

**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)

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**Department of Environmental Health,
Faculty of Health Sciences,
Flinders University**

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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

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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
<i>k</i>	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
<i>P</i>	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