



A multi-omics based ecological analysis of coastal marine sediments from Gladstone, in Australia's Central Queensland, and Heron Island, a nearby fringing platform reef



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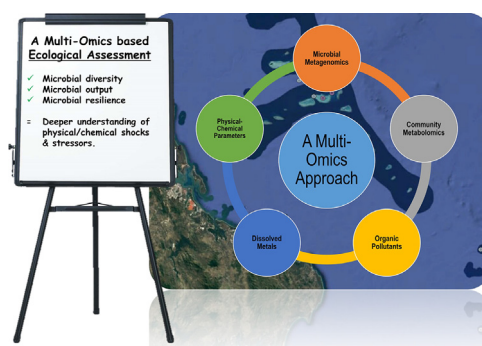
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HIGHLIGHTS

- Multi-omics provides insight into ecosystem health and anthropogenic impacts.
- Contaminants affect bacterial communities in terms of distribution, resilience and robustness.
- Multi-omics provides additional information that complements traditional monitoring data.

GRAPHICAL ABSTRACT



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ABSTRACT

The impact of anthropogenic factors arising from point and non-point pollution sources at a multi commodity marine port and its surrounding ecosystems were studied using sediment samples collected from a number of onshore (Gladstone Harbour and Facing Island) and offshore (Heron Island and Fitzroy Reefs) sites in Australia's Central Queensland. Sediment samples were analyzed for trace metals, organic carbon, polycyclic aromatic hydrocarbons (PAH), emerging chemicals of concern (ECC) and sterols. Similarly, the biological and biochemical interaction between the reef and its environment was analyzed by the multi-omic tools of next-generation sequencing characterization of the bacterial community and microbial community metabolic profiling. Overall, the trace elements were observed at the lower end of the Australian environmental guideline values at the offshore sites, while higher values were observed for the onshore locations. Nickel and copper were observed above the high trigger value threshold at the onshore sites. The levels of PAH were below limits of detection across all sites. However, some of the ECC and sterols were observed at higher concentrations at both onshore and offshore locations, notably, the cholesterol family sterols and 17 α -ethynylestradiol. Multi-omic analyses also indicated possible thermal and photo irradiation stressors on the bacterial communities at all the

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tested sites. The observed populations of γ -proteobacteria were found in combination with an increased pool of fatty acids that indicate fatty acid synthesis and utilisation of the intermediates of the shikimate pathways. This study demonstrates the value of applying a multi-omics approach for ecological assessments, in which a more detailed assessment of physical and chemical contaminants and their impact on the community bacterial biome is obtained.

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1. Introduction

More than 60% of the world's urban population is located within coastal zones, relying heavily on the surrounding estuarine and marine ecosystems to sustain quality of life through the provision of food and recreational activities (*i.e.*, fishing and tourism) (Delhez and Barth, 2011). However, population densification has resulted in negative impacts and degradation to the surrounding ecosystem(s) that these communities rely upon (Barbier et al., 2011; Wilkinson and Salvat, 2012). Compounding the issue further, coastal ports and harbours with high levels of shipping traffic, and urban and industrial activities are considered to be particularly problematic (Sakellariadou, 2015). These marine basins sites, through topography, bathymetry and/or construction, are characterized as closed or semi-closed systems with limited water circulation. Ultimately, this leads to pollution hotspots or areas of stagnation, where chemical pollution is high and often extends beyond the contaminated port or harbour to nearby and surrounding estuaries, seabeds, and reefs (Sakellariadou, 2015).

Port Curtis (located in Gladstone Harbour, Queensland, Australia) has been extensively studied and is of importance to communities in Central Queensland (Hutchings et al., 2005; Jones et al., 2005; Melville et al., 2009; Taylor and Rasheed, 2011). Port Curtis is one of Australia's leading commercial ports used extensively for exporting coal and other commodities, and is adjacent to the Great Barrier Reef (GBR) Marine Park, which is of particular global interest to environmentalists, politicians, and tourism industries. As a result of coastal urbanization and agricultural activities at Port Curtis and Gladstone, the GBR is being exposed to increasingly more nutrient, sediments, and land-based chemical and microbial contaminants. As is the case with the majority of the studies undertaken at Port Curtis (Angel et al., 2012; DERM, 2011), the limitation of monitoring approaches is that they are focused on a finite number of analytes (physical and/or chemical) collected from specific sites and specified locations. Such monitoring programs measure only environmental proxies, with limited information regarding the human impacts and the extent of land-based pollution, such as dredging or runoff, on the overall health and resilience of the aquatic environment in the harbour or the nearby estuaries and adjacent reefs. As a consequence, policy decisions are often based on limited data which may hinder policy development and appropriate management strategies for proper mitigation. Monitoring programs should be a component of a larger process of either conservation-oriented science or management, where monitoring is continual and undertaken with sufficient detail in order to provide the evidence to support policy (Nichols and Williams, 2006). In addition, monitoring and assessment programs should employ indicators that are sensitive to anthropogenic change against natural variability of the system studied (Hallett et al., 2016).

One approach to overcome current monitoring shortages is through the application of multi-omics-based approaches (*i.e.*, metagenomics, transcriptomics, proteomics, and metabolomics) in conjunction with the collection of standard physical and chemical data. Such an approach enables a system-wide assessment of site-specific microbial communities, their metabolic output and their underlying physicochemical disturbances in order to assess the health and resilience of the coastal marine ecosystems (Desai et al., 2010). This approach requires a deeper analysis of each site using high-throughput next-generation techniques that facilitates a data-driven analysis for

analyzing, visualizing, and evaluating complex metabolic dynamics and networks in sediment microbial ecosystems (Yoshida et al., 2014).

The advancement of omics-based techniques and their integration (multi-omics) have contributed towards advancements in the fields of environmental and molecular biology, thereby pushing the boundaries of our understanding of microbial physiology (Thakur et al., 2008). To date, such studies have focused on specific pollution events (*e.g.*, the Deepwater Horizon oil spill (Kimes et al., 2013), survey of urban waterways (Beale et al., 2017)), the assessment of biotechnology/bioremediation (*e.g.*, bioremediation of steroids in the environment (Yang et al., 2016)), or have been used to characterize well-controlled engineered systems (*e.g.*, anaerobic bioreactors (Beale et al., 2016)). To the best of our knowledge, such a tandem approach has not been used to characterize a system as part of a sediment monitoring survey. However, the application of metagenomics or metabolomics in isolation has been applied within this context with some success (Beale et al., 2013a; Gomez-Alvarez et al., 2012; Tan et al., 2015; Van Rossum et al., 2015).

The aim of this study was to use multi-omics-based approaches in order to investigate the ecological impact arising from anthropogenic activities within the Port Curtis and Gladstone Harbour marine basin in addition to the immediate ecosystems outside the harbour and the nearby fringing platform reefs near Heron Island (approximately 80 km away). Using multivariate statistics, bacterial metagenomics and microbial community metabolomics datasets were merged with additional physico-chemical data, thus providing a multi-omics-based approach to investigate marine sediments.

2. Materials and methods

2.1. Sediment sampling sites

Marine sediment samples were collected from four different ecosystems surrounding Port Curtis in May 2015. Fig. 1 shows a map of the Gladstone Harbour/Heron Island region overlaid with the Queensland land use (primary classes) data, with the sediment sampling sites annotated with 'yellow' stars and numbered. Two samples (1 and 2) were collected from inside of a coral reef lagoon, adjacent to the fringing platform reef (Fitzroy Reef), where the dominant influence is presumably from material trapped within the lagoon. The reef comprises significant biodiversity, supporting around 900 of the 1500 fish species and 72% of the coral species found on the GBR (DERM, 2011). Two sediment samples (3 and 4) were collected from just outside of a fringing reef (Heron Reef) surrounding Heron Island. This area is influenced by material from several sources, including terrestrial runoff, export from the fringing reef and lagoon, upwelling from the Coral Sea, and tourist activity. Two sediment samples (5 and 6) were collected from sites within the immediate Port Curtis/Gladstone Harbour region in the channel between Curtis Island and Facing Island, where the biochemistry is influenced by harbour seagrasses and export from the southern channel. Lastly, an additional three sediment samples (7, 8 and 9) were collected from sites on the north side of Facing Island (immediately outside of Gladstone Harbour), which is predominantly influenced by dredge spoil and export activities. It is important to note. Heron Island and Fitzroy reef sediments are predominantly carbonate-based, whereas those in Gladstone harbour will have a greater terrigenous composition.

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