

DECLARATION

This dissertation was supervised by Dr. J. Rossouw and Prof. J.A. Freat.

I hereby declare that this dissertation, submitted in fulfilment of the requirements for Magister Scientiae at the University of the Witwatersrand, Johannesburg, is the result of my own investigation unless acknowledged to the contrary within the text.

Adrienne N Saif

Date

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SUMMARY

The Detection of *Burkholderia* spp. and pathogenic *Leptospira* spp. in South Africa

By

Adrienne N. Saif

Leptospirosis is a zoonosis of ubiquitous distribution and causes a wide spectrum of disease. *Burkholderia* species are important plant and human pathogens. Little or no investigation has been performed on any clinically-relevant *Burkholderia* or *Leptospira* species in Johannesburg. Environmental samples were taken from different sites in Johannesburg along the Jukskei River. These were subjected to culturing for *Burkholderia* spp. and polymerase chain reaction (PCR) for *Burkholderia* and *Leptospira* spp. Human serum, animal serum and kidney samples were also subjected to PCR for both organisms. A *Leptospira* IgM ELISA was also performed on human serum samples. More *Burkholderia* spp. were isolated by culture from soil samples than water samples. The PCR yielded a significantly higher PCR positive from soil samples ($p = 0.015$). There was a high prevalence of pathogenic *Leptospira* spp. in soil samples. The ELISA yielded only 7.8% (26/332) positive samples. There were no human or animal positive PCR results for either organism. There is an environmental presence of both leptospires and *Burkholderia* in the area sampled. More studies are needed to establish how both organisms might affect patients with compromised immune systems, and how often both infections are incorrectly or under-diagnosed.

CONTENTS

Acknowledgements	ii
Summary	iii
Contents	iv
List of Abbreviations	vii
List of Figures	ix
List of Tables	xi
Chapter 1 Literature Review	1
1.1 General Introduction	1
1.2 <i>Burkholderia</i>	2
1.2.1 Background and History	2
1.2.2 Morphology	3
1.2.3 Life Cycle	4
1.2.4 Epidemiology	5
1.2.5 Pathogenesis	6
1.2.6 Clinical Disease	8
1.2.7 Diagnostics	9
1.2.8 Treatment	10
1.2.9 Importance	12
1.3 <i>Leptospira</i>	13
1.3.1 Background and History	13
1.3.2 Morphology	14
1.3.3 Life Cycle	15
1.3.4 Epidemiology	16
1.3.5 Pathogenesis	18
1.3.6 Clinical Disease	19
1.3.7 Diagnosis	21
1.3.8 Treatment	22
1.3.9 Clinical Importance	23

1.3.10 Human leptospirosis in South Africa.....	23
1.4 Objectives of Study.....	25
Chapter 2 Materials and Methods	26
2.1 Sample Collection.....	26
2.1.1 Environmental Samples.....	26
2.1.2 Human Samples.....	30
2.1.3 Rodent Samples.....	30
2.2 Culture of <i>Burkholderia</i> species.....	31
2.2.1 Media Selection.....	31
2.2.2 Processing of Environmental Samples.....	32
2.2.3 Colony Identification.....	32
2.2.3.1 Gram Stain and Microscopy.....	33
2.2.3.2 Oxidase.....	33
2.2.3.3 Catalase.....	33
2.2.3.4 API® 20NE.....	34
2.3 DNA Extraction.....	36
2.3.1 DNA Extraction from Environmental Samples.....	36
2.3.2 DNA Extraction from Bacterial Cultures.....	37
2.3.3 DNA Extraction from Human and Rodent Samples.....	38
2.3.4 DNA Quantification.....	38
2.4 PCR Amplification.....	39
2.4.1 <i>Burkholderia</i> spp. <i>RecA</i> Gene.....	41
2.4.2 <i>Burkholderia pseudomallei lpxO</i> Gene.....	41
2.4.3 <i>Leptospira</i> 16S rDNA Gene.....	42
2.5 PCR Optimization using Taguchi Technique.....	43
2.6 Agarose Gel Electrophoresis.....	43
2.7 Visualisation and Documentation.....	44
2.8 <i>Leptospira</i> IgM ELISA.....	44
2.9 Data Analysis.....	46

Chapter 3 Results.....	47
3.1 Environmental Samples.....	47
3.1.1 <i>Burkholderia</i> Culture.....	47
3.1.2 PCR.....	49
3.1.2.1 <i>Burkholderia</i> PCR.....	49
3.1.2.2 <i>Leptospira</i> 16S rDNA PCR.....	53
3.2 Human and Animal Samples.....	58
3.2.1 Animal.....	58
3.2.2 Human.....	59
3.3 <i>Leptospira</i> IgM ELISA.....	59
Chapter 4 Discussion and Conclusion.....	60
References.....	70
Appendices.....	84

LIST OF ABBREVIATIONS

AESC	Animal Ethics Screening Committee
AIDS	Acquired-immunodeficiency syndrome
Amp	Amperes
API	Analytical Profile Index
ATP	Adenosine triphosphate
ATCC	American Type Culture Collection
Bcc	<i>Burkholderia cepacia</i> vcomplex
BCSA	<i>Burkholderia cepacia</i> selective agar
Bp	Base pairs
ca	Approximately
CF	Cystic fibrosis
DMP	Diagnostic Media Products
DNA	Deoxyribonucleic acid
dNTP	Deoxyribonucleic triphosphate
EDTA	Ethylene
ELISA	Enzyme linked immunosorbent assay
EPS	Exopolysaccharide
<i>et al</i>	et alii (and others; abbreviation from Latin)
EtBr	Ethidium Bromide
g	Gram/s
HIV	Human immuno-deficiency virus
HRP	Horseradish peroxidase
HREC	Human Research Ethics committee
IL	Inter-leukin
IgM	Immunoglobulin M
IHA	Indirect haemagglutination
LPS	Lipopolysaccharide
<i>lpxO</i>	Dioxygenase gene
MAT	Microscopic agglutination test
Min	Minute
M	Molar
µg/ml	Microgram per milliliter
UHQ	Ultra high quality
µl	Microlitre/s
ml	Milliliter
mM	Milli-molar
mm ³	Cubic millimeter
mol	Mole
ms	Millisecond
NICD	National Institute for Communicable Diseases
NHLS	National Health Laboratory Services
n	Sample number
nm	Nanometer
n-PCR	Nested PCR
OFBBL	Oxidation-fermentation bactitracin lactose agar
PCA	<i>Pseudomonas cepacia</i> agar

PCR	Polymerase Chain Reaction
pmol/μl	Pico-moles per microliter
<i>recA</i>	Recombination gene
rRNA	Ribosomal RNA
rpm	Revolutions per minute
s	Second
SBPRU	Special Bacterial Pathogens Reference Unit
spp.	Species
16S rRNA	16 small sub-unit ribosomal RNA
TAE	Tris/acetate/EDTA
TNFα	Tumour necrosis factor alpha
U	Units
U/μl	Units per microliter
μM	Micromolar
μm	Micro-meters
UV	Ultra-violet
V	Volts
Wits	University of the Witwatersrand
w/v	Weight per volume
v/v	Volume per volume

LIST OF FIGURES

Figure 1.1	(A) An electron scanning micrograph of <i>Burkholderia cepacia</i> and (B) a photograph of a <i>Burkholderia cepacia</i> culture on McConkey agar.....	3
Figure 1.2	Ecology of <i>B. pseudomallei</i> and the interactions between environmental <i>Burkholderia</i> spp. and human-animal hosts.....	5
Figure 1.3	Morphology of a leptospire.....	14
Figure 1.4	The transmission cycles of pathogenic leptospirosis.....	16
Figure 1.5	Global annual incidence of human leptospirosis.....	17
Figure 1.6	(A) Conjunctival suffusion and (B) purpuric rash, symptoms of <i>Leptospira</i> infection.....	20
Figure 2.1	The drainage basin of the Jukskei River in Gauteng showing the respective sampling sites (black stars).....	29
Figure 2.2	Map illustrating the regions of the City of Johannesburg.....	31
Figure 2.3	API [®] 20NE strips illustrating positive (A) and negative (B) reactions.....	35
Figure 3.1	The percentage of five most frequent organisms isolated in soil and water samples collected in the Jukskei River catchment during 2010 and 2011.....	48
Figure 3.2	The percentages of <i>Burkholderia</i> organisms isolated by culture from soil and water samples collected in the Jukskei River catchment during 2010 and 2011.....	49
Figure 3.3	Agarose gel analysis showing the results of a <i>Burkholderia</i> spp. <i>RecA</i> PCR on cultured isolates from the API positives.....	50
Figure 3.4	Taguchi optimization performed on <i>Burkholderia LpxO</i> PCR using a control strain of <i>Burkholderia pseudomallei</i>	50
Figure 3.5	The percentages of <i>Burkholderia</i> species detected with <i>Burkholderia</i> spp. <i>RecA</i> PCR amplification from water and soil samples collected from Jukskei River catchment during 2010 and 2011.....	51
Figure 3.6	Agarose gel analysis of <i>Burkholderia</i> spp. <i>RecA</i> PCR soil samples from the Modderfontein Dam no.4.....	52
Figure 3.7	The percentage of <i>Burkholderia</i> culture and PCR positive samples.....	53

Figure 3.8	Agarose gel analysis of the products of the first round <i>Leptospira</i> 16S rDNA Taguchi optimization.....	54
Figure 3.9	Percentage of total leptospires found in soil and water by site.....	55
Figure 3.10	Agarose gel analysis showing the first round <i>Leptospira</i> 16S results.....	55
Figure 3.11	Percentage of pathogenic leptospires isolated in soil samples.....	56
Figure 3.12	Agarose gel analysis showing the results from a second round <i>Leptospira</i> 16S rDNA.....	57
Figure 3.13	Percentage of pathogenic to the total amount of leptospires found in soil samples.....	58
Figure 3.14	Shows the percentage positive and negative samples for the <i>Leptospira</i> IgM ELISA for 2010 and 2011.....	59

LIST OF TABLES

Table 2.1	Environmental samples collected in the Juskskei River catchment during 2010 and 2011.	28
Table 2.2	List of Primers used in this study.	40
Table 2.3	The components which make up a Taguchi square, optimized with at least three different concentrations.	43
Table 2.4	The interpretation of test results of the Panbio <i>Leptospira</i> IgM ELISA.	46