Variation%
	Salinity (Bottom)	1	-0.1 * x + 47.1	0.0002	0.009	0.02	0.0002	0.0004	2.1
	$\delta^{13}C$	1	-0.2 * x + -29.1	0.06	1.2	4.9	0.1	0.3	1.1
	$\delta^{15}N$	1	-0.2 * x + 4.3	-0.01	0.5	12.3	0.01	0.4	2.3
Wilson	Sand	1.46	0.4 * x + 26.4	-0.4	35.9	80.5	0.4	0.8	62.7
	Mud	1.46	0.4 * x + 33.5	0.4	35.9	64.7	0.4	0.7	49.6
	Porosity	1	0.3 * x + 52.0	0.07	17.0	24.1	0.1	0.1	15.9
	Secchi	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
	TCO2 Flux	1	-0.1 * x + 8.9	0.5	9.7	128.5	0.5	6.9	21.8
	Sorting	1	-0.03 * x + 3.6	-0.08	1.4	41.2	0.1	2.2	15.3
	Kurtosis	5.65	0.136 * x + 3.0	-0.2	3.1	79.3	0.2	4.1	43.9
	Skewness	5.18	0.3 * x + -0.3	0.03	0.8	155.4	0.02	4.9	90.3
	Mean	1	0.3 * x + 69.8	-1.1	132.5	113.8	1.1	0.9	71.5
	N	2.59	0.4 * x + 0.3	-0.0003	0.3	67.6	0.0003	0.1	53.4
	C	2.35	0.4 * x + 2.8	0.02	3.1	68.1	0.02	0.5	53.5
	D13c	1	0.4 * x + -13.2	-0.02	0.5	2.5	0.02	0.1	1.9
	D15n	2.80	0.2 * x + 3.8	-0.01	0.6	14.3	0.01	0.3	6.0
Leschenault	Sand	1	0.2 * x + 32.5	-0.7	22.4	57.3	0.7	1.7	24.4
	Mud	1	0.2 * x + 50.1	0.7	22.4	36.9	0.7	1.1	15.3
	Porosity	1.26	0.1 * x + 64.5	0.1	9.8	13.2	0.1	0.2	6.0
	Secchi	1.93	0.9 * x + 0.9	0.02	0.5	44.5	0.02	1.9	20.6
	TCO2 Flux	1	0.05 * x + 11.6	-0.2	9.2	74.2	0.2	2.0	27.7
	Sorting	1	0.1 * x + 4.4	-0.02	1.3	26.0	0.02	0.5	11.4
	Kurtosis	1	-0.06 * x + 3.3	-0.01	2.6	80.6	0.01	0.4	12.0
	Skewness	1	-0.002 * x + -0.5	-0.001	0.7	146.7	0.001	0.3	42.1

Estuary	Variable	Optimal Power	Regression Function	Mean	Relative Mean Square Error	Relative Root Mean Square Error %	Mean Absolute Error	Relative Mean Absolute Error %	Coefficient of Variation%
	Mean	1	-0.04 * x + 59.1	-0.9	71.6	122.0	0.9	1.6	25.4
	N	1.54	0.38 * x + 0.1	-0.0003	0.1	54.92319	0.0003	0.1	52.4
	C	1.15	0.2 * x + 1.7	-0.02	1.2	52.01	0.02	0.9	33.7
	Temp (Surface)	1	-0.01* x + 19.7	0.02	1.3	6.7	0.02	0.09	1.8
	Temp (Bottom)	1.08	0.04 * x + 18.001	-0.03	2.9	15.4	0.03	0.2	5.6
	DO (Surface)	1	-0.02 * x + 7.3	-0.001	0.8	10.8	0.001	0.01	3.8
	DO (Bottom)	1	-0.09 * x + 7.8	0.003	1.1	15.8	0.003	0.05	3.8
	Salinity (Surface)	1.46	0.2 * x + 33.3	0.05	2.1	5.4	0.05	0.1	2.7
	Salinity (Bottom)	1.93	0.3 * x + 28.0	0.05	1.9	4.8	0.05	0.1	3.2
	D13c	3.01	0.5 * x + -8.3	0.02	0.7	4.2	0.02	0.1	5.2
	D15n	5.61	0.1 * x + 2.1	0.03	0.8	37.6	0.03	1.2	16.8
Wellstead	Sand	1.63	0.1 * x + 23.8	-1.2	25.7	86.6	1.2	4.1	36.7
	Mud	1.63	0.09 * x + 66.7	1.2	25.7	36.5	1.2	1.7	14.6
	Porosity	1.39	-0.03 * x + 89.6	1.03	9.3	10.9	1.04	1.2	2.9
	Secchi	1	-0.06 * x + 0.7	0.009	0.1	14.4	0.01	1.3	3.01
	TCO2 Flux	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
	Sorting	1	-0.1 * x + 4.6	-0.02	0.9	23.2	0.02	0.5	5.3
	Kurtosis	1.76	0.2 * x + 2.2	-0.07	1.1	34.2	0.07	2.3	19.0
	Skewness	1.99	0.2 * x + -0.2	0.03	0.6	190.1	0.03	8.7	102.1
	Mean	1.91	0.2 * x + 25.8	-1.4	34.7	94.1	1.4	3.9	48.1
	N	1.61	0.009 * x + 0.8	0.03	0.4	52.9	0.03	4.5	16.2

Estuary	**Variable**	**Optimal Power**	**Regression Function**	**Mean**	**Relative Mean Square Error**	**Relative Root Mean Square Error %**	**Mean Absolute Error**	**Relative Mean Absolute Error %**	**Coefficient of Variation%**
	C	1.36	-0.03 * x + 7.2	0.4	3.3	50.4	0.4	5.7	12.6
	Temp (Surface)	3.55	0.2 * x + 17.6	-0.05	1.4	6.7	0.05	0.2	3.6
	Temp (Bottom)	2.71	0.2 * x + 16.6	-0.02	0.9	4.3	0.02	0.1	2.5
	DO (Surface)	2.78	0.3 * x + 4.3	-0.03	0.4	7.1	0.03	0.6	4.8
	DO (Bottom)	3.27	0.3 * x + 4.2	-0.09	0.6	9.6	0.09	1.6	7.7
	Salinity (Surface)	4.201	0.7 * x + 13.4	-0.05	1.1	2.7	0.05	0.1	3.8
	Salinity (Bottom)	4.27	0.8 * x + 6.7	-0.01	0.5	1.4	0.01	0.03	3.6
	$\delta^{13}c$	2	0.4 * x + -11.4	0.08	1.1	5.6	0.08	0.4	4.8
	$\delta^{15}n$	3.32	0.4 * x + 0.8	-0.05	0.6	36.4	0.05	2.9	34.2

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