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Different resource limitation by carbon, nitrogen and phosphorus between base flow and high flow conditions for estuarine bacteria and phytoplankton

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ABSTRACT

Freshwater inflows can deliver substantial inputs of allochthonous organic carbon to estuaries. The role that allochthonous DOC has on structuring bacterial and phytoplankton communities is still not well understood. We performed a series of 1.25 L bioassay limitation experiments on the Bega and Clyde River estuaries in NSW, Australia, examining what resources limit bacteria and phytoplankton growth. We hypothesized that during base flow conditions bacteria would be carbon limited, and after high flow conditions they would be nutrient limited. A full factorial design was used with additions of carbon (glucose), nitrogen (KNO_3) and phosphorus (KH_2PO_4). During the experiments that took place during base flow conditions bacteria were always primarily C-limited. After high flow conditions, bacteria were P-limited on the Clyde River, and remained C-limited on the Bega River. Phytoplankton growth was limited at all times in each estuary, tending toward N-limitation on the Bega River and P-limitation on the Clyde river. During high flow conditions on the Clyde River, when bacteria and phytoplankton were both primarily P-limited, it appeared that bacteria was able to outcompete phytoplankton for nutrients. These results suggest that freshwater inflows and allochthonous DOC maybe important in structuring estuarine microbial ecosystems and individual estuaries may behave differently in terms of their limiting resources.

1. Introduction

Understanding the ways in which essential substrates such DOC and nutrients regulate bacterial production is crucial for understanding how estuarine systems function. In marine systems, bacterial productivity is closely coupled with primary productivity as bacterial populations are primarily reliant on autochthonous carbon (Cole et al., 1988; Tranvik, 1992). In estuarine and coastal environments this relationship has been shown to be less consistent due to the subsidy of allochthonous carbon (Almeida et al., 2005; Ameryk et al., 2005).

The delivery of allochthonous DOC to estuaries can be conceptualized as two compartments, base flow and high flow (Hinton et al., 1997; Buffam et al., 2001). During base flow conditions water generally travels through lower mineral dominated soils collecting more refractory carbon (Ivarsson and Jansson, 1994; Frank et al., 2000). High flows, often occurring during periods of precipitation, differ with flow paths through the litter and upper soil horizons (Aitkenhead-Peterson et al., 2003) with some evidence that this allochthonous carbon is more labile than base flow materials (Kaplan and Newbold, 1995; Wikner et al., 1999). Further, high flows have been found to deliver greater total annual loads of DOC than that of base flows to estuaries (Wiegner et al., 2009; Raymond and Saiers, 2010).

Resource bioavailability and stoichiometry are important for understanding resource limitation (Thingstad and Lignell, 1997; Jansson et al., 2006; Kankaala et al., 2010). Estuarine bacterial growth has been shown to be limited by carbon (Hoikkala et al., 2009; Hitchcock et al., 2010), nutrients (Cotner et al., 2000), co-limited by a combination of carbon and nutrients (Pinhassi et al., 2006), and limitation may vary both spatially and temporally (Sala et al., 2002; Pinhassi et al., 2006). The 'bacteria-phytoplankton paradox' suggests that because bacteria rely on phytoplankton for DOC they are in-essence controlled by primary production (Bratbak and Thingstad, 1985; Mindl et al., 2005). However, in the presence of excess allochthonous DOC, this reliance may be removed or

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diminished and in doing so change the relationship between phytoplankton and bacteria from one of commensalism to competition. In situations where bacteria and phytoplankton are competing for the same limiting nutrient, bacteria have been shown to be able to outcompete phytoplankton due to the greater surface area to volume ratio and the high affinity of bacteria for P, potentially suppressing primary production (Currie and Kalff, 1984; Jansson, 1993; Thingstad et al., 1993).

Enrichment bioassays are commonly used to investigate factors limiting phytoplankton (Hecky and Kilham, 1988) and bacterial (Kuparinen and Heinänen, 1993; Zweifel et al., 1993; Pomeroy et al., 1995) growth in relation to substrate availability (Smith and Kemp, 2003). Whilst a number of studies examining the differences between base and high flow have taken place in freshwater systems (Leff and Meyer, 1991; Buffam et al., 2001; Wiegner et al., 2009), few have been conducted in estuaries (Wikner et al., 1999).

In this study, we performed bioassay limitation experiments in two south-eastern Australian estuaries with the aim of determining which key resources limit bacterial and phytoplankton production and how limitation may change following periods of high flow. To our knowledge this is the first bioassay limitation study that explicitly investigates changes in estuarine resource limitation between base and high flow conditions. With freshwater inflows to estuaries greatly reduced around the world and changing patterns of precipitation due to climate change, understanding this key question is of particular importance.

2. Materials and methods

2.1. Study sites

The Bega River estuary, NSW, Australia, (−36° 42′ 43.64″, +149° 54′ 8.76″) is a wave dominated riverine estuary with an open/intermittently closed barrier (Fig. 1). It drains a catchment 1941 km^2. The catchment is split into two sub-catchments, the Brogo River, regulated by Brogo Dam and Bega River which has two smaller dams, Cochrane and Candello. The upper sections of the catchment consist of mainly forested areas, whilst the middle catchment is mostly cleared for agriculture, dominated by the dairy industry. The freshwater tidal section has been mostly cleared for agriculture and irrigated farmland, whilst the catchment surrounding the estuary itself is largely unmodified. Broad scale land clearing since colonization has drastically altered the river and estuary via the transportation of sandy top soils into the river channel (Brooks and Brierley, 1997). The Clyde River estuary, NSW, Australia, (−35° 33′ 11.44″, +150° 11′ 12.11″) is a tidally dominated drowned river valley estuary, with its middle and upper estuary riverine dominated (Fig. 1). It drains a catchment 1791 km^2 and is unregulated, with no significant structures or water extraction. The catchment is almost entirely forested, with some forestry activities in the upper catchment. The lower sections of the estuary have a small urban settlement as well as oyster and other fisheries activities.

2.2. Experimental design

In each estuary a site was chosen at the lowest end of the freshwater tidal region where all experiments were conducted. Sites were chosen in the freshwater tidal zone as this is the first region that processes allochthonous material in the estuary (Muylaert et al., 2005). Four bioassay enrichment experiments were conducted in each estuary between November and January in both 2010 and 2011 under various discharge conditions (Fig. 2). This period was chosen as it is the time of maximum extraction within the Bega River estuary, and thus the period when the results are most pertinent to water management in the catchment. Experiments took place using 1.25 L clear PET bottles, containing estuary water collected from a depth of approximately 0.2–0.5 m which

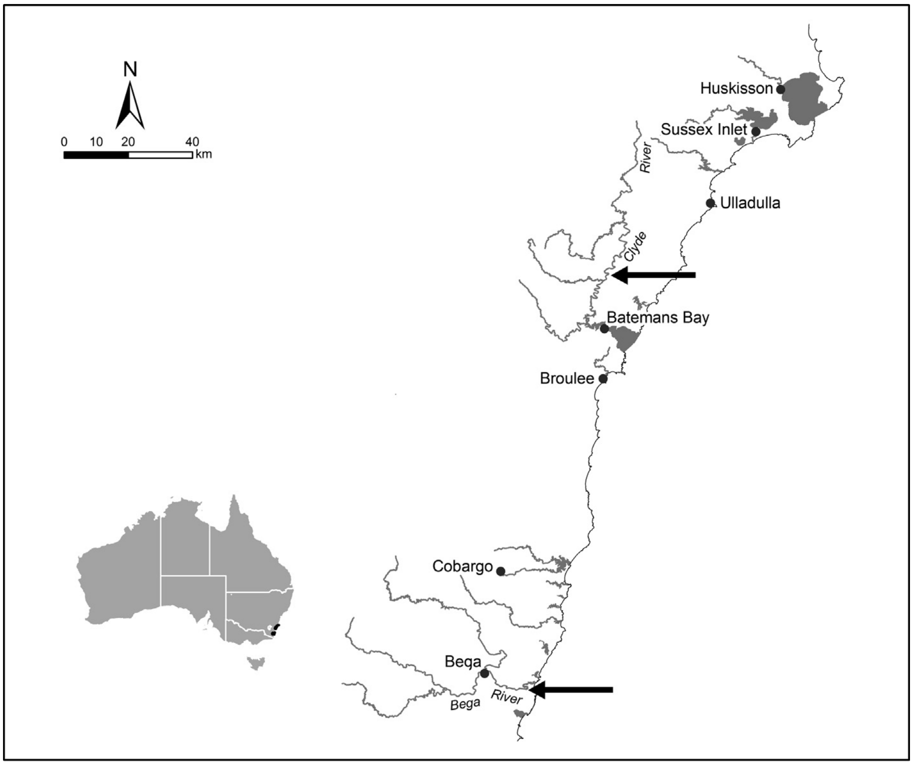

Fig. 1. Map of the Bega and Clyde River catchments, NSW, Australia. Arrows indicate location of experimental sites.