CHEMICAL PROFILING OF EXHALED BREATH FROM CYSTIC FIBROSIS SUBJECTS USING COMPREHENSIVE TWO-DIMENSIONAL GAS CHROMATOGRAPHY

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A thesis submitted for the
Degree of Doctor of Philosophy (Science)
University of Technology Sydney
January 2021

CERTIFICATE OF AUTHORSHIP AND ORIGINALITY

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I certify that the work in this thesis has not previously been submitted for any other degree/s nor has it been submitted as part of the requirements for a degree/s except as fully acknowledged within the text.

I also certify that the thesis has been written by me. Any help that I have received in my research work and the preparation of the thesis itself has been acknowledged. In addition, I certify that all the information sources and literature used are indicated in the thesis.

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

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DEDICATION

To my parents

And

My younger brother and his everyday struggle to survive through terminal illness

Love you and always with you until I am alive

ACKNOWLEDGEMENT

Firstly, thanks and regards to my primary supervisor, Dr. Shari L. Forbes for her continuous support and for sharing her knowledge. I am also grateful to Dr. Tapan Rai (my second primary supervisor), Dr. Katie Nizio (co-supervisor), Dr Maiken Ueland (co-supervisor), and Dr Peter G Middleton (external supervisor) for their great support all throughout my PhD study. I am especially grateful to all the participants of my study both from the outpatient Cystic Fibrosis Clinic, Westmead Hospital and from the Faculty of Science, University of Technology Sydney. Without their generous participation the project wouldn't be possible. I am also grateful to my co-workers in the Gas Chromatography Lab here in UTS. In particular, Amber Brown, Nicole Cattarossi, and Vitor Cesar Taranto. In addition, my special thanks for technical staff; Dr. Ronald Shimmon, Dr Regina V. Taudte, and Dr. Dayanne Bordin. I am also very fortunate to be a part of such a lovely research group and have so many fun memories.

Finally, I am thankful to my family, especially my lovely wife Ayesha and a beautiful boy (Zyad) we are blessed with. Her relentless support and numerous sacrifices made this PhD battle possible. I am also asking forgiveness for not giving enough time to my wife, my boy, my brother, and above all my parents. I do feel guilty of that.

Most importantly, my gratitude to the Australian Government and the Taxpayers, because it is their money allocated to me as tuition fee waivers and living allowances. Without that I can't even dream of pursuing this PhD degree.

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LIST OF ABBREVIATIONS AND SYMBOLS

(Listed alphabetically)

ACFDR	Australian Cystic Fibrosis Data Registry
BAL	Bronchoalveolar lavage
CAR	Carboxen
CF	Cystic fibrosis
CFTR	Cystic fibrosis transmembrane conductance regulator
DMS	Dimethyldisulfide
DVB	Divinylbenzene
EBC	Exhaled breath condensate
GC×GC	Comprehensive two-dimensional gas chromatography
GC×GC-TOFMS	Comprehensive two-dimensional gas chromatography time-of-flight mass spectrometry
GC–FID	Gas chromatography flame ionisation detection
GC–MS	Gas chromatography mass spectrometry
GC–TOFMS	Gas chromatography time-of-flight mass spectrometry
HCN	Hydrogen cyanide
HS-SPME	Headspace–solid-phase microextraction
IMR–MS	Ion-molecule reaction mass spectrometry
IMS	Ion mobility spectrometry
LDA	Linear discriminant analysis
LD	Linear discriminant
LOOCV	Leave-one-out cross-validation
NMR	Nuclear magnetic resonance
PDMS	Polydimethylsiloxane
ppb	Parts-per-billion
PTR–MS	Proton transfer reaction mass spectrometry

SIFT-MS	Selected ion flow tube mass spectrometry
SPME	Solid phase microextraction
STs	Sorbent tubes
TB	Tuberculosis
VAP	Ventilator-associated pneumonia
VOCs	Volatile organic compounds

PUBLICATIONS

1. Forensic decomposition odour profiling: A review of experimental designs and analytical techniques. MA Iqbal, KD Nizio, M Ueland, SL Forbes. ***TrAC Trends in Analytical Chemistry 91, 2017, 112-124.***
2. Recent advances in the estimation of post-mortem interval in forensic taphonomy. MA Iqbal, M Ueland, SL Forbes ***Australian Journal of Forensic Sciences, 52, 2020, 107-123.***

CONFERENCE PROCEEDINGS

1. Poster presentation in the 24th Australian Society of Medical Research (ASMR) NSW ANNUAL SCIENTIFIC MEETING held by Australian Society for Medical Research during 6th June 2016 at Powerhouse Museum, Sydney, Australia.
2. Co-presented in the Combined Health Science Conference Sydney New Horizons held during 21st November 2016 at the University of Technology Sydney, Australia.
3. Poster presentation in the 35th Combined Health Science Conference Sydney New Horizons 2018, 19-20 November 2018, Kolling Institute, St Leonards, Australia.

AWARDS

1. Winner of April 2017 Paper of the Month for SCHOOL OF MATHEMATICAL AND PHYSICAL SCIENCES (MAPS), University of Technology Sydney.

ABSTRACT

Chronic lung infections are the leading cause of death in subjects with cystic fibrosis (CF). To date, sputum culture is the most common technique for the diagnosis of lung infections in adult CF subjects. However, it requires several days or longer to obtain culture results. Therefore, a rapid diagnostic technique for lung infections would significantly improve CF healthcare. During recent decades, exhaled breath analysis has attracted interest as a rapid and non-invasive tool for the diagnosis of non-communicable diseases such as cancers and heart diseases. However, there is limited progress in the diagnosis of infectious diseases such as lung infections in CF subjects using volatile organic compounds (VOCs) as biomarkers of infection.

In this study, sputum and breath samples were collected from CF subjects and healthy controls (only breath) and profiled for VOCs using comprehensive two-dimensional gas chromatography time-of-flight mass spectrometry (GC×GC–TOFMS). Multivariate analyses (e.g. principal component analysis and linear discriminant analysis (LDA)) were then performed to allow differentiation between: (i) CF subjects and healthy controls and (ii) CF subjects with/without *Pseudomonas aeruginosa* infections and those with no known lung infections as confirmed using their sputum culture results. This study identified a set of 16 VOCs which allowed differentiation between CF subjects and healthy controls. In particular, healthy controls were classified with 98% accuracy, while CF subjects were classified with 92% accuracy. It is important to note that all of the CF subjects that participated in this study are significantly different from control groups, not only in terms of their lung infection status but also in terms of numerous other factors (e.g. diet, lifestyle, medications, and other health complications). These factors can also impact the breath profiles obtained from the study group (CF subjects).

The analysis of matching sputum and breath samples collected from CF subjects provided a set of 24 core VOCs common between both sample types. LDA performed using these VOCs provided accurate classification of CF subjects according to their lung infection status (i.e. CF subjects with/without *Pseudomonas aeruginosa* infection). The outcome of LDA also showed that these common VOCs have better classification accuracy than the entire profile of the VOCs detected in sputum and breath samples. Finally, the comparison of breath profiles between CF subjects with/without *Pseudomonas aeruginosa* infection and those with no known lung infection showed that it is also

possible to allow differentiation between these contrasting groups using breath VOCs profiles.