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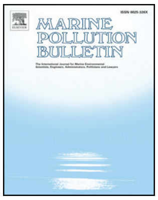

Wastewater nitrogen and trace metal uptake by biota on a high-energy rocky shore detected using stable isotopes

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ABSTRACT

On high-energy rocky shores receiving treated wastewater, impacts are difficult to distinguish against a highly variable background and are localised due to rapid dilution. We demonstrate that nitrogen stable isotope values ($\delta^{15}N$) of rocky shore biota are highly sensitive to wastewater inputs. For macroalgae (*Ulva lactuca* and *Endarachne binghamiae*), grazing snails (*Bembicium nanum* and *Nerita atramentosa*), and predatory snails (*Morula marginalba*), $\delta^{15}N$ was enriched near a wastewater outfall and declined with distance, returning to background levels within 290 m. Any of these species therefore indicates the extent of influence of wastewater, allowing identification of an appropriate scale for studies of ecosystem impacts. For *M. marginalba*, significant regressions between $\delta^{15}N$ and tissue copper, manganese, and zinc concentrations indicate a possible wastewater source for these metals. This suggests that $\delta^{15}N$ is a proxy for exposure to wastewater contaminants, and may help to attribute variations in rocky shore communities to wastewater impacts.

1. Introduction

Sewage treatment plants are a major source of anthropogenic input to coastal environments (Costanzo et al., 2001). Depending on the level of treatment, the wastewater released from sewage treatment plants may contain a variety of contaminants, including nutrients, particulate organic matter, and trace metals (Costanzo et al., 2001; Cabral-Oliveira et al., 2015). This has led to concern regarding the impact of treated wastewater on receiving environments.

Wastewater is often discharged to estuaries and coastal environments and a number of studies have assessed its impact on marine communities. These have shown that the effect of wastewater can be species-specific. Whereas more tolerant species may demonstrate increased productivity (Cabral-Oliveira et al., 2014) and recruitment (Bellgrove et al., 1997), the health of more sensitive species and/or individuals may be adversely impacted (e.g., Schlacher et al., 2007), causing them to become less dominant (López-Gappa et al., 1990). These changes can be reflected in altered population structure (Hindell and Quinn, 2000) and diversity (Fairweather, 1990; López-Gappa et al., 1990), and the species composition and biomass of communities in the receiving environment (Littler and Murray, 1975; López-Gappa et al., 1990; Bellgrove et al., 1997).

The impacts of wastewater on marine communities are more difficult to detect where treated wastewater is discharged to high-energy coastal environments such as rocky shores. Due to rapid dilution and dissipation of wastewater in these environments, impacts can be highly localised (e.g., Fairweather, 1990), and/or difficult to detect against a background of high environmental and community variability (Smith, 1994; O'Connor, 2013). Sampling at an inappropriate scale can lead to an inability to identify impacts or incorrect assessment of impacts (Bishop et al., 2002). To counteract these limitations, it has been suggested that studies aiming to determine wastewater impacts on rocky shores should sample at multiple scales (Bishop et al., 2002), and/or include multiple control sites (Fairweather, 1990; Hindell and Quinn, 2000; Cabral-Oliveira et al., 2015). However, where the ability to adopt these recommendations is limited by the additional effort and cost involved and/or availability of suitable controls, it would be valuable to ascertain, a priori, the extent of influence of the wastewater plume, and the likely exposure of rocky shore communities to treated wastewater and its contaminants.

A relatively inexpensive, simple method to detect the distribution of anthropogenic inputs within aquatic environments is the analysis of stable isotopes (Oakes et al., 2010). In the case of wastewater, preferential removal of ^{14}N via ammonia volatilisation during secondary and tertiary treatment leads to wastewater-derived nitrogen typically having a nitrogen isotope signature ($\delta^{15}N$) that is relatively enriched in the rare, heavy isotope, ^{15}N (Heaton, 1986; Costanzo et al., 2001). This is reflected in enriched $\delta^{15}N$ values for nutrients within discharged wastewater, which is readily assimilated by primary producers (Costanzo et al., 2001; Cole et al., 2004). Consumers that feed upon these primary producers also acquire an enriched $\delta^{15}N$ 'wastewater signal', and this is then passed to higher consumers via trophic transfer (e.g., Connolly et al., 2013). Fractionation usually results in further

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enrichment of δ^{15}N values with each trophic transfer (Minagawa and Wada, 1984; McCutchan et al., 2003). Depending on tissue turnover time, analysis of δ^{15}N values of flora and fauna gives a temporally-integrated indication of wastewater nitrogen inputs, compared to analysis of δ^{15}N within water column nutrients, which can vary rapidly with changes in discharge rate and environmental conditions (Hood et al., 2014). Stable nitrogen isotope analysis has previously been used to investigate wastewater nitrogen uptake by biota in a variety of aquatic environments but, to our knowledge, has not previously been applied in a high-energy, rocky shore environment receiving wastewater input.

In the current study, we aimed to determine (1) if δ^{15}N values could be used to detect wastewater input in a high-energy rocky shore environment (sensitivity), and (2) how well δ^{15}N values of species across multiple trophic levels reflected wastewater input (suitability of species as indicators). Based on this, we further aimed to determine (3) the extent of influence of wastewater input to a rocky shore environment, and (4) if variations in wastewater exposure (based on δ^{15}N values) explained variations in trace metal concentrations within biota (i.e., if stable isotopes are a proxy for the spread of other contaminants).

2. Materials and methods

2.1. Study site

The study was done in mid-2009 in northern New South Wales, Australia. Impacted sites were located in the intertidal zone approximately 10 m, 40 m, 60 m, 100 m, 150 m, and 290 m north of an outfall pipe which discharges secondary-treated effluent from the Lennox Head Wastewater Treatment Plant just below low tide level at the base of a cliff at Skennars Head (28°49′28″S, 153°36′25″E; Fig. 1). Longshore drift is north. A generally flat basaltic intertidal platform approximately 1700 m south of the outfall (Flat Rock) was selected as the non-impacted control site (Fig. 1). The study sites are exposed to the open ocean and are subject to high wave energy, depending on tidal height.

At the time of the study, treated wastewater from the Lennox Head Wastewater Treatment Plant had average concentrations of ammonium, nitrate, total nitrogen, and total phosphorous of 35.7, 149.9, 285.6 and 164.6 μmol L^{-1}, respectively (ranges of 0.7–464.1, 6.4–359.8, 128.5–778.2, and 51.6–298.6 μmol L^{-1}). Data regarding metal concentrations in treated wastewater are not available for the time when the study was done, or for all metals. However, over 12 months from September 2007 to 2008 average (± standard error) concentrations for some metals dissolved in the treated wastewater were as follows: cadmium $<0.001 \pm <0.001$ mg L^{-1}, chromium $<0.01 \pm <0.001$ mg L^{-1}, copper $0.02 \pm <0.01$ mg L^{-1}, iron 0.10 ± 0.02 mg L^{-1}, lead $<0.01 \pm 0.00$ mg L^{-1}, zinc $0.01 \pm <0.01$ mg L^{-1}, boron 0.15 ± 0.02 mg L^{-1}, copper 0.02 ± 0.01 mg L^{-1}, calcium 42.6 ± 1.5 mg L^{-1}, magnesium 25.2 ± 1.2 mg L^{-1}, potassium 19.9 ± 0.8 mg L^{-1}, sodium 192.1 ± 10.0 mg L^{-1}, and sulphate 84.5 ± 4.4 mg L^{-1}. Some treated wastewater from the treatment plant is re-used locally, with the volume re-used determined by weather conditions (more during dry periods) and the surplus discharged to the ocean at a rate of up to 100 L s^{-1}. Daily flow from the outfall varies with weather conditions. For example, under similar operating conditions over 6 months from December 2007 to May 2008, the daily flow ranged from 4.4 to 1036.6 ML d^{-1} (average 45.4 ± 12.7 ML d^{-1}).

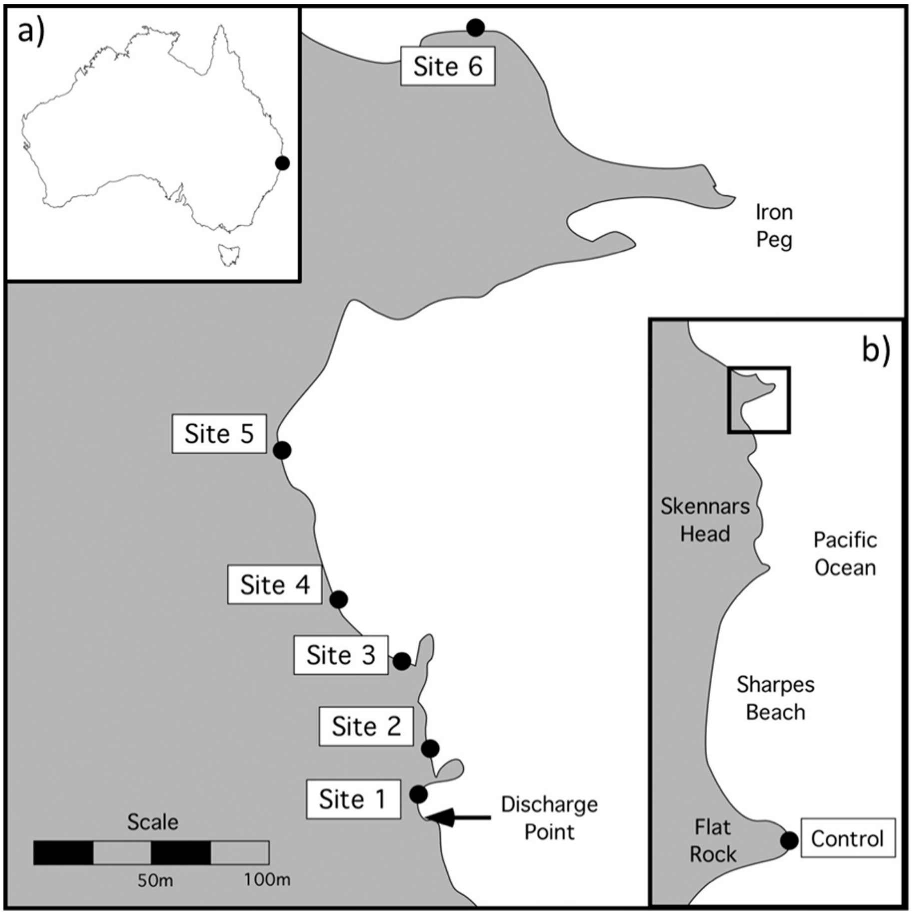

Fig. 1. Sampling locations at Skennars Head. Inset shows control site at Flat Rock and area encompassed by main map (defined by square). Approximate location of the wastewater outfall is indicated by an arrow.

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