2012

Anthropogenic effects on benthic metabolism and nutrient cycling in Currambene Creek Sanctuary Zone, Jervis Bay Marine Park, NSW

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A thesis submitted in partial fulfilment of the requirements of the Degree of Master of Science Research at the Wollongong University

October 2012

Declaration of Originality

This thesis (Anthropogenic effects on benthic metabolism and nutrient cycling in Currambene Creek sanctuary zone, Jervis Bay Marine Park, NSW, by Kristen A Lee 2011) is my original work and has not been submitted, in whole or in part for a degree at any National or International University. Under no exception does it contain any material published or written by any other person that has not been acknowledged in the text.

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Abstract

Currambene Creek sanctuary zone is located within Jervis Bay Marine Park, New South Wales (NSW). In NSW, sanctuary zones are established with the aim of conserving biodiversity and maintaining ecological processes. While these aims are well defined by the Marine Parks Act 1997, no study yet has been undertaken to establish whether sanctuary zones do maintain ecological processes. Ecological processes that include benthic metabolism and nutrient cycling, control fundamental sediment–water interactions. The aim of this study was to investigate the affect sanctuary zone protection, compared to an experimental disturbance, had on the benthic metabolism and nutrients in a tidal mudflat to provide benchmarks to guide the management of NSW estuaries and sanctuary zones.

Benthic flux measurements of oxygen (O_2), ammonium (NH_4^+), nitrite and nitrate (NO_x), dissolved inorganic nitrogen (DIN) and phosphorous (PO_4^{3-}) were estimated using the sediment core incubation technique. Two sites were exposed to three different experimental disturbances subjected to both dark and light conditions. The three disturbances were control (undisturbed), trample (trampled sediment) and trample bait-pumping (trampled sediment and bait-pumping of the common ghost shrimp, *Biffarius arenosus*). These disturbances are typical human activities that would take place on an intertidal mudflat. Additionally, chlorophyll *a* was also examined.

Benthic metabolism displayed autotrophic characteristics as gross primary production (GPP) exceeded respiration. Benthic community respiration (BCR) was significantly

different among treatments with both trample and trample pump disturbances influencing respiration. The presence or absence of macrofauna such as *B. arenosus* and other taxa was shown to partly cause the differences found in BCR. The presence of macrofauna in samples from the control treatment consumed more O_2, while in the disturbed treatments there was a reduced number of macrofauna, therefore accounting for the reduced consumption of O_2.

In general, nutrient fluxes did not typically display diel variations, with the sediments being a net sink for all nutrients. Dark and light differences were attributed to the dominating photosynthetic activities of benthic microalgae (BMA). Once again, the presence or absence of macrofauna influenced the difference between NH_4^+ in the disturbed and undisturbed treatments. The metabolic excretion of NH_4^+ from the macrofauna had the biggest measurable influence on uptake in the disturbed treatments. However, BMA dominated the sediments and had a significant influence over all nutrients.

Benthic metabolism and nutrient fluxes in Currambene Creek were dominated by BMA and the experimental disturbances had a limited affect because of their activities. Nutrient budgets showed that the sediment could not meet the demands of the BMA mostly because the BMA sequestered nutrients from the water column. While the experimental disturbances had a significant influence on macrofaunal abundance (data from a parallel study), that consequently had an influence on respiration and some nutrients, it was difficult to determine whether ecological processes were maintained in the MPA.

This study demonstrated that trampling and bait pumping impact upon the Currambene Creek sanctuary zone benthic metabolism, nutrient cycling, respiration rates and some nutrients. Whereas, production and other nutrients (e.g. DIN) were not affected by the disturbances. A dramatic change to these ecological processes would have seen the disturbances subsequently change the net autotrophic characteristics of the sediments into net heterotrophic sediment, however this did not happen.

Changes in benthic metabolism and nutrient cycling due to the experimental disturbances suggest that in the Currambene Creek sanctuary zone BMA influence ecological processes more than benthic macrofauna. However, the macrofauna still had a significant influence of respiration and NH_4^+. The nutrient fluxes of Currambene Creek provide essential nutrient benchmarks for the management of NSW estuaries from which future changes or disturbances can be assessed.

Acknowledgements

Dr. Katarina Mikac (primary supervisor): You are no long my supervisor, you are my friend. Your dedication to your work and students goes way beyond anyone I have come across. I am forever grateful for all the time, effort and commitment you gave to this thesis. Jervis Bay will always be filled with memories of Husky bakery pig outs, fresh made gnocchi and pavlova, you cooking seafood and holding a beer in hand while I'm about to lose it because the incubator won't work and you jamming to crazy music in the car. Thank you for being there at all hours of the night and day, for listening to me ask stupid questions and all your hard work leading up to the final weeks. Thank you for making this happen!

Jaimie Potts (secondary supervisor): Thank you for all your expertise and guidance. I know "I am the worst student you have had for sending drafts and asking questions", but your enthusiasm and expertise inspired me to figure things out on my own, that way I was sure to finally understand what you were on about. Thank you for lending me your contraptions (incubators), that almost turned me into a mad scientist like yourself. I am grateful for the opportunity to have participated in volunteer work with you and the boys up the coast, I hope there are more fishing trips ahead. Your relaxed nature always calmed me down and gave me the confidence that I so needed.

Tye Hunt: Without you this would not have been possible. You were so encouraging and helpful, in particular during the incubation set up. Also, to the delicious food you cooked and the times we spent eating at Husky bakery. A special thank you to all your buff friends for helping with core collections.

Aidan Johnson: Thank you for your countless input, especially reading my drafts.

Dr. Nathan Knott: Thank you for your expertise in experimental design and statistics. Your supportiveness and direction throughout this research is greatly appreciated. A special thank you for making your intern come out into the field to brighten up our day and to all the guys at JBMP for allowing us to borrow equipment.

DECCW: To all the guys in the lab that helped me find my way. I appreciate all the time and patience you gave me, especially when Jaimie decided he wasn't coming into today.

Recovery Works (Carl Jacobs): Carl, you worked magic on my back and neck every week during my writing. Cramped over my computer, living in a 'cave' for days on end, you always found the time to treat and fix my pains. Without you I would be a hunched over mess probably unable to walk. Thank you!

GeoGAS: To everyone in the team, I greatly appreciate the flexibility you gave me when I would call up and say I couldn't come into day because I was "doing uni work". Seriously, knowing that uni work came first was a great relief and I wouldn't have been able to complete my research without all your positive and helpful attitudes.

Stingrays: To my disappearing acts to my anti-social behaviour, I appreciate you all putting up with me throughout this difficult year. I have been locked away in this 'cave' for way to long. At training you allowed me to vent my frustrations and release the energy I built up from sitting at my computer for days on end. But most of all I

would just like to say thank you for just being there every Wednesday, Friday and Sunday. You all distracted me just enough from uni work to allow me to continue on.

Ammy Lewis: From the random messages to phone calls "saying I can't do this", you have always supported me and never given up on me. I am truly thankfully for all the motivation you gave me. You listened to my endless complaining and ideas of how I could put off doing work. While, yes you did distract me from a lot you are one of the reason why I achieved as much as I did. You pushed me and made me get started, despite my hardest attempts to once again put it off. For all the drafts you read over and the edits you made, I cannot thank you enough. You, Linda, Bindee and Mimi always brought a smile to my face even if I didn't show it. I am forever grateful.

This thesis is dedicated to:

My Family: Yes it has been six years of university and you have all been there for every battle, including this thesis. For all your support and belief in me, I am forever grateful. Thank you for shrugging off my constant complaining, bad moods and strange behaviour. You made living in my 'cave' just bearable where I was able to go on. In particular Jacina, you are my best friend. You bring a calm over me whenever you are near and a feeling of comfort which lets me know everything will be alright. I appreciate you refraining from distracting me as much as you could. But for the numerous dance parties, I always welcomed those. They made me laugh and gave me the energy to keep writing. A big thank you for all the help you gave me editing and referencing as well. I am blessed to have you in my life. Jacob, for your help with Illustrator and Photoshop. I know you had better things to do with your time but you

always made sure I could use your computer and that if I needed help you were there. Pickles, despite that I think you go outside way too much too pee (I know you really just go outside to eat random things and run on the road), I think I needed those trips outside to get fresh air and a break from writing. You always sat beside me, never doubting me and giving me that look I needed to know that you cared.

Ashleigh Connor: Your thank you comes last because I struggle to find the words I need to say. I sat here for over a year writing away about a topic I knew nothing about and I still find it easier than finding the courage to accept that this thankyou comes from a heartbroken place. I thank you for giving me the courage to finish this thesis. You never strayed away from talking to me, despite my sometimes-awkward silent nature. Your encouraging spirit was such a comfort. From one freckle to another, I will always be proud that when I wrap a cheeseburger I can say that you were my teacher and that you are my friend.

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Abbreviations

ANOVA	Analysis of Variance
BCR	Benthic community respiration
BMA	Benthic microalgae
BTSI	Benthic Trophic State Index
C	Carbon
Chl –*a*	Chlorophyll *a*
DECCW	Department of Environment, Climate Change, and Water
DIN	Dissolved Inorganic Nitrogen
hr	Hour
GPP	Gross Primary Production
MPAs	Marine Protected Areas
N	Nitrogen
NEM	Net Ecosystem Metabolism
NH_4^+	Ammonium
NO_2^-	Nitrite
NO_3^-	Nitrate
NO_x	Nitrite and Nitrate
NPP	Net Primary Productivity
ns	no significance
O	Oxygen
OM	Organic matter
P	Phosphorus
P-I	Photosynthetic-Irradiance

PO_4^{3-}	Phosphorus
RM-ANOVA	Repeated Measure Analysis of Variance
SE	Standard Errors
SWI	Sediment-water interface
T_0	Initial time
T_1	Time one
TN	Total Nitrogen
TP	Total Phosphorus

Chapter 1

Introduction

Investigating benthic nutrient cycling in protected estuarine soft-sediments

Preamble

The following review discusses benthic nutrient cycling in protected temperate intertidal mudflats and is divided into three main sections: 1) marine protected areas (MPAS); 2) ecosystem processes; and 3) methods for determining benthic processes in estuarine soft-sediments. Section one outlines the importance of MPAs, their definitions and the natural disturbances they face in protected areas. In section two nitrogen (N) and phosphorus (P), the key nutrient that aid ecosystem processes, are discussed with regards to estuarine soft-sediments. The third and final section summarises the main methods (e.g. benthic chambers) used for determining and measuring benthic processes.

1.1 Marine Protected Areas (MPAs)

MPAs definition and types

Many terms and definitions have been used to describe MPAs (Nicholls 1998). However, in Australia MPAs are defined by the Australian Government Department of Sustainability, Environment, Water, Population and Communities' definition (which has been adapted from the IUCN definition) as *"an area of land and/or sea especially dedication to protection and maintenance of biological diversity and of natural and associated cultural resources, and managed through legal or other*

effective means" (Department of Sustainability, Environment, Water, Population and Communities 2010).

There are many different types of MPAs and they range from small no take zones (sanctuary zones) which are heavily protected, to large multifarious regions that allow various recreational/commercial activities, and offer different levels of protection (Woodley *et al.* 2010). For instance, the Solitary Island Marine Park (New South Wales, Australia) and the Jervis Bay Marine Park (NSW) are multi-zoned parks, separated into four main zones: 1) sanctuary; 2) habitat protection; 3) general use; and 4) special purpose. Sanctuary zones offer the highest level of protection in a marine park and prohibit all forms of fishing and collection, but permit activities that do not harm the flora, fauna and their habitats. Habitat protection zones are implemented to protect habitats, reduce harmful activities (fishing, bait collecting etc) and allow restricted recreational and commercial fishing. The general use zones offer the lowest form of protection and allow a wide range of activities, including recreational and commercial fishing. Finally, the special purpose zones cater for existing infrastructure, such as public wharves and jetties that require special management (Marine Park Authority 2011a,b). Despite a range in terminology the majority of MPAs all aim to conserve biodiversity, cultural and historical features, provide tourism, education, recreation and research for present and future generations (Roberts *et al.* 2003).

Marine Protected Areas: Intertidal soft-sediments

Why protect intertidal soft-sediments?

Chemical, biological and physical processes, such as nutrient cycling, predation and tidal changes, can influence the ecology of intertidal estuarine soft-sediments. These habitats are among the most productive ecosystems in the world, and represent important transitional zones, where fresh and marine ecosystems interact through various ecological processes (Wall *et al.* 2001). Many such habitats have been recognised as ecosystems that warrant conservation by their zoning as MPAs (e.g. Currambene Creek MPA JBMP). However, the establishment of MPAs is biased towards conserving habitats that support numerous vertebrate communities and other more noticeable ecosystems (e.g. coral reefs). For example, Winberg *et al.* (2008) found from 682 peer-reviewed papers on MPAs, 174 focused on coral reefs, 27 on rocky reefs and only four on the macrofauna (invertebrates) of soft sediments such as mudflats. Of these studies, 283 were carried out on fish, 86 on whole assemblages, and only 30 studies focused on invertebrates, mostly of commercial importance (e.g. crayfish, abalone, etc.). Few studies in Australia and internationally, apart from Winberg *et al.* (2008), have specifically focused on marine protected intertidal estuarine mudflats, including investigating their ecological processes and associated macrofaunal regardless of their commercial value.

Estuarine soft-sediments and tidal mudflats are constantly exposed to a diverse range of anthropogenic disturbances, such as runoff from residential and industrial development, recreational activities and tourism. Marine environments, especially MPAs draw large numbers of tourists, who engage in recreational activities such as scuba diving, swimming, snorkelling and bait collecting for recreational fishing

(Rossi *et al.* 2007). These activities exert pressure on soft-sediment ecosystems in a range of negative ways and the most widely studied of these activities is bait collecting or bait pumping (Wynberg and Branch 1991, 1994; Contessa and Bird 2004; Winberg *et al.* 2007; Winberg 2008; Hunt 2011) and the associated trampling of the sediments that occurs (Peterson 1977; Wynberg and Branch 1997; Dernie *et al.* 2003; Rossi *et al.* 2007; Hunt 2011). Research has shown that disturbance from bait pumping and trampling result in a decrease in macrofaunal biodiversity (Contessa and Bird 2004; Dunn 2004; Rossi *et al.* 2007), which consequently shifts the dominance of particular species in these communities and thus alters the patterns of ecological processes (Underwood and Peterson 1988). Ecological processes that occur in soft-sediment and mudflats are rarely defined in a single study, but the synthesis of numerous research has lead to an understanding that it is a processes that plays an essential role in maintaining ecosystem integrity through the cycling of water, the nutrients (nitrogen and phosphorus), the flow of energy and biological diversity (Gürel *et al.* 2005).

Anthropogenic disturbances

Bait pumping is a popular activity by recreational fishers that involves the collection of macrofauna for bait and burley, either using hand operated suction pumps (bait-pumps) or by physically digging and churning sediments. The harvesting of bait, by bait pumping and its associated trampling, not only acts to remove macrofaunal indiscriminately, thereby reducing overall assemblage numbers, but also greatly affects habitats by altering their sediment structure (Contessa and Bird 2004) and presumably their ecological processes. A study conducted by Wynberg and Branch (1994) in South Africa, demonstrated that the intensive removal of the ghost shrimps

Callianassa kraussi and *Upogebia africana* had an obvious and significant detrimental impact on their abundance. Collection of *C. kraussi* by bait pumping reduced populations by 70%. Similar results were found by Contessa and Bird (2004) in Coronet Bay, Victoria, when investigating the effects of bait pumping on *Trypaea australiensis*. During their three month bait-pumping experiments, abundance decreased significantly with additional changes in the physical sediment properties noted over the same time period. The authors argued that the population decline was due to sediment compaction (collapsed burrows smothered the organisms) and changes to sediment porosity. What either of the aforementioned studies failed to investigate was the affect that bait-collection (pumping and trampling) had on the ecological processes operating in the soft-sediments. However, Contessa and Bird (2004) did note that bait pumping deceased sediment porosity, organic carbon concentrations and redox potential, with their results showing an increase in chlorophyll *a* concentrations (a surrogate for BMA). Nevertheless the direct effect of bait pumping on ecological parameters is rarely studied. The presence of macrofauna alone impacts (both negatively and positively) on ecological processes (e.g. benthic metabolism and benthic nutrients) (refer to section, '*Biological factors affecting the SWI – Macrofauna'*), therefore it is fair to assume that bait pumping would affect not only the macrofauna that are removed but also the ecological processes that govern the habitat or ecosystem that is being disturbed.

Trampling is usually considered an associated effect of bait pumping, however increased interest in leisurely mudflat walks (i.e. trampling) are now becoming a popular tourist activity (Rossi *et al.* 2007). It is important that walking/trampling is recognised in its own right as a threatening process and not simply an act associated

with bait pumping as is usually reported (e.g. Contessa and Bird 2004). The physical sediment disturbance that occurs through trampling can: alter the exchange of oxygen and nutrients at the sediment water interface through compaction (Contessa and Bird 2004); cause a decrease in macrofaunal densities; destroy macrofaunal burrows; and cause macrofaunal destruction/death (Peterson 1977; Wynberg and Branch 1997; Dernie *et al.* 2003; Contessa and Bird 2004; Rossi *et al.* 2007; Hunt 2011).

In a parallel study to this study, Hunt (2011) demonstrated in the MPA Currambene Creek in Jervis Bay Marine Park (NSW) that trampling had a far larger impact on soft-sediment macrofauna compared with bait pumping alone or (where sediments were disturbed but macrofauna were not removed) bait pumping and the removal of macrofauna. Bait pumping and trampling treatments showed a decrease in the total abundance, and species richness of macrofauna, with a 90% effect size caused by trampling and only 10% accounted for by bait pumping. A considerable effect from trampling was also found by Peterson (1977) who illustrated that an intertidal population of the ghost shrimp *Neotrypaea californiensis* was completely destroyed by trampling during experimentation.

While such studies focus on the impact of trampling Rossi *et al.* (2004) also investigated the effect of trampling on ammonium (NH_4^+) and nitrate (NO_3^-) concentrations in the pore waters, water content and biomass of microphytobenthos. NH_4^+ concentrations and microphytobenthos biomass were correlated with the number of footsteps taken, indicating that trampling had an impact on the abiotic variables (Rossi *et al.* 2004). Further Rossi *et al.* (2004) also highlighted the negative effect that trampling had on the abundance of the clam *Macoma balthica* and the

cockle *Cerastoderma edule.* While the studies above demonstrate the affect that bait pumping and trampling have on macrofauna directly, they rarely investigate how bait pumping and trampling impact/influence ecological processes, whether indirectly or directly. Therefore, there is a need to experimentally demonstrate a relationship between bait pumping and trampling and the change in ecological processes as demonstrated by a change in physico-chemical and nutrient sediment properties.

1.2 Ecosystem processes – nutrient cycling in estuarine soft-sediments

Sediment-water interface

The sediment-water interface (SWI) separates a mixture of solid sediment and interstitial water from an overlying body of water (Lerman 1978). It is a region where the exchange of solutes can occur between the sediment and the water column mediated by a diffusion gradient (Wakeham 2002). The exchange of solutes is an important factor influencing the concentration of oxygen (O), nitrogen (N), phosphorus (P), carbon (C), and silicon (Si) (Asmus *et al.* 1998; Tengberg *et al.* 2003).

Nutrient sources come from both natural and anthropogenic inputs. Natural sources of nutrients include aquatic (e.g. rivers, oceans, etc.), terrestrial inputs (e.g. soil leaching) and through atmospheric processes (Fogg 1982; Tappin 2002). Anthropogenic sources come from urbanization and industrial run-off (Tappin 2002; Tengberg *et al.* 2003; Galloway *et al.* 2004). Nutrients are transferred between the SWI through gases in both inorganic and organic forms. Within the sediments inorganic nutrients are adsorbed to the surface of sediment particles, dissolved in sediment pore water and stored in the sediment matrix. The relationship among nutrient transport,

transformation, adsorption, desorption and biological processes affect the behaviour of nutrients at the SWI and in general the ecological processes in the habitats in which they occur (Furnas 1992; Kennish 2001).

Also, primary producers at the SWI drive diel nutrient patterns, creating an influx of nutrients during the dark and efflux in the light (Blakey 2005; Eyre and Ferguson 2005; Potts *et al.* 2005). Nutrient cycling in estuarine ecosystems has been extensively studied, with particular research focused on N processes (Heap *et al.* 2001; Eyre and Ferguson 2002; Ryan *et al.* 2003).

In estuaries the exchange of nutrients between the SWI is strong and as such calculating ammonium (NH_4^+) fluxes out of sediments based on measured rates of net ecosystem metabolism (NEM) is possible (Hopkinson *et al.* 1999). However, total dissolved inorganic nitrogen (DIN) flux may be more influential to the stoichiometry of NEM, whereas in oxic sediments nitrification plays an essential function in the transformation of NH_4^+ into nitrate (NO_3^-). This is important because physical, chemical and biological variables influence the chemical exchange at the SWI (Furnas 1992) (Figure 1.1) and this in turn impacts upon nutrient cycling in estuaries (Spagnoli and Bergaminin 1997). For the purpose of this study and to narrow the focus of this review the proceeding sections will focus on the chemical and biological ecological processes that affect the SWI only.

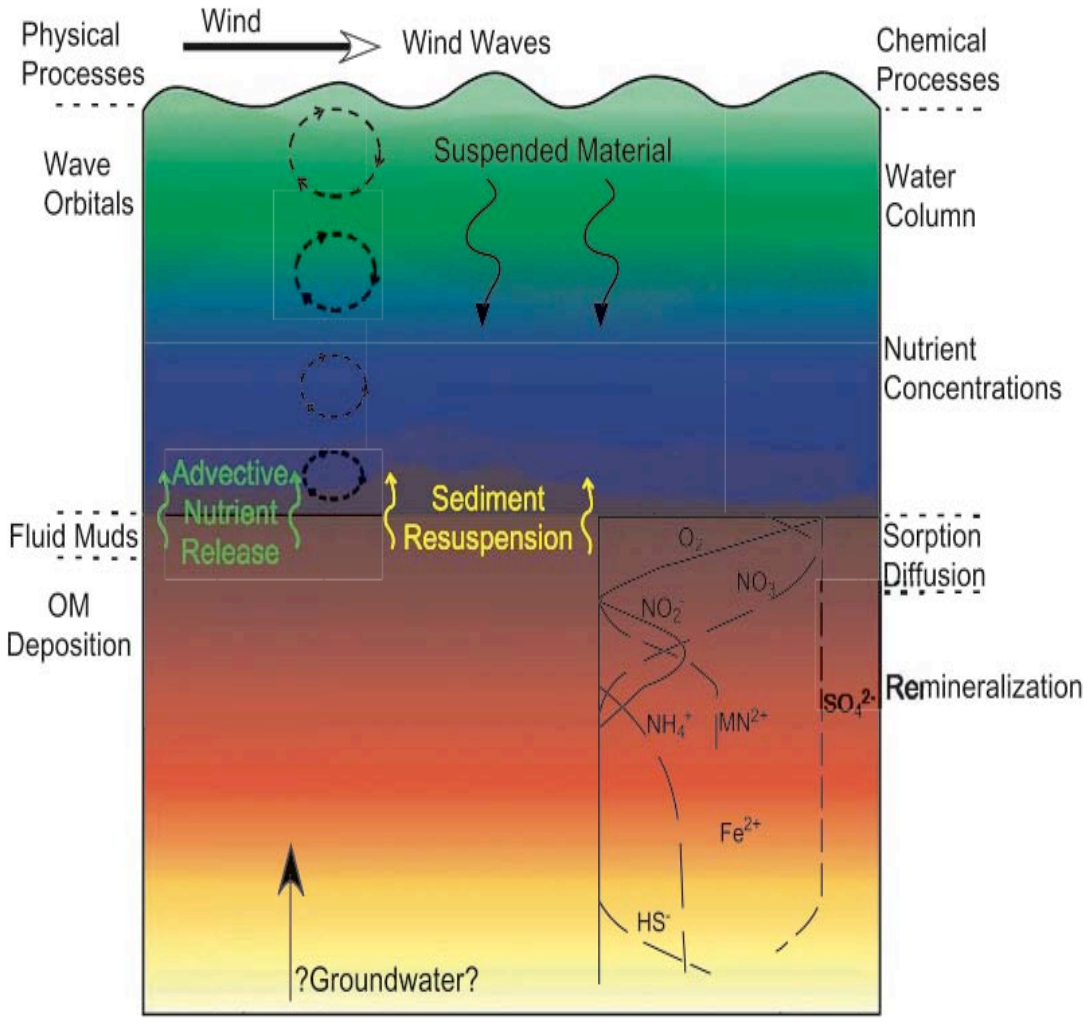

Figure 1.1 The physico-chemical and biological factors affecting the SWI. Nutrients are introduced to the water column from the sediment through passive (i.e. diffusion and desorption) and dynamic (i.e. resuspension, groundwater advection and biological mixing) processes (taken from Giffin and Corbett (2003) without modification).

Chemical factors and processes affecting the SWI

Nitrogen

Nitrogen is a key constituent of all living matter and has been the subject of thorough study within marine ecosystems. Literature reviews outlining nitrogen cycling within coastal ecosystems have been published by Fogg (1982), Furnas (1992) and Herbert (1999). Both physico-chemical and biological factors (e.g. macrofauna) mediate the complex regulatory mechanisms and interactions of nitrogen cycling in marine ecosystems (Gürel *et al.* 2005). In estuarine environments the nitrogen forms, which are important biologically, include ammonia/ammonium (NH_3/NH_4^+), nitrite (NO_2^-), nitrate (NO_3^-), and nitrogen gas (N_2) (Gürel *et al.* 2005). Ammonium, NO_2^- and NO_3^- comprise of DIN, which is used for growth by phytoplankton. Typical concentrations of NH_4^+ and NO_3^- range from <1-10µM in estuarine ecosystems (Postma *et al.* 1984). The important benthic processes associated with the nitrogen cycle include: nitrogen consumption; nitrogen fixation; organic-nitrogen synthesis from inorganic NH_4^+; organic-nitrogen mineralization to NH_4^+, nitrification, dissimilatory nitrate reduction (denitrification and nitrate ammonification (dissimilatory nitrate reduction to ammonium (DNRA))), and sediment-water inorganic nitrogen fluxes (Qu 2004; Dunn 2009).

The different forms of nitrogen in different oxidative states undergo oxidation and reduction reactions (Figure 1.2). A diverse range of microorganisms that are influenced by physico-chemical and sediment conditions mediate these oxidation/reduction reactions (Figure 1.2). Nitrogen has been recognised as the key nutrient involved in all aspects of marine ecology and is mediated by various organisms (Qu 2004).

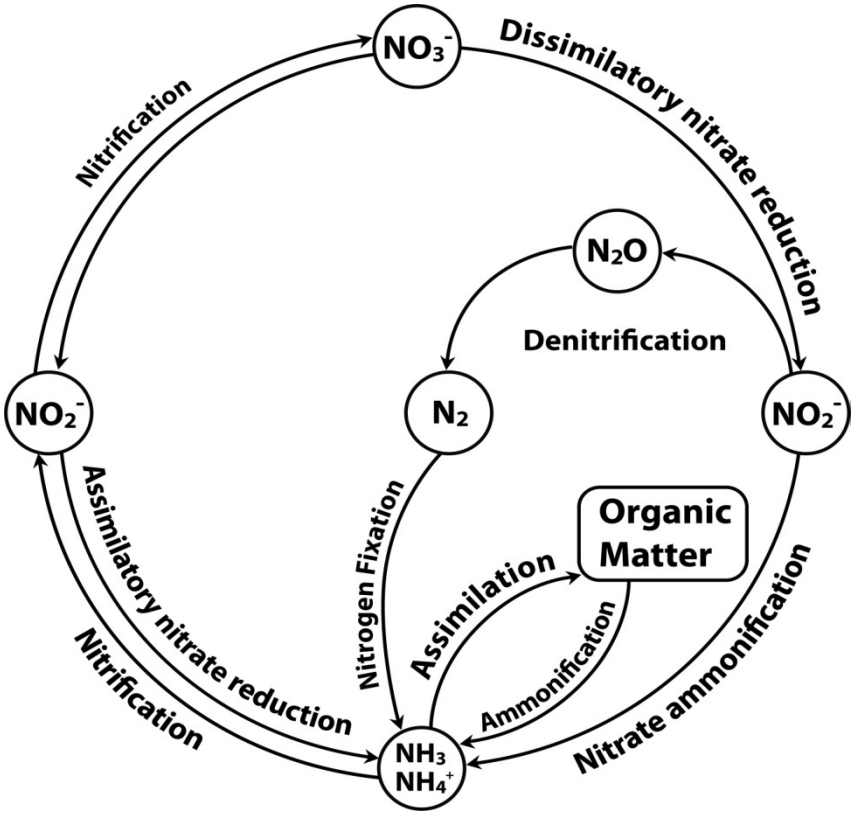

Figure 1.2 The complex interactions and processes involved in the biogeochemical cycling of nitrogen (taken from Herbert (1999) without modification).

These organisms are important mediators of nitrogen between the sediment, water column and ecological processes that are important links or sinks in the nitrogen cycle within estuaries (Seitzinger 1988).

Nitrogen consumption

The availability of NH_4^+ and NO_3^- regulates the growth of primary production in estuarine soft-sediment habitats (Gürel *et al.* 2005). Concentrations of NH_3 above 1-2 µM have the potential to inhibit the assimilation of other nitrogen forms (Postma *et al.* 1984). Assimilatory NO_3^- reduction requires more energy and if NO_3^- is to be assimilated it should first be reduced by enzymes to NH_3 (Valiela 1995). This is important because nitrogen contributes significantly to primary production and the nitrogen requirements of organisms within the marine system (Dunn 2009).

Nitrogen concentrations in shallow marine systems are relatively low because organisms store large amounts of nitrogen and metabolise it slowly (Bonin and Raymond 1990). Rysgaard *et al.* (1996) showed that the consumption of nitrogen by the seagrass *Zostera noltii* was so significant that the consumption was more important as a nitrogen sink than denitrification.

Organic-nitrogen synthesis from inorganic ammonium (NH_4^+), nitrate (NO_3^-), dinitrogen gas (N_2), and organic-nitrogen mineralization to NH_4^+

The mineralisation of organic nitrogen results in the production of NH_3/NH_4^+ (Gürel *et al.* 2005). Organic compounds excreted by animals and the decomposition of organic matter by microbes results in the regeneration of NH_4^+. Excretion contributes mostly to NH_4^+ regeneration in water, whereas the decomposition of organic matter

contributes largely to the sediment budget (Postma *et al.* 1984; Herbert 1999). Shallow marine sediments are important sites for mineralisation, as the shallow depth and relatively rapid settling rates allow for a significant portion of primary production to be transferred into the upper layer of sediment (Nixon 1981; Souchu *et al.* 1998). Aerobic and anaerobic respiration processes mineralise organic compounds. Rapid depletion of oxygen is caused by aerobic respiration, which takes place in the upper surface sediments (Herbert 1999; Qu 2004; Gürel *et al.* 2005; Dunn 2009). In all instances, production of NH_3/NH_4^+ occurs through the mineralisation of organic nitrogen compounds (Nixon 1981; Joye and Hollibaugh 1995).

Nitrogen cycling can also be influenced by benthic metabolism through the consumption and production of nitrogen (Gürel *et al.* 2005). Autotrophs produce organic matter for nitrogen consumption and the production of nitrogen occurs during the respiratory destruction of organic material (Postma *et al.* 1984; Herbert 1999; Qu 2004). The following equation (further describes the processes of primary production and respiration:

$$106\ CO_2 + 106\ H_2O + 16\ NH_4^+ + HPO_4^{2-}\ (+ \text{trace metals} + \text{energy})$$

$$\xrightleftharpoons[R]{P}$$

$$\{CH_2O\}_{106}(NH_3)_{16}(H_3PO_4) + 14\ H^+ + 106\ CO_2 \quad \text{Eqn (1) (Herbert 1999)}$$

This equation is based on a simplified Redfield model in which it assumes that NH_4^+ is the source of nitrogen for assimilation and organic matter consists of only carbohydrates (Valiela 1995). Variables such as light, nutrients and different primary

producers (e.g. benthic microalgae) limit and affect both benthic metabolism and primary production (Eyre and Ferguson 2002; Gay 2002). As discussed above, the use of this equation has helped estimate/calculate productivity and respiration rates in soft-sediments and can thus be used as an estimate of ecological processes in soft-sediments habitats.

Nitrification

Autotrophic microorganism energy requirements are met by the nitrification process that oxidises NH_4^+ to NO_2^- and then to NO_3^- under aerobic conditions. Nitrification (Eqn 4) is mediated by bacteria (*Nitrosomonas* spp. and *Nitrobacter* spp.), that oxidise NH_4^+ to NO_2^- to NO_3^- between the sediment and the water in a two-step process (Eqn 2 and 3).

The first step involves the oxidation of ammonium to nitrite:

$NH_4^+ + 1½O_2 \leftrightarrow 2H^+ + NO_2^- + H_2O$. Eqn (2) (Gürel *et al.* 2004; Qu 2004).

The second step involves the oxidation of nitrite to nitrate:

$NO_2^- + ½O_2 \leftrightarrow NO_3^-$. Eqn (3) (Gürel *et al.* 2004; Qu 2004).

The final chemical reaction and result of nitrification is:

$NH_4^+ + 2O_2 \leftrightarrow NO_3^- + H_2O + 2H^+$. Eqn (4) (Gürel *et al.* 2004; Qu 2004).

Biological and physico-chemical factors all influence the process of nitrification within estuaries (Henriksen and Kemp 1988; Herbert 1999). Nitrification in turn influences marine primary productivity by passing nitrogen to the denitrification pathway, competing with heterotrophic bacteria for limited dissolved oxygen supplies and converting forms of nitrogen released during decomposition. The NO_3^- produced

from nitrification diffuses either up to the water column or down to the anoxic zone, where denitrification takes place (Henrisken and Kemp 1988; Herbert 1999; Gürel *et al.* 2004). Nitrification is an important process that is mediated by both biological and physico-chemical factors. Through nitrification, organismal energy requirements are met and any change (e.g. experimental disturbances) in its process can interrupt organismal needs and alter the system (Gürel *et al*. 2004; Qu 2004; Dunn 2009).

Denitrification

Generally denitrification is an anaerobic process, however this process can occur in the presence of minimal oxygen concentration (Seitzinger 1988; Rysgaard *et al.* 1996), where bacteria reduce nitrate (NO_3^-) to nitrogen gas (N_2):

$$NO_3^- \rightarrow NO_2^- \rightarrow NO \rightarrow N_2O \rightarrow N_2$$ Eqn (5) (Gürel *et al.* 2005)

Coupled nitrification-denitrification (oxidation of NH_4^+ to NO_3^-) and NO_3^- distributed from the water column are the main sources of NO_3^- for denitrification where its processes take place in anoxic waters and sediments (Seitzinger 1988). Nitrification and denitrification are separate processes but are often coupled by NO_3^- which is generated by nitrification that diffuses into the sediment.

Just like nitrification, denitrification is influenced by physico-chemical and biological factors (Herbert 1999). The rates of denitrification are also largely influenced by nitrification and its ability to supply NO_3^- within the sediments. Nitrate is found three to four times greater concentration in sediment layers than in the overlying water column (Seitzinger 1988; Gürel *et al.* 2005).

Nitrogen gas is substantially removed from the ecosystem through denitrification and is therefore mostly unavailable to support primary production. Nitrogen is a vital element for primary production and its removal from the benthic sediment and water into the atmosphere can potentially neutralise the effects of eutrophication (Smith and Hollibaugh 1997; Herbert 1999). With increased rates of eutrophication from anthropogenic inputs and the importance of primary production, it is vital to understand the denitrification process within estuarine habitats (Bonin and Raymond 1990; Rysgaard *et al.* 1996; Herbert 1999) and also understand how anthropogenic processes (bait pumping, trampling, etc.) can affect and change it.

Phosphorus

Phosphorus, similar to nitrogen, is a limiting nutrient available for the growth of microorganisms. However, the concentrations of phosphorus needed for the growth of microorganisms are much smaller than those of nitrogen (Gürel *et al.* 2005). The various forms of phosphorus, its distribution, sources and transport within estuarine systems are multifaceted as a result of its complex interaction and reactions (Dunn 2009) (Figure 1.3).

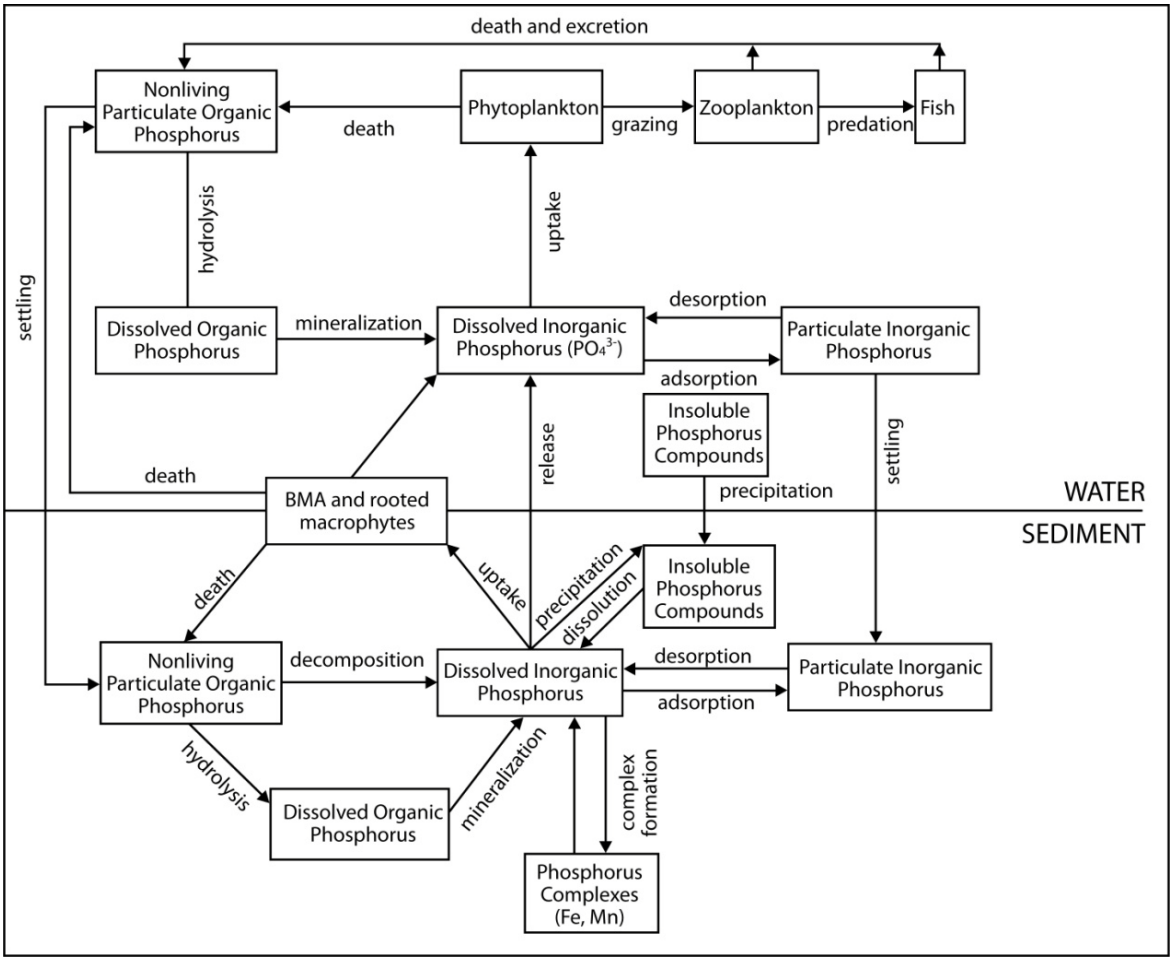

Figure 1.3 The transformation mechanisms and complexity of the phosphorus cycle between the sediment-water interface in marine ecosystems (modified from Gürel *et al.* (2005)).

The external sources of phosphorus related to estuarine systems include but are not limited to: wastewater discharge from households and industry; fertiliser runoff from agriculture; and internal natural sources of phosphorus including benthic and pelagic regeneration (Valiela 1995; Qu 2004; Gürel *et al*. 2005; Dunn 2009). Particulate and dissolved describe the two forms of phosphorus found within the water column (Lebo 1991). Particulate phosphorus can be in the form of plant and animal matter, and trace elements. Dissolved phosphorus is composed mainly of orthophosphate (PO_4^{3-}), which is the most significant form available for phytoplankton growth (Kemp and Boynton 1984; Clavero *et al.* 2000).

Phosphoric acid (H_3PO_4) and its dissociation products ($H_2PO_4^-$, HPO_4^{2-}, PO_4^{3-}) are all forms of orthophosphate ions (Blakey 2005; Gürel *et al.* 2005). Phosphorus undergoes various reactions, depending on environmental conditions, and can be taken up by phytoplankton and incorporated into cells during growth, dissolved into organic phosphorus (DIP) or released as dissolved organic phosphate (DOP) (Wetzel 1983; Clavero *et al.* 2000). A detailed review of the literature outlining the phosphorus cycle within aquatic ecosystems is beyond the scope of this thesis but has been conducted by Heap *et al.* (2001), Lillebø *et al.* (2004) and Wang *et al.* (2007). Nevertheless, phosphorus is an important part of the ecological processes occurring within estuarine soft-sediments because it is required by all biota. However, the impact that bait pumping/trampling has on phosphorus has not been investigated.

The consumption and production of phosphorus from the sediments into the water column plays an important role in water quality and consequently its movement between the two regions has been the subject of multiple studies (Boers *et al.* 1998;

Clavero *et al.* 2000; Heap *et al.* 2001). Phosphorus is bound predominantly within the sediments and less in dissolved sediment pore waters. Phosphorus is produced from sediments in the form of inorganic phosphate (PO_4^{3-}) (Eqn 6).

$$106CO_2 + 16NH_4^+ + PO_4^{3-} + 106H_2O \leftrightarrow (CH_2O)_{106}(NH_4^+)_{16}(PO_4^{3-}) + 53\ O_2$$

Eqn (6) (Boers *et al.* 1998)

Phosphorus is produced into the water column through desorption, dissolution and decomposing organic matter, where these processes affect phosphorus concentration and bioavailability in the water column (Boers *et al.* 1998; Wang *et al.* 2007). The exchange of phosphorus across the SWI is regulated by physico-chemical processes and is affected by enzymatic reactions, redox potential, temperature and microbial activity (Clavero *et al.* 2000). Nixon (1981) reported average rates of phosphorus production from various sediments are approximately -15 - 50 $\mu mol.m^{-2}\ hr^{-1}$.

The Significance of Nitrogen:Phosphorus (N:P) ratios to soft-sediments and ecological processes

The ratio of total nitrogen to total phosphorus in sediment nutrient inputs and the dynamics of internal biogeochemical processes can differ due to changes between these ratios (Smith 1984; Herbert 1999; Heap *et al.* 2001). The Redfield ratio states that the mean ratio of C:N:P in algal cells is 106:16:1. Therefore, it has been assumed that the cells require C, N, and P in these ratios. However, the ratio of these elements in the water can vary extensively and elements in surplus cannot be utilized (Correll 1998; Wang 2003; Dunn 2009).

In aquatic systems, net primary production is nitrogen limited, but phosphorus can also be a limiting nutrient (Herbert 1999). The transition from freshwater to coastal waters in estuaries can bring about a shift from phosphorus limitation to nitrogen limitation. This shift can be attributed to large amounts of nitrogen loss to the atmosphere through denitrification and an increase in the efficiency of phosphorus recycling (Boers *et al.* 1998; Wang 2003). However, using the ratios (106:16:1) to indicate nutrient limitation has problems as different types of algae have different N:P ratios (e.g. 10:1, 30:1) and nutrient loading does not occur at a steady rate. For instance, more than one type of algae may be present and some algae can grow using dissolved phosphate concentrations from stored nutrients (Nixon 1981; Qu 2004; Gürel *et al.* 2005). Nevertheless, N:P in estuaries is important because the behaviour of the nutrients for that particular system can be based on this ratio. Anything outside of the ratio suggests that either the sediment nutrient or water column nutrient sources are insufficient in meeting the nutrient demands of the system. This is vital information as it can determine if the entire system is autotrophic or heterotrophic.

Biological factors affecting the SWI

Benthic Microalgae

Estuarine habitats contain various types of benthic photoautotrophs, that play a vital role in the systems productivity (McGlathery *et al.* 2004) (Table 1.1). Inhabiting the benthos are communities of benthic microalgae (BMA) that consist of diatoms and cyanobacteria. These BMA communities are highly productive (Evans 2005) and the importance of BMA production and their affects on nutrient cycling in estuarine habitats is well documented (Underwood and Kromkamp 1999). BMA have fundamental effects on nutrient processes (Sand-Jensen and Nielsen 2004), with the

effects on nitrogen cycling mostly studied from temperate estuaries (e.g. Risgaard-Petersen *et al.* 1994; Sundback and Miles 2000; Dalsgaard 2003) (Table 1.1). The top few millimetres of sediments contain BMA that respire and photosynthesise, functioning as filters and indirectly affecting nutrient cycling processes. As filter feeders, the BMA are capable of stopping the dispersal of sediment-derived nutrients into the water column (Krom 1991; Risgaard-Petersen *et al.* 1994; Sundback *et al.* 2000).

As a function of indirect alterations to the nutrient process, BMA production has been shown to alter O_2 concentrations and penetration depth (Revsbech *et al.* 1983; Risgaard-Petersen *et al.* 1994), nutrient concentrations and depth distribution (e.g. Krom 1991; Rysgaard *et al.* 1995; McGlathery *et al.* 2001) at the SWI. Sediment to water nutrient effluxes can also decrease due to BMA production. In the presence of NH_4^+, decreases in DIN fluxes to the water column are attributed to O_2 production stimulating nitrogen removal through coupled nitrification-denitrification (Risgaard-Petersen *et al.* 1994; Dalsgaard 2003; Sundback *et al.* 2004). During light conditions, stimulated nitrification from photosynthetic O_2 production generally supports higher rates of coupled nitrification-denitrification. Nevertheless, nitrogen limitation can occur due to insufficient nitrogen from the lack of light stimulation. Light stimulated nitrogen fixation rates are higher in the sediments relative to those in the water column. In contrast some studies have established that BMA production increase sediment O_2 concentrations through inhibiting denitrifies, resulting in decreasing nitrogen removal by denitrification (Tiedje *et al.* 1989; Risgaard-Petersen *et al.* 1994; An and Joye 2001).

Table 1.1 Summary of benthic oxygen O_2 flux (µmol.m^{-2} hr^{-1}) rates reported in shallow marine ecosystems for the past decade using incubated cores only. O_2 light fluxes = Net Primary Production, O_2 dark fluxes = Benthic Community Respiration; Gross PP = Gross Primary Production; nd = no data (modified from Qu 2004).

Study location	Sediment	Primary producers	Water depth (m)	O_2 light	O_2 dark	Gross PP	References
Kattegat (Sweden)	Sandy, silt-silty sand	BMA	0.2-0.7	-208-3291	-4916 ~ 166	41-4499	Sundback *et al.* (2000)
Marennes-Oleron Bay (France)	Fine mud	BMA	Intertidal	nd	-458 ~ 0	nd	Laima *et al.* (2002)
Langstone & Chichester Harbours (UK)	Clay-fine sand	BMA	Intertidal	nd	-49499 ~ -18208 (-23999)	2291-18499	Trimmer *et al.* (2000)
Tagus Estuary (Portugal)	Mud to sand	BMA	Intertidal	-1749-1041	-4791~ 124	333-1499	Cabrita and Brotas (2000)
Tolo Harbour (Hong Kong)	N/A	BMA	12	nd	-2249 ~ -749	nd	Hu *et al.* (2001)
Huon Estuary (Australia)	Find sand – silt	BMA	Intertidal	-749-5749	-2583 -541	291-6666	Cook (2003)
Brunswick-Simpsons & Sandon estuary (Australia)	Mud	BMA	0.75	-749-2999	-2208-0	249-3999	Ferguson (2002)
6 temperate costal lagoons (Australia)	N/A	Different PP	0.5-3.0	-249-833	-1791-499	499-1749	Eyre and Ferguson (2002)

BMA is an important sink for nutrients in all types of shallow-water ecosystems (e.g. Sundback and Miles 2000; Eyre and Ferguson 2002; Thornton *et al.* 2002). In nitrogen limited habitats, BMA have demonstrated the ability to turn sediments into nitrogen sinks, particularly in winter and spring (Sundback and Miles 2000). The role of BMA in primary production and nutrient cycling at the ecosystem level is poorly understood and it is difficult to predict the role that BMA will have on nutrient turnover (McGlathery *et al.* 2004; Evans 2005) with each system unique in the influence that BMA will have on nutrient turnover. It is therefore difficult to predict the influence that disturbances such as bait pumping and trampling will have on BMA.

Macrofauna

Benthic macrofaunal communities are critical to the maintenance of marine sediment health (Wallace and Webster 1996; Webb and Eyre 2004). In soft sediment ecosystems, benthic macrofaunal communities significantly influence microbial activity and geochemical processes at the SWI through respiration, excretion, bioturbation and bioirrigation (Table 1.2) (Qu 2004; Dunn 2009). For instance, Nedwell and Walker (1995) showed that in the absence of bioturbation O_2 consumption was reduced by 33% and the sediment production of NH_4^+ was reduced by 50%.

It is well known that burrowing macrofauna introduce O_2 deeper into their burrows, oxidizing the surrounding sediment (Bertics *et al.* 2010). Due to this enhanced O_2 penetration, the oxidised sediment surface area spreads wider and deeper into the sediment layer favouring nitrification. Correspondingly, denitrification is stimulated

by an increase of NO_3^- exchanges (Dunn 2009). The increase of NO_3^- is facilitated by bacteria in the overlying water and nitrification within the sediment. Additionally, burrows created by polychaetes and crustaceans enhance the mixing of oxic and anoxic micro-environments and this strengthens the exchange between nitrification and denitrification (Gilbert *et al.* 1998). Pelegri *et al.* (1994) reported through the use of 15N isotopes that the mud shrimp *Corophium volutator* enhanced denitrification within the sediment and water and that O_2 consumption increased by 2-, 3- and 5-fold respectively. In oxic layers, the NO_3^- produced from nitrification is passed to anoxic sediment where it is lost through denitrification as gas (N_2) or into the overlying water column. In a much earlier study Henrisksen *et al.* (1983) showed that *C. volutator* greatly increased nitrification rates in burrow linings compared to the surrounding sediment.

Also, bioirrigation and sediment particle reworking activities by macrofauna have the potential to enhance the accumulation and removal of nutrients and microbial growth rates, by creating substrate-rich environments. These microniches containing both oxic and anoxic sections can facilitate areas of increased microbial activity (Bertics *et al.* 2010) (Table 1.2). Water entering the sediment through ventilation from burrows, distributes O_2, NO_3^- and SO_4^{-2} and removes NH_4^+ and H_2S inhibitory metabolites (Henrisksen *et al.*1983; Bertics *et al.* 2010). Ammonium flux is influenced substantially by the presence or absence of macrofauna through excretion. Ammonium excreted in the burrows is produced directly into the overlying water column by the ventilation current, or is oxidised by autotrophic nitrifying bacteria (Pelegri and Blackburn 1994).

Table 1.2 The effects of macrofaunal bioturbation on sediment metabolism, nutrient fluxes and nutrient cycling (ecological) processes using different methods of investigation in marine sediments from the last decade only (modified from Dunn 2009). NH_4^+ = ammonium, NO_3^- = nitrate, 0_2 = oxygen, PO_4^{3-} = phosphate and NO_x = nitrate and nitrite.

Methods	Benthic macrofauna	Approx. density	Effect (↑ & ↓ flux/process)	Reference
Microcosm (40cm× 8 cm i.d.) Homogenised sediments	*Nereis succinea*	0, 1600, 3200	↑ NH_4^+ efflux; ↑ NO_3^- flux; ↑ Denitrification	Bartoli *et al.* (2000)
Microcosm (33 cm l × 8 cm i.d.) Homogenised sediments	*Pestarella tyrrhena*	200	↑ O_2 flux; ↑ NH_4^+ efflux; ↓ NO_3^- flux	Papaspyrou *et al.* (2004)
Microcosm (28 cm l × 10 cm i.d.) Homogenised sediments	*Cerastoderma edule*	250	↑ O_2 flux; ↑ NH_4^+, NO_3^- & PO_4^{3-} efflux	Mermillod- Blondin *et al.* (2005)
	Corophium volutator	5100		
	Nereis diversicolor	640		
Microcosm (33 cm l × 8 cm i.d.) Homogenised sediments	*Arenicola marina*	200	↑ O_2 & NO_3^- flux	Papaspyrou *et al.* (2007)
Microcosm (30cm l × 8 cm i.d.) Intact sediment cores	*Nereis species*	800-4000	↑ O_2 flux; ↑ NO_3^- flux; ↑ NH_4^+ efflux; ↑ soluble reactive phosphorus efflux & flux; ↑ Denitrification	Nizzoli *et al.* (2007)
In situ micocosm (30 cm l × 30 cm w × 35 cm h)	*Arenicola marina*	2580- 3160	↑ NO_3^- flux; ↑ NH_4^+ efflux;	Papaspyrou *et al.* (2007)
In situ microcosm (29 cm l× 29 cm w× 20 cm h)	*Trypaea australiensis*	2580-3160	↑ O_2 & NOx flux; ↓ NH_4^+ efflux; ↑ Denitrification	Webb and Eyre (2004)

For instance, Henriksen *et al.* (1983) reported that excretion from *C. volutator* communities of approximately 6000 ind.m^{-2} densities accounted for a 125 to 150% increase in NH_4^+ efflux in sediments. Macrofaunal communities serve as valuable marine health indicators and the various roles performed by these macrofaunal communities in soft sediment marine ecosystems emphasise their importance to ecosystem processes and their need for conservation (Wallace and Webster 1996). The influence of bait pumping and trampling on macrofauna in Australia has only been investigated by a few studies (Contessa and Bird 2004; Winberg 2008; Hunt 2011). With macrofauna playing a critical role in many aspects of estuarine soft-sediments, it is vital to understand how common practices such as bait pumping and trampling influence the macrofauna and how this in turn affects ecological processes.

1.3 Measuring benthic processes in marine ecosystems

Comparison of methods for measuring benthic metabolism

Many studies have reported on the range of techniques used for measuring benthic metabolism (production or respiration) in marine systems including their advantages and disadvantages (Table 1.3) and range of values (Table 1.4). Some common methods include ^{14}C fixation, calculations, oxygen microelectrode, oxygen and TCO_2 exchange and each technique is characterised by its own assumptions and limitations. Confidence in estimating various benthic measurements can be achieved and improved through the implementation of multiple methods simultaneously (Hammond *et al.* 1985; Knox 1986). However, it is often difficult to implement more than one method, whether due to financial restrictions, equipment or time restrictions. Also, Callender and Hummond (1982) and Kuwae *et al.* (1998) argue that different methods yield very important differences.

Table 1.3 Summary of the advantages and disadvantages of different methods used to measure benthic metabolism.

Method/Technique	Advantages	Limitations/Errors	References
^{14}C fixation	Oxygenic & anoxygenic photosynthesis measured	- Expensive - Uneven diffusion of HCO_3^-	-Revsbech and Jorgensen (1981) -Cook (2003) -Jitts and Scott (1961) -Arthur and Rigler (1967) -Peterson (1980) -Beardall and Light (1994)
Calculations	Free from limitations faced by cores & benthic chambers	- Assumes only chlorophyll & light limit rate of photosynthesis - Only estimates benthic primary production for BMA - Difficult to accurately estimate sediment chl-*a* concentrations	-McGlathery *et al.* (2001) -Webster *et al.* (2002)
Oxygen microelectrode	Non-destructive & repeatable measures	- Time consuming - Controversy scaling up measurements to larger rates	-Revsbech and Jorgensen (1986) -Glud *et al.* (1992) -Rasmussen and Jorgensen (1992) -Underwood and Kromkamp (1999)
Oxygen exchange	Simple and inexpensive	- Cannot measure anaerobic respiration - Difference between light & dark respiration & oxidation rates	-Dalsgaard *et al.* (2000) -Blakey (2005); Potts *et al.* (2005) -Revsbech and Jorgensen (1981)
TCO_2 exchange	Measures aerobic & anaerobic	- Not mentioned	-Anderson *et al.* (1986)

Table 1.4 Summary of the advantages and disadvantages of different techniques used to measure nutrient fluxes across SWI.

Method/technique	Advantages	Limitations/Errors	References
Sediment core incubations	- Inexpensive, - Less complex incubation conditions, - Regulated easily	- Potential for disturbing cores, - Complex variables	-Kemp *et al.* (1992) -Cowan *et al.* (1996) -Moore *et al.* (1998) -Fisher and Reddy (2001) -Blakey (2005) -Potts (2005) -Qu *et al.* (2005)
Benthic chambers	- Useful for interdisciplinary research, - Incorporates natural variables	- Expensive to construct, - Disturbance during instillation, - Sealing of chamber, - Mixing of chamber waters, - Pressure gradients, - Correction for stirring effects	-Chanton and Martens (1987) -Bolalek and Graca (1996) -Miller-Way *et al.* (1994) -Berelson *et al.* (1998) -Forja and Gomez-Parra (1998) -Clavero *et al.* (2000) -Fisher and Reddy (2001)
Pore water profiles	- Inexpensive	- Complicated calculations, - Disturbance during sampling	-Kuwae *et al.* (1998) -Moore *et al*. (1998) -Qu *et al.* (2005)

Comparison of flux measurement methods

The primary methods for estimating nutrient fluxes all use concentration gradients between the sediment and water column to determine nutrient fluxes (Qu 2004). While benthic chambers are the preferred method for estimating nutrient fluxes, it should be noted that they are expensive and time consuming to build because of this do not allow for simultaneous and multiple site assessments. Additionally, there is no uniformity in their design, chamber anchorage, collection of samples and agitation of incubated water and they often require the assistance of a diver to set up and use (Berelson *et al*. 1998; Eyre and Ferguson 2002; Qu *et al.* 2003). However, they are preferred because of their many advantages and because the method provides a realistic view of both diffusion and faunal bioturbation effects (Santschi *et al.* 1990; Nicholson and Longmore 1999).

Incubated cores, are less expensive than other methods, but studies have reported highly variable flux measurements (Cowan *et al.* 1996; Moore *et al.* 1998). However, they can provide cost-effective data at a large number of sites through the recovery of intact cores, while enabling accurate monitoring of environmental variables (Eyre and Ferguson 2002; Qu *et al.* 2003). Therefore, during the past decade many studies have used incubated cores to measure nutrient fluxes between the SWI and a range of nutrient values have been reported (Table 1.5).

Table 1.5 Summary of past decade (2000 – 2010) benthic N and P fluxes (μmol.m^{-2} hr^{-1}) measured under dark and light incubations in shallow marine ecosystems using incubated cores (modified from Qu 2004).

Study location	NH_4^+ dark flux	NH_4^+ light flux	NOx dark flux	NOx light flux	DIN dark flux	DIN light flux	References
Huon Estuary (Australia)	-10~95	-28~75	-14~5	-40~5	-25~100	-42~75	Cook (2003)
Brunswick-Simpson & Sandon estuary (Australia	-100~220	-200~70	-130~0	-80~2	-80~150	-220~10	Ferguson (2002)
Six temperate lagoons (Australia)	-5~100	-200~5	-50~0	-125~0			Eyre and Ferguson (2002)
Tolo harbour (Hong Kong)	41~613		73~96		60~640		Hu *et al.* (2001)
Langstone & Chichester harbours (UK)	-20~290	-80~220	-980~10	-1400 ~ 20			Trimmer *et al.* (2000)
NE Kattegat (Sweden)	5~70	-30~30	-10~50	-60~20	-20~80	-70~20	Sundback *et al.* (2000)
Risgarde Bredning & Bight of Aarhus estuary (Denmark)	-50~175		-250~8				Risgaard-Petersen and Ottosen (2000)
Marennes-Oleron Bay (France)	29~138		-69~-21				Laima *et al.* (2002)
Bassin d'Arcachon (France)	-700~-50	-650~ -180	-1000~ -75	-2200~ -100	-900~ -750	-2400~ -800	Welsh *et al.* (2000)
	493±65	84±31	6±10	-3±12	507	89	
Tagus estuary (Portugal)	18~94		-806 ~ 1761		-790 ~ 1797		Cabrita and Brotas (2000)

Despite studies that have shown variation between benthic chambers and incubated cores, numerous other studies have been conducted that show similar nutrient results (Qu *et al.* 2003, 2004). For example, Fisher and Reddy (2001) conducted a comparative study using benthic chambers and incubated cores, to estimate P fluxes from sediments to the overlying water column, with the two methods that yielded similar results. Additionally, Kemp *et al.* (1992) found similar results when measuring O_2 and NH_4^+ fluxes measured in Chesapeake Bay (USA) using benthic chambers and incubated cores.

A similar situation is described for pore water profiles. Elderfield *et al.* (1981) and Qu *et al.* (2005) established that there was no significant difference between nutrient fluxes using incubated cores and pore water profile techniques. However, Fisher and Reddy (2001) and Callender and Hammond (1982) reported that pore water profiles estimated nutrient fluxes of a lower magnitude compared to benthic chamber and incubator core methods. It has been stated that accurate estimates of benthic fluxes can be achieved through the implementation of two or more methods (Hammond *et al.* 1985; Knox 1986). Nonetheless, it is sometimes unfeasible as not all methods are suitable for research budgets, experiment objectives or environments. The three most common methods (incubated cores, benthic chambers and pore water profiles) all use concentration gradients between the sediment and water column to determine nutrient fluxes. Each method is characterised by its own advantages and disadvantages that need to be acknowledged and understood in the context of the planned study (Table 1.4).

1.4 Conclusion

In NSW, sanctuary zones are established with the aim of conserving biodiversity and maintaining ecological processes. While these aims are well defined by the Marine Parks Act 1997, no study has yet been undertaken to establish whether sanctuary zones maintain ecological processes or how biodiversity and these processes are interrelated. Benthic metabolism and nutrient cycling are vital ecological processes that influence the complex interactions at the SWI. Nutrient cycling involves interconnected and complicated interactions between microalgal assimilation, heterotrophic activity and bioturbation by macrofauna. Macrofauna can play a fundamental role in these interactions. Considerable changes in the abundances of macrofauna in estuarine intertidal mudflats, indicates that protection may not only affect biodiversity but also ecological processes.

1.5 Aims and objectives

This project offers a unique opportunity to assess the effect that protection has on benthic nutrient cycling and provide benchmarks to guide the management of NSW estuaries and sanctuary zones. The aim of this project is to investigate benthic nutrient cycling in Currambene Creek sanctuary zone in Jervis Bay Marine Park to determine if the ecological processes are being maintained there under an experimental anthropogenic disturbance. A secondary aim is to gain a greater understanding of the interrelationships between benthic biodiversity and sediment water nutrient cycling in estuarine intertidal mudflats in sanctuary zones.

The specific objectives of this study are to:

1. Provide essential nutrient cycling benchmarks for the management of NSW estuaries and sanctuary zones from which future changes in human pressures or climate change can be assessed and effectively managed;
2. Assess potential changes in benthic metabolism and nutrient cycling due to trampling and bait pumping on/of the estuarine intertidal mudflats; and
3. Evaluate the relationship between benthic biodiversity and nutrient cycling in estuarine intertidal mudflats.

Chapter 2

Materials and Methods

2.1 Study Site

Jervis Bay Marine Park is a semi-enclosed temperate marine park spanning 220 square kilometres of coastal and marine habitat and over 100 kilometres of coastline. The marine park extends from Sussex Inlet in the south to Kinghorn Point in the North (Marine Parks Authority 2008a). The marine park encompasses various habitats, including beaches, temperate reefs, deep-water cliffs, rock platforms, seagrass and kelp forests, mangroves and small estuaries. The marine park is a multi-zoned park, partitioned into sanctuary zones (20%), habitat protection zones (73%), general use zones (6%) and Special purpose zones (0.2%) (Marine Parks Authority 2008b). The MPA study location Currambene Creek is a permanently open estuary that has a low open wave and low energy entrance, entering Jervis Bay north of the township of Huskisson (Figure 2.1). Currambene Creek comprises of 0.943 km^2 of mangroves, 0.252 km^2 of seagrass and 0.267 km^2 of saltmarsh, with the majority of the lower estuary in pristine condition undisturbed by human impact (Marine Parks Authority 2008b). The sediment of Currambene Creek is comprised of 1300 individual macrofauna, representing three Phyla, eight families and five species. A comprehensive study on the macrofauna assemblages from the same study location can be obtained from Hunt (2011). Currambene Creek has a surface area of approximately 2.2 km^2, with an average depth of 1.1 m and water temperatures ranging from 15°C during winter to 23°C during summer (Marine Parks Authority 2008b).

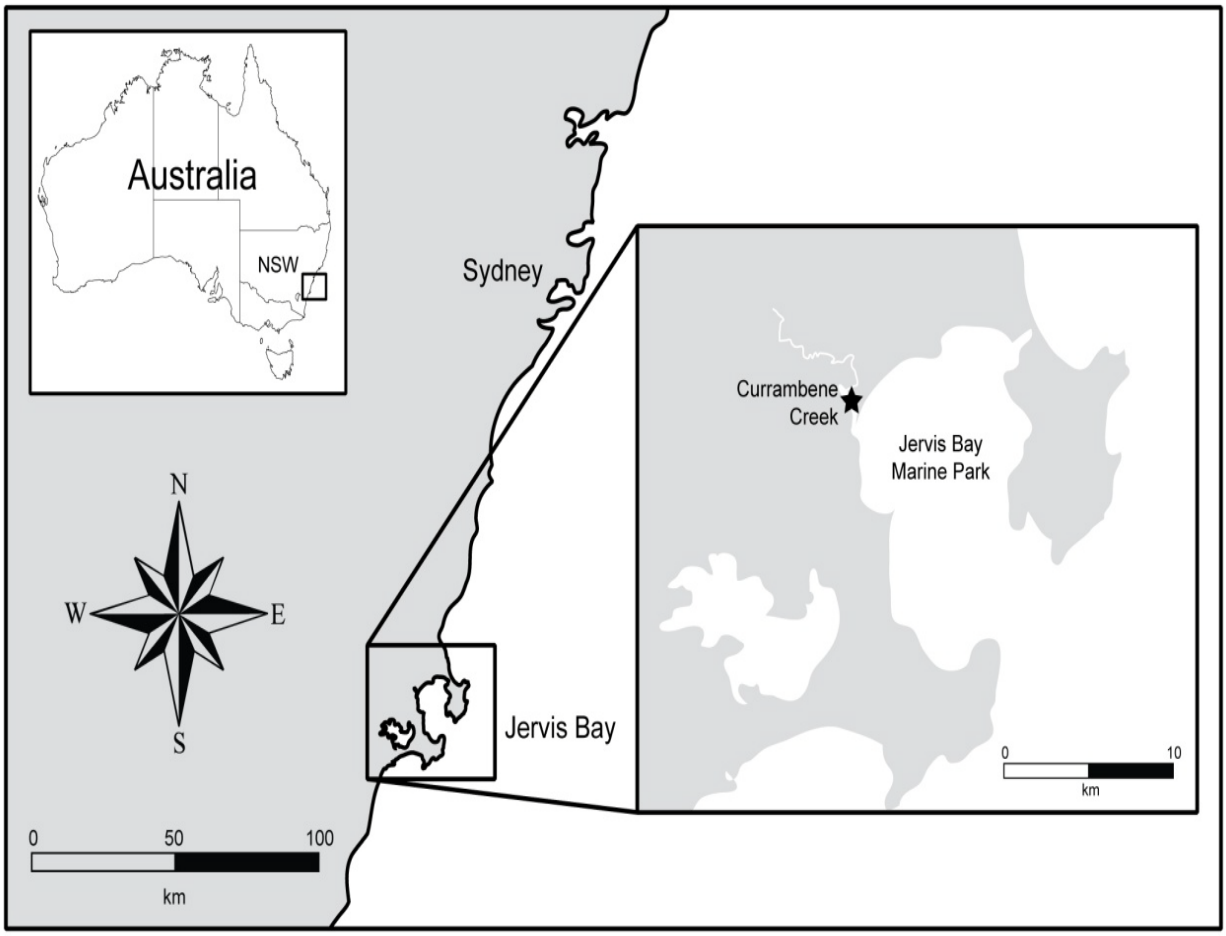

Figure 2.1 Location of Jervis Bay Marine Park and the study location Currambene Creek in New South Wales, Australia.

Two study sites were chosen within the Currambene Creek sanctuary zone approximately 1 km from the estuary mouth. The two tidal flat study sites contained intermittently occurring seagrass (*Zostera capricorni*) beds but were primarily unvegetated soft-sediments/mudflats. Activities such as walking, fishing and bait pumping were prohibited for approximately nine years in Currambene Creek prior to the study taking place (i.e. sanctuary zone protection began in October 2002). Winberg (2008) showed that considerable changes in biological diversity occurred after the sanctuary zone protection was implemented. Thus, this location and sites therein were specifically chosen to complement and build on from the findings of Winberg (2008).

2.2 Experimental Design

The bait pumping and trampling experiment was carried out from December 2010 to January 2011, during the peak Christmas holiday season when the majority of intense bait pumping was known to occur (Hunt 2011). Replicate study sites were investigated and separated by 20 m to minimise spatial variation in macrofauna, which is known to occur over small distances at this particular site (Winberg 2008). The two independent study sites (20 m x 14 m) consisted of eight replicate plots (1m x 1m), with three experimental treatments (control, trample, and trample pump) undertaken in each site (Figure 2.2). This study was conducted in parallel with a study on the macrofauna and for which eight independent replicate plots were marked out for each experiment treatment, however only six replicate treatments plots were used in the present study because of restricted number of available spaces for cores in subsequent incubation chambers (Figure 2.2). Each treatment plot was marked out by four, one metre length (20 mm diameter) PVC pipes, colour coded respectively

(control: white; trample: gold; trample pump: blue). A two metre buffer zone was left around each treatment plot to minimize edge effects and to allow access into the treatment plots.

To replicate the typical disturbance a soft-sediment/mudflat would experience from bait pumping, five ghost shrimp burrows were targeted in the trample pump treatment plots and pumped three times per burrow (Rotherham and West 2003). This number of bait pumps was representative of the intensity of this activity previously observed during peak holiday periods (Hunt 2011). The trample treatment plots were trampled using 15 steps, to imitate the same number of steps taken while bait pumping (Hunt 2011). Each treatment was applied once daily at low tide consecutively for 14 days, while the control plots were left undisturbed for the duration of the experiment.

Nutrient flux measurements

Sediment core collection

Sediment samples were collected over a three day period in three batches immediately following the 14 day experimental disturbance (i.e. the first batch consisted of 24 sediment cores and was collected and incubated overnight, followed by the 2nd and 3rd batches on consecutive days). Transparent polycarbonate cores (80mm x 500mm) were used to collect surface sediments randomly from each treatment plot (control, trample and trample pump) (Figure 2.2). For each treatment a total of 12 cores were collected from each site (i.e. two from each plot). The number of samples taken were limited by the experimental equipment (see section *Incubation System*). The cores were pushed into the sediment so that an overlying water column of approximately

0.165 meters remained above the sediment. To stop the drainage of both sediment and water, cores were capped on the top and bottom with rubber stoppers.

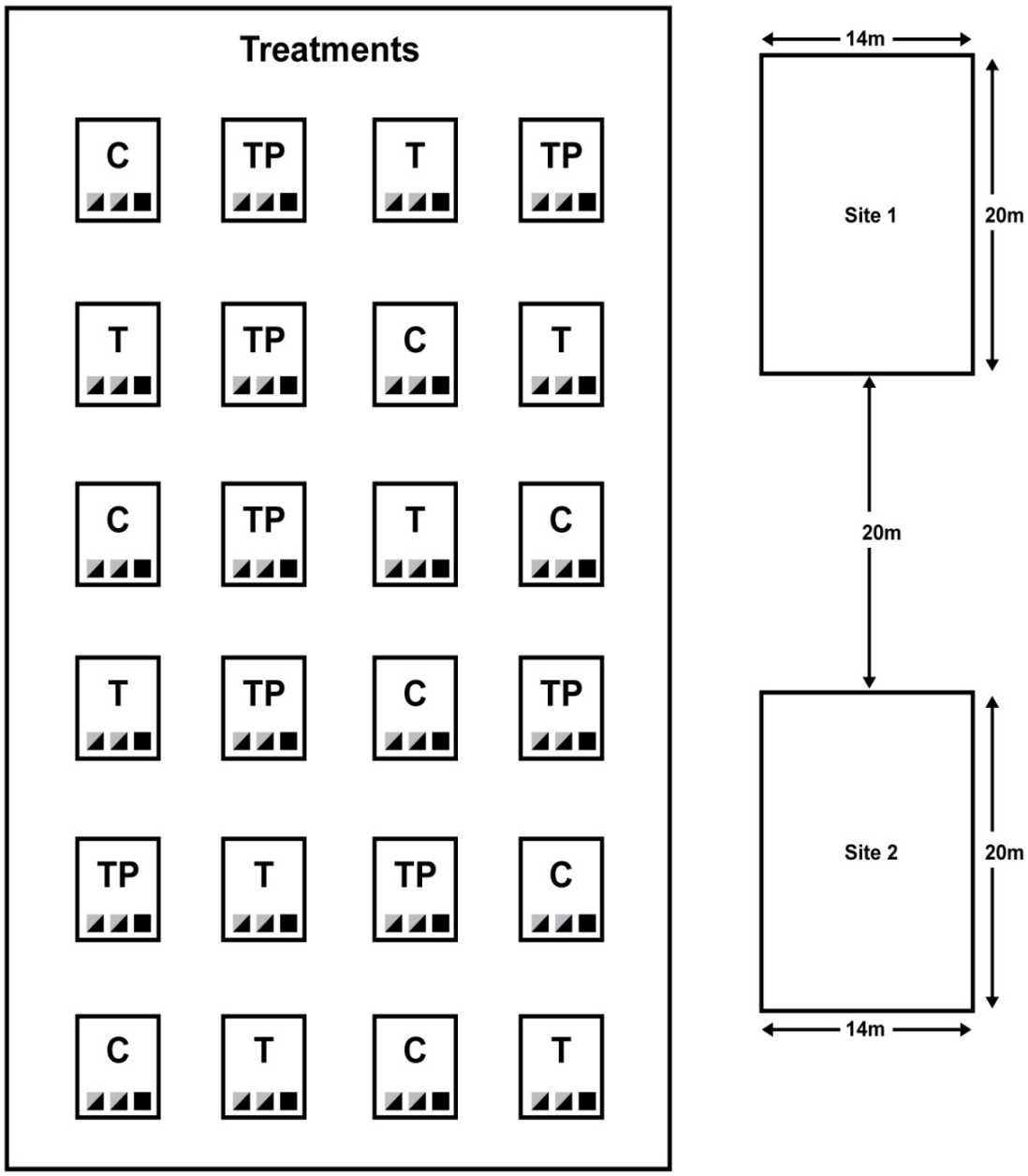

Figure 2.2 Experimental design consisting of three treatments: control (C), trample (T) and trample pump (TP). Eight treatment plots were marked out with only six plots used per site in this study. Three replicate sediment cores were collected from each treatment plot (black squares), with only two replicates taken for physico-chemical analyses (gray/black squares).

Immediately after sample collection the capped sediment and water cores were returned to shore to minimise any further disturbances.

Incubation system

For sediment-water nutrient (i.e. N, P etc) and oxygen flux estimates to be made for each treatment, cores were incubated under controlled laboratory conditions following the modified methods of Dalsgaard *et al*. (2000), Blakey (2005) and Potts (unpublished data). Cores were transferred to the cylindrical incubation chamber and placed randomly in the incubation chambers. Each chamber contained 15 cores including 12 sediment cores and three blank samples, resulting in a total of 30 cores per incubation. This limited the number of cores that could be sampled to 24 per treatment batch. The plastic cap at the top of each core was replaced with a modified perspex cap that had a central cavity to allow the insertion of a temperature/oxygen probe (Figure 2.3 D). A small intake and out-take tube was positioned on either side of the probe and used to facilitate initial flushing and to extract water samples for nutrient analysis during incubation (Figure 2.3 D).

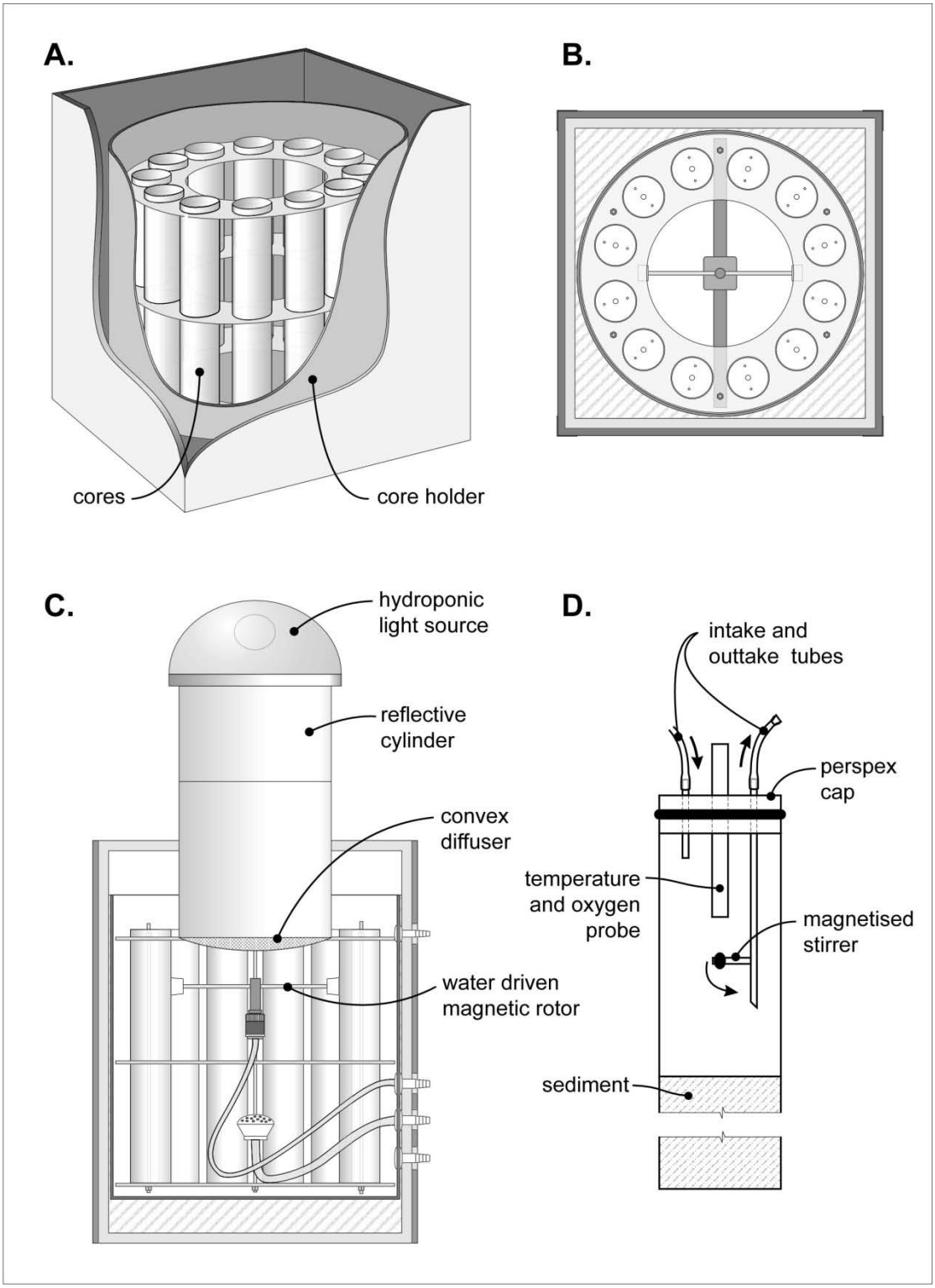

Figure 2.3 Three views of the core incubation chamber: A. Side view of core incubation chamber showing cylindrical core holder with cores; B. Aerial view of core incubation chamber; C. Side view of core incubation chamber with magnetic rotor, light source with a highly reflective cylinder and a convex diffuser. D: Core showing modified perspex cap with oxygen probe, intake and out-take tubes and magnetised stirrer.

To ensure the overlying water column in the cores were continually homogenised a small magnetised lever attached to the out-take tube was regularly moved (~60 rpm) by two magnets mounted on a rotating arm in the incubation chamber (Figures 2.3 C and D).

Flushing

At the commencement of the study 1200 L of *in situ* water was collected from Currambene Creek and transported to the laboratory for use in the incubations. The water was collected using a bilge pump and hose at a depth of approximately 0.25 m above the sediment. While it can be seen to reason that the water should have been collected during every set of cores collected and used for those particular incubations, it was impractical to transport large amounts of water so frequently and potentially at night as incubations were run during night and day. Also, it is unlikely that this method will influence the outcomes of the incubations.

The *in situ* water collected from the sampled site was transferred to a 250-litre drum and mixed to ensure homogeneity. The water was then pumped using an in line bilge pump to a 40-litre header tank situated one metre above the incubation chamber. Water from the header tank was gravity transferred to the incubator, where fresh water poured into the top of the cores via the intake tube on the modified perspex caps and this forced water to escape via the out-take tubes. A thermostatically controlled pump kept the cores at a constant *in situ* temperature (20 - 25˚ C) by circulating water through the chamber. Cores capped with the modified perspex caps were slowly flushed with *in situ* water prior to day/night incubation (Figures 2.1 A and B). The cores stored overnight were unplugged to allow contact with the ambient air. To

establish standardised initial conditions and to restore the *in situ* nutrient concentrations within the cores that were stored overnight, the cores were flushed the following morning at a rate of approximately 2.2 litres/hour for one hour prior to incubation.

Incubation

After one hour of flushing, 60-millilitre initial (time zero or T_0) nutrient concentrations were taken from each core using a syringe. The water samples were passed through a Sartorius glass filter (0.45-micron) into two 30-ml V-bottom vials and immediately frozen. All cores were then sealed, incubated for four hours in the chambers and again sampled for nutrient concentrations (time one or T_1). Dissolved oxygen measurements of each core were taken at the beginning of the four-hour incubation (time zero or (T_0) and after the cores were incubated for four hours (time one or T_1) for both light and dark incubations. Dissolved oxygen was logged for each core using a Membrane Inlet Mass Spectrometer (MIMS).

Dark/Light Treatment

To replicate the circadian rhythm of biota all dark incubations were performed at night immediately after core collection in the evening, while light incubations were carried out the following day during daylight hours (between 9 am and 12 pm). Cores and blanks were illuminated in the chamber by piping photosynthetically active radiation (PAR) light from a hydroponic light source down a highly reflective cylinder and a convex diffuser (Figure 2.3C and 2.4). A blank core was removed and replaced with a light meter to measure light intensity. Adjusting the height of the lamp allowed for light intensity modification.

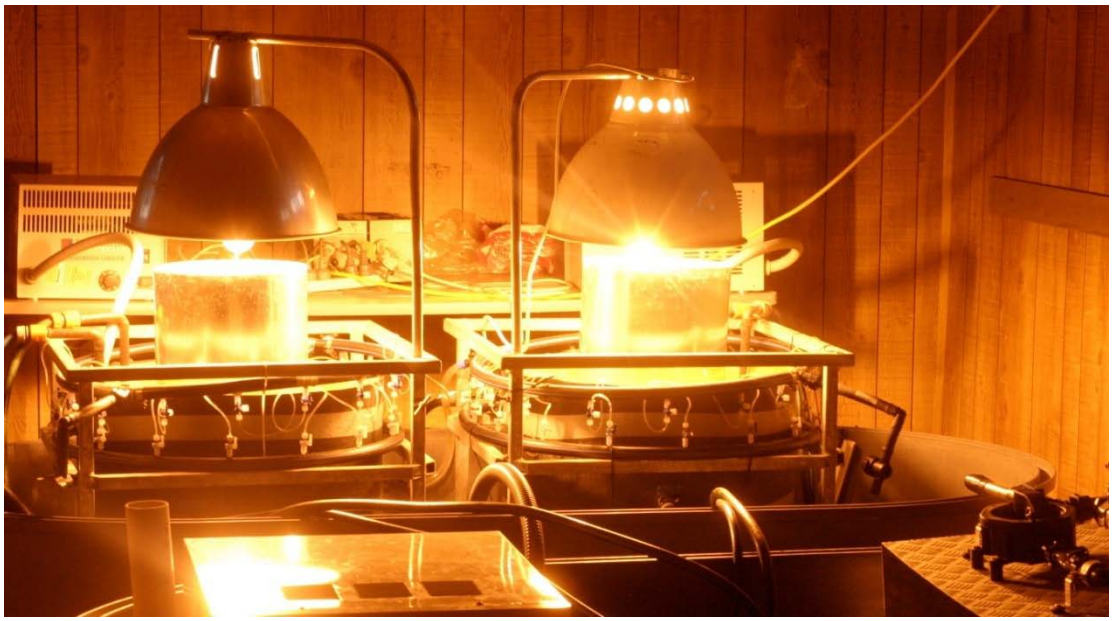

Figure 2.4 Sediment core incubators during the light incubation undertaken in this study.

Once the preferred light intensity was reached, the light meter was removed and replaced with the blank core. As per pilot study results, cores were incubated at a maximum light intensity of 450 $\mu E.m^{-2}s^{-1}$.

Samples taken for dark incubation were processed using the same incubation procedures as outlined for daytime measurements. The light intensity of 0 $\mu E.m^{-2}s^{-1}$ (complete darkness) was established by concealing the incubator with a thick rubber mat. After a set of light and dark incubations the cores were removed from the incubation chambers and the top 5mm of sediment were removed and placed in a centrifuge vile, covered with foil and frozen for later chlorophyll *a* analysis. Cores were then passed onto Tye Hunt to undertake parallel research on the macrofaunal communities found within the cores (e.g. Hunt 2011).

Nutrient Analysis

Concentrations of nutrients (O2, $NH4^{+}$, NO_x, DIN and PO_4^{3-}) were established using standardised methods (Table 2.1). All analyses were conducted at the former NSW Department of Environment, Climate Change and Water (DECCW) laboratories. Flow injection analysis (FIA) was performed on the initial (T_0) and final (T_1) concentrations of nutrients for each core, including blanks. Ammonium was analysed by the automated phenate method, PO_4^{3-} by the automated ascorbic acid reduction method and NO_x (nitrite and nitrate) by the automated cadmium reduction method (Table 2.1).

Table 2.1 Methods used for nutrient analyses and detection limits.

Test ID	Methods used during testing	Reagents	Units	Detection limit (µg/L)
NH_4^+	APHA Method 4500 – NH_3 H	Phenate	µg NH_3-N.L^{-1}	2
NO_x	APHA Method 4500 – NO_3 F (Modified)	Cadmium	µg NOx-N.L^{-1}	1
PO_4^{3-}	APHA Method 4500 – P F	Ascorbic acid	µg PO_4-P.L^{-1}	1

Nutrient flux rate calculations

Core and blank flux rates (μmol.m^{-2}.hr^{-1}) were defined as the difference between the final and initial concentration in the water column after four hours of incubation using the formula:

$$Fx = \frac{(Cf - Ci)\,x\,V}{A\,xt}$$ Eqn (7) (Potts *et al*. 2005)

Where:

Fx = flux of nutrient species (μmol.m^{-2}.hr^{-1});

Cf = final concentration (μM);

Ci = initial concentration (μM);

V = volume of water (l);

A = surface area (m^2); and

t = incubation time (h).

Daily nutrient flux rates for each core were calculated by multiplying hourly flux rates obtained during light and dark incubations, by the average number of light and dark hours during a 24 hour period.

Benthic community respiration (BCR) and net primary production (NPP) calculations

For each core, including the blank, the net primary production and respiration rates (μg/L/hr) were determined by DO measured using MIMS, during both light and dark conditions. The benthic net primary production (NPP) and respiration rates (BCR) for each core were calculated by subtracting the average water blank NPP rate/respiration rates from the net production rate/respiration rates measured in the cores. The NPP

and BCR rates for each core were converted to areal units (O_2 μmol.m^{-2}.hr^{-1}) using the formula:

$$\overline{X_B} = \frac{\left(\overline{X_b}\, x V\right)}{\left(32\, x A\right)}$$ Eqn (8) (Potts *et al*. 2005)

Where:

$\overline{X_B}$ = average benthic net primary production or respiration rates (O_2 μmol.m^{-2}.hr^{-1});

$\overline{X_b}$ = average benthic net primary production or respiration rates (μg.l.hr^{-1});

V = volume of water (l); and

A = sediment surface area (m^2).

32 is the molecular weight of dioxygen

Benthic gross primary production (GPP) for each treatment was then calculated by the following formula:

$$\overline{GPP_B} = \overline{NPP_B} - \overline{R_B}$$ Eqn (9) (Potts *et al.* 2005)

Where:

$\overline{GPP_B}$ = benthic gross primary production

$\overline{NPP_B}$ = benthic net primary production

$\overline{R_B}$ = benthic respiration

Chlorophyll a

As an indication of benthic algae biomass chlorophyll *a* (chl-*a*) concentrations from the surface sediments were analysed (Qu 2004). Immediately after light incubation, the surface sediments (top 5mm) of each core were removed and placed in 30 mm centrifuge vials, individually per core, and covered with foil and frozen pending analysis. The samples were then transported frozen and coved by foil to DECCW laboratories for analysis. Sediment chl-*a* was determined using standard spectrophotometric methods (Dalsgaard *et al.* 2000) where all analyses were carried out under low light. Samples were weighed for wet weight, freeze dried then weighted again for dry weight. Samples were then sub sampled (approximately 3-5g) and 95% acetone was added and incubated over night at 4°C. The samples were then centrifuged for 20 minutes at 5000 rmp and a sub-sample of the supernatant analysed using spectrophotometer. Spectrophotometric readings were taken for each sample before and after adding 100uL of 0.1 M HCL between 620 and 760 nm. Values for chl-*a* were then calculated according to the following standard equation:

$$\text{Chl-}a = \frac{A \times K \times [(665b - 750b) - (665a - 750a)] \times v}{a \times l}$$ Eqn (10) (Dalsgaard *et al.* 2000)

Where:

Chl-*a* = chlorophyll *a* (mg chl-*a* m^{-2});

A = absorption coefficient of chl-*a*, 11.0;

K = factor to equate the reduction in absorbency to initial chl-*a* concentrations 2.43;

665_b = the extinction at 665 nm before acidification;

665_a = the extinction at 665 nm after acidification;

750_b = the extinction at 750 nm before acidification;

750_a = the extinction at 750 nm after acidification;

v = volume of acetone extracted (ml);

a = area of samples (m^2); and

l = path length of the cuvette (cm).

2.3 Statistical analyses

Benthic Metabolism and Nutrient statistical analyses

NCSS 2007 statistical software for windows (Hintze 1998) was used to calculate a Repeated Measures Analysis of Variance (RM-ANOVA) at the 95% probability level as the same sediment cores and blanks were used to measure treatment effects under dark and light conditions for O_2, NH_4^+, NO_x, DIN and PO_4^{3-}. Disturbance type was the between cores factor, while 'dark and light' was the within cores factor. The use of RM-ANOVA reduces unsystematic variability in the design generating greater power to detect effects. However, RM-ANOVA is of concern since it violates the basic ANOVA assumption of independence (i.e. variable). Despite this, various reputable studies have used RM-ANOVA when investigating differences in benthic and nutrient fluxes (i.e. Blakey 2005; Dunn 2009; Potts unpublished data). A central assumption of the RM-ANOVA is that of sphericity. A case of circularity assumptions, sphericity verifies whether the variance/covariance matrix of the between conditions are equal (Field 1998). However, sphericity is only relevant if the within factor has more than two levels, therefore in this study the assumption of sphericity is not applicable as there are only two within factors (dark and light). Additionally, the BCR, GPP, NEM and chl-*a* variables were assessed in NCSS 2007 statistical software using a one-way ANOVA. It should be noted that output of the RM-ANOVA reveals the average (net) between BCR and NPP. Ultimately this is

calculated as the NEM. For consistency and accuracy a one-way ANOVA was additionally run on NEM. All post-hoc multi-comparisons of means were made using the Student-Newman-Keuls (SNK) test.

Chapter 3

Results

3.1 Sediment properties

Surface sediment chlorophyll a (chl-a) concentration

Chl-*a* concentration in the control sediment were significantly higher in concentration than in the trample and trample pump treatments (Table 3.1 and Figure 3.1). The SNK test revealed that the undisturbed control sediments had larger concentrations of chl-*a* than both the trample and trample pump treatment sediments. The decrease in biomass of BMA from the control and trample sediments had only half (4.7 mg m^{-2}) of the decrease noted between the trample and trample pump sediments (8.6 mg m^{-2}) (Figure 3.1). These concentrations suggest that the greater the disturbance the greater the decrease in chl-*a* concentration found (Figure 3.1).

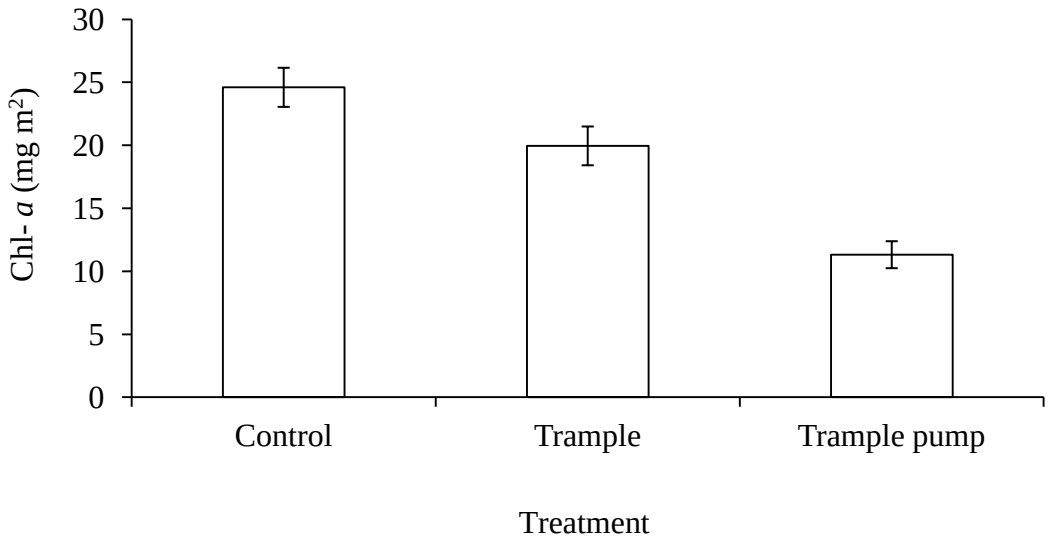

Figure 3.1 Sediment chl-*a* concentrations across the three different treatments (control, trample, trample pump) pooled across sites. Error bars represent standard error (mean ± 1 SE).

Table 3.1 Summary of the ANOVA analyses of benthic metabolism, nutrient and sediment characteristic variables from control, trample and trample pump sediments. ns: p > 0.05, *: P < 0.05, [1] : One-way ANOVA, [2]: RM- ANOVA.

Variable	Source of Variation	df	MS	F	P
Chl-*a*	Treatment[2]	2	353.89	5.13	*
O_2	Dark/Light[2]	1	7.35	986.74	*
	Treatment[2]	2	2399207	3.35	*
	Dark/Light x Treatment[2]	2	1273647	1.71	ns
	BCR[1]	2	628438.1	7.76	*
	GPP[1]	2	2547293	1.71	ns
	NEM[1]	2	2399207	3.35	*
NH_4^+	Dark/Light[2]	1	2706.48	3.94	*
	Treatment[2]	2	1225.96	2.97	*
	Dark/Light x Treatment[2]	2	142.1063	0.21	ns
NO_x	Dark/Light[2]	1	1260.73	33.65	*
	Treatment[2]	2	2.15	0.08	ns
	Dark/Light x Treatment[2]	2	18.36043	0.49	ns
DIN	Dark/Light[2]	1	7568.70	9.96	*
	Treatment[2]	2	1307.13	2.56	ns
	Dark/Light x Treatment[2]	2	192.9223	0.25	ns
PO_4^{3-}	Dark/Light[2]	1	3.84	0.89	ns
	Treatment[2]	2	13.04	4.18	*
	Dark/Light x Treatment[2]	2	1.762072	0.41	ns

3.2 Benthic Metabolism

Gross primary production (GPP) and benthic community respiration (BCR) rates occurring at the SWI are referred to as benthic metabolism. GPP occurs during the day, while BCR occurs during the night. These measures signify the basic carbon processes in marine ecosystems. Rates of GPP and BCR are determined from the production and consumption of dissolved oxygen in estuary water. GPP as opposed to NPP was analysed in this study because GPP has less uncertainty than NPP (Gürel *et al*. 2005; Qu 2004). Dark/light and treatment type were all found to have a significant effect on O_2 production, however no interaction was found between the two treatments and therefore will be discussed separately.

Dark/light conditions

Dark/light condition had a significant effect on O_2 production. O_2 was consumed by the sediments under dark conditions, while under light conditions O_2 was produced from the sediments into the water column (Table 3.1 and Figure 3.3).

Benthic Community Respiration (BCR)

BCR is the respiration occurring from primary produces and BCR rates are derived from measuring primary producers respiring at night, mostly measured as consumption of dissolved oxygen.

In this study the sediments consistently demonstrated consumption of O_2 for each treatment during the dark phase. There was a significant difference in BCR between the control and both the trample and trample pump sediments (Table 3.1 and Figure 3.3). BCR varied among treatments but was lowest in the control followed by the

trample and trample pump treatments. The greatest difference in BCR was noted between the control and both disturbances (trample and trample pump) (Figure 3.3).

Gross Primary Production (GPP)

GPP is the rate of oxygen produced and measured as the production of dissolved oxygen in a marine system. The GPP of the control sediments was 4540.8 μmol.m^{-2}.hr^{-1}, while the disturbed sediment of the trample and trample pump were 4833.6 μmol.m^{-2}.hr^{-1} and 4134.3 μmol.m^{-2}.hr^{-1} respectively (Figure 3.3). Sediments consistently demonstrated production of O_2 among treatments with no significant difference found between the control, trample and trample pump treatment sediments (Table 3.1 and Figure 3.3).

Net Ecosystem Metabolism (NEM)

NEM is the overall or net result of production (GPP) and respiration (BCR). A positive NEM value indicates that production is exceeding respiration (autotrophic). While, a negative value indicates that respiration exceeds production (heterotrophic). NEM values in the current study showed that despite O_2 being consumed during respiration and produced during productivity, the overall system was producing O_2 (Figure 3.2). This implies that at the time of sampling the mudflat was autotrophic because more O_2 was produced than consumed. O_2 was consumed and produced from the three different treatment types at significantly different rates (Table 3.1 and Figure 3.2). The NEM of the control sediments (1598.3 μmol.m^{-2}.hr^{-1}) and trample pump (1673.7 μmol.m^{-2}.hr^{-1}) sediments were similar however, the NEM of the trample treatments (2025.9 μmol.m^{-2}.hr^{-1}) was significantly higher (Figure 3.2).

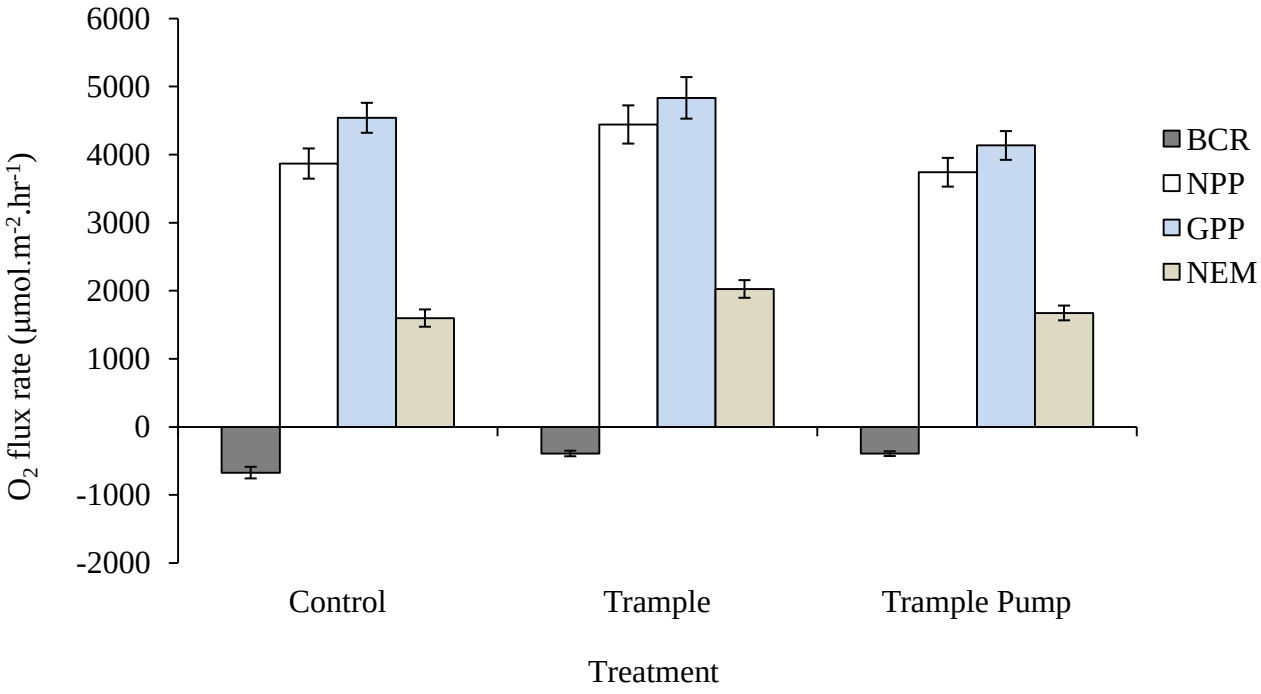

Figure 3.2 Benthic metabolism (O_2) from the three different treatments (control, trample, trample pump) pooled across sites. BCR = Benthic community respiration. NPP = Net primary production. GPP = Gross primary production. NEM = Net ecosystem metabolism. Error bars represent standard error (mean ± 1 SE).

3.3 Benthic nutrient flux rates

Ammonium (NH_4^+) flux rates

The dark/light condition and treatment type were all found to have a significant effect on NH_4^+ production, without a significant interaction noted between the two factors (Table 3.1 and Figure 3.3). Similarly, the three treatments displayed a net consumption of NH_4^+ from the sediments when exposed to dark and light conditions (Figure 3.3). NH_4^+ fluxes showed dark and light variation, with enhanced consumption by the sediments during light conditions. For instance, the dark flux varied from -9.1 $\mu mol.m^{-2}.hr^{-1}$ (control) to -17.1 $\mu mol.m^{-2}.hr^{-1}$ (trample). While a significantly higher consumption rate occurred during light fluxes which ranged from -14.2 $\mu mol.m^{-2}.hr^{-1}$ (control) to -26.3 $\mu mol.m^{-2}.hr^{-1}$ (trample) (Figure 3.3).

The SNK test revealed that the control sediment consumption of NH_4^+ was significantly less than for both the trample and trample pump sediments. The control sediment NH_4^+ flux was -11.7 $\mu mol.m^{-2}.hr^{-1}$, while the trample and trample pump NH_4^+ fluxes were -21.7 $\mu mol.m^{-2}.hr^{-1}$ and -19.7 $\mu mol.m^{-2}.hr^{-1}$ respectively (Figure 3.3).

An overall net consumption of NH_4^+ during dark and light conditions for all treatments was also observed with an obvious trend noted in the control sediments which consumed far less NH_4^+ during both dark and light conditions (Figure 3.3).

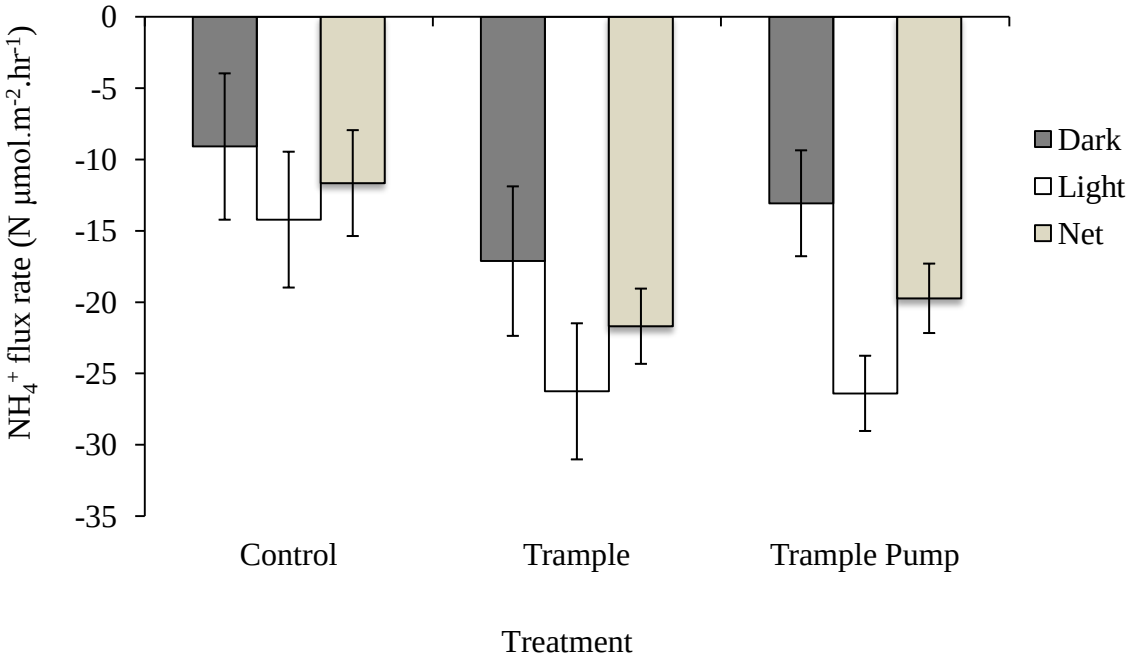

Figure 3.3 NH_4^+ flux rate from the three different treatments pooled across sites.

Error bars represent standard error (mean ± 1 SE).

Nitrite and Nitrate (NO_x) flux rates

Dark/light condition had a significant influence on NO_x fluxes while treatment type did not and no significant interaction between the two variables was found (Table 3.1 and Figure 3.4).

NO_x was produced from the sediments during the dark, while consumption of NO_x by the sediments occurred during light conditions for all treatments. Under dark incubations the NO_x flux rates consumed by the sediments ranged from 0.52 $\mu mol.m^{-2}.hr^{-1}$ (control) to 1.32 $\mu mol.m^{-2}.hr^{-1}$ (trample), while during light incubations NO_x fluxes significantly reduced to rates ranging from -4.45 $\mu mol.m^{-2}.hr^{-1}$ (control) to -6.07 $\mu mol.m^{-2}.hr^{-1}$ (trample) (Figure 3.4). However, the net NO_x flux rates showed an overall consumption by the sediment during dark and light conditions for all treatments, suggesting that at the time of sampling sediments were a net sink for NO_x (Figure 3.4).

The treatments did not appear to affect NO_x cycling between the SWI (Figure 3.4). NO_x was produced and consumed from the different treatments at similar rates, with the undisturbed control (-1.97 $\mu mol.m^{-2}.hr^{-1}$) having similar flux rates to the disturbed trample (-2.37 $\mu mol.m^{-2}.hr^{-1}$) and trample pump (-2.29 $\mu mol.m^{-2}.hr^{-1}$) sediments (Table 3.1 and Figure 3.4).

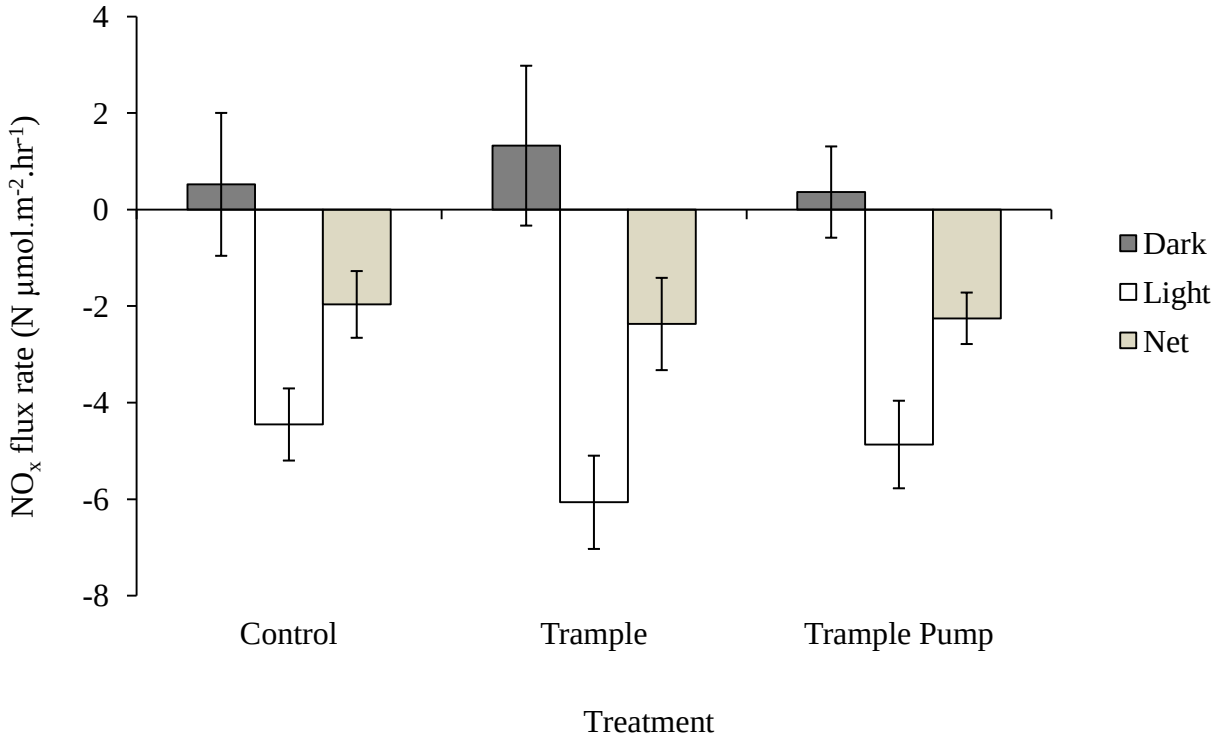

Figure 3.4 Nitrate and Nitrite (NO_x) flux rates from the three different treatments pooled across sites. Error bars represent standard error (mean ± 1 SE).

Dissolved Inorganic Nitrogen (DIN) flux rates

The benthic flux of DIN was dominated by NH_4^+. NH_4^+ was approximately 20 times greater than the flux of NO_x, with enhanced consumption noted during light conditions for both nutrients and sediment treatments (Figure 3.5). DIN fluxes demonstrated that the dark/light condition had a significant affect on DIN production, with neither an effect on production noted by the three treatments or a significant interaction found between the two variables (dark/light) (Table 3.1 and Figure 3.5). The majority of DIN was consumed during light conditions with rates ranging from -18.7 $\mu mol.m^{-2}.hr^{-1}$ (control) to -32.2 $\mu mol.m^{-2}.hr^{-1}$ (trample). The sediments consumed a reduced rate of DIN during dark incubations (e.g. -8.6 $\mu mol.m^{-2}.hr^{-1}$ (control) to -15 $\mu mol.m^{-2}.hr^{-1}$ (trample)). DIN was consumed from the sediment at similar rates for all three treatments (e.g. -13.6 $\mu mol.m^{-2}.hr^{-1}$ (control), -24.0 $\mu mol.m^{-2}.hr^{-1}$ (trample) -20.7 $\mu mol.m^{-2}.hr^{-1}$ (trample pump). A net consumption during dark and light conditions for all treatments was observed, illustrating that the sediments regardless of treatment were a net sink for DIN (Figure 3.5).

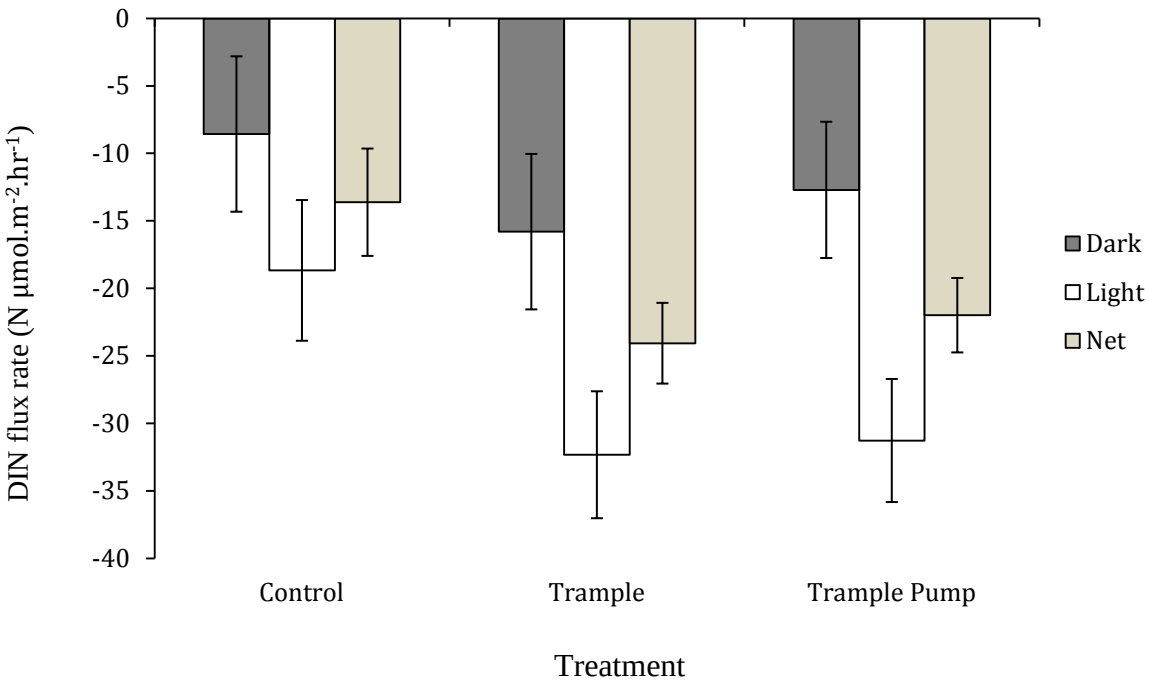

Figure 3.5 Dissolved inorganic nitrogen (DIN) flux rates from the three different treatments pooled across sites. Error bars represent standard error (mean ± 1 SE).

Phosphorus (PO_4^{3-}) flux rates

Fluxes measured under dark and light conditions had no significant affect on PO_4^{3-}, however treatment type did (Table 3.1 and Figure 3.6). No interaction between the two factors was revealed.

Under dark conditions consumption of PO_4^{3-} by the sediment was measured at -0.45 $\mu mol.m^{-2}.hr^{-1}$ (control) to -1.78 $\mu mol.m^{-2}.hr^{-1}$ (trample), while under light conditions the consumption was only minimally reduced to a range of -0.29 $\mu mol.m^{-2}.hr^{-1}$ (control) to -1.00 $\mu mol\ m^{-2}h^{-1}$ (trample) (Figure 3.6).

Conversely, PO_4^{3-} fluxes measured across different treatments showed a discernible affect on PO_4^{3-} production with the SNK multiple comparison test indicating that the control and trample treatments were significantly different from each other (Table 3.1 and Figure 3.6). PO_4^{3-} consumption by the sediment in the control was at a rate of -0.37 $\mu mol.m^{-2}.hr^{-1}$, while the trample sediment consumption of PO_4^{3-} was recorded at -1.39 $\mu mol.m^{-2}.hr^{-1}$. A net consumption was noted and again indicated that at the time of sampling the sediments were a net sink for PO_4^{3-}, regardless of treatment type (Figure 3.6).

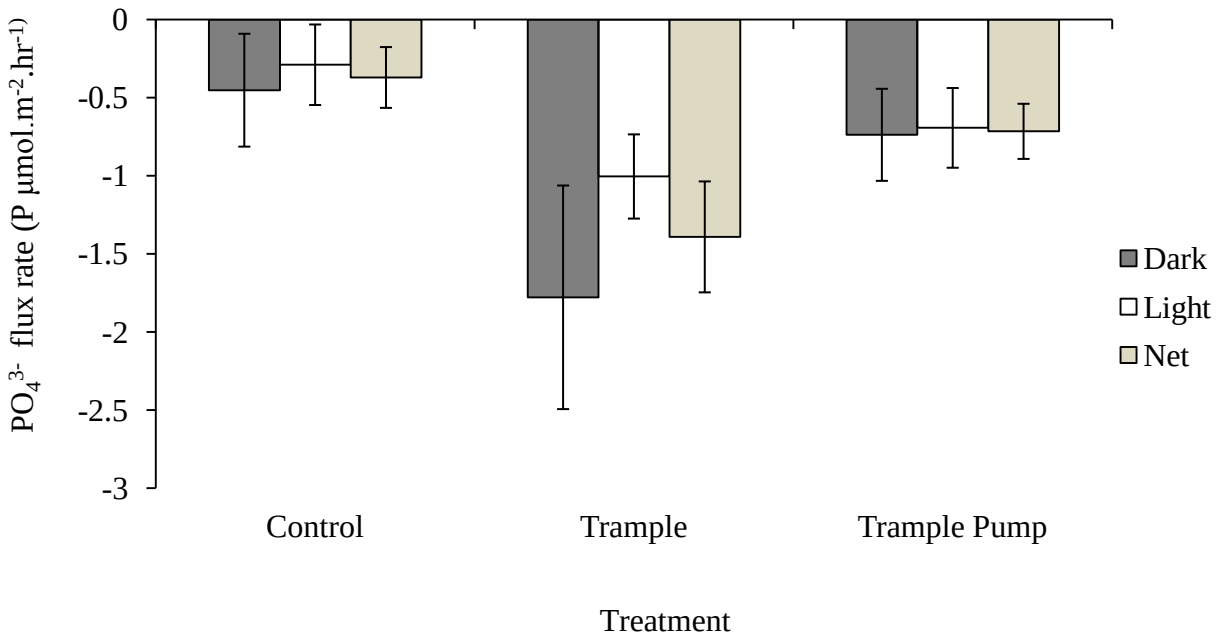

Figure 3.6 Phosphorous (PO_4^{3-}) flux rates from three the different treatments pooled across sites. Error bars represent standard error (mean ± 1 SE).

Chapter 4

Discussion

This study was the first to investigate the relationship between the effect of disturbance on macrofauna and nutrient cycling in estuarine intertidal mudflats in an MPA. The sediments from Currambene Creek sanctuary zone show autotrophic characteristics with the sediments being a net sink for all nutrients. Increasing the disturbances substantially decreased chl-*a* concentrations. Respiration was significantly affected by treatments, while productivity remained unaffected. In general, the trample treatment had the greatest impact upon benthic metabolism. Nutrient rates varied in regards to the influences from dark/light conditions and treatment type. All nutrients were influenced by the dark/light conditions, with the exception of PO_4^{3-}. The majority of nutrients were also influenced by treatment type with the exception of NO_x and DIN. Again, the trample treatment had the largest influence on the measured nutrient rates. Of note in the study was that even if no significant difference was found among the nutrient rates and treatment type, the trample treatment still seemed to vary the most from the control. Also, because there was no interaction effect between dark/light and treatments each physico-chemical variable will be discussed for dark/light and treatments separately.

4.1 Benthic Metabolism

Difference between dark/light conditions for Benthic Metabolism

The Currambene Creek soft-sediments investigated consistently demonstrated consumption of O_2 for each treatment during dark conditions, whereas during light

conditions O_2 was produced in each treatment (Figure 3.2). The O_2 flux of these predominately sandy sediments (Hunt 2011) were comparable to Cook *et al.* (2004a). These authors investigated C and N cycling in a temperate Australian intertidal mudflat (in the Huon Estuary, Tas) using *ex situ* incubations to measure fluxes over four seasons. During summer, the dark O_2 flux rate was approximately -1000 $\mu mol.m^{-2}hr^{-1}$ while the light flux rate was 4000 $\mu mol.m^{-2}hr^{-1}$. These results are similar to Currambene Creek where under dark conditions sedimentary consumption was -700 $\mu mol.m^{-2}hr^{-1}$ and under light conditions sedimentary production was ~3900 $\mu mol.m^{-2}hr^{-1}$. Compared to sub-tropical estuaries in northern NSW, the soft-sediments of Currambene Creek showed lower rates of production. For instance, the subtropical Brunswick-Simpson and Sandon estuaries (in NSW) showed higher rates of production with the highest rate of GPP ~8000 $\mu mol.m^{-2}hr^{-1}$ (Ferguson *et al.* 2004) compared to just ~4800 $\mu mol.m^{-2}hr^{-1}$ for Currambene Creek GPP.

The presence of BMA has been shown to alter oxygen concentrations at the sediment-water interface (Revsbech *et al.*1983, Risgaard-Petersen *et al.*1994) with approximately 20-30% of production contributed to BMA (Ferguson *et al.* 2004).
In Currambene Creek BMA were present at the sediment surface as indicated by chl-*a* concentrations and by the reduced consumption of O_2 by the sediments during light incubations compared to dark incubations. The presence of BMA that influence the differences between dark and light O_2 fluxes illustrates that the system is autotrophic as more O_2 was being produced than consumed and more C is fixed than is respired. Oxygen fluxes were positive under light conditions reflecting low inputs of organic matter to the sediment from internal sources and therefore the system required external sources of nutrients (Dunn *et al.* 2008). The presence of O_2 can enhance

coupled nitrification-denitrification by altering redox potential and facilitating the oxidation of phosphorous (Eyre and Ferguson 2002).

Primary producers such as BMA respire at night and produce during the day. During respiration O_2 is consumed and CO_2 is released, while during production inorganic nutrients and CO_2 are assimilated into their biomass for growth, and O_2 is released as a by product (Hauxwell and Valiela 2004). The relationship between respiration and production indicates that soft-sediments, which are colonised by BMA, fix more C than what they respired. Also, the sediments are producing O_2 during daylight hours and respiring (consuming) O_2 during dark/night hours.

During light incubations BMA are capable of modifying benthic flux rates through direct and indirect processes. Directly, BMA can interrupt the diffusion of nutrients to meet their autotrophic needs and indirectly as filter feeders enhancing the concentration and penetration of O_2 in the sediments as a product of photosynthesis (Potts *et al.* 2005). The overall composition of the BMA community can alter with seasonal temperature changes and can influence nutrient flux rates (Wolff 1987; Underwood 1994). Consequently the trophic status of a system can change seasonally as BMA change and sediments with organic matter content are modified (Engelsen *et al.* 2008). Peaks in primary production have generally been observed during spring (MacIntyre *et al.* 1996). So while Currambene Creek has been classified as autotrophic during summer when the study was undertaken, it has the potential to become heterotrophic in a different season, and this warrants further investigation.

Difference between treatments for Benthic Metabolism

Difference between BCR among treatments

The sediments of Currambene Creek actively consumed O_2 (respiration) for all three treatments. BCR rates were significantly greater in the control treatments sediments than both the trample and trample pump treatments. Nevertheless BCR values were within the range reported from other Australian estuaries (Eyre and Ferguson 2002; Ferguson *et al.* 2004; Blakey 2004; Cook *et al.* 2004a; Webb and Eyre 2004; Potts *et al.* 2005).

Overall in the trample and trample pump treatments sediments benthic O_2 consumption reduced, when compared to the control by approximately 58%. In the parallel study by Hunt (2011) the abundance of macrofauna decreased from 634 m^{-2} in the control sediment to 396 m^{-2} (trample) and 270 m^{-2} (trample pump) in the disturbed sediments. Hunt (2011) suggested that macrofaunal abundances were different among treatments due to the disturbances causing sediment compaction, burrow collapse, damage to microhabitats, sediment agitation and by the direct removal of the crustacean *B. arenosus* a well known sediment engineer. Other studies have suggested similar explanations for the reduction in macrofaunal abundance due to trampling and bait pumping in soft-sediments (Wynberg and Branch 1994; Chandrasekara and Frid 1996; Contessa and Bird 2004; Casu *et al.* 2006; Rossi *et al.* 2007). The presence of macrofauna can mediate both direct and indirect influences on BCR. Directly the macrofauna influence BCR through their physiological activities and metabolic processes e.g. respiration and excretion of dissolved nutrients (Hewitt *et al.* 2004; Dunn *et al*. 2008). During respiration the macrofauna consume O_2, and produce water and CO_2 (Valiela 1995; Dunn 2009). In the absence of these organisms

it is fair to assume that less respiration occurred in the disturbed sediments. Indeed this was demonstrated by a 58% reduction of sedimentary O_2 consumption in the trample and trample pump treatments in Currambene Creek.

Nedwell and Walker (1995) found that by removing large amphipods from the sediment, benthic O_2 consumption rates decreased by 33%. Also, Webb and Eyre (2004) demonstrated that natural populations of a common ghost shrimp, *Trypaea australiensis* (60 to > 200 m^{-2}) influenced benthic metabolism, nutrient flux, dentirificaton and irrigation rates using *in situ* benthic chambers in eastern Australian estuaries. The presence of *T. australiensis* accounted for an 81% increase in sediment O_2 demand compared to the control sediments (where no *T. australiensis* was found), with approximately 15% of this consumption used by the organism for respiration. The remaining 85% was accounted for by microbial respiration and oxidation reactions within the burrows. Fry *et al*. (1982) reported that high tubificid Polychaete densities accounted for 100% of sedimentary O_2 consumption, while generally the O_2 consumption ranges reported 40% - 80% depending upon sediment type and macrofaunal species (Kikuchi 1986; Blackburn 1987; Binnerup *et al*. 1992; Pusch and Schworbel 1994; Marshall and Hall 2004).

Evidently macrofauna can directly influence benthic metabolism and exchange rates, however, it's their indirect activities (e.g. bioturbation) that are known to have a greater influence on the SWI (Edwards and Rolley 1965; Nedwell and Walker 1995; Webb and Eyre 2004; Dunn 2009). For example, Edwards and Rolley (1965) showed how macrofaunal bioturbation enhanced sedimentary O_2 consumption more than the total of the macrofaunal's respiration. Laboratory studies averaged the rate of O_2

consumption from sediment cores with and without animals and compared them to earlier experiments. They established that approximately 40% of O_2 consumption was due to animal respiration, with the physical activity of macrofauna and microbial activity accounting for the remaining 60% (Edwards and Rolley 1965). This is comparable to Webb and Eyre's (2004) study that demonstrated only 15% of sedimentary O_2 consumption was due to respiration with the remaining 85% roughly attributed to macrofaunal bioturbation.

Bioturbation (mixing of particles) and bioirrigation (the mixing and/or movement of fluids) (Bertics *et al*. 2010) increase sediment porosity, enhance O_2 penetration, increase solute exchange between the sediment and the overlying water (Gilbert *et al.* 1998), increases O_2 supply to the sediment through ventilation and amplify the mixing of deeper anoxic layers into the upper oxic layers (Pelegri and Blackburn 1994). Pelegri and Blackburn (1994) established that densities of 3000-6000 m^{-2}*C. volutator* enhanced O_2 fluxes by approximately 10 to 20%, with the amphipods metabolism accounting for 2-3% of sediment oxygen demand and the remaining 97-98% to bioturbation, bioirrigation and microbial activity. The physical activities of macrofauna communities significantly enhance O_2 availability and therefore in their absence from the trample pump sediments in this study helps explain the large reduction (58%) of sedimentary O_2 consumption found.

In this study there was no evaluation of whether macrofaunal communities influenced O_2 in the sediment and water column through direct metabolism or indirect physical activities. However it is apparent that their reduced abundance in the disturbed sediments had a substantial influence on O_2 concentrations. As a decrease in

macrofauna abundance decreased O_2 availability and therefore the reduction of sedimentary O_2 consumption in the trample and trample pump sediments.

Various studies have established that trampling negatively affects macrofauna and influence the exchange of nutrients and O_2 rates (Contessa and Bird 2004; Rossi *et al.* 2007). It is well established that trampling can destroy organism burrows and cause sediment compaction, entice deeper burrowing organisms to the surface, disrupt benthic biofilms, and alter biological interactions (Peterson 1977; Wynberg and Branch 1994, 1997; Chandrasekara and Frid 1996; Rossi and Chapman 2003; Cruz-Motta *et al.* 2003; Contessa and Bird 2004; Rossi *et al.* 2007; Hunt 2011). Chandrasekara and Frid (1996) investigated human trampling on the abundance of macrofauna inhabiting a tidal flat trampled by pilgrims visiting a Holy place in Lindisfarne (UK). After a lengthy spatial and temporal study the authors found that macrofaunal abundances in the disturbed locations decreased because of direct or indirect mortality. Directly the trampling could crush the organisms, whereas indirectly death could have been caused by sediment compaction leading to the collapse of their burrows. It is obvious that in the present study the reduced abundance of macrofauna in the trample sediment had a significant influence on O_2 concentration. In the absence of macrofauna a significant reduction in O_2 consumption in the trample sediment was found compared to the control sediment where macrofauna were left untouched.

Difference between GPP among treatments

Contrary to the results for BCR, GPP in this study showed no significant difference between the control, trample and trample pump sediments. The GPP of Currambene

Creek had comparable productivity rates to those reported from other Australian estuaries (e.g. Cook *et al*. 2004a; Webb and Eyre 2004).

Up to 30-50% of primary production in estuarine systems is provided by BMA (Underwood and Patterson 1993; de Jonge and Colijn 1994; MacIntyre *et al.* 1996; Underwood and Kromkamp 1999). As discussed, BMA in Currambene Creek were present at the sediment surface in all treatments in this study as indicated by the reduced consumption of O_2 during light incubations (compared to dark incubations) and by the chl-*a* concentrations. After a disturbance, BMA communities can quickly recolonise sediment through immigration, recruitment and regrowth (Reice 1994). It is likely that the prompt recolonisation of BMA (as measured by chl-*a*) after the disturbances of trampling and pumping in this study could explain non-significant difference found among treatments for GPP. Plante *et al*. (2010) established that BMA recovery occurred in less than three hours, with migration from underlying sediment a dominant mechanism of recolonisation. It is possible that an increase in productivity of fewer BMA cells is due to the rapid growth phase that follows a disturbance.

In heavily mixed soft-sediments, the resuspension of BMA that are attached to sediment particles also contribute extensively to productivity (MacIntyre *et al*. 1996). For instance, Forhead (2006) simulated a disturbance by raking subtidal sediments to investigate what influence this had on BMA resuspension and subsequently its influence on the sediment water fluxes of nutrients and oxygen. The resuspension associated with the disturbance interrupted adequate chl-*a* to increase the concentration of the water column by 2.44$\mu g.L^{-1}$, or 148$\mu g.L^{-1}$ of organic carbon,

indicating that resuspension was significant in escalating the productivity of the water column. The trampling and trample pumping experiments in Currambene Creek interrupted the BMA layer that inhabits the top few millimetres of the sediment, possibly resuspending the attached BMA consequently increasing in productivity in the trample and trample pump treatment sediments. In such situations, this could counterbalance the damaging effects that the experimental disturbances have on productivity and account for the non-significant productivity established among the experimental treatments investigated in Currambene Creek.

Productivity in Currambene Creek is predominately influenced by the presence of BMA and to a lesser extent macrofauna in the sediments. BMA contribute substantially to productivity in estuarine systems and the present study confirmed this as not even the disturbances of (trampling and pumping) interrupted their ability to contribute to consistent productivity across all of the treatments.

Difference between NEM among treatments

NEM showed significant difference between the control and trample sediments. In the parallel study by Hunt (2011) the effect of trampling reduced macrofauna (from the control) by 90% with only a 10% affect size attributed to the trample pump treatment; a result supported by other studies (Wynberg and Branch 1994; Contessa and Bird 2004). Generally trampling and trample pumping had a limited influence on productivity but a considerable influence on respiration for reasons discussed in section '*Difference between BCR among treatments and Difference between GPP among treatments'*.

NEM is the overall or net result of production (GPP) and respiration (BCR). A positive NEM value indicates that production is exceeding respiration and that the system is autotrophic. While, a negative NEM value indicates that respiration exceeds production and so the system is heterotrophic (Qu 2004; Gürel *et al.* 2005). The balance between oxygen production and consumption in surface sediments is known as benthic trophic status (heterotrophy vs autotrophy) (Viaroli and Christian 2003; Cook *et al.* 2004a; Engelsen *et al.* 2008). The trophic status is a versatile indicator for: assessing water quality (Rizzo *et al.* 1996); ecosystem stability (de Wit *et al.* 2001); whether sediments are net sources or sinks of nutrients; and nutrient cycling (Eyre and Ferguson 2002; Risgaard-Petersen 2003). A number of indices have been developed to classify trophic status (Cook *et al.* 2004a). Some of the most common indices are: production: respiration ratios (P: R ratios) (Eyre and Ferguson 2002; Cook *et al.* 2004a); trophic oxygen state indicator (TOSI) (Viaroli *et al.* 1996); and the benthic trophic state index (BTSI: Rizzo *et al.* 1996). BTSI and P:R indexes were applied to the O_2 flux data in this study (Table 4.1). The BTSI and P:R ratios both classify the sediments of Currambene Creek as highly autotrophic (BTSI 3 and P:R > 1.0). Sediment that appears to be autotrophic based exclusively on the BTSI are heterotrophic over a diurnal cycle. The BTSI does not incorporate the status level over a diurnal cycle and therefore the use of P: R ratios takes this into account, especially as this study was conducted in a temperate region where day length fluctuates seasonably (Eyre and Ferguson 2002; Cook *et al.* 2004a).

In Currambene Creek the soft-sediments were classified as autotrophic and the sediment was composed of 92.4% sand (Hunt 2011). Similar results were found by Rizzo *et al.* (1996), who examined factors influencing the BTSI and determined that

predominately autotrophic sediments were composed of coarser sandy sediments. Subsequently, Rizzo *et al.* (1996) postulated that this was predominately caused by organic-rich, finer sediments with increased respiration rates. Additionally, Blakey (2005) investigated the benthic fluxes of sandy and muddy sediments from a temperate permanently open coastal lagoon (Lake Illawarra) north of Currambene Creek. Blakey (2005) determined that sandy sediments were all autotrophic, while all muddy sediments were heterotrophic.

The sediments of Currambene Creek showed autotrophic characteristics as determined by the NEM as production exceeded respiration. While production rates were high, respiration rates were low and essentially were influenced more by the presence or absence of macrofauna.

Table 4.1 Classification of shallow sediments by benthic trophic state index (BTSI) as defined by Rizzo *et al.* (1996) and the ranges of gross P: R associated with each BTSI value. Criteria are based on mean O_2 fluxes in light and dark for each sampling event and assume 12 hours of daylight for P:R ratios. The P:R ratios were calculated in accordance with the calculations in Cook *et al.* (2004a).

BTSI	P:R	Description	Criteria
0	NA	Totally heterotrophic	O_2flux light $\leq$ O_2flux < dark 0
1	> 0.0.5	Net heterotrophic	O_2flux light > O_2flux dark < 0
2	0.5-1.0	Net autotrophic	0 < O_2flux light < | O_2flux dark |
3	> 1.0	Highly autotrophic	0 < | O_2flux dark | < O_2flux light

4.2 Benthic nutrients

Ammonium (NH_4^+) fluxes

Difference between NH_4^+ in dark/light conditions

Under dark and light conditions among all treatments the sediments were a net sink for NH_4^+. Dark fluxes across all treatments showed lower sedimentary NH_4^+ consumption compared to light fluxes which showed enhanced sedimentary NH_4^+ consumption. Similar negative NH_4^+ fluxes have been reported by various other studies (Simon 1988; Rowe and Phoel 1992; Lohse *et al*. 1996; Baric *et al*. 2002; Magalhães *et al*. 2002). In particular Baric *et al*. (2002), reported similar results to Currambene Creek for NH_4^+ fluxes measured at the Krka (NH_4^+ ranged from -4.9 to -17.9 $\mu mol.m^{-2}.hr^{-1}$) and Neretva estuaries in Croatia (NH_4^+ ranged from -9.1 to -26.4 $\mu mol.m^{-2}.hr^{-1}$). Numerous studies have also established net effluxes of NH_4^+ from estuarine sediments (Blackburn and Henriksen 1983; Hansen and Kristensen 1997; Bartoli *et al*. 2003; Cook *et al*. 2004b; Jordan *et al*. 2009; Sandwell *et al*. 2009).

While diel variation in fluxes due to BMA communities have been established (Risgaard-Petersen 2003; Qu *et al*. 2003, 2004; Cook *et al*. 2004a; Eyre and Ferguson 2009), results from this study demonstrate general sedimentary consumption of nutrients during dark (night) and light (day) conditions. As discussed in Chapter three: '*Surface sediment chlorophyll a (chl-a) levels'*, BMA were present at the sediment surface in all treatments. BMA have the capability to metabolise under dark and light conditions as Rysgaard *et al*. (1993) reported that BMA could assimilate NH_4^+ at high rates, approximately 150 $\mu mol.m^{-2}.hr^{-1}$, up to 60 hours after the exclusion of a light source. BMA are located in the top few millimetres of the sediment and can prevent the production of sediment-derived nutrients to the water column through filtration (Risgaard-Petersen *et al*. 1994; Sundback and Miles 2000). Therefore it is likely that

BMA in Currambene Creek are contributing to the NH_4^+ sedimentary consumption during both dark and light conditions.

Another possible explanation for the removal of NH_4^+ from the water column may be due to nitrification. That is, BMA produce O_2 that provides improved conditions for nitrification and for the general influx of NH_4^+. Thus potentially enhancing NH_4^+ consumption during the light incubations conducted in this study (e.g. Magalhães *et al*. 2002). The O_2 requirements for nitrification were measured by the consumption of NH_4^+ (Magalhães *et al*. 2002). The authors where used atomic ratios of O_2 assimilation of NH_4^+ consumption and compared these with the O_2 required to generate NO_3^- during nitrification. Magalhães *et al*. (2002) verified that despite when values are similar to expected O_2:NH_4^+ (1.8) ratios, methods other than aerobic autotrophic nitrification must be responsible for the consumption Therefore, NH_4^+ consumption rates may not only be accounted for by the process of nitrification but also because of anaerobic NH_4^+ oxidation (nitrate serves as electron acceptors when NH_4^+ is oxidized) (Mulder *et al*. 1995; Magalhães *et al*. 2002). Additionally, a correlation between NH_4^+ and PO_4^{3-} fluxes were established by Baric *et al*. (2002). The positive correlation between NH_4^+ and PO_4^{3-} consumption signifies the removal of NH_4^+ from the pore-water by adsorption or nitrification. This is comparable to this study as similar negative fluxes of NH_4^+ and PO_4^{3-} were found. Further if NH_4^+ is being removed from the pore water, the BMA potentially intercept this nitrogen source and prevent its production from the sediment.

The expected lower NH_4^+ consumption during dark incubations and the enhanced consumption of NH_4^+ under light conditions can be explained once again by the

influence of BMA (Sundback *et al*. 1991; Asmus *et al*. 1998). BMA are photosynthetically active organisms utilizing light for metabolic processes and preferably consuming NH_4^+ during illumination (Valiela 1995). However BMA can adapt to low light levels and local conditions (Sundback and Jonsson 1988; Blakey 2005). Recent studies in southeast Australia on nutrient cycling within various shallow waterways have consistently revealed that light exposure results in enhanced consumption by the sediments (Qu 2004; Potts *et al*. 2005). The BMA modulate nutrient flux rates under light conditions by directly interrupting the diffusion of nutrients to meet their autotrophic needs and indirectly as filter feeders enhancing the concentration and penetration of O_2 in the sediments as a product of photosynthesis (Potts *et al*. 2005). Nonetheless, unaided NH_4^+ consumption might not complete the requirements of benthic primary producers, signifying that nutrients regenerated within the sediments may also supply primary producers (Magalhães *et al*. 2002).

As discussed previously, Currambene Creek sediments were categorised as autotrophic. Autotrophic sediments remove nutrients from the overlying water column through BMA, while heterotrophic sediments supply nutrients to the overlying water column (Engelsen *et al*. 2008). In this study nutrient influxes to the sediment occurred under both dark and light conditions, which is contrary to findings on nutrient diel fluxes by Qu (2004) and Dunn (2009). However, the chl-*a* results indicate the presence of BMA in the sediment, which use the nutrients for metabolic processes. Given the presence of BMA and their ability to function under both dark and light conditions, they can account for lower sedimentary NH_4^+ consumption during dark conditions as they function at a reduced rate during such conditions. While during light conditions the BMA function optimally, thus attributing to the

enhanced sedimentary NH_4^+ consumption found in Currambene Creek sediments under light conditions.

Difference in NH_4^+ among treatments

NH_4^+ consumption occurred across all treatments, with NH_4^+ in the control sediments significantly different to both trample and trample pump sediments. As discussed macrofauna influence nutrient fluxes and in this study significantly differed in abundance among treatments. In particular reduced abundances were found in trample and trample pump sediments (Hunt 2011). For example, *B. arenosus* were directly removed from the trample pump sediments through bait pumping and indirectly it was assumed that macrofauna were killed in the trampled sediments through the compaction of sediment. With the exception of Contessa and Bird (2004) who assessed the disturbances bait pumping had on redox potential, no other study apart from this one has investigated the influence trampling or bait pumping has on benthic nutrients. However, several studies have evaluated the influence similar disturbances have had on macrofaunal assemblages and the presence or absence these organisms have on benthic nutrients (Wynberg and Branch 1994, 1997; Chandrasekara and Frid 1996; Contessa and Bird 2004; Casu *et al*. 2006; Rossi *et al*. 2007). Nevertheless, this is the first study to demonstrate a clear link between the affect of disturbance on macrofauna and the benthic nutrients in an MPA.

Macrofauna have an essential affect on NH_4^+ fluxes, generally increasing it considerably through burrow irrigation /ventilation, stimulating microbial nitrogen transformations in the microenvironment surrounding the burrows and direct excretion (Blackburn and Henriksen 1983; Hansen and Kristensen 1997; Svensson

1997; Nizzoli and Welsh 1999; Mortimer *et al*. 1999; Tuominen *et al*. 1999; Bartoli *et al*. 2000; Papaspyrou *et al*. 2004; Mermillod-Blondin *et al*. 2005; Michaud *et al*. 2006; Hietanen *et al*. 2007; Karlson *et al*. 2007; Nizzoli *et al* 2007; Papaspyrou *et al*. 2007). Burrows stimulate irrigation and ventilation of surface water into the sediment by supplying electron-acceptors (e.g. O_2) and possibly removing inhibitory metabolites (e.g. NH_4^+) (Pelegri and Blackburn 1994). The irrigation activity of macrofaunal species can enhance the solute flux out of the sediment as reported by both in situ and laboratory experiments (Kristensen and Blackburn 1987; Kristensen *et al*. 1992; Kristensen and Hansen 1999). For instance, the transport enhancement of pore water solutes of TCO_2 and NH_4^+ and the influence of the polychaete *Nereis diversicolor* on the solute flux was investigated by Kristensen and Hansen (1999). CO_2 and NH_4^+ fluxes were enhanced 1.5 - 5 times due to the presence of these irrigating polychaetes. Similarly, the removal of large bioturbating amphipods in a shallow marine environment over 6 months resulted in a 50% decrease in the sedimentary production of NH_4^+ (Nedwell and Walker 1995). This reduction indicates the significant influence amphipod burrows have on nutrient production from the sediment through irrigation and ventilation. Also, *in situ* nutrient flux experiments containing different densities of the bivalve *Austrovenus stuchburyi* established that NH_4^+ was released from the sediments and increased in densities by a factor of 7 (Sandwell *et al*. 2009).

The reduced consumption rate of NH_4^+ in the control treatment in Currambene Creek included the presence of the bioturbating *B. arenosus*, which like other macrofauna excretes NH_4^+. Ventilation and irrigation through *B. arenosus* burrows would have stimulated the transfer of some excreted NH_4^+ from the sediment into the overlying

water column. Analogous results by Pelegri and Blackburn (1994) show that the consumption of NH_4^+ decreased by approximately 60% and 90% in incubations inhibited by amphipod densities of 3000 and 6000 ind m^{-2} respectively. The increase in NH_4^+ excretion and its subsequent flushing of the overlying water column presumably accounts for the decreased NH_4^+ consumption found in the control treatment sediments in Currambene Creek. On the contrary, an increase in sediment NH_4^+ consumption was seen in the trample and trample pump sediments. The removal of *B. arenosus* in the trample pump sediments and potential killing of macrofauna through sediment compaction in the trample sediments would decrease excreted NH_4^+. Consequently a reduction in the transport of excreted NH_4^+ to the overlying water due the collapse of macrofaunal burrows in the trample and trample pump treatments was anticipated. While excreted NH_4^+ can influence flux rates it has the potential to only contribute to the overall NH_4^+ flux minimally. For instance, while, 60% of NH_4^+ was produced from cores containing amphipods (*M. affinis)* only 5 – 10% of the NH_4^+ was from their excretion, with the remaining attributed to other mechanisms (Tuominen *et al.* 1999).

NH_4^+ can be enhanced by stimulated microbial nitrogen transformations in the microenvironment surrounding the burrows (Blackburn and Henriksen 1983; Henriksen *et al*. 1983) and sediment resuspension as proposed by Simon (1988). Therefore it is possible that enhanced NH_4^+ consumption found in the trample and trample pump sediments could have been because of mechanisms other than excretion. Nevertheless, it is more likely that the excretion of NH_4^+ from the macrofauna is responsible for the difference between the control, trample and trample pump sediments in Currambene Creek. Ultimately production rates across all

treatments were consistent thus suggesting that the difference between NH_4^+ fluxes in the control, trample and trample pump sediments is associated with macrofaunal excretion.

Nitrite and Nitrate (NO_x) fluxes

Difference between NO_x dark/light conditions

Studies have reported a clear diel variation in fluxes due to the presence of BMA communities (Risgaard-Petersen 2003; Qu *et al*. 2003; Cook *et al*. 2004a; Eyre and Ferguson 2002). In this study, sedimentary consumption of all nutrients occurred during both dark and light conditions with the exception of NO_x. An obvious difference between NO_x dark and light fluxes occurred with dark producing and light consuming NO_x. In Lake Illawarra, NSW Qu (2004) demonstrated similar results where dark NO_x fluxes were produced and light NO_x fluxes consumed by the sediments. Rysgaard *et al*. (1994) suggested that during light conditions, nitrogen assimilation is higher and benthic O_2 production creates extended oxic zones, enhancing diffusion thus reducing NO_3^- concentrations in the water column. Nitrogen can be assimilated by BMA as any form (e.g. NO_2^-, NO_3^-, NH_4^+ or N_2) with the preferred form being NH_4^+(Tandeau de Marsac and Houmard 1993; Hart and Grace 2000). BMA do not typically assimilate alternative nitrogen sources when NH_4^+ is available (Turpin 1991). As NH_4^+ was consumed during all light incubations in this study it is thought that NH_4^+ was not in excess for BMA requirements. Therefore, assimilation of NO_x occurred during light conditions. If there was production of NH_4^+ it would imply that there was an excess of nitrogen and as a result other nitrogen sources would not be utilised by BMA. Also as NH_4^+ is the preferred nitrogen source it was taken up by BMA considerably more compared to NO_x as shown in this study.

Contrary to light fluxes, NO_x was produced from the sediment during dark conditions. During dark conditions NO_x should be another nitrogen source for BMA as consumption of NH_4^+ indicates that the nitrogen requirements of BMA are not being met as also shown in this study. Others have shown that during night conditions however, nitrogen assimilation was low and additionally the oxic zone was small due to O_2 consumption increasing the potential for NO_x effluxes (e.g. Rysgaard *et al*. 1994). As NH_4^+ is the preferred source of N and BMA metabolise in the dark at reduced rates.

Therefore it is possible that BMA are assimilating NH_4^+ at full capacity and are not capable of assimilating NO_x as an additional source of nitrogen due to night conditions, causing a small production of NO_x from the sediments. Alternatively, as this small production of NO_x could be considered as consumption, the BMA are assimilating the available NO_x as an additional nitrogen source because the consumption of NH_4^+ is still not meeting their nitrogen needs. NO_x contributed minimally to DIN compared to NH_4^+ and the consumption of both NH_4^+ and NO_x reflects the ability of BMA to assimilate additional nitrogen sources. Throughout this study BMA have demonstrated their role in influencing the variation found between dark and light for nitrogen fluxes in Currambene Creek.

Differences in NO_x among treatments

In this study NO_x fluxes did not differ among the control, trample and trample pump sediments. Net NO_x sedimentary consumption occurred across all treatments. Various authors have reported similar consumption rates found in Currambene Creek for NO_x (Ogailve *et al*. 1997; Asmus *et al*. 1998, Cabrita and Brotas 2000; Baric *et al*. 2002;

Qu 2004). The consumption of NO_x indicates denitrification is occurring within the sediment of Currambene Creek (Baric *et al*. 2002; Magalhães *et al*. 2002). While production generally indicates nitrification is an important process occurring within the sediments (Dunn 2004). The activities and presence of macrofauna are known to stimulate nitrification-denitrification coupling (Pelegri *et al*. 1994) and enhance NO_3^- supply (Kristensen *et al*. 1991). Through sediment reworking and bioirrigation macrofauna increase solute exchanges and sediment porosity (Gilbert *et al*. 1998). Nitrification is favoured when oxidised sediment surfaces are enhanced by O_2 penetration of sediments facilitated by macrofauna.

Also, denitrification is stimulated through enhanced NO_3^- exchange by macrofaunal sediment reworking providing more NO_3^- for bacteria (Kristensen *et al.* 1991). Nitrification-denitrification coupling is enhanced when macrofauna increase the proximity of oxic and anoxic microenvironments through sediment reworking (Pelegri *et al*. 1994). It would have been expected that since the control sediment in the present study contained more macrofauna than the trample and trample pump sediments that bioirrigation and sediment reworking in the control sediments would have enhanced denitrification through greater nutrient availability (e.g. NO_2^- and NO_3^-) (Gilbert *et al*. 1998). For instance, Pelegri and Blackburn (1994) showed that NO_3^- within the sediment was enhanced by 20 to 70% by the 3000 to 6000 ind m^{-2} densities of amphipods. However, no difference was found between sediments containing macrofauna and sediments containing reduced numbers of macrofauna in Currambene Creek.

Therefore, there was no evidence of an increase in nutrient availability due to macrofaunal activities of bioturbation and sediments reworking. This could potentially be due to the rapid consumption of nutrients by denitrification (Sayama and Kurihara 1983; Huttel 1990) but also NO_3^- flux measurements may have been hindered by low water column concentrations and the presence of denitrifying bacteria in the anoxic layer of the sediments (Sandwell *et al*. 2009). There was a minor decrease in NO_x consumption in the control sediment suggesting that nitrification in bioturbated sediment was enhanced more than denitrification, as interpreted from the increased consumption of NH_4^+ relative to the decreased consumption of NO_x in these sediments (Pelegri and Blackburn 1994).

While the activities of the macrofauna are said to enhance nitrification-denitrification coupling by mixing or enhancing the proximity of oxic and anoxic sediment layers this was not seen because the physical disturbance of trampling and trample pumping alone mixed the two sediment layers, concealing any potential influence on nutrients that macrofauna may have had in this study.

An alternative explanation for the difference in NO_x among treatments is the presence of BMA and their ability to assimilate various forms of N (Tandeau de Marsac and Houmard 1993; Hart and Grace 2000). The presence of BMA across all treatments could explain the consistent consumption of NO_x. Despite the reduced biomass of BMA in the disturbed treatment sediments, production was similar across all three treatments as BMA have the ability to grow and recolonise rapidly. For instance, Plante *et al*. (2010) established that BMA recovery was significant in less than three hours, with migration from underlying sediment a dominant mechanism of

recolonisation. Therefore, BMA in the present study were likely to assimilate NO_x consistently across all treatments, as the disturbances do not seem to influence their assimilation potential.

Net NO_x fluxes demonstrated consistent sedimentary consumption across all treatments. The presence or absence of macrofauna had little influence on the NO_x fluxes. The consistent consumption across all treatments is attributed to potential denitrification rapidly consuming nutrients, low nutrients recorded due to denitrifying bacteria in the anoxic layer and low water column concentrations preventing or reducing measurable NO_x rates.

Dissolved Inorganic Nitrogen (DIN) fluxes

Difference between DIN dark/light conditions

The strong autotrophic metabolism of the sediments drove the consumption of nutrients under both dark and light conditions. The relative percentages of the different forms of nitrogen consumption by the sediments were quite consistent and were dominated by the influx of NH_4^+. NH_4^+ represented approximately 76.1% to 97. 3%, compared to NO_x that accounted for only 2.7% to 23.9% of the mean total of DIN across the treatments. NH_4^+ has been shown to constitute a substantial part of benthic N flux in estuarine systems (Blackburn and Henriksen 1983; Blakey 2005; Qu 2004; Dunn 2009). Mean DIN fluxes ranged between -8.6 and -32.3 $\mu mol.m^{-2}.hr^{-1}$ and a significant difference was observed between DIN fluxes during dark and light incubations in this study. Sedimentary DIN consumption during dark conditions was low compared to the approximate 38% - 46% increase in consumption during light conditions in Currambene Creek. Similar DIN rates were estimated by Tyler (2002) in

temperate coastal lagoons from Virginia, USA where DIN consumption by sediments whereas significantly higher in the light than in the dark. In particular, the author showed DIN net consumption of approximately -20 μmol.m^{-2}.hr^{-1}, and pooled data from across all experiments showed an average dark flux of -3.91μmol.m^{-2}.hr^{-1} and average light flux of -13.91 μmol.m^{-2}.hr^{-1}. Tyler (2002) speculated that BMA were responsible for the dark/light differences in sedimentary DIN consumption.

The net autotrophic conditions characterizing Currambene Creek suggest an active microalgal community in the sediment (Dunn 2009). Also, the presence of BMA were confirmed by chl-*a* which presumably accounted for the difference in the dark and light consumption of DIN. BMA can assimilate NO^{-3} and NH_4^+ at elevated rates; up to 60 hours after the darkening of the sediment (Rysgaard *et al*. 1993). However, BMA are photosynthetically active organisms that function and metabolize optimally under light conditions (Valiela 1995). The reduced rate of metabolizing BMA under dark conditions exemplifies the reduced sedimentary DIN consumption during dark conditions compared to increased sedimentary DIN consumption under light conditions where BMA function optimally.

Furthermore, light exposure has been shown to enhance DIN consumption by the sediment, contributing to the increased consumption of DIN under light conditions (Qu 2004; Potts *et al*. 2005). Also, the current study was conducted during the summer months, where enhanced sunlight availability was expected. Various studies have shown that DIN concentrations are generally lower in summer and autumn than those in spring and winter when productivity and light are usually at their maximum (Qu 2004; Blakey 2005; Potts *et al*. 2005; Dunn 2009). Alternatively, the deficit of

DIN relative to respiration could be due to the removal of nitrogen from dissolved organic nitrogen (DON) production or restriction into microbial biomass (Hammond *et al*. 1999; Ferguson 2002).

A comprehensive study on the influence of seagrass and BMA on nutrient fluxes from Lake Illawarra, NSW, illustrated that DIN production by cores incubated with seagrass and algae shifted to DIN consumption when compared to cores incubated without algae (Qu 2004). These results combined with the results from this present study indicate that BMA act as a filter reducing the flux of DIN and nutrients to the water column (Qu 2004; Potts *et al*. 2005).

Nitrifying bacteria and BMA compete for nitrogen substances when nutrient cycling in euphotic sediments are influenced by autotrophic assimilation. Subsequently leading to competition for N substrates between heterotrophic and autotrophic assemblages (Sundback and Miles 2000). Heterotrophic bacteria incorporate DIN and generally nitrogen for growth and protein synthesis subsequently facilitating the completion and potentially reducing effluxes from the sediments (van Duyl *et al*. 1993; Caraco *et al*. 1998).

The dark/light difference in DIN consumption could also be possible because of the redox "filter" formed by photosynthetic O_2 production preventing NH_4^+ effluxes (Sundback and Graneli 1988). The nitrogen mineralization at Currambene Creek may not have been enough to sustain both BMA demand and the redox "filter" effect, and consequently resulted in the consumption of NH_4^+ from the sediments.

Differences in DIN among treatments

Large DIN fluxes in sediments are associated with greater organic matter content (Jensen *et al*. 1990; Caffrey and Miller 1995) and with muddy compared to sandy sediments (Fisher *et al*. 1982; Sundback *et al*. 1991; Reay *et al*. 1995). DIN consumption occurred across all treatments with no significant difference between DIN fluxes in the control, trample and trample pump sediments of Currambene Creek.

Macrofauna can directly and indirectly contribute to DIN fluxes through excretion, respiration, irrigation and bioturbation (Fisher *et al*. 1982; Jensen *et al*. 1990; Kemp *et al*. 1990; Rizzo *et al*. 1992; Yoon and Benner 1992; Pelegri and Blackburn 1995; Rysgaard *et al*. 1995, 1996; Lohse *et al*. 1996; Conley *et al*. 1997; Berelson *et al*. 1998; Risgarrd-Petersen *et al* 1998; Asmus *et al*. 2000; Bartoli *et al*. 2000; Cabrita and Brotas 2000; Risgarrd-Petersen and Ottosen 2000; Sundback *et al*. 2000; Trimmer *et al*. 2000; Welsh *et al*. 2000; Hu *et al*. 2001; Eyre and Ferguson 2002; Ferguson 2002; Laima *et al*. 2002; Cook 2003; Contessa and Bird 2004; Webb and Eyre 2004; Kristensen and Kostka 2005; Nizzoli *et al*. 2007; Dunn 2009). A decrease in sedimentary consumption was significantly correlated (data not shown) with an increase in macrofaunal abundance. These results are comparable to Dunn (2009) where a noticeable trend of higher macrofaunal densities resulted in increase in DIN production. Although production was not recorded in the present study a decrease in sedimentary consumption was noted in the control sediment. Thus suggesting that the DIN flux had the potential to turn from consumption to production due to the presence of macrofaunal. Despite this trend no significant difference among the control, trample and trample pump sediments were evident.

Anderson and Kristensen (1988) demonstrated similar results and suggested that increases in DIN were mainly from an increase in NH_4^+ and the production of DIN decreased significantly due to consumption by BMA. The presence of BMA has had a profound influence on the benthic nutrients in the sediments from Currambene Creek and did not influence DIN rates among the control, trample and trample pump sediments. Ferguson *et al*. (2004) demonstrated that net DIN assimilation by macrofauna was significantly lower than that for BMA and consistently accounted for the bulk of assimilated (73 to 231%) DIN in the estuaries of northern NSW, investigated.

Additionally, Kuwae *et al*. (1998) found that DIN was recycled within the sediments and BMA played a significant responsibility in restraining the production of DIN from the sediment. The total BMA consumption of DIN estimated from O_2 productivity and the Redfield ratio was 573.4 $\mu mol.m^{-2}.hr^{-1}$ (Kuwae *et al.* 1998). This is similar to the present study where it was found that BMA played a significant role in suppressing the production of DIN and this could account for the treatments having no influence on DIN fluxes.

Phosphorus (PO_4^{3-}) fluxes

Difference between PO_4^{3-} in dark/light conditions

The sediments of Currambene Creek sequestered PO_4^{3-} during both dark and light conditions. There was no significant difference between dark and light PO_4^{3-}fluxes. Negative PO_4^{3-} fluxes have been reported from various shallow intertidal marine ecosystems (Hartwig 1978; Klump and Martens 1981; Fisher *et al*. 1982; Rizzo 1990; Zimmerman and Benner 1994; Reay *et al*. 1995; Berelson *et al*. 1996; Baric *et al*.

2002; Magalhães *et al*. 2002). The fate, source, transport and distribution of PO_4^{3-} within estuarine and marine systems are relatively complex. During nutrient loading periods, PO_4^{3-} may accumulate in the sediments.

Distinct from nitrogen compounds, a significant amount of PO_4^{3-} is bound nearly permanently within the sediment, while a smaller portion is found dissolved in the sediment pore waters (Benitez-Nelson 2000). PO_4^{3-} can be released into the overlying water column through dissolution, desorption and decomposition of organic matter. Also, dissolved PO_4^{3-} can be removed from the water column or pore water through biotic consumption, adsorption and authigenic precipitation (Lebo 1991). Typically sediment properties and chemical reactions affect the fluxes of PO_4^{3-} (Sundby *et al*. 1992; Mazouni *et al*. 1996).

Regenerated PO_4^{3-} can be absorbed in oxidising sediments, which reduces the flux into the overlying water (Sundby *et al*. 1992). Under favourable conditions PO_4^{3-} can be released into the overlying water from sediments where it is often trapped in oxidised iron compounds (Lavery and McComb 1991). PO_4^{3-} generally remains fixed to sediment particles except if the particles become anoxic or resuspended (Nedwell *et al*. 1999). Both non-biological and biological mechanisms of PO_4^{3-} binding are capable of reducing its production from sediments (Froelich *et al*. 1982; Benitez-Nelson 2000). PO_4^{3-} in the sediment can act as a buffing mechanism, where the sediments have the ability to control PO_4^{3-} in the water column and the buffering capacity is often a result of abiotic geochemical (precipitation/dissolution) and physical (adsorption/desorption) processes.

However, as previously discussed BMA also play a significant role, as PO_4^{3-} is an essential nutrient for biota (Froelich 1988). Consumption of PO_4^{3-} occurred consistently under both dark and light conditions. BMA would be expected to absorb enhanced PO_4^{3-} during the light period similar to the nitrogen fluxes. However, the lack of difference between PO_4^{3-} fluxes under dark and light conditions implies that photosynthesis by BMA was not directly accountable for the flux rates observed (Crisholm and Stross 1976; Magalhães *et al*. 2002; Spears *et al*. 2008). Sediment water fluxes are not affected by photosynthesis when PO_4^{3-} concentrations within the sediments are in excess of BMA requirements (Rizzo 1990). Therefore, as a non-biotic process PO_4^{3-} was bound to the sediment. Thus maintaining consistent PO_4^{3-} adsorption was probably due to the permanently aerobic sediment layer that resulted from the combination of the oxygenated water column and autotrophic nature (Bordalo 1991) of Currambene Creek sediments. In addition to PO_4^{3-} binding to sediments, the formation of Fe(III)-oxide and oxygenation surfaces may have stimulated PO_4^{3-} adsorption and prevented the removal of PO_4^{3-} from pore water, resulting in reduced PO_4^{3-} fluxes across SWI in Currambene Creek (Sundby *et al*. 1992).

It seems unlikely that BMA are responsible for the majority of nitrogen assimilation and take no part in phosphorus assimilation. The significant negative correlation between PO_4^{3-} and NPP rates suggests that the current PO_4^{3-} rates are influenced by BMA assimilation and oxygen (data not shown). Similar correlations were established by Engelsen *et al*. (2008) who argued that BMA and oxygen content from shallow marine ecosystems off the west coast of Sweden control PO_4^{3-} fluxes.

Eyre and Ferguson (2002) established that PO_4^{3-} can constantly (day and night) be absorbed by BMA while Gürel *et al*. (2005) suggested that PO_4^{3-} can be stored by BMA as it is a non toxic nutrient. Although there is no detectable difference between dark and light in Currambene Creek for PO_4^{3-} fluxes, there was a noticeable enhanced consumption of PO_4^{3-} during dark conditions. These PO_4^{3-} concentrations probably reflect BMA's ability to assimilate and store PO_4^{3-} during dark conditions and reflect the overnight maintenance of the PO_4^{3-} binding oxic layer (Eyre and Ferguson 2002).

A laboratory study by Sundback and Graneli (1988) found that when the amount of light was $\geq$ 10 $\mu E\ m^{-}2s^{-1}$ the BMA prevent the production of PO_4^{3-} from the sediment. Similar results were established by Spears *et al*. (2008) where high light attenuation resulted in no difference between dark and light PO_4^{3-} fluxes. In the present study light incubations were conducted under 450 $\mu E\ m^{-2}s^{-1}$ and therefore BMA under these light conditions could aid in the consumption of PO_4^{3-} from the sediment as production of PO_4^{3-} from the sediment was prevented. Additionally, Webb and Eyre (2004) explained that consumption of PO_4^{3-} occurred during light conditions due to the assimilation of BMA, as BMA are considered a major mediator of PO_4^{3-} fluxes.

Differences in PO_4^{3-} among treatments

PO_4^{3-} consumption occurred across all treatments, with PO_4^{3-} in only the trample sediments significantly different to the control sediments in Currambene Creek. While the trample pump sediments did demonstrate increased PO_4^{3-} consumption compared to the control, it was not statistically different from either the control or trampled sediments. The trample sediment showed a 73.4% enhanced consumption of PO_4^{3-} and trample pump sediment increased PO_4^{3-} consumption by 51.3%.

The production and consumption of PO_4^{3-} in sediments is determined by various mechanisms (Wang 2003). PO_4^{3-} is a redox sensitive nutrient where changes to the redox potential at the SWI influence nutrient flux between sediment pore waters and the overlying water column (Paytan and McLaughlin 2007; Sondergaard 2007). Sediment redox potential influences oxidation conditions that subsequently impact the chemical binding of PO_4^{3-} (Sondergaard 2007). The binding potential of iron and manganese under varying redox conditions control sediment PO_4^{3-} production, with oxidised conditions supporting iron in its particulate oxidised form (Fe (III)) that possesses a high affinity for binding PO_4^{3-}.

In contrast, anoxic conditions convert iron to a dissolved form (Fe(II)) so that adsorbed PO_4^{3-} becomes dissolved and is ultimately released (Penn *et al*. 2000; Paytan and McLaughlin 2007). A study on the impact of bait pumping in Coronet Bay, Victoria, highlighted that the destruction of burrows and compaction of sediments due to trampling and bait pumping created reducing conditions to depths of 20 cm in the sediment (Contessa and Bird 2004). The reducing conditions remobilised previously bound PO_4^{3-} releasing it into the pore water from the reduction of iron oxides (Sundby *et al*. 1992; Jensen *et al*. 1995). However, the presence of BMA at the sediment surface potentially intercepts the PO_4^{3-} and prevents it from releasing into the water column.

Redox potential in the present study was not measured and it is uncertain whether the disturbances caused any changes to it. Therefore it is possible that despite the disturbances causing mixing of oxic and anoxic environments the sediments could have sustained low but sufficient iron in an oxidised form binding the PO_4^{3-} and

preventing its production from the sediments. Also, the mixing from bait pumping may have enhanced iron from sediments particles from O_2 depleted environments and mixed them up into O_2 enriched sediments increasing the binding potential of PO_4^{3-} (Sondergaard 2007).

A more probable explanation for the enhanced consumption of PO_4^{3-} in the trample sediments is the influence that the absence of macrofauna had on the PO_4^{3-} flux. The macrofauna in the trample sediments were potentially killed due to burrows collapsing and sediment compaction. As discussed previously the burrows of macrofauna increase transport rates of nutrients, in this case PO_4^{3-} from the sediment to the overlying water through the pumping of water and remixing of organic matter (Hansen and Kristensen 1998; Devine and Vanni 2002). Without the burrows and the macrofauna the transport of PO_4^{3-} from sediment to the overlying water is drastically reduced, accounting for the enhanced sedimentary consumption of PO_4^{3-} as it cannot be transported to the overlying water and is retained within the sediments.

For instance, Webb and Eyre (2004) established that a net production of PO_4^{3-} was seen in sediments containing the crustacean *T. australiensis* compared to sediments in their absence. They postulated that PO_4^{3-} production stimulated 5-fold greater irrigation rates, where transport resulted in net source of PO_4^{3-} to the water column. It is well known that micro-environments adjacent to the burrow walls influence microbial activity and solute distribution (Papaspyrou *et al*. 2007). Microbial processes can increase transport rates due to gas ebullition. These bubbles physically bring up nutrient containing water rising through the sediments (Kamp-Nielsen 1975). The lack of microbial activity due to burrows collapsing and the absence of

macrofauna contribute to the retention of PO_4^{3-} within the sediments and consequently the enhanced sedimentary consumption of PO_4^{3-} in the trample and trample pump sediment.

Finally, production rates in the trample sediments showed a slight increase compared to the control and trample pump sediments. Therefore as productivity was slightly higher the demand for nutrients was also potentially higher, hence accounting for the enhanced consumption of PO_4^{3-} in the trampled sediments of Currambene Creek.

Chapter 5

Thesis synopsis, management implications and future research

5.1 Synopsis

Benthic Metabolism

The benthic metabolism results signify the vital importance that macrofauna and especially BMA have on O_2 exchange rates at the SWI in Currambene Creek. BMA are important ecological contributors in estuarine soft-sediments, providing approximately 30-50% of primary production in such systems (Underwood and Paterson 1993; de Jonge and Colijn 1994; MacIntyre *et al.* 1996; Underwood and Kromkamp 1999). BMA were present at the sediment surface as indicated by chl-*a* concentrations and by the reduced consumption of O_2 by the sediments during light incubations compared to dark incubations. Also, of macrofauna can influence benthic respiration and production rates directly through their physiological and physical activities (Nedwell and Walker 1995; Dunn 2009). This reduces O_2 that is available for sedimentary consumption and therefore explains the difference between undisturbed and disturbed treatments for BCR, where the control treatment had an increased level of sedimentary O_2 consumption. In contrast to BCR, GPP showed no difference between undisturbed and disturbed sediments due to the presence of BMA and their rapid growth and recolonisation rates. Further, the resuspension of the BMA attached to sediments caused by bait pumping and trampling could have contributed to similar rates of production between the control and disturbed soft-sediments.

Overall, NEM showed significant differences between the control and the disturbed soft-sediments, with the largest difference found between the control and trample treatments. Hunt (2011) also showed this result but for macrofauna, where macrofauna decreased by 90% in trampled plots while a decrease of only 10% was found in trample pump versus control treatments. These results were further supported by Wynberg and Branch (1994) and Contessa and Bird (2004) in similar studies investigating the effects of bait pumping and trampling. With GPP exceeding respiration across all treatments the Currambene Creek sand flat was deemed as autotrophic. Generally, the disturbances of trample and trample pump had a limited influence on productivity but considerable influence on respiration. Based on the results from this study a conceptual model was developed for benthic metabolism in Currambene Creek illustrating the effects that disturbances such as bait pumping and trampling (i.e. common recreational activities) have on respiration and production (Figure 5.1).

Benthic Nutrients

The different forms of nitrogen consumed by the soft-sediments was consistent and dominated by NH_4^+. Also, sedimentary consumption of all nutrients occurred during both dark (night) and light (day) conditions. NO_x was the only exception as under dark conditions a small release was seen and under light conditions consumption occurred.

Once more, BMA illustrate their dominance in influencing nutrient fluxes with their presence confirmed by the chl-*a* concentrations. All forms of nitrogen showed significant differences between dark and light fluxes and the BMA were accountable for some of the differences between dark and light fluxes for all nitrogen fluxes.

Nitrogen can be assimilated by BMA as NO_2^-, NO_3^-, NH_4^+ or N_2 with the preferred form being NH_4^+ (Tandeau de Marsac and Houmard 1993; Hart and Grace 2000). BMA have the capacity to metabolise under dark and light conditions (e.g. Rysgaard *et al*. 1993) and can prevent the release of sediment-derived nutrients to the water column by directly interrupting the diffusion of nutrients to meet their autotrophic needs (Potts *et al*. 2005).

As discussed, the only form of nitrogen to show a difference between control and disturbed treatments was NH_4^+. BMA has been discussed as a likely cause of the difference but the reduced numbers of macrofauna in the disturbed sediments could have also contributed. The excretion of NH_4^+ from the macrofauna was proposed for the difference between control and disturbed sediments. While the results appear to suggest that greater consumption occurred in the disturbed treatments, there was in fact less NH_4^+ excretion due to a decrease in macrofauna and subsequently NH_4^+ was being removed from the sediments into the water column.

NO_x and DIN fluxes demonstrated consistent sedimentary consumption across all treatments. The presence or absence of macrofauna seemed to have little influence on these N forms. Ferguson *et al*. (2004) demonstrated that DIN assimilation by macrofauna was significantly lower than that for BMA. As discussed throughout this thesis the presence of BMA has had a profound influence on the benthic nutrients in the sediments from Currambene Creek and possibly accounted for the lack of variation found between NO_x and DIN fluxes in the control and disturbed sediments. Furthermore, production rates across all treatments were consistent, facilitating the similar DIN and NO_x consumption rates among the control and disturbed sediments.

Unlike nitrogen, phosphorus fluxes showed no difference between dark and light fluxes. Distinct from nitrogen compounds, a significant amount of PO_4^{3-} is bound nearly permanently within the sediment, while a slighter portion is found dissolved in sediment pore waters (Benitez-Nelson 2000). Both non-biological and biological mechanisms of phosphorus binding are capable of reducing its release from sediments (Froelich *et al*. 1982; Benitez-Nelson 2000). It was thought that the similarity between dark and light phosphorus fluxes was because the phosphorus was bound to the sediment. Maintaining consistent phosphorus adsorption was probably due to the permanently aerobic sediment layer resulting from the combination of the oxygenated water column and autotrophic nature (Bordalo 1991) of the Currambene Creek intertidal sediments. Finally, phosphorus can constantly (day and night) be absorbed and stored by BMA (Eyre and Ferguson 2002) as it beneficial in trace concentrations (Gürel *et al*. 2005).

In summary nitrogen and phosphorus consumption occurred across all treatments, with NO_x the only exception, as under dark conditions a small release occurred. Nitrogen showed significant variation between dark and light fluxes, while phosphorus showed no significant difference between dark and light fluxes. NH_4^+ was the only form of nitrogen that showed a significant difference between the control and treatment sediments. While BMA has been discussed as a likely cause of the difference, it is likely that the reduced numbers of macrofauna in the treatment sediments could also have contributed to the differences found.

In contrast to nitrogen, phosphorus in the trample treatments was significantly different to the control and the actual physical disturbance of the trampling rather than the presence or absence of macrofauna was the suspected cause of variation found. Based on the results from this study conceptual models were developed for the Currambene Creek benthic metabolism (Figure 5.1) and nutrients (Figures 5.2 – 5.5), illustrating the affects the disturbed treatments had on nutrient fluxes.

Currambene Creek Nutrient Budget

The tidal mudflats of Currambene Creek are a net sink for both nitrogen and phosphorus. Nutrient assimilation by the BMA reduced the production of remineralised nutrients from the sediments to the overlying water, effectively decoupling nutrient turnover with the sediments from the water column processes. It was concluded that the BMA were completely filtering out or intercepting the nutrient fluxes across the SWI and subsequently reducing the nutrient availability for bacteria, macroalgal and phytoplankton.

The behaviour of nutrients within estuarine systems is based on the molar ratio of material likely to be reacting in the system. It is assumed that this material can be described by the Redfield C:N:P ratio as 106:16:1 (Qu 2004; Eyre and Ferguson 2005). Nitrogen to phosphorus stoichiometric estimates based on these ratios normally see the sediments approaching the conventional Redfeild ratio at N:P = 16. In the present study, all treatments had N: P ratios (N:P =12) slightly lower than expected (Table 5.1). This suggests that the sediments were accumulating both nitrogen and phosphorus and the BMA were growing at slower than expected.

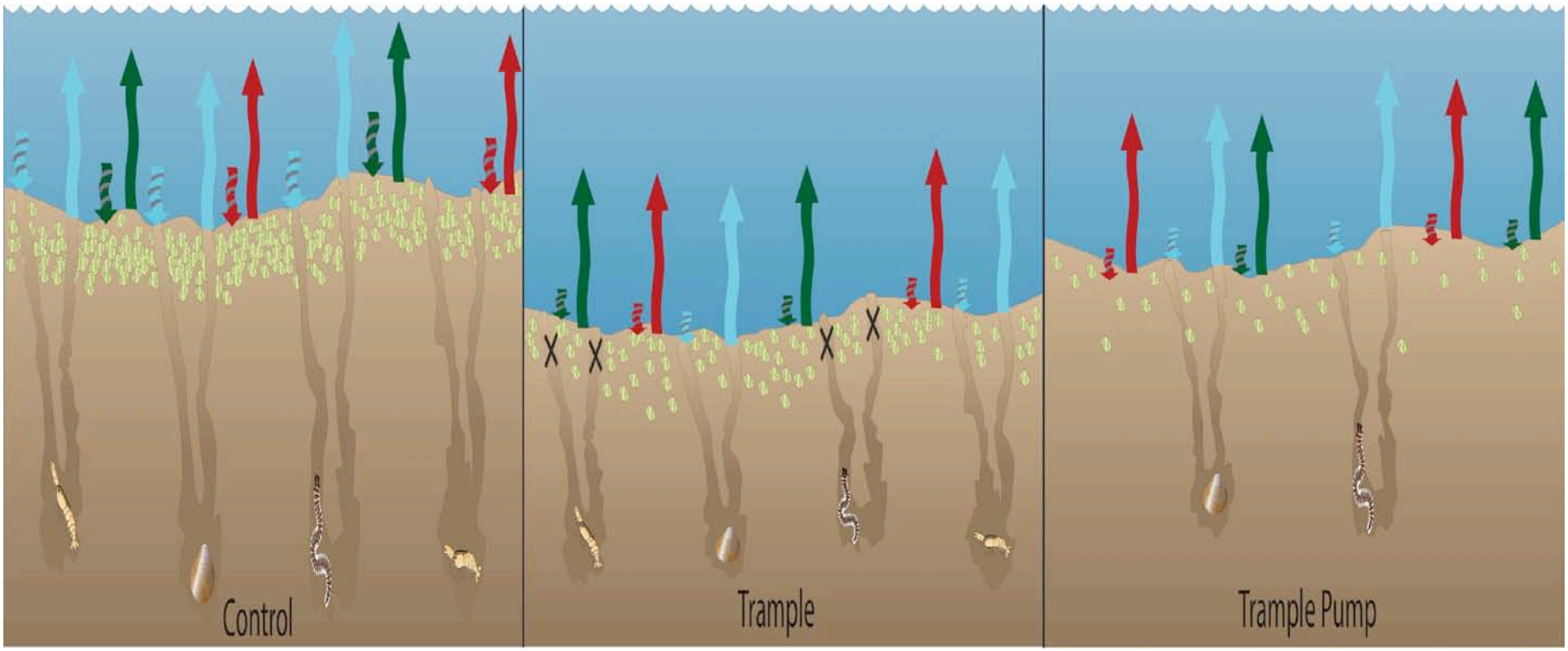

Figure 5.1 Conceptual diagram of treatment affects on O_2 fluxes. The control treatment had greater BCR rates due to increased number of macrofauna. The trample and trample pump treatments had reduced BCR rates due to the reduced number of macrofauna. GPP rates are similar for all treatments due to BMA. Arrows with strips = BCR. Arrows without strips = GPP. Blue arrows = macrofaunal O_2. Red arrows = microbial O_2. Green arrows = BMA O_2. X = closed burrows due to compaction.

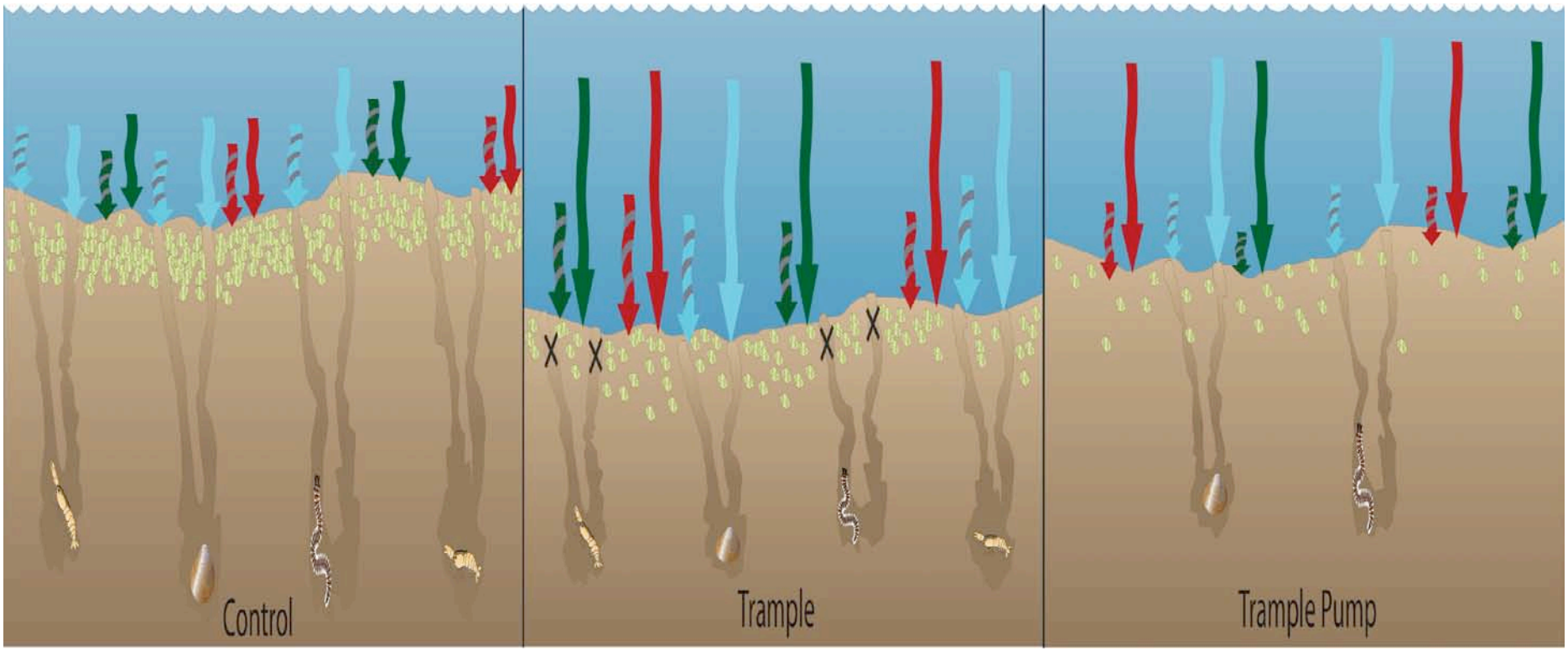

Figure 5.2 Conceptual diagram of treatment affects on NH_4^+ fluxes during dark (striped arrows) and light (arrow without stripes) conditions. The control treatment had reduced consumption due to the increased number of macrofauna excreting NH_4^+. Trample and trample pump treatments had enhanced NH_4^+ consumption due to the reduced number of macrofauna excreting NH_4^+ in the sediment and therefore sequestering NH_4^+ from the water column. Blue arrows = macrofaunal NH_4^+. Red arrows = microbial NH_4^+. Green arrows = BMA NH_4^+. X = closed burrows due to compaction.

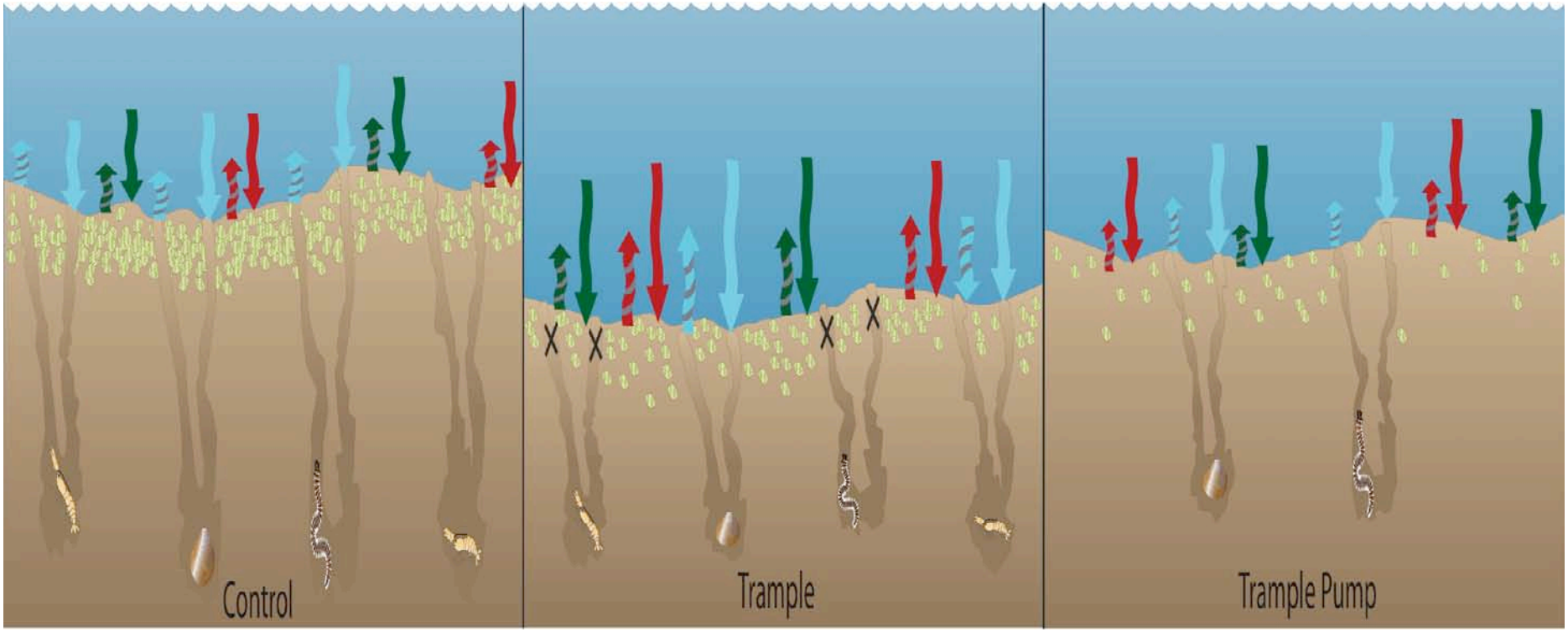

Figure 5.3 Conceptual diagram of treatment affects on NO_x fluxes during dark (striped arrows) and light (arrow without stripes) conditions. The presence of BMA dominated the shallow system and probably caused a lack of variation between NO_x fluxes from the undisturbed (control) and disturbed (trample and trample pump) treatments. A Significant difference between dark and light fluxes was also attributed to BMA. Blue arrows = macrofaunal NO_x. Red arrows = microbial NO_x. Green arrows = BMA NO_x. X = closed burrows due to compaction.

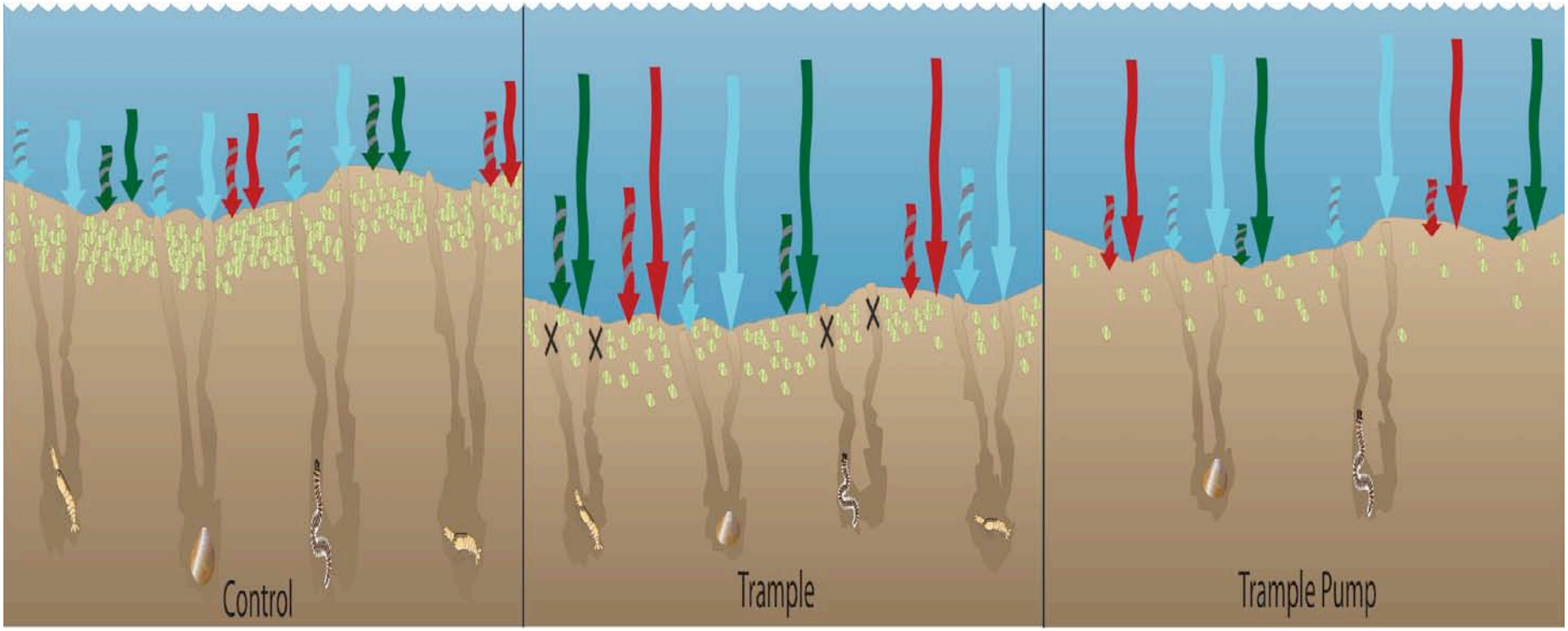

Figure 5.4 Conceptual diagram of treatment affects on DIN fluxes during dark (striped arrows) and light (arrow without stripes) conditions. The presence of BMA dominated the shallow system and probably caused a lack of variation between DIN fluxes from the undisturbed (control) and disturbed (trample and trample pump) treatments. A significant difference between dark and light fluxes was also attributed to BMA. Blue arrows = macrofaunal DIN. Red arrows = microbial DIN. Green arrows = BMA DIN. X = closed burrows due to compaction.

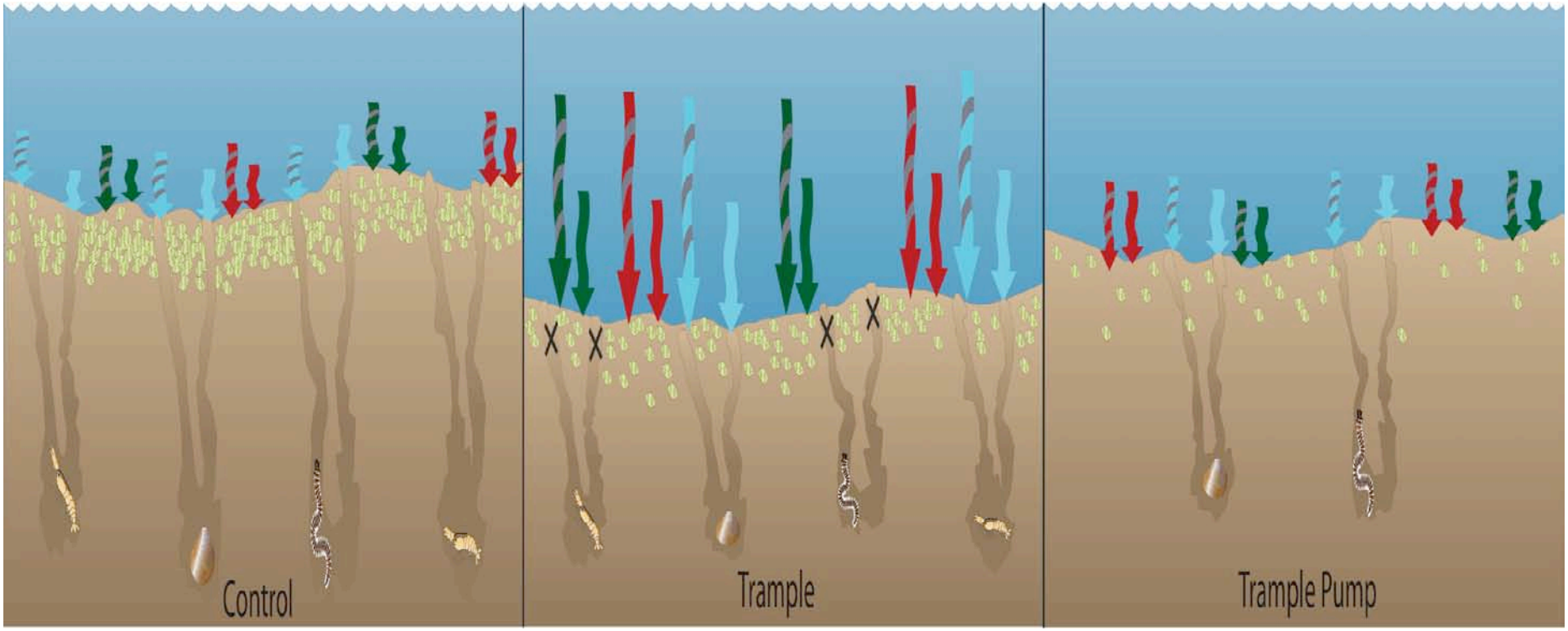

Figure 5.5 Conceptual diagram of treatment affects on PO_4^{3-} fluxes during dark (striped arrows) and light (arrow without stripes) conditions. The lack of difference between dark and light PO_4^{3-} fluxes was due to it being bound to the sediment and sequestered by BMA. PO_4^{+} in only the trample sediment was significantly different to the control sediments. This was attributed to the physical trampling disturbance altering the redox potential and subsequent the binding affinity of the PO_4^{+} to sediment particles. Blue arrows = macrofaunal PO_4^{3-}. Red arrows = microbial PO_4^{3-}. Green arrows = BMA PO_4^{3-}. X = closed burrows due to compaction.

The net requirements of the BMA were significantly higher than what the sediments could produce for both nitrogen and phosphorus across all treatments (Table 5.1).

Consequently, the sediment nutrient sources were insufficient in meeting the demands of BMA, thereby creating a downward flux of nutrients from the water column to the benthic community. The sediments supplied only 8 – 17% of the required nitrogen and phosphorus nutrients in Currambene Creek across all treatments. Whereas, the water column nutrients were sequestered to aid in the nutrient demands of the sediments. The water column nutrients supplied 82 – 91% of the nitrogen and phosphorus across all treatments to the tidal flat sediments.

Generally nitrogen and phosphorus showed reduced consumption during dark conditions and enhanced consumption under light conditions. During dark conditions nutrients were in less demand due to the breakdown of organic matter to produce nitrogen and phosphorus. Therefore, during dark conditions nutrients are produced and less are required from the overlying water column, accounting for the reduced consumption of nutrients during dark incubations. Alternatively, during light conditions the requirement of inorganic nitrogen and phosphorus are high, ultimately accounting for the enhanced consumption from the water column as nutrients are drawn out of the water column.

The trample treatment had the highest production rates. As a result, the high level of production requires enhanced concentrations of nitrogen and phosphorus. This was seen in the enhanced consumption of nitrogen and phosphorus in the disturbed trample treatment in comparison to the undisturbed control and disturbed trample

pump treatments. Further, nutrient budget calculations (Table 5.1) show that the two disturbed sediment types are producing less nutrients than the undisturbed control, but ultimately require more than the control sediments. The competition for nitrogen and phosphorus resources in the disturbed treatments is enhanced and the sediments appear to be increasing the limited nutrient. This active competition for limited nitrogen resources suggests recycling conservation of nutrients in Currambene Creek.

The net daily demand across all treatments is markedly higher than the sediment can produce (Table 5.1), as the sediments are nutrient limited in respect to both nitrogen and phosphorus. This is not surprising as Australian estuaries naturally have predominately low inputs of nutrients due to its terrestrial soils having low nutrient concentrations as discussed in Blakey (2005).

Table 5.1 Nutrient budgets for nitrogen and phosphorus of undisturbed and disturbed sediments.

		Net Remineralised	Net Assimilated	Net Produced	Net Required	Net daily demand
Control	N	74.7	-429.86	896.1	-5158.3	**-4262.2**
	P	6.3	-36.5	76.1	-437.9	**-361.9**
Trample	N	43.4	-493.6	521.2	-5923.6	**-5402.4**
	P	3.6	-41.9	44.3	-502.9	**-458.7**
Trample pump	N	43.7	-415.6	524.6	-4987.7	**-4463.1**
	P	3.7	-35.3	44.5	-423.5	**-378.9**

5.2 Management Implications

Currambene Creek sanctuary zone is managed under the Marine Parks Act 1997 in which it aims to conserve biodiversity and maintain ecological processes. Currently only Winberg (2008) has investigated the affect that establishing a sanctuary zone has had on conserving the intertidal soft sediment biodiversity of Currambene Creek. Sampling on the tidal flat by Winberg (2008) began one year before and two years after the development of the sanctuary zone (BACI design) (Underwood 1997).

The implementation of the sanctuary zone demonstrated that there was a dramatic increase in macrofaunal assemblages. In a parallel study, Hunt (2011) assessed the influence trampling and bait pumping had on the same soft sediment assemblages as Winberg (2008) and found a significant treatment affect, where trampling then bait pumping had the greatest impact on macrofauna. These studies demonstrate that establishing the sanctuary zone has assisted in conserving macrofaunal biodiversity. Allowing human activity on the intertidal flat now would impact upon the soft sediment macrofauna communities and the sediments and potentially place the soft sediments and their benthic communities at risk. While these studies focus on conserving biodiversity, there is still limited research on whether the ecological processes of the soft-sediments are being maintained within sanctuary zones. Ecological processes are complex interactions that are difficult to measure/investigate but individually are vital to the health of a sanctuary zones and as such management strategies that address them must be drafted (Bennett *et al*. 2009).

This study demonstrated that trampling and bait pumping (common recreational activities) impacted upon the Currambene Creek sanctuary zone ecological processes,

of benthic metabolism, nutrient cycling, respiration rates and on some nutrients. Whereas, production and other nutrient (e.g. DIN) were not affected by the disturbances. Moreover, a dramatic change to these ecological processes would have seen the disturbances change the net autotrophic characteristics of the sediments into net heterotrophic sediment, though this did not happen during the study.

These complex and varied results give no definitive answer as to whether the sanctuary zone is maintaining these processes. Despite this, there are several reasons that could have diluted the effects the disturbances had on the sediments of Currambene Creek. The sanctuary zone was established in October 2002 and has been an anthropogenically undisturbed ecosystem since. This extended period free from disturbances has allowed the system to build up a resistance to disturbance and potentially buffer against the experimental disturbances implemented in the current study. It is speculated that increasing the duration of the experimental disturbances could dramatically shift and change the benthic metabolism and nutrient cycling within the MPA. Additionally, the presence of BMA in the shallow sandy sediments dominated productivity. If experimentations were untaken in deeper water where BMA productivity was less dominant, then there may have been a discernable effect on benthic metabolism and nutrient cycling, particularly in nitrificaiton-denitrification pathways.

While it is difficult to establish whether the Currambene Creek sanctuary zone is helping maintain all of its ecological processes, there is no doubt that trampling and bait pumping had a dramatic influence on the soft sediment macrofaunal communities during the study (Winberg 2008; Hunt 2011). Current management of the zone

completely prohibits all activities that remove organisms or interferes with their habitats. This management is effectively conserving biodiversity (Winberg 2008; Hunt 2011) and it is proposed that it is maintaining ecological processes. However, to be completely certain further investigation is required and replication in other protected habitats is recommended.

5.3 Research implications

The work conducted in the Currambene Creek sanctuary zone in this thesis provided essential nutrient cycling benchmarks for the management of NSW estuaries and sanctuary zones from which future changes or disturbances can be assessed. This data can also be used for baseline referencing for surrounding habitats as intertidal estuaries are often used to evaluate the health of their neighboring marine ecosystems (Kaiser *et al*. 2001; Contessa and Bird, 2004; Mikac *et al*. 2007). Trampling of the Currambene Creek mudflats had a significant influence on BCR rates and macrofaunal abundance. These results are important from a management perspective as allowing people to walk on these areas can cause just as much, if not more damage to the mudflat habitat than the bait pumping itself.

Further, changes in benthic metabolism and nutrient cycling due to the experimental disturbances suggested that in Currambene Creek sanctuary zone BMA influence biogeochemical processes more than benthic macrofauna. However, the macrofauna still had a significant influence on respiration and NH_4^+ rates. Although, evaluating the relationship between benthic biodiversity and nutrient cycling in estuarine intertidal mudflats is complicated and the methods and aims presented in the current study can nevertheless be used as a guide for future research.

5.4 Future Research

The investigation of ecological processes in soft-sediment needs further work. Due to the resilient nature of the sanctuary zone examined in this study the duration that experimental disturbances are implemented for can effectively determine the threshold or buffering capacity of the system. Also, extending the scope of the study to include multiple sites within and outside sanctuary zones would give a comprehensive analysis of the differences between protected and non-protected areas. The link between macrofauna and ecological processes are complex and significantly influence the biogeochemical processes at the SWI (Qu 2004; Dunn 2009). The current study showed that macrofaunal communities had a limited affect on benthic metabolism and nutrient cycling due to the dominant presence of BMA. In future studies core samples taken from deeper water where BMA productivity is less may see discernable effects on nitrogen and phosphorus cycling, particular nitrification and denitrification pathways in regards to the presence or absence of macrofauna.

Further, determining the recovery periods of these site specific macrofaunal assemblages will help assist natural resource managers estimate predicted recovery rates from anthropogenic disturbances specifically bait pumping and trampling. If recovery rates are rapid, it may encourage partial opening of the sanctuary zone for human activity throughout the year.

5.5 Final remarks

This study presents a snapshot of the sediment properties, benthic metabolism and benthic nutrients in Currambene Creek sanctuary zone tidal flat during summer; with

specific focus on the influence human disturbances in the form of trampling and bait pumping have on these variables. The nutrient fluxes provide essential nutrient cycling benchmarks for the management of NSW estuaries and sanctuary zones. From this data, effective management decisions can be made and future changes from human pressures can be assessed accordingly. The main conclusions of this study are (Table 5.2):

1. The soft-sediment of Currambene Creek sanctuary zone tidal flat were net autotrophic and a net sink for both nitrogen and phosphorus for both undisturbed and disturbed treatments;
2. The disturbances of trampling and bait pumping had a significant influence on benthic community respiration, which were due to a reduction in macrofaunal abundance;
3. The disturbances had a limited affect on gross primary productivity and BMA production;
4. BMA production dominated nitrogen and phosphorus cycling and consequently the disturbances had little effect on nitrogen and phosphorus cycling; and
5. Sanctuary zones are vital for the conservation of soft sediment benthic macrofaunal communities as the disturbances greatly reduced abundances. However, further investigation is needed to completely verify if sanctuary zones are maintaining ecological processes.

Table 5.2 Summary of measured sediment variables and biogeochemical processes studied in undisturbed and disturbed sediments in Currambene Creek sanctuary zone. All O, N and P fluxes are measured in µmol.$m^{-2}hr^{-1}$. * = Refer to parallel study by Hunt (2011).

Treatment	Control	Trample	Trample pump	Notes	Thesis Section
Sediment types	Sand	Sand	Sand	92.4 % sand, 6.3% silt and 1.3% clay	Hunt (2011)*
Chl-*a* rates (mg m^2)	24.6	19.9	11.3	C ≠ T = TP	Ch 3 pg. 52
Macrofaunal abundances (m^{-2})	634	396	270	92% Mollusc	Hunt (2011)*
BCR flux	-672.1	-390.9	-393.5	C ≠ T = TP	Ch 3 pg. 54
GPP flux	4540.8	4833.6	4134.3	C = T = TP	Ch 3 pg. 55
NEM flux	1598.3	2025.9	1673.7	C ≠ T = TP, Autotrophic	Ch 3 pg. 55
NH_4^+ Dark (Light) flux	-9.1 (-14.1)	-17.1 (-26.3)	-13.1 (-26.4)	D ≠ L, No diel variation	Ch 3 pg. 57
NH_4^+ Net flux	-11.7	-21.7	-19.7	C ≠ T = TP, Sediment sink	Ch 3 pg. 57
NO_x Dark (Light) flux	0.5 (-4.5)	1.3 (6.1)	0.4 (-4.9)	D ≠ L, Small diel variations	Ch 3 pg. 59
NO_x Net flux	-1.9	-2.4	-2.3	C = T = TP, Sediment sink	Ch 3 pg. 59
DIN Dark (Light) flux	-8.6 (-18.7)	-15.8 (-32.3)	-12.7 (-31.3)	D ≠ L, No diel variation	Ch 3 pg. 61
DIN Net flux	-13.6	-24.1	-21.9	C = T = TP, Sediment sink	Ch 3 pg. 61
PO_4^{3-} Dark (Light) flux	-0.5 (-0.3)	-1.8 (-1.0)	-0.7 (-0.7)	D = L, No diel variation	Ch 3 pg. 63
PO_4^{3-} Net flux	-0.4	-1.4	-0.7	C = TP ≠ T, Sediment sink	Ch 3 pg. 63

References

An S, Joye SB (2001) Enhancement of coupled denitrification by benthic photosynthesis in shallow subtidal estuarine sediments. Limnology and Oceanography 46: 62-74.

Arthur CR, Rigler FH (1967) A Possible Source of Error in the 14C Method of Measuring Primary Productivity. Limnology and Oceanography 12: 121-124.

Asmus RM, Jensen MH, Jensen KM, Kristensen E, Asmus H, Wille A (1998) The role of water movement and spatial scaling for the measurement of dissolved inorganic nitrogen fluxes in intertidal sediments. Estuarine, Coastal and Shelf Science 46: 221-232.

Asmus RM, Sprung M, Asmus H (2000) Nutrient fluxes un intertidal communities of a South European lagoon (Ria Formosa) - similarities and differences with a northern Wadden sea Bay (Sylt-Romo Bay). Hydrobiologia 436: 217-235.

Baric A, Kuspilic G, Matijevic S (2002) Nutrient (N, P, Si) fluxes between marine sediments and water column in coastal and open Adriatic. Hydrobiology 475/476, 151-159. Hydrobiologia 475/476: 151-159.

Bartoli M, Nizzoli D, Viaroli P (2003) Microphytobenthos activity and fluxes at the sediment-water interface: interactions and spatial variability. Aquatic Ecology 37: 341-349.

Bartoli M, Nizzoli D, Welsh DT, Viaroli P (2000) Short-term influence of recolonisation by the polychaete worm *Nereis succinea* on oxygen and nitrogen fluxes and denitrification: a microcosm simulation. Hydrobiologia 431: 165-174.

Beardall J, Light B (1994) Biomass, Productivity and Nutrient Requirements of Microphytobenthos. CSIRO. Melbourne: 20 p.

Benitez-Nelson CR (2000) The biogeochemical cycling of phosphorus in marine systems. Earth- Science Reviews 51: 109-135.

Bennett AF, Haslem A, Cheal DC, Clarke MF, Jones RN, Koehn JD, Lake PS, Lumsden LF, Lunt ID, Mackey BG, Mac Nally R, Menkhorst PW, New TR, Newell GR, O'Hara T, Quinn GP, Radford JQ, Robinson D, Watson JEM, Yen AL (2009) Ecological processes: A key element in strategies for nature conservation. Ecological Management and Restoration 10: 192-199.

Berelson W, Heggie D, Longmore A, Kilgore T, Nicholson G, Skyring G (1998) Benthic nutrient recycling in Port Phillip Bay, Australia. Estuarine, Coastal and Shelf Science 46: 917-934.

Berelson WM, Kilgore T, Heggie D, Skyring G, Ford P (1996) Benthic Chamber, Nutrient Fluxes and the Biogeochemistry of the Seafloor of Port Phillip Bay 1995 & 1996. Australian Geological Survey Organization. Canberra. Pp. 89.

Bertics VJ, Sohm JA, Treude T, Chow CT, Capone D, Fuhrman JA, Ziebis W (2010) Burrowing deeper into benthic nitrogen cycling: the impact of bioturbation on nitrogen fixation coupled to sulfate reduction. Marine Ecology Progress Series 409: 1-15.

Binnerup SJ, Jensen KM, Revsbech NP, Jensen MH, Sørensen J (1992) Denitrification, dissimilatory reduction of nitrate to ammonium, and nitrification in a bioturbated estuarine sediment as measured with 15N and microsensor techniques. Applied and Environmental Microbiology 58: 303-313.

Blackburn TH, Henriksen K (1983) Nitrogen cycling in different types of sediments from Danish waters. Limnology and Oceanography 28: 477-493.

Blackburn TH (1987) Microbial food webs in sediments in Sleigh MA (editor) Microbes in the sea. New York: Wiley. 39-58 pp.

Blakey C (2005) The significance of sediment type and light in the flux of nutrients from the sediment to the water column in Bellambi Lagoon. Honours Thesis. University of Wollongong, Australia.

Boers P, Van Raaphorst W, Van der Molen DT (1998) Phosphorus retention in sediment Water Science and Technology 37: 31-39.

Bolalek J, Graca B (1996) Ammonium nitrogen at the water-sediment interface in Puck By (Baltic Sea). Estuarine Coastal and Shelf Science 43: 767-779.

Bonin P, Raymond N (1990) Effects of oxygen on denitrification in marine sediments. Hydrobiologia 207: 115-122.

Bordalo AA (1991) Ecology of River Douro estuary. Spatial and temporal relationships in the microbial compartment. PhD thesis, University of Porto, Portugal.

Bridgewater P, Ivanovici A (1993) Achieving a representative system of marine and estuarine protected areas for Australia in Ivanovici AM, Tarte D, Olsen M (editors) Protection of Marine and Estuarine Areas - A Challenge for All Australians. Proceedings of the Fourth Fenner Conference on the Environment, Canberra. 9-1 October 1991.

Cabrita MT, Brotas V (2000) Seasonal variation in denitrification and dissolved nitrogen fluxes in intertidal sediments of the Tagus estuary, Portugal. Marine Ecology Progress Series 202: 51-65.

Caffrey JM, Miller LG (1995) A comparison of two nitrification inhibitors used to measure nitrification rates in estuarine sediments. FEMS Microbial Ecology 17: 213-220.

Callender E, Hammond DE (1982) Nutrient exchange across the sediment-water interface in the Potomac River estuary. Estuarine Coastal and Shelf Science 15: 395-413.

Caraco NF, Lampman G, Cole JJ, Limburg KE, Pace ML, Fischer D (1998) Microbial assimilation of DIN in a nitrogen rich estuary: implications for food quality and

isotope studies. Marine Ecology Progress Series 167: 59-71.

Casu D, Ceccherelli G, Castelli A (2006) Immediate Effects of Experimental Human Trampling on Mid-Upper Intertidal benthic Invertebrates at the Asinara Island MPA (NW Mediterranean). Hydrobiologia 555: 271-279.

Chandrasekara WU, Frid CLJ (1996) Effects of human trampling on tidal flat infauna. Aquatic Conservation: Marine and Freshwater Ecosystems 6: 299-311.

Chanton JP, Martens CS (1987) Biogeochemical cycling in an organic-rich coastal marine basin. 7. Sulfur mass balance, oxygen uptake and sulfide retention. Geochemica et Cosmochimica Acta 51(1): 1187-1199.

Clavero V, Izquierdo JJ, Fernandez JA, Niell FX (2000) Seasonal fluxes of phosphate and ammonium across the sediment-water interface in a shallow small estuary (Palmones River, southern Spain). Marine Ecology Progress Series 198: 51-60.

Conley DJ, Stockenberg A, Carman R, Johnstone RW, Rahm L, Wulff F (1997) Sediment-water nutrient fluxes in the Gulf of Finland, Baltic Sea. Estuarine Coastal and Shelf Science 45: 591-598.

Contessa L, Bird FL (2004) The impact of bait-pumping on populations of the ghost shrimp *Trypaea australiensis* Dana (Decapoda: Callianassidae) and the sediment environment. Journal of Experimental Marine Biology and Ecology 302: 75-97.

Cook PLM, Butler ECV, Eyre BD (2004a) Carbon and nitrogen cycling on intertidal mudflats of a temperate Australian estuary. I. Benthic metabolism. Marine Ecology Progress Series 280: 25-38.

Cook PLM, Revill AT, Butler ECV, Eyre BD (2004b) Carbon and nitrogen cycling on intertidal mudflats of a temperate Australian estuary. II. Nitrogen cycling. Marine Ecology Progress Series 280: 39-54.

Cook PLM (2003) Carbon and nitrogen cycling on intertidal mudflats in a temperate Australian estuary. PhD. University of Tasmania, Australia.

Correll DJ (1998) The role of Phosphorus in the eutrophication of receiving waters: a review. Journal of Environmental Quality 27: 261-266.

Cowan JLW, Pennock JR, Boynton WR (1996) Seasonal and inter-annual patterns of sediment-water nutrient and oxygen fluxes in Mobile Bay, Alabama (USA): regulating factors and ecological significance. Marine Ecology Progress Series 141: 229-245.

Crisholm SW, Stross RG (1976) Phosphate uptake kinetics in *Euglena gracillis* (Euglenophyceae) grown on light/dark cycles. I. Synchronized batch cultures. Journal of Phycology 12: 210-217.

Cruz-Motta JJ, Underwood AJ, Chapman MG, Rossi F (2003) Benthic assemblages in sediments associated with intertidal boulder-Welds. Journal of Experimental Marine

Biology and Ecology 285: 383-401.

Dalsgaard T (2003) Benthic primary production and nutrient cycling in sediments with benthic microalgae and transient accumulation of macroalgae. Limnology and Oceanography 48: 2138-2150.

Dalsgaard T, Nielsen LP, Brotas V, Viaoli P, Underwood AJ, Nedwell DB, Sundback K, Rysgaard S. (2000) Protocol Handbook for NICE- Nitrogen Cycling in Estuaries. Ministry of Environment and Energy. Silkeborg, Denmark. 62 p.

de Jonge VN, Colijn F (1994) Dynamics of microphytobenthos biomass in the Ems estuary measured as chlorophyll *a* and carbon. Marine Ecology Progress Series 104: 185-196.

Department of Sustainability, Environment, Water, Population, and, Communities (2010) Marine Protected Areas. Accessed November 20th 2010, http://www.environment.gov.au/coasts/mpa/about/index.html#1

Dernie KM, Kaiser MJ, Warwick RM (2003) Recovery rates of benthic communities following physical disturbances. Journal of Animal Ecology 72: 1043-1056.

De Roach RJ, Rate AW, Knott B, M. DP (2002) Denitrification activity in sediment surrounding polychaete (*Ceratonereis aequisetis*) burrows. Mar. Freshwater Res. 53: 35-41.

Devine JA, Vanni MJ (2002) Spatial and seasonal variation in nutrient excretion by

benthic invertebrates in a eutrophic reservoir. Freshwater Biology 47: 1107-1121.

de Wit R, Stl LJ, Lomstein BA, Herbert RA, Van Gemerden H, Viaroli P, Ceccherelli VU, Rodriguez-Valera F, Bartoli M, Giordani G, Azzoni R, Schaub B, Welsh DT, Donnelly A, Cifuentes A, Anton J, Finster K, Nielsen LB, Underlien Pedersen A-E (2001) ROBUST: the role of buffering capacities in stabilising coastal lagoon ecosystems. Continental Shelf Research 21: 2021-2041.

Dunn RJK (2009) Biogeochemical processes of a sub-tropical coastal lagoon (Coombabah Lake, southern Moreton Bay, Australia): with emphasis on organic matter and nutrient dynamics. PhD thesis, Griffith University, Australia.

Dunn RJK, Welsh DT, Lee SY, Lemckert CJ, Teasdale PR, Meziane T (2008) Investigating the distribution and sources of organic matter in surface sediment of Coombabah Lake (Australia) using elemental, isotopic and fatty acid biomarkers. Continental Shelf Research 28: 2535-2549.

Edwards RW, Rolley HLJ (1965) Oxygen consumption of river muds. Journal of Ecology 53: 1-19.

Elderfield H, Luedtke NA, MCCaffret RJ, Bender M (1981) Benthic flux studies in Narragansett Bay. American Journal of Science 281: 768-787.

Engelsen A, Hulth S, Phil L, Sundback K (2008) Benthic trophic status and nutrient fluxes in shallow-water sediments. Estuarine Coastal and Shelf Science 78: 783-795.

Evans JLB (2005) The effect of benthic microalgal photosynthetic oxygen production on nitrogen fluxes across the sediment-water interface in a shallow, sub-tropical estuary. Masters thesis, University of Maryland, USA.

Eyre BD, Ferguson AJP (2002) Comparison of carbon production and decomposition, benthic nutrient fluxes and denitrification in seagrass, phytoplankton, benthic microalgae- and macroalgae- dominated warm-temperate Australian lagoons. Marine Ecology Progress Series 229: 43-59.

Eyre BD, Ferguson AJP (2005) Benthic metabolism and nitrogen cycling in a sub-tropical east Australian estuary (Brunswick): Temporal variability and controlling factors. Limnology and Oceanography 50: 81-96.

Eyre BD, Ferguson AJP (2009) Denitrification efficiency for defining critical loads of carbon in shallow coastal ecosystems. Hydrobiologia 629: 137-146.

Field, A. 1998. A bluffer's guide to sphericity. The British Psychological Society: Mathematical, Statistical and Computing Section (BPS–MSC) Newsletter, 6 (1):13–22.

Ferguson AJP, Eyre BD, Gay JM (2004) Benthic nutrient fluxes in euphotic sediments along shallow sub-tropical estuaries, northern NSW, Australia. Aquatic Microbial Ecology 37: 219-235.

Ferguson AJP (2002) A comparison of benthic nutrient cycling in the sub-tropical

Sandon and Brunswick Estuaries. PhD Thesis. Southern Cross University, Australia.

Fisher MM, Reddy KR (2001) Phosphorus Flux from Wetland Soils Affected by Long-Term Nutrient Loading. Journal of Environmental Quality 30: 261-271.

Fisher TR, Carlson PR, Barber RT (1982) Carbon and nitrogen primary productivity in three North Carolina estuaries. Estuarine, Coastal and Shelf Science 15: 621-644.

Fogg GE (1982) Nitrogen cycling in sea water. Philosophical Transactions Royal Society London 296: 511-520.

Forja JM, Gomez-Parra A (1998) Measuring Nutrient Flux Across the Sediment-Water Interface Using Benthic Chambers. Marine Ecology Progress Series 164: 95-105.

Froelich PN, Luedtke NA, Heap A, DeVries T (1982) The marine phosphorus cycle. American Journal of Science 282: 475-511.

Froelich PN (1988) Kinetic control of dissolved phosphate in natural rivers and estuaries: A primer on the phosphate buffer mechanism. Limnology and Oceanography 33: 649-668.

Fry B, Lutes R, Northam M, Parker PL, Ogden J (1982) A 13C/12C comparison of food webs in Caribbean seagrass meadows and coral reefs. Aquatic Botany 14: 389-398.

Furnas MJ (1992) The behaviour of nutrients in tropical aquatic ecosystems. In: Connerll DW, Hawker DW (Editors) Pollution in tropical aquatic systems. CRC Press, Inc.: Florida. 30-53 pp.

Gay JM (2002) Pelagic and benthic metabolism in three sub-tropical Australian estuaries. PhD Thesis, Southern Cross University, Australia.

Galloway JN, Dentener FJ, Capone DG, Boyer EW, Howarth RW, Seitzinger SP, Asner GP, Cleveland CC, Green PA, Holland EA, Karl DM, Michaels AF, Porter JH, Townsend AR, Voosmarty CJ (2004) Spatial distribution of sediment particulate organic matter on two estuarine intertidal mudflats: A comparison between Marennes-Oleron Bay (France) and the Humber Estuary (UK). Continental Shelf Research 20: 1199-1217.

Giffin D, Corbett DR (2003) Evaluation of sediment dynamics in coastal systems via short-lived isotopes. Journal of Marine Systems 42: 83-96.

Gilbert F, Stora G, Bonin P (1998) Influence of bioturbation on denitrification activity in Mediterranean coastal sediments: an *in situ* experimental approach. Marine Ecology Progress Series 163: 99-107.

Glud RN, Ramsing NB, Revsbech NP (1992) Photosynthesis and photosynthesis-coupled respiration in natural biofilms quantified with oxygen microsensors. Journal of Phycology 28: 51-60.

Gürel M, Tanik A, Russo RC, Gönenç IE (2005) Biogeochemical cycles in Gönenç IE, Wolfin JP (editors) Coastal lagoons: ecosystem processes and modelling for sustainable use and development. CRC Press: Boca Raton. 79- 192 pp.

Hammond D, Fuller C, Harmon D, Hartman B, Korosec M, Miller L, Rea R, Warren S, Berelson W, Hager SW (1985) Benthic fluxes in San Francisco Bay. Hydrobiologia 129: 69-90.

Hammond DE, Giordani P, Berelson WM, Poletti R (1999) Diagenesis of carbon and nutrients and benthic exchange in sediments of the Northern Adriatic Sea. Marine Chemistry 66: 53-79.

Hansen K, Kristensen E (1997) Impact of macrofaunal recolonization on benthic metabolism and nutrient fluxes in a shallow marine sediment previously overgrown with macroalgal mats. Estuarine Coastal and Shelf Science 45: 613-628.

Hansen K, Kristensen E (1998) The impact of the polychaete *Nereis diversicolor* and enrichment with the macroalgal (*Chaetomorpha linum*) detritus on benthic metabolism and nutrient dynamics in organic-poor and organic rich sediment. Journal of Experimental Marine Biology and Ecology 231: 201-223.

Hart BT, Grace MR (2000) Nitrogen workshop 2000: Sources transformations, effects and management of nitrogen in freshwater ecosystems. Land and Water Australia: Canberra.

Hartwig EO (1978) Factors affecting respiration and photosynthesis by the benthic community of a subtidal siliceous sediment. Marine Biology 46: 283-293.

Hauxwell J, Valiela I (2004) Effects of nutrient loading on shallow seagrass-dominated coastal systems: patterns and processes in S. Nielsen, Banta G, Pedersen M (editors), Estuarine Nutrient Cycling: the Influence of Primary Producers. Kluwer Academic Publishers, the Netherlands: 59-92 pp.

Henriksen K, Kemp WM (1988) Nitrification in estuarine and coastal marine sediments in Blackburn TH, Sφrensen J (editors). Nitrogen cycling in coastal marine environments (vol. 33). Chichester: John Wiley and Sons Ltd. 207-249 pp.

Henrisksen K, Rasmussen MB, Jensen A (1983) Effect of bioturbation on microbial nitrogen transformations in the sediment and fluxes of ammonium and nitrate to the overlying water. Ecological Bulletins 35: 193-205.

Herbert RA (1999) Nitrogen cycling in coastal marine ecosystems. FEMS Microbiology Reviews 23: 563-590.

Hewitt JE, Thrush SF, Legendre P, Funnell GA, Ellis J, Morrison M (2004) Mapping of marine soft sediment communities: integrated sampling for ecological interpretation. Ecological Applications 14: 1203-1216.

Hietanen S, Laine AO, Lukkari K (2007) The complex effects of the invasive polychaetes Marenzelleria spp. on benthic nutrient dynamics. Journal of Experimental

Marine Biology and Ecology 352: 89-102.

Hintze JL (1998) User's Guide for NCSS 2000 Statistical Systems for Windows. Number Cruncher Statistical Systems Press. Utah: 1716 p.

Hopkinson CS, Giblin AE, Tucker J, Garrit RH (1999) Benthic metabolism and nutrient cycling along an estuarine salinity gradient. Estuaries 22: 863-881.

Howarth RW, Jensen HS, Marino R, Postma H (1995) Transport to and processing of P in near-shore and oceanic waters in Tiessen H, (ed.) Phosphorus in the Global Environment: Transfers, Cycles, and Management. New York: John Wiley and Sons. 323–345 pp.

Hunt TA (2011) The effect of bait-pumping and trampling on inter-tidal soft-sediment macrofaunal communities in Jervis Bay Marine Park, New South Wales. Honours thesis, University of Wollongong, Australia.

Huttel M (1990) Influence of the lugworm *Arenicola marina* on porewater nutrient profiles of sand flat sediments. Marine Ecology Progress Series 62: 241-247.

Jensen HS, Andersen FØ (1992) Importance of temperature, nitrate and pH for phosphorus release from aerobic sediments of four shallow eutrophic lakes. Limnology and Oceanography 37: 577-589.

Jensen KM, Jensen MH, Kristensen E (1996) Nitrification and denitrification in Wadden Sea sediments (Königshafen, Island of Sylt, Germany) as measured by

nitrogen isotope pairing and isotope dilution. Aquatic Microbial Ecology 11: 181-191.

Jensen MH, Lomstein E, Sφrensen (1990) Benthic NH_4^+ and NO_3^- flux following sedimentation of a spring phytoplankton bloom in Aarhus Bight, Denmark. Marine Ecology Progress Series 61: 155-162.

Jitts HR, Scott BD (1961) The determination of zero-thickness activity in Geiger counting of C14 solutions using in marine productivity studies. Limnology and Oceanography 6: 116-123.

Jordan MA, Welsh DT, Dunn RJK, Teasdale PR (2009) Influence of *Trypaea australiensis* population density on benthic metabolism and nitrogen dynamics in sandy estuarine sediment: A mesocosm simulation. Journal of Sea Research 61: 144-152.

Joye SB, Hollibaugh JT (1995) Influence of sulfide inhibition of nitrification on nitrogen regeneration in sediments. Science 270: 623-625.

Kaiser MJ, Broad G, Hall SJ (2001) Disturbance of intertidal soft-sediment benthic communities by cockle hand raking. Journal of Sea Research 45: 119-130.

Karlson K, Hulth S, Rosenberg R (2007) Density of *Monoporeia affinis* and biogeochemistry in Baltic Sea sediments. Journal of Experimental Marine Biology and Ecology 344: 123-135.

Kemp WM, Boynton WR (1984) Spatial and temporal coupling of nutrient inputs to

estuarine primary production: the role of particulate transport and decomposition. Bulletin of Marine Science 33: 522-535.

Kemp WM, Sampou P, Caffrey J, Mayer M (1990) Ammonium recycling versus denitrification in Chesapeake Bay sediments. Limnology and Oceanography 35: 1545-1563.

Kemp WM, Sampou PA, Garber J, Tuttle J, Boynton WR (1992) Seasonal depletion of oxygen from bottom waters of Chesapeake Bay: roles of benthic and planktonic respiration and physical exchange processes. Marine Ecology Progress Series 85: 137-152.

Kennish MJ (2001) Ecology of estuaries: Physical and chemical aspects. CRC Press Inc.: Boca Raton, USA. 254 pp.

Kikuchi E (1986) Contribution of the polychaete, *Neanthes* japonica (Izuka) to the oxygen uptake and carbon dioxide production of an intertidal mud-flat of the Nanakita River Estuary, Japan. Journal of Experimental Marine Biology and Ecology 97: 81-93.

Klump JV, Martens CS (1981) Biogeochemical cycling in an organic rich coastal marine basin-II. Nutrient sediment-water exchange processes. Geochemica et Cosmochimica Acta 45: 101-121.

Knox GA (1986) Estuarine Ecosystems: A System Approach Volume 2. CRC Press,

Inc. Boca Raton: 230 pp.

Kristensen E, Andersen FØ, Blackburn TH (1992) Effects of benthic macrofauna and temperature on degradation of macroalgal detritus: The fate of organic carbon. Limnology and Oceanography 37: 1404-1419.

Kristensen E, Blackburn TH (1987) The fate of organic carbon and nitrogen in experimental marine sediment systems: Influence of bioturbation and anoxia. Journal of Marine Research 45: 231-257.

Kristensen E, Jensen MH, Aller RC (1991) Direct measurement of dissolved inorganic nitrogen exchange and denitrification in individual polychaete (*Nereis virens*) burrows. Journal of Marine Research 49: 355-377.

Kristensen E, Kostka JE (2005) Macrofaunal burrows and irrigation in marine sediment: microbiological and biogeochemical interactions in Kristensen E, Kostka JE, Haese, R (editors) Interactions between Macro- and Microorganisms in Marine Sediments. American Geophysical Union: Washington, DC.

Kristensen E (1985) Oxygen and inorganic nitrogen ex- change in a *Nereis virens* (Polychaeta) bioturbated sediment-water system. Journal of Coastal Research 1: 109-116.

Krom MD (1991) Importance of benthic productivity in controlling the flux of dissolved inorganic nitrogen through the sediment-water interface in a hypertrophic

marine ecosystem. Marine Ecology Progress Series 78: 163-172.

Kuwae T, Hosokawa Y, Eguchi N (1998) Dissolved inorganic nitrogen cycling in Banzu intertidal sand-flat, Japan. Mangroves and Salt Marshes 2: 167-175.

Laima M, Brissard D, Sauriau PG, Girard M, Richard P, Gouleau D, Joassard L (2002) The influence of long emersion on biota, ammonium fluxes and nitrification in intertidal sediments of Marennes-Oleronon Bay, France. Marine Environmental Research 53: 381-402.

Lavery PS, McComb AJ (1991) Macroalgal-sediment nutrient interactions and their importance to macroalgal nutrition in a eutrophic estuary. Estuarine Coastal and Shelf Science 32: 281-295.

Lebo ME (1991) Particle-bound phosphorus along an urbanized coastal plain estuary. Marine Chemistry 34: 225-246.

Lerman A (1978) Chemical exchange across sediment-water interface. Annual Review of Earth and Planetary Sciences 6: 281-303.

Lillebø AI, Neto JM, Flindt MR, Marques JC, Pardal MA (2004) Phosphorous dynamics in a temperate intertidal estuary. Estuarine, Coastal and Shelf Science 61: 101-109.

Lohse L, Kloosterhuis HT, Raaphorst W, Heldet W (1996) Denitrification rates as

measured by the isotope pairing method and by the acetylene inhibition technique in continental shelf sediments of the North Sea. Marine Ecology Progress Series 132: 169-179.

MacIntyre HL, Geider RJ, Miller DC (1996) Micro- phytobenthos: The ecological role of the 'secret garden' of unvegetated, shallow-water marine habitats. I. Distribution, abundance and primary production. Estuaries 19: 186-201.

Magalhães C, Bordalo AA, Wiebe WJ (2002) Temporal and spatial patterns of intertidal sediment-water nutrient and oxygen fluxes in the Douro River estuary, Portugal. Marine Ecology Progress Series 233: 55-71.

Marine Parks Authority (2011a) Jervis Bay Marine Park User guide. in Department of sustainability E, Water, Population and Communities, ed. Canberra.

Marine Parks Authority (2011b) Solitary Islands Marine Park and Solitary Island Marine Reserve (Commonwealth Waters) User guide in Department of sustainability Environment, Water, Population and Communities.

Marshall MC, Hall RO (2004) Hyporheic invertebrates affect N cycling and respiration in stream sediment microcosms. Journal of the North American Benthological Society 23: 416-428

McGlathery KJ, Anderson IC, Tyler AC (2001) Magnitude and variability of benthic and pelagic metabolism in a temperate coastal lagoon. Marine Ecology Progress

Series 6: 1-15.

McGlathery KJ, Sundback K, Anderson C (2004) The importance of primary producers for benthic nitrogen and phosphorus cycling in Nielsen SL, Banta GT, Pedersen MF (editors) Estuarine nutrient cycling: the influence of primary producers, Kluwer Academic Publishers, China.

Mazouni N, Gaertner JC, Deslous-Paoli JM, Landrein S, d'Oedenberg M (1996) Nutrient and oxygen exchanges at the water-sediment interface in a shellfish farming lagoon (Thau, France). Journal of Experimental Marine Biology and Ecology 205: 91-113.

Mermillod-Blondin F, François-Carcaillet F, Rosenberg R (2005) Biodiversity of benthic invertebrates and organic matter processing in shallow marine sediments: an experimental study. Journal of Experimental Marine Biology and Ecology 315: 187-209.

Michaud E, Desrosiers G, Mermillod-Blondin F, Sundby B, Stora G (2006) The functional group approach to bioturbation: II. The effects of the *Macoma balthica* community on fluxes of nutrients and dissolved organic carbon across the sediment-water interface. Journal of Experimental Marine Biology and Ecology 337: 178-189.

Mikac KM, Maher WA, Jones AR (2007) Do physiochemical sediments variables and their soft sediment macrofauna differ among microsize coastal lagoons with forested and urbanised catchments? Estuarine Coastal and Shelf Science 72: 308-318.

Miller-Way T, Boland GS, Rowe GT, Twilley RR (1994) Sediment oxygen consumption and benthic nutrient fluxes on the Louisiana continental shelf: A methodological comparison. Estuaries 17: 809-815.

Moore PA, Reddy KR, Fisher MM (1998) Phosphorus Flux Between Sediment and Overlying Water in Lake Okeechobee, Florida: Spatial and Temporal Variations. Journal of Environmental Quality 27: 1428-1439.

Mortimer RJG, Davey JT, Krom MD, Watson PG, Frickers PE, Clifton RJ (1999) The effect of macrofauna on porewater profiles and nutrient fluxes in the intertidal zone of the Humber estuary. Estuarine, Coastal and Shelf Science 48: 683-699.

Mulder A, van de Graaf AA, Robertson LA, Kuenen JG (1995) Anaerobic ammonium oxidation discovered in a denitrifying fluidized bed reactor. FEMS Microbial Ecology 16: 177-184.

Nedwell DB, Jickells JD, Trimmer M, Sanders R (1999) Nutrients in estuaries. In: Advances in Ecological Research: Estuaries 29 142-192. Advances in Ecological Research: Estuaries 29: 142-192.

Nedwell DB, Walker TR (1995) Sediment-water fluxes of nutrients in an Antarctic coastal environment: influences of bioturbation. Polar Biology 15: 57-64.

Nicholls HB (1998) Canadian east coast marine-protected areas: A review. Ocean and Coastal Management 39: 87-96.

Nicholson GJ, Longmore AR (1999) Causes of the observed temporal variability of nutrient fluxes from a southern Australian marine embayment. Marine Freshwater Research 50: 581-588.

Nixon S (1981) Remineralization and nutrient cycling in coastal marine ecosystems in Neilson BJ, Cronin LE (editors) Estuaries and nutrients. Humana press, Clifton, New Jersey. 111-138 pp.

Nizzoli D, Bartoli M, Cooper M, Welsh DT, Underwood GJC, Viaoli P (2007) Implications for oxygen, nutrient fluxes and denitrification rates during the early stage of sediment colonization by the polychaete *Nereis* spp. in four estuaries. Estuarine Coastal and Shelf Science 75: 125-134.

Nizzoli D, Welsh DT (1999) Influence of the polychaete worm *Nereis succinea* on fluxes of oxygen, inorganic and organic nitrogen and rates of denitrification and coupled nitrification-denitrification in experimental mesocosms. Limnology and Oceanography 13: 319-328.

Papaspyrou S, Kristensen E, Christensen B (2007) *Arenicola marina* (Polychaeta) and organic matter mineralisation in sandy marine sediments: In situ and microcosm comparison. Estuarine Coastal and Shelf Science 72: 213-222.

Papaspyrou S, Thessalou-Legaki M, Kristensen E (2004) Impact of *Pestarella tyrrhena* (Decapoda: Thalassinidea) on benthic metabolism in sediment microcosms

enriched with seagrass and macroalgal detritus. Marine Ecology Progress Series 281: 165-179.

Paytan A, McLaughlin K (2007) The Oceanic Phosphorus Cycle. Chemical Reviews 107: 563-576.

Pelegri SP, Blackburn TH (1994) Pelegrí SP, Blackburn TH (1994) Bioturbation effects of the amphipod *Corophium volutator* on microbial nitrogen transformations in marine sediments. Marine Biology 121: 253-258.

Pelegri SP, Blackburn TH (1995) Effect of bioturbation by *Nereis* sp., *Mya arenaria* and *Cerastoderma* sp. on nitrification and denitrification in estuarine sediments. Ophelia 42: 289-299.

Pelegri SP, Neilsen LP, Blackburn TH (1994) Denitrification in estuarine sediment stimulated by the irrigation activity of the amphipod *Corophium volutator*. Marine Ecology Progress Series 105: 285-290.

Penn MR, Auer MT, Doerr SM, Driscoll CT, Brooks CM, Effler SW (2000) Seasonality in phosphorous release rates from the sediment of a hypereutrophic lake under a matrix of pH and redox conditions. Canadian Journal of Fisheries and Aquatic Science 57: 1033-1041.

Peterson BJ (1980) Aquatic primary productivity and the 14C-CO2 method: A history of the productivity problem. Annual Review of Ecological Systems 11: 359-385.

Peterson CH (1977) Competitive organization of soft-bottom macrobenthic communities of Southern-California lagoons. . Marine Biology 43: 343-359.

Plante CJ, Feipel S, Wilkie JL (2010) Disturbance effects of deposit feeding on microalgal community structure and mechanisms of recolonization. Journal of Phycology 46: 907-916.

Postma H, Kemp WM, Colebrook JM, Horwood J, Joint IR, Lampitt R, Nixon SW, Pilson MEQ, Wulff F (1984) Nutrient cycling in estuarine and coastal marine ecosystems, in Fashman MJR (editor) Flows of Energy and Materials in Marine Ecosystems Theory and Practical. Plenum Press, New York. 651 p.

Potts J, Blakey C, Watson R (2005) Nutrient fluxes in 5 lagoons along the Illawarra coast. Report of Wollongong City Council, Department of Environment and Conservation, Sydney.

Pusch M, Schmoerbel J (1994) Community respiration in hyporheic sediments of a mountain stream (Steina, Black Forest). Hydrobiologia 130: 35-52.

Qu WC, Morrison RJ, West RJ, C. S (2005) Diagenetic Stoichiometry and Benthic Nutrient Fluxes at the Sediment—Water Interface of Lake Illawarra, Australia. Hydrobiologia 537: 249-264.

Qu WC, Morrison RJ, West RJ (2003) Inorganic nutrient and oxygen fluxes across

the sediment-water interface in the inshore macrophyte areas of a shallow estuary (Lake Illawarra, Australia). Hydrobiologia 492: 119-127.

Qu WC (2004) Studies on nitrogen cycling processes in Lake Illawarra, New South Wales, Australia. PhD thesis, University of Wollongong, Australia.

Rasmussen H, Jorgensen BB (1992) Microelectrode studies of seasonal oxygen uptake in a coastal sediment: role of molecular diffusion. Marine Ecology Progress Series 81: 289-303.

Reay WG, Gallagher DL, Simmons GM (1995) Sediment-water column oxygen and nutrient fluxes in nearshore environments of the lower Delmarva Peninsula, USA. Marine Ecology Progress Series 118: 215-227.

Reice SR (1994) Nonequilibrium determinants of biological community structure. American Journal of Science 82: 424-435.

Revsbech NP, Jorgensen BB (1981) Primary production of microalgae in sediments measured by oxygen profile, H14CO3- fixation, and oxygen exchange methods. Limnology and Oceanography 26: 717-730.

Revsbech NP, Jφrgensen BB (1983) Photosynthesis of benthic microflora measured with high spatial resolution by the oxygen profile method: Capabilities and limitations of the method. Limnology and Oceanography 28: 749-756.

Revsbech NP, Jorgensen BB (1986) Microelectrodes: Their use in microbial ecology. Advances in Microbial Ecology 9: 293-352.

Risgaard-Petersen N, Dalsgaard T, Rysgaard S, Christensen PB, Borum J, McClathery K, Nielsen LP (1998) Nitrogen balance of a temperate eelgrass *Zostera marina* bed. Marine ecology Progress Series 174: 281-291.

Risgaard-Petersen N, Ottosen LD (2000) Nitrogen cycling in two temperate *Zostera marina* beds: seasonal variation. Marine Ecology Progress Series 198: 93-107.

Risgaard-Petersen N, Rysgaard S, Nielsen LP, Revsbech NP (1994) Diurnal variation of denitrification and nitrification in sediments colonised by benthic microphytes. Limnology and Oceanography 39: 573-579.

Risgaard-Petersen N (2003) Coupled nitrification- denitrification in autotrophic and heterotrophic sediments: on the influence of benthic microalgae. Limnology and Oceanography 48: 93-105.

Rizzo WM, Berry BE, Wetzel RL, Dailey SK, Lackey GJ, Christian RR (1996) A metabolism-based trophic index for comparing the ecological values of shallow-water sediment habitats. Estuaries 19: 247-256.

Rizzo WM, Lackey GJ, Christian RR (1992) Significance of euphotic, subtidal sediments to oxygen and nutrient cycling in a temperate estuary. Marine Ecology Progress Series 86: 51-61.

Rizzo WM (1990) Nutrient exchanges between the water column and a subtidal benthic microalgal community. Estuaries 13: 219-226.

Roberts CM, Andelman S, Branch G, Bustamante RH, Castilla JS, Dugan J, Halpern BS, Lafferty KD, Leslie H, Lubchenco J, McArdie D, Possingham HP, Ruckelshaus M, Warner RR (2003) Application of ecological criteria in selecting marine reserves and developing reserves networks. Ecological Applications 13: 199-214.

Rossi F, Chapman, MG (2003) Influence of sediment on burrowing by the soldier crab *Mictyris longicarpus* Latreille. Journal of Experimental Marine Biology and Ecology 289: 181-195.

Rossi F, Forster RM, Montserrat F, Ponti M, Terlizzi A, Ysebaert T, Middelburg JJ (2007) Human trampling as short-term disturbances on intertidal mudflats: effects on macrofauna biodiversity and population dynamics of bivalves. Marine Biology 151: 2077-2090.

Rotherham D, West RJ (2003) Comparison of methods for sampling populations of ghost shrimp, *Trypaea australiensis* (Decapoda: Thalassinidea: Callianassidae). Fisheries Research 60: 585-591.

Rowe GT, Phoel WC (1992) Nutrient regeneration and oxygen demand in Bering Sea continental shelf sediments. Continental Shelf Research 12: 439-449.

Rysgaard S, Christensen PB, Nielsen PL (1995) Seasonal variation in nitrification and denitrification in estuarine sediment colonised by benthic microalgae and bioturbating infauna. Marine Ecology Progress Series 126: 111-121.

Rysgaard S, Risgaard-Petersen N, Nielsen LP, Revsbech NP (1993) Nitrification and denitrification in lake and estuarine sediments measured by the 15N dilution technique and isotope pairing. Applied and Environmental Microbiology 59: 2093-2098.

Rysgaard S, Risgaard-Petersen N, Sloth NP, Jensen K, Nielsen LP (1994) Oxygen regulation of nitrification and denitrification in sediments. Limnology and Oceanography 39: 1643-1652.

Rysgaard S, Risgaard-Petersen N, Sloth NP (1996) Nitrification, denitrification, and nitrate ammonification in sediments of two coastal lagoons in southern France. Hydrobiologia 329: 133-141.

Sand-Jensen K, Nielsen SL (2004) Estuarine primary producers in Nielsen SL, Banta GT, Pedersen MF (editors) Estuarine nutrient cycling: the influence of primary producers, Vol 2. Kluwer Academic Publishers, London. 17-57 pp.

Sandwell DR, Pilditch CA, Lohrer AM (2009) Density dependent effects of an infaunal suspension-feeding bivalve (*Austrovenus stutchburyi)* on sandflat nutrient fluxes and microphytobenthic productivity. Journal of Experimental Marine Biology and Ecology 373: 16-25.

Santschi P, Hohener P, Benoit G, Buchholtz-ten, Brink B (1990) Chemical processes at the sediment-water interface. Marine Chemistry 30: 269-315.

Sayama M, Kurihara Y (1983) Relationship between burrowing activity of the polychaetous annelid, *Neanthes japonica* (Izuka) and nitrification-denitrification processes in the sediment. Journal of Experimental Marine Biology and Ecology 72: 233-242.

Seitzinger S (1988) Denitrification in freshwater and coastal marine ecosystems: Ecological and geochemical significance. Limnology and Oceanography 33: 702-724.

Simon NS (1988) Nitrogen cycling between sediment and the shallow-water column in the transition zone of the Potomac River and estuary. I. Nitrate and ammonium fluxes. Estuarine Coastal and Shelf Science 26: 483-497.

Smith SV, Hollibaugh JT (1997) Annual cycle and interannual variability of ecosystem metabolism in a temperate climate embayment. Ecological Monographs 67: 509-533.

Smith SV (1984) Phosphorus versus nitrogen limitation in the marine environment Limnology and Oceanography 29: 1149-1160.

Sondergaard M (2007) Nutrient dynamics in lakes - with emphasis on phosphorus, sediment and lake restorations. PhD thesis, University of Arhus, Denmark.

Souchu P, Gasc A, Collos Y, Vaquer A, Tournier H, Bibent B, Deslous-Paoli JM (1998) Biogeochemical aspects of bottom anoxia in a Mediterranean lagoon (Thau, France). Marine Ecology Progress Series 164: 135-146

Spagnoli F, Bergamini MC (1997) Water-sediment exchange of nutrients during early diagenesis and resuspension of anoxic sediments from the northern Adriatic Sea shelf. Water, Air and Soil Pollution 99: 541-556.

Spears B, Carvalho L, Perkins R, Paterson D (2008) Effects of light on sediment nutrient flux and water column nutrient stoichiometry in a shallow lake. Water Research 42: 977-986.

Sundback K, Enoksson V, Graneli W, Pettersson K (1991) Influence of sublittoral microphytobenthos on the oxygen and nutrient flux between sediment and water: a laboratory continuous-flow study. Marine Ecology Progress Series 74: 263-279.

Sundback K, Graneli W (1988) Influence of microphytobenthos on the nutrient flux between sediment and water: laboratory study. Marine Ecology Progress Series 43: 63-69.

Sundback K, Jonsson B (1988) Microphytobenthic productivity and biomass in sublittoral sediments of a stratified bay, southeastern Kattegat. Journal of Experimental Marine Biology and Ecology 122: 63-81.

Sundback K, Miles A (2000) Balance between denitrification and microalgal

incorporation of nitrogen in microtidal sediments, NE Kattegat. Aquatic Microbial Ecology 22: 291-300.

Sundback K, Rysgaard S (2000) Protocol Handbook for NICE- Nitrogen Cycling in Estuaries. Ministry of Environment and Energy. Silkeborg, Denmark. 62 p.

Sundby B, Gobeil C, Silverberg N, Mucci A (1992) The phosphorus cycle in coastal marine-sediments. Limnology and Oceanography 37: 1129-1145.

Tandeau de Marsac N, Houmard J (1993) Adaptation of cyanobacteria to environmental stimuli: new steps towards molecular mechanisms. FEMS Microbial Ecology 140: 119-190.

Tappin AD (2002) An examination of the fluxes of nitrogen and phosphorus in temperate and tropical estuaries; Current estimates and uncertainties. Estuarine Coastal and Shelf Science 55: 885-901.

Tengberg A, Almroth E, Hall P (2003) Resuspension and its effects on organic carbon recycling and nutrient exchange in coastal sediments: in situ measurements using new experimental technology. Journal of Experimental Marine Biology and Ecology 285/286: 119-142.

Thornton DCO, Dong LF, Underwood AJ, Nedwell DB (2002) Factors affecting microphytobenthic biomass, species composition and production in the Colne Estuary (UK). Aquatic Microbial Biology 27: 285-300.

Tiedje JM, Simkins S, Groffman PM (1989) Perspectives on measurement of denitrification in the field including recommended protocols for acetylene based methods. Plant and Soil 115: 261-284.

Trimmer M, Nedwell DB, Sivyer DB, Malcolm SJ (2000) Seasonal organic mineralization and denitrification in intertidal sediments and their relationship to the abundance of *Enteromorpha* sp. and *Ulva* sp. Marine Ecology Progress Series 203: 67-80.

Tuominen L, Mäkelä K, Lehtonen KK, Haathi H, S H, Kuparinen J (1999) Nutrient fluxes, porewater profiles and denitrification in sediment influenced by algal sedimentation and bioturbation by *Monoporeia affinis*. Estuarine Coastal and Shelf Science 49: 83-97.

Tyler AC, McGlathery KJ (2003) Benthic algae control sediment-water column fluxes of nitrogen in a temperate lagoon. Limnology and Oceanography 48: 2125-2137.

Tyler AC (2002) Impact of Benthic Algae on Dissolved Organic Nitrogen in a Temperate, Coastal Lagoon. PhD thesis, University of Virginia, USA.

Underwood AJ, Kromkamp J (1999) Primary production by phyto- plankton and microphytobenthos in estuaries. Advances in Ecological Research 29: 93-153.

Underwood AJ, Paterson DM (1993) Recovery of intertidal benthic diatoms from biocide treatment and associated sediment dynamics. Journal of the Marine Biological

Association of the United

Underwood AJ, Peterson CH (1988) Towards an ecological framework for investigating pollutions. Marine Ecology Progress Series 46: 227-234.

Underwood AJ (1997) Experiments in Ecology: Their Logical Design and Interpretation Using Analysis of Variance, Cambridge University Press, Cambridge. 505pp.

Underwood G (1994) Seasonal and spatial variation in epipelic diatom assemblages in the Severn Estuary. Diatom Research 9: 451-472.

Valiela I (1995) Nutrient cycles and ecosystem stoichiometry, in Marine Ecological Processes (second edition). Springer-Verlag, New York. 425- 466 pp.

van Duyl FC, van Raaphorst W, Kop AJ (1993) Benthic bacterial production and nutrient sediment-water exchange in sandy North Sea sediments. Marine Ecology Progress Series 100: 85-95.

Viaroli P, Bartoli M, Bondavalli C, Christian RR, Giordani G, Naldi M (1996) Macrophyte communities and their impact on benthic fluxes of oxygen, sulphide and nutrients in shallow eutrophic environments. Hydrobiologia 329: 105-119.

Viaroli P, Christian RR (2003) Description of trophic status of a eutrophic coastal lagoon through potential oxygen production and consumption: defining

hyperautotrophy and dystrophy. Ecological Indicators 3: 237-250.

Wakeham S (2002) Diagenesis of organic matter at the water-sediment interface in Gianguzza A, Pelizzetti E, Sammartano S (editors) Chemistry of marine water and sediments. Springer-Verlag: Germany. 147-164 pp.

Wall DH, Palmer MA, Sneigrove PVR (2001) Biodiversity in critical transition zones between terrestrial, freshwater, and marine soils and sediments: Processes, linkages, and management implications. Ecosystems 4: 418-420.

Wallace JB, Webster JR (1996) The role of macroinvertebrates in stream ecosystem function. Annual Review of Entomology 41: 115-139.

Wang H (2003) Modelling of phosphorus dynamics in aquatic sediments: I-model development. Water Research 37: 3928-3938.

Wang D, Chen Z, Wang J, Xu S, Yang H, Chen H, Yang L, Hu L (2007) Summer-time denitrification and nitrous oxide exchange in the intertidal zone of the Yangtze Estuary. Estuarine Coastal and Shelf Science 73: 43-53.

Webb AP, Eyre BD (2004) Effect of natural populations of burrowing thalassinidean shrimp on sediment irrigation, benthic metabolism, nutrient fluxes and denitrification. Marine Ecology Progress Series 268: 205-220.

Webb KL (1981) Conceptual models and processes of nutrient cycling in estuaries in

Neilson BJ, Cronin LE (editors) Estuaries and nutrients. Human Press: Clifton. 25-46 pp.

Webster IT, Ford PW, Hodgson B (2002) Microphytobenthos contribution to nutrient-phytoplankton dynamics in a shallow coastal lagoon. Estuaries 25: 540-551.

Welsh DT, Bartoli M, Nizzoli D, Castaldelli G, Riou SA, Viaroli P (2000) Denitrification, nitrogen fixation, community primary productivity and inorganic-N and oxygen fluxes in an intertidal *Zostera noltii* meadow. Marine Ecology Progress Series 208: 65-77.

Wetzel RG (1983) Limnology (second edition). Saunders College Publishing, Philadelphia.

West RJ, Thorogood C, Walford T, Williams RJ (1985) An estuarine inventory for New South Wales. Fisheries Bulletin No. 2. (NSW Department of Agriculture). 140 pp.

Winberg PC (2008) Confronting the challenges of tidal flat conservation: spatial patterns and human impacts in a marine protected area in southern NSW, Australia, PhD thesis, University of Wollongong, Australia.

Winberg PC, Lynch TP, Murray A, Jones AR, Davis AR (2007) The importance of spatial scale for the conservation of tidal flat macrobenthis: An example from New South Wales, Australia. Biological Conservation 134: 310-320.

Wolff WJ (1987) Flora and macrofauna of intertidal sediments. Biological surveys of estuaries and coasts. J. M. Baker and W. J. Wolff. Cambridge, Cambridge University Press.81 – 105 pp.

Woodley S, Loneragan N, Babcock R (2010) Report on the scientific basis for and the role of marine sanctuaries in marine planning. Prepared for the Inter Departmental Committee for the Management of the States Marine Protected Areas Perth Western Australia.

Wynberg RP, Branch GM (1991) An assessment of bait-collecting for *Callianassa kraussi* Stebbing in Langebaan Lagoon, Western Cape, and of associated avian predation. South African Journal of Marine Science 11: 141-152.

Wynberg RP, Branch GM (1994) Disturbance associated with bait-collection for sandprawns (*Callianassa kraussi*) and mudprawns (*Upogebia africana*): Long-term effects on the biota of the intertidal sandflats. Journal of Marine Research 52: 523-558.

Wynberg RP, Branch GM (1997) Trampling associated with bait- collection for sandprawns *Callianassa kraussi* Stebbing: effects on the biota of an intertidal sandflat. Environmental Conservation 24: 139-148.

Yoon WB, Benner R (1992) Denitrification and oxygen consumption in sediments of two south Texas estuaries. Marine Ecology Progress Series 90: 157-167.

Zimmerman AR, Benner R (1994) Denitrification, nutrient regeneration and carbon mineralization in sediments of Galveston Bay, Texas, USA. Marine Ecology Progress Series 114: 275-288.