

## Water and the environment

### Assessing ecosystem health through aquatic microbial diversity

**A completed Water Research Commission (WRC) study sought to use Next Generation Sequencing technology to characterise microbial communities in four Eastern Cape estuaries.**

#### Background

Estuaries are ecologically and economically important aquatic systems, functioning as feeding/stages sites for migratory birds, as nurseries for marine fish, and as repositories of high biodiversity. They are also important for the tourism industry and serve as sites for productive fish and invertebrate fisheries and aquaculture.

Over the past decade, there has been a dramatic increase in urban, agricultural and industrial development along the southern African coastline, particularly in the vicinity of estuaries, resulting in an escalation in anthropogenic stresses on these delicate ecosystems. While many studies have been carried out on the macro-fauna and macro-flora in South African estuaries, very few have been done to assess the microbial foodweb.

#### Potential of microbial diversity to detect ecosystem health

Microbes are extremely abundant and diverse in aquatic ecosystems and play critical roles in regulating key biogeochemical cycles such as the carbon, nitrogen and sulphur cycles. Furthermore, due to their small size and rapid proliferation rates, prokaryotes exhibit rapid response rates to changes in nutrient availability and physico-chemical changes in the environment, such as those that may be induced by pollution.

Characterisation of the microbial population within a target system would therefore provide an excellent assessment of ecosystem health and analysis of such data would flag the presence of potential microbial pathogens.

This WRC project sought to develop protocols for sample collection, template preparation for high throughput Next Generation Sequencing (NSG) and analysis of 16S rRNA sequences in estuarine water and sediment; to use NGS technology to characterise the microbial communities in four Eastern Cape estuarine systems; and to establish the links between the physico-chemical characteristics and the diversity and structure of microbial diversity within each estuary.

#### Methodology

Four permanently open Eastern Cape estuaries, with diverse hydrodynamics and anthropogenic impacts were selected for this study. The Kariega Estuary is a freshwater-deprived, marine-dominated system with a relatively pristine catchment area.

The Kowie Estuary is also marine-dominated, but in contrast with the Kariega, has extensive settlement along the banks and a small harbour at its mouth, with increased urban impacts due to influx of large volumes of sewage and sewerage into the system.

The freshwater dominated Sundays Estuary is impacted by commercial agriculture in its catchment area, and it is supplemented by the interbasin transfer scheme from the Gariep Dam, which increases the freshwater inflow into this estuary.

Finally, the Swartkops Estuary is also freshwater dominated and was selected because it is highly urbanised and severely impacted by anthropogenic pollution inflicted on this estuary by both urban and industrial activities.

In this study, the water column and the sediment were sampled at three sites along each of these estuaries. The physico-chemical properties of the water column at each of the sampling sites within the target estuaries were ascertained.

For characterisation of the microbial communities, a region within the eubacterial 16S rRNA gene was analysed using 454-pyrosequencing of amplicon libraries constructed from each of the samples. Approximately 179 000 sequence reads were analysed to determine the relative abundances of each of the phylotypes as well as identify those bacteria which occur in low abundances (i.e. rare).

## Results

The data revealed that the bacterial communities within South African estuaries differ significantly from estuaries reported in the literature. Principle component analysis showed a close relationship between the Kowie and the Kariega estuaries that likely reflects their strong marine influence.

The Sundays Estuary did not cluster with any of the other estuaries. With respect to the Swartkops Estuary, the upper reaches sampling site clustered more closely with the Sundays Estuary while the middle and lower reaches clustered more closely with the Kowie and Kariega estuaries.

The analysis revealed different microbial assemblages in the water column versus the sediment. There was a distinct spatial distribution of dominant taxa in the sediment along the length of the estuaries, even in estuaries with relatively small physico-chemical gradients from the mouth to the upper reaches.

All the estuaries were found to be net-heterotrophic while unicellular algae were responsible for the majority of the phototrophy within these ecosystems. The impact of sewage effluent, and potential subsequent nutrient spikes, into the Kowie Estuary was highlighted by the occurrence of a mono-specific incidence of the cyanobacteria *Synechococcus*, which is indicative of a bloom remnant or early bloom-formation.

The dominant phyla represented in the water columns of these estuaries included Bacteroidetes, Gammaproteobacteria and Betaproteobacteria, while the sediments had elevated levels of Deltaproteobacteria with reduced Betaproteobacterial populations. The species diversity found in the sediment was found to be several-fold higher than that in the water column. This is indicative of the more homogenous character of the water column compared to the more complex environment found within the sediment samples.

## Conclusions

This study has demonstrated the power of NGS technologies in the analysis of aquatic microbial communities. The results have shown that the diversity and structure of microbial communities reflect the physico-chemical characteristics of aquatic systems and provide important insight into the functioning of estuarine ecosystems.

### Further reading:

To order the report, *Aquatic microbial diversity: A sensitive and robust tool for assessing ecosystem health and functioning* (Report No. 2038/1/14) contact Publications at Tel: (012) 330-0340, Email: [orders@wrc.org.za](mailto:orders@wrc.org.za) or Visit: [www.wrc.org.za](http://www.wrc.org.za) to download a free copy.