Use of microcosm and in-situ studies for the estimation of exposure risk from recreational coastal waters and sediments

Duncan L. Craig

B. App. Sci. (Environmental Health); B. Sc. (Hons)

August 2005

Department of Environmental Health,

Faculty of Health Sciences,

Flinders University

TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

LIST OF PLATES

SUMMARY

The interaction of microorganisms with sediments can enhance their survival by reducing exposure to various stressors and thus marine sediments may act as reservoirs for pathogenic microorganisms. In coastal waters there can be an increased risk of infection to humans due to the possible re-suspension of these microorganisms during recreational activities. This research attempts to more accurately identify environmental exposure in the first stage of a health risk assessment in recreational coastal waters.

Techniques were developed to successfully separate microorganisms from sediment particles. Of the methods investigated, subjecting diluted sediment samples to a sonication bath for 10 minutes was found to be the most efficient separation technique over a range of sediment types. This method was therefore used in the subsequent studies to enumerate organisms from the surface sediment layer, as distinct from the water column.

Faecal coliforms were enumerated by membrane filtration in both water and sediment from three Adelaide metropolitan recreational coastal sites, chosen to represent different physical sediment characteristics, over a 12-month period. All sites investigated met current National Health and Medical Research Council Guidelines for primary contact recreation. Faecal coliform concentrations were generally greater in sediment compared with overlying water for all samples. This was most evident in sediment consisting of greater silt/clay and organic carbon content (with up to 1000 times higher concentrations in the surface sediment layer compared with overlying water). For coastal recreational sites impacted by stormwater or river discharges, high faecal coliform concentrations were found to be associated with rainfall.

A laboratory-based microcosm study utilising intact sediment cores was undertaken to determine the decay rates of faecal indicator organisms (*E. coli*, enterococci and somatic coliphage) and pathogens (*Salmonella derby* and *S.*

typhimurium) in both overlying water and in various sediment types. For all organisms tested, temperature had an inverse relationship with survival. Greater decay was observed in the overlying water compared to the surface sediment layer. Small particle size and high organic carbon content was found to be more conducive to microbial survival. In general, decay rates of *E. coli* were significantly greater than enterococci and coliphage. Although no significant correlations were observed between decay rates of the pathogens and indicator organisms, decay of *Salmonella* spp. in overlying water more closely resembled that of *E. coli* than that of other indicators.

Using decay rates measured in the microcosm study and available dose-response data, a quantitative microbial risk assessment (QMRA) utilising Monte Carlo simulation was undertaken to estimate the risk of infection to *Salmonella* spp. and rotavirus following exposure to recreational coastal water subject to a range of faecal contamination levels. For modelling purposes, the assumption was made that rotavirus decay was equivalent to coliphage decay. The probability of infection from rotavirus due to exposure to contaminated recreational coastal water was greater than that for *Salmonella* spp. under all scenarios. This increased probability of infection is linked to the high infectivity of rotavirus compared to *Salmonella* spp.

Results of this research highlight the limited effectiveness of using prescribed faecal coliform concentrations in the water column alone to estimate the risk of exposure to pathogenic microorganisms during recreational activity at coastal areas. It demonstrated that coastal sediments act as a reservoir for both indicator and pathogenic organisms released into the coastal environment. This suggests an increased exposure risk if these organisms are resuspended back into the water column during recreational activity. A combined risk-based monitoring program would provide a more robust and reliable estimate of health risk associated with coastal recreational areas.

DECLARATION

I certify that this thesis does not incorporate without acknowledgement any material previously submitted for a degree or diploma at any university; and that to the best of my knowledge and belief it does not contain any material previously published or written by another person except where due reference is made in the text.

Duncan Craig

ACKNOWEDGEMENTS

This thesis would not have been possible without the help and assistance of the following people:

Nancy Cromar: For your support and guidance from Day 1 ($N_t = 0$) of my PhD adventure.

Howard Fallowfield: What can I say mate…..cruel but fair! I really appreciated your dry sense of humour, love of Coopers Pale Ale and your "pearls of wisdom" along the way.

Biomedical Engineering, Flinders Medical Centre: You blokes are legends, especially Laurie and Brian. Despite our whacky ideas, you always came up with (sometimes workable) solutions. I hope you both enjoy a well-earned retirement.

Dianne Davos, Salmonella Reference Laboratory, IMVS: For supplying the S. *derby* culture and general advice.

The Bureau of Meteorology, Adelaide: For providing meteorological data.

The Commonwealth Government, under the Public Health Education Research Program (Environmental Health Specialty Program) for funding the majority of this research.

And a special thanks to:

All the staff and students at the Department of Environmental Health, especially:

Alana Hanson: You must dread whenever there's a new post-grad student! Without doubt, you play a major role in the success of the Department. You were always there whenever help was needed. *John Edwards* (and to a lesser extent *Richard Bentham*…but I will explain that later), thank you for being there to bounce ideas off and providing general advice. Although coming late in the picture, *Richard*….you are a clown! You certainly made a lasting impression. And last, if not least, thank you to all the admin people, especially *Trish Amee*, for providing support throughout my time at Flinders.

Megge and *Michelle*: There is no doubt we had a great time at Flinders. I don't think Howard and Nancy knew what had hit them when we all joined forces in the lab!

I would especially like to thank all the volunteer staff at the *Flinders Medical Centre Coffee Shop*. Without opening my mouth, you had my mug of coffee and slice of carrot cake at the ready!

And of course, I would like to thank my family *Mum*, *Dad*, *Gav*, *Nath*, *Andrew* and *Leanne* (aka *Nell*) for "keepin' me real" throughout my PhD. The support you all gave has been the essence of my achievement. Thank you.

ABBREVIATIONS

AFR	Accidental faecal release
AFRI	Acute febrile respiratory illness
AGI	Acute gastrointestinal illness
ANOVA	Analysis of variance
APHA	American Public Health Association
CFU	Colony forming units
d	Days
DVC	Direct viable count
FISH	Fluorescent in-situ hybridisation
FITC	Fluorescein isothiocyanate
h	Hours
k	Decay rate
min	Minutes
MPN	Most probable number
N_0	Initial number of bacteria
N_t	Bacterial survival after time t
NHMRC	National Health and Medical Research Council
NLV	Norwalk-like virus
P	Significance compared to control
PBS	Phosphate buffered saline
PCR	Polymerase chain reaction
PI	Propidium iodide
QMRA	Quantitative microbial risk assessment
rev	Revolutions
SD	Standard deviation
s	Seconds
TDS	Total dissolved solids
UV	Ultra violet
VBNC	Viable but not culturable
WHO	World Health Organisation
WWTP	Wastewater treatment plant