CONTRIBUTIONS OF INSHORE AND OFFSHORE SOURCES OF PRIMARY PRODUCTION TO THE FOODWEB, AND THE TROPHIC CONNECTIVITY BETWEEN VARIOUS HABITATS ALONG A DEPTH-GRADIENT, IN SODWANA BAY, KWAZULU-NATAL, SOUTH AFRICA

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Abstract

Sodwana Bay, situated within the iSimangaliso Wetland Park, is ecologically important as it contains high-latitude corals and the most southerly known population of coelacanths. This thesis utilised stable isotope and lipid analyses to investigate the trophic ecology of the area, in particular, understanding the relative contribution of inshore and offshore primary production to consumers inhabiting intertidal and shallow subtidal, coral reef, deep reef, canyon head and pelagic habitats.

Seaweeds, excluding certain species of red seaweeds with highly depleted carbon signatures, and phytoplankton, such as diatoms, were found to be the principal sources of primary production for all consumers. Offshore production was typified by dinoflagellates.

Particulate organic matter (POM) was spatio-temporally variable. Three distinct productivity periods related to nutrient cycling were noted with enriched carbon signatures and higher organic matter loads associated with warmer water.

Inshore primary production was an important source of carbon to consumers in all habitats with the exception of zooplankton that were more reliant on pelagic primary production. Benthic invertebrates reflected a gradient in the utilisation of inshore production, due to the reduced availability of this source further offshore. Consumers at the furthest sites offshore were found to include a substantial quantity of inshore-derived production in their diets. Fishes, which are more mobile, were found to incorporate a similar proportion of inshore production into their diets regardless of where they were collected from.

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Chapter 1

General Introduction

1.1 Trophic studies

Food web studies aim to understand the trophic linkages between organisms from a community perspective, from the basal trophic level, primary producers, to primary consumers and higher level organisms (Carpenter *et al.* 1987). Within the marine environment there are a range of potential sources of organic matter available to consumers. In coastal waters these include algae, both phytoplankton (Duggins *et al.* 1989) and seaweeds (Bustamante & Branch 1996), seagrasses (Suchanek *et al.* 1985), and where large rivers drain into the ocean, terrestrial and riverine inputs (Gaston *et al.* 2006). These sources of organic matter are incorporated into marine foodwebs either directly through grazing organisms (Vine 1974) or via detrital consumption (Mann 1988).

Investigating the origin and importance of organic matter, including particulate organic matter (POM) that incorporates living phytoplankton as well as detritus particles from phytoplankton, macrophytes and zooplankton, will facilitate a better understanding of the flow of energy through an ecosystem. The analysis of POM has been used to investigate the spatio-temporal variations in the sources of nitrogen, in terms of the newly fixed nitrates and regenerated ammonia, and carbon, in terms of carbon dioxide, carbonates, and bicarbonates, utilised by primary producers (Dugdale & Goering 1967, Deuser *et al.* 1968, Wada & Hattori 1976). POM analyses may also indicate relative rates of primary production, and the proportions of detritus, across spatio-temporal scales within a system (Gu *et al.* 2006, Lara *et al.* 2010). Traditional approaches to food web analyses have included direct observations in either the field or the laboratory, gut content analysis, or tracer methods such as radio-

(Rounick & Winterbourn 1986) and stable isotope analyses (Corbisier *et al.* 2006, Tamelander *et al.* 2006, West *et al.* 2006, Layman *et al.* 2011).

1.2 Stable isotope analysis

Direct dietary observations (Uchima 1988, Clements & Choat 1993, Muñoz & Ojeda 1997, Sala & Ballesteros 1997) are useful in investigating high resolution short-term trophic linkages but unfortunately provide limited data on the assimilation of different food sources (Kling *et al.* 1992). Dietary observations, such as gut content analysis, can also be biased toward less easily digested material leading to the potential under-estimation of the importance of soft-bodied prey (Duffy & Jackson 1986). Stable isotope analysis provides an alternative tool to observational trophic studies as it provides information on assimilation rather than purely ingestion (DeNiro & Epstein 1978) with tissue-dependent turnover rates providing dietary information at different temporal scales (Suring & Wing 2009). Despite its lower resolution compared to dietary observations, in that trophic-interactions at the species level are generally not discernible, it is arguably the most logistically practical technique to use if trophic interactions are to be investigated at a larger, ecosystem scale beyond a few species. Stable isotopes indicate the trophic ordering of organisms in a food web and where isotopically distinct sources of production exist their relative contributions to higher trophic levels can be estimated (Parnell *et al.* 2010). At a finer-scale, where trophic interactions between individual species are of concern, stable isotope and observational techniques such as gut content analysis may complement each other, with each method highlighting details not provided by the other (Kadye & Booth 2012).

Most ecologically important elements occur naturally as at least two stable isotopes, for example ^{13}C and ^{12}C, and ^{15}N and ^{14}N, whose ratios provide an elegant solution to tracing

energy flows and estimating an organism's dependency on certain food sources and habitats (Bustamante & Branch 1996). The use of stable isotopes in dietary studies is based on the assumption that animal tissues have an element-dependent fixed isotopic enrichment, or depletion, of the heavier isotope relative to the diet (Ehleringer *et al.* 1986). This discrimination is referred to as fractionation (Ehleringer *et al.* 1986, Peterson & Fry 1987). A consumer's carbon signature tends to be similar to its diet with carbon fractionation usually being < 1 ‰ and is typically used to indicate dietary sources (DeNiro & Epstein 1978, Rau *et al.* 1983, Post 2002). These small levels of carbon enrichment are caused by fractionation during assimilation or respiration processes (Peterson & Fry 1987). Details of a consumer's diet can be inferred from stable carbon isotope ratios if potential food sources possess different $^{13}C/^{12}C$ ratios (DeNiro & Epstein 1981). Dietary determination, using stable isotopes, involves estimating the $\delta^{13}C$ value of the overall diet from the $\delta^{13}C$ value of the consumer's carbon signature and then mathematically determining the relative contribution of individual diet sources of known $\delta^{13}C$ values that in combination can explain the mixed-diet signature of the consumer (DeNiro & Epstein 1978, Parnell *et al.* 2010). Interpretation becomes difficult where more than two potential diet sources are present and the sample exhibits an intermediate isotopic value (Peterson & Fry 1987). Primary producers with overlapping carbon signatures hinder clear interpretation of energy flows (Dunton & Schell 1987). Nitrogen isotope fractionation between trophic levels (3 – 5 ‰) is larger than for carbon and is generally used as an indicator of the trophic level of an organism (DeNiro & Epstein 1981, Minagawa & Wada 1984, Peterson & Fry 1987, Post 2002). This stepwise trophic-level enrichment has been found to be relatively uniform for a multitude of food webs, from both terrestrial and marine environments (Minagawa & Wada 1984). Enrichment of ^{15}N occurs primarily through the excretion of isotopically lighter nitrogen isotopes (^{14}N) in urea (Peterson & Fry 1987).

1.3 Sodwana Bay

Sodwana Bay is situated approximately 300 km north east of Durban and approximately 80 km south of the South Africa-Mozambique border. It is situated within the iSimangaliso Wetland Park (IWP), formerly the Greater St Lucia Wetland Park, a declared World Heritage Site (Figure 1.1). The IWP incorporates a 155 × 5 km marine conservation area with the southernmost boundary at Cape Vidal, 160 km north of Durban, extending to the Mozambican border in the north (UNEP 2005). The importance of the northern KwaZulu-Natal region of the South African coast in general, and the IWP in particular, with respect to its conservation value is high because it contains both the southernmost distribution of corals in African waters (Ramsay & Mason 1990) and a viable population of the endangered IUCN red data list species *Latimeria chalumnae* (Venter *et al.* 2000). Coelacanths were discovered in submarine canyons a few kilometres offshore of Sodwana Bay in 2000 and are considered to be the southernmost population of the species (Hissmann *et al.* 2006).

Past marine-based studies in Sodwana Bay have largely focussed on its coral reefs (Ramsay & Mason 1990, Riegl *et al.* 1995, Celliers & Schleyer 2002, Schleyer & Celliers 2003a, b, Floros *et al.* 2004, Schleyer & Celliers 2005, Glassom *et al.* 2006, Celliers & Schleyer 2008). More recently, however, following the discovery of the coelacanth population, research emphasis has changed to include deeper habitats (Heemstra *et al.* 2006, Hissmann *et al.* 2006, Ramsay & Miller 2006, Sink *et al.* 2006, Uken & Green 2006, Green *et al.* 2007). Exploratory studies using submersibles and remotely operated vehicles (ROVs) in the canyon habitats were undertaken to better understand these deeper environments (Hissmann *et al.* 2006). While there is a reasonably good understanding of the physical environment in which coelacanths live (Hissmann *et al.* 2006) only limited biological data are available. Apart from data from video footage, from which potential predator-prey interactions have been inferred

(Heemstra *et al.* 2006), there is a lack of knowledge of ecological interactions involving the coelacanth and other species. Overall, the broader ecology of Sodwana Bay has been poorly studied and no literature exists regarding its trophic ecology.

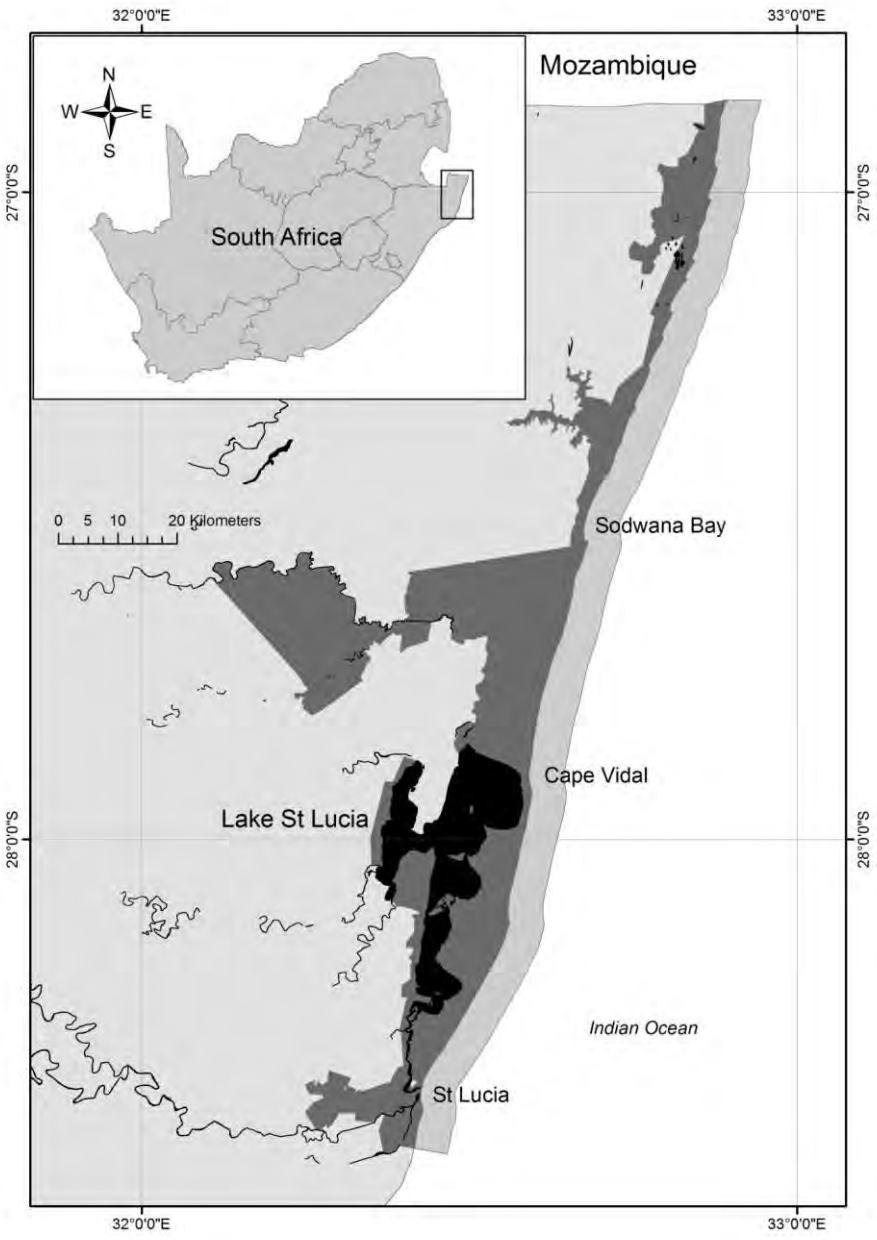


Figure 1.1: Map of iSimangaliso Wetland Park (IWP; dark grey region), indicating the position of Sodwana Bay. The shaded oceanic region represents the marine component of the IWP.

Northern KwaZulu-Natal is situated between tropical and subtropical zones and experiences warm summers and mild winters. It receives summer rainfall with annual precipitation ranging between 1 200 and 1 300 mm (UNEP 2005). Sodwana Bay possesses warm clear waters carrying a low phytoplankton load (Ramsay & Mason 1990). Surface water temperatures reach a minimum of ~ 22 ^{o}C in winter and a maximum of ~ 28 ^{o}C in summer (De Clerck *et al.* 2005). Warm water temperatures are maintained by the close proximity of the south-flowing Agulhas Current to the coast because of a narrow (~ 3 km wide) continental shelf (Schumann 1988, Ramsay 1994). The salinity of the subtropical surface water is relatively high (> 35.5ppt) due to high evaporation rates (Schumann 1988). Higher summer temperatures are caused by increased insolation with reduced salinities due to precipitation (Schumann 1988). Seasonal temperature variations are not evident below a depth of 50 m (Schumann 1988).

The southward flowing western boundary current, the Agulhas Current, is the most important large-scale oceanographic feature in the area (Viana *et al.* 1998). The current is the result of converging flows from the Madagascar current and south-moving eddies passing through the Mozambique Channel (Ridderinkhof *et al.* 2001, Lutjeharms 2006). The clear subtropical water of the Agulhas Current (Flemming 1981, Viana *et al.* 1998), and the lack of nearby silt laden rivers result in a relatively deep euphotic zone, facilitating the occurrence of corals in the region (Ramsay & Mason 1990). In the absence of riverine input seaweeds are thought to be important sources of organic matter, however the relative contribution of seaweeds, among other potential sources of production, to the marine food web are unknown (De Clerck *et al.* 2005). The marine flora of the Park includes 325 known species of seaweed, constituting 78 % of all seaweeds known to occur in Kwazulu-Natal (UNEP 2005).

At Sodwana Bay, a number of different habitats occur along a depth gradient from inshore to offshore (Figure 1.2). The nearshore/intertidal region is predominantly sandy-sediment, with a rocky point at the southern end of the bay, providing little suitable substrata for seaweeds. Immediately to the south of the bay are a stretch of rocky ledges in the intertidal region. Situated between approximately 100 m and 1 km offshore is a coral reef complex in water depths of between 6 and 40 m (Ramsay & Mason 1990), with diverse invertebrate and ichthyofauna possessing almost 85 % of reef fish species endemic to the Western Indian Ocean region (UNEP 2005). Beyond the coral reef are deeper suspension feeder reefs flanked by subaqueous dunes on the seaward and landward side (Ramsay 1994). In northern KwaZulu-Natal a shelf break occurs at a distance of between 2.1 and 4.1 km offshore (Ramsay 1994), corresponding to depths of between 45 and 70 m, and is incised by 12 submarine canyons (Viana *et al.* 1998) three of which occur off Sodwana Bay (Ramsay 1994, Hissmann *et al.* 2006). Caves found within the canyons, which are inhabited by coelacanths, are erosional features indicating that this zone was previously in the intertidal during the last glacial maximum (Green & Uken 2005). The shelf of the northern KwaZulu-Natal coastline is on average < 65 m deep (Ramsay 1994).

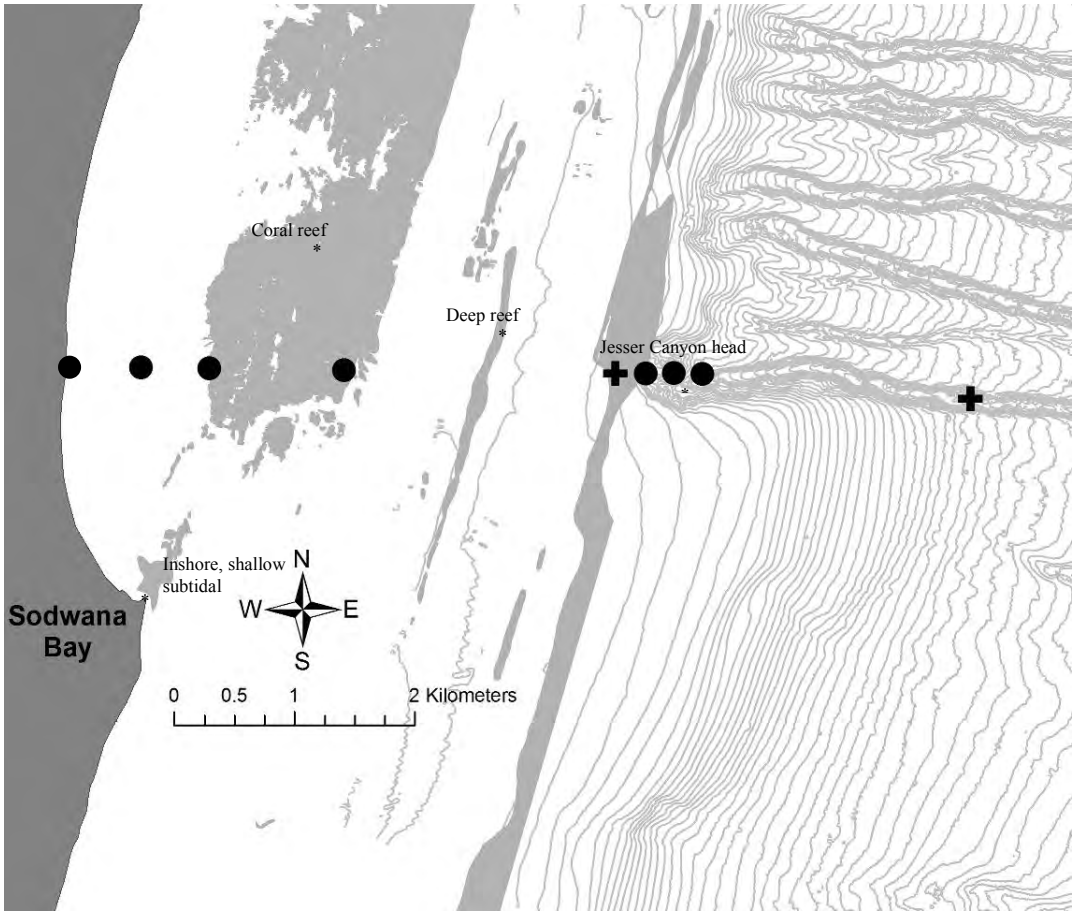


Figure 1.2: Map of the study site, indicating the position of the various habitats, and particulate organic matter sampling stations (Chapter 2). Circles indicate position of stations sampled between August and December 2010, crosses indicate the additional stations sampled thereafter.

The shallower, inshore region includes the shallow subtidal and rocky intertidal habitats and the coral reefs. The nearshore/intertidal rocky habitats along the stretch of coastline incorporating Sodwana Bay tend to predominantly include filter-feeding and grazing invertebrates (Sink *et al.* 2005). Coral reefs are highly productive regions, supporting diverse flora and fauna (Odum & Odum 1955), and provide many goods and ecological services (Moberg & Folke 1999). The ecological role of coral reefs is varied, from providing a habitat and substrate to fishes, invertebrates and primary producers to protecting the coast against

wave exposure and erosion (Jennings & Kaiser 1998). Sodwana Bay's reefs have been suggested to offer coral-organisms a refuge against periodic elevated water temperatures because of the upwelling of cold, deep water during periods where water temperatures could have otherwise caused corals to bleach (Riegl & Piller 2003). Unfished coral reefs have a high secondary production potential, in terms of fish biomass, with fishing pressure drastically reducing this (Friedlander & DeMartini 2002). Due to the existence of corals in warm, clear water the abundance of phytoplankton is generally low, despite this large amounts of plankton are filtered from water passing over the coral reef (Odum & Odum 1955). With the low phytoplankton abundance, and clear waters allowing for a deep euphotic zone (Ramsay & Mason 1990), the production by benthic algae is likely to be important to the consumers occurring on the coral reefs, and inshore of this zone.

The seaweeds occurring in the region are classified as predominantly tropical Indo-West Pacific (De Clerck *et al.* 2005). Within the inshore habitats there may be a high biomass of seaweeds, with a large proportion of the consumers occurring there reliant on them as a source of production (Bustamante & Branch 1996). It was shown, around the coast of South Africa, that the biomass of intertidal herbivorous grazers was correlated with the productivity of *in situ* algal producers (Bustamante *et al.* 1995). Filter-feeder biomass, while not influenced by rates of production by intertidal producers, were related to the nearshore primary production rates (Bustamante *et al.* 1995). Phytoplankton contribution to nearshore consumers has been shown to be limited (Hill 2007). The nearshore habitats in Sodwana Bay are likely to have a higher availability of seaweed production than deeper habitats, with a depth gradient of seaweed biomass already noted (Anderson *et al.* 2005). Seagrass, *Thallassodendron ciliatum*, occurs near the rocky point at the southern end of the bay. Due to the refractory nature of seagrasses in general, they have been found to be of little value as a

carbon source to consumers (Stephenson *et al.* 1986). The export of nearshore production to deeper habitats is possible.

Little is known of the biota occurring in the deeper habitats because of a lack of focussed scientific exploration in this zone (Sink *et al.* 2006). At depths greater than 25 m, coral cover is reduced, with increasing dominance by non-photosynthetic organisms including sponges, ascidians and sea-fans (Riegl *et al.* 1995). Due to the reduced occurrence of photosynthetic organisms, consumers in the deeper habitats, including the deep reefs and canyons, are likely to rely on allochthonous sources of production, that may be available in various forms of organic matter from dissolved form to macrozooplankton, derived from primary production occurring in the overlying surface waters or imported from shallower habitats that is transported to the deeper benthos (Bosley *et al.* 2004, Duineveld *et al.* 2004, Carlier *et al.* 2009). Deep reef and canyon consumers in Sodwana Bay likely derive a large proportion of their organic matter from allochthonous sources, originating partly from particulate matter in the water column. This particulate matter may include phytoplankton as well as seaweed-derived organic matter.

1.4 Aims and objectives

The management and conservation of marine ecosystems, both those which are exploited and those which are not, requires an understanding of their habitats and their trophic structure. Gaining an insight into the trophic dynamics and energy flows within a marine ecosystem is vital and may contribute to the informed management of the system through an ecosystem-based approach (Forget *et al.* 2011).

This thesis aims to address some of the deficiencies regarding our knowledge of energy flows, from the major primary producers through to higher trophic levels, including species that are of general interest that occur off Sodwana Bay such as the coelacanth and large pelagic game fish that are both major scientific and tourist attractions to this stretch of coastline. The degree of connectivity between the different habitats from the intertidal zone down to the canyons together with shallow and deep reefs and the pelagic zone is unknown. With certain forms of marine resource exploitation allowed in this section of the iSimangaliso Wetland Park, such as pelagic game fishing and, arguably, SCUBA diving (Schleyer & Tomalin 2000), quantifying the connectivity between habitats and the species occurring within them is of significance in understanding how consumptive practices may influence seemingly unrelated organisms, within the different habitats. This thesis therefore aims to expand on the knowledge of the ecology of the Sodwana Bay marine ecosystem, especially in terms of its trophic ecology and the relative importance of the potential sources of primary production to higher trophic levels at the different depth and habitat zones.

1.5 Thesis outline

This thesis has been divided into four chapters. Chapter 2 investigates the spatial and temporal dynamics of the particulate organic matter (POM) available to consumers along a transect from the intertidal to a submarine canyon utilising both stable isotope and fatty-acid analyses. It was hypothesised that there would be a gradient in the type of organic matter, derived from different production sources, available to the consumers in the different habitats. These gradients are hypothesised to be related to light availability at different depths and to be influenced by primary producer composition, abundance and productivity. The deep reef and canyon habitats were hypothesised to rely predominantly on allochthonous sources of production being imported either from inshore, where benthic producers were within the

euphotic zone, or from pelagic production occurring in the water column above the deeper habitats. Potential causal factors for the observed spatial and temporal variation in the POM are discussed.

Chapter 3 examines trophic relationships between organisms from the intertidal and shallow subtidal, coral reefs, deep reefs, canyon and pelagic zones. It was hypothesised that the utilisation of organic matter by consumers at each of the habitat zones would reflect the availability of organic matter, evident from the POM analysis in Chapter 2. The major sources of primary production utilised by the consumers at the different depth zones are discussed as well as the potential connectivity between inshore and deeper habitats.

To conclude, Chapter 4 discusses the importance of the gradient observed in organic matter sources, and the utilisation of this primary production in the context of the study area. Being a conservation area, potential threats to the habitats and species that rely on them are discussed. The impact of the recreational activities allowed in the Park, namely fishing for pelagic fishes and SCUBA diving, are considered from a trophic ecology perspective, addressing the potential impacts, observed in other studies, which may occur here.

Chapter 2

Particulate organic matter dynamics along a nearshore-offshore gradient off Sodwana Bay, South Africa

2.1 Introduction

Organic matter within the aquatic environment occurs as both particulate organic matter (POM) and dissolved organic matter (DOM). POM comprises suspended material that includes living phytoplankton as well as phytoplankton-, macrophyte- and zooplankton-derived detritus. POM can provide a detailed and integrated record of energy derived from autochthonous and allochthonous sources. As a consequence it is important in food web studies that aim to understand the trophic linkages between organisms within a community from primary production through to higher trophic level consumers (Carpenter *et al.* 1987). These studies have typically used dual stable isotope tracers of carbon and nitrogen, $\delta^{13}C$ and $\delta^{15}N$, such that the energy sources and trophic positions, respectively, of organisms can be inferred (DeNiro & Epstein 1981, Vander Zanden & Rasmussen 2001, Post 2002). An alternative approach is fatty acid analysis. Fatty acids are synthesised by primary producers and may serve as biomarkers, if they are rare or unique to a given taxon, or as indicators, if they are elevated in certain taxa and/or absent in others (Kelly & Scheibling 2012). Both their presence and relative abundance are, therefore, important (Kelly & Scheibling 2012). Fatty acids are particularly useful in investigating pelagic foodwebs and especially as a means of identifying the composition of POM/phytoplankton (Dalsgaard *et al.* 2003). Ratios of certain fatty acids may indicate the relative proportion of certain taxa in a mixed-phytoplankton sample, for example diatoms and dinoflagellates (Budge & Parrish 1998). If there are shared fatty acids between primary producers, for example seaweeds and phytoplankton, these ratios

may be ambiguous. The dynamics of the POM stable isotope signatures and fatty acids have also been used to understand some of the intricacies of carbon and nitrogen cycling, both of which have important effects on primary production, but are difficult processes to investigate.

One important component of POM is phytoplankton. Phytoplankton species composition and abundance varies in relation to the prevailing environmental conditions such as temperature, nutrient availability and mixing (Müren *et al.* 2005). Temperature changes affect the planktonic foodweb with phytoplankton species assemblages and growth rates varying with changes in temperature and the abundance of heterotrophic organisms generally increasing with temperature (Müren *et al.* 2005). Such shifts in species composition and growth rates are reflected in the POM's stable isotope signatures (Gu *et al.* 2011). Diatom blooms, for example, are characterised by enriched carbon signatures (Fry & Wainright 1991) and when nutrient concentrations are low, in warm, stable water columns, cyanobacteria have been found in abundance (Müren *et al.* 2005). Blooms of cyanobacteria account for most of the nitrogen fixation in oceanic waters (Howarth *et al.* 1988). Nitrogen fixation facilitates primary production even when nitrogen sources are otherwise limiting (Howarth *et al.* 1988), and may be discerned from the $\delta^{15}N$ signatures of the POM.

Carbon stable isotopes of POM generally reflect variations in carbon sources that include carbon dioxide, carbonate and bicarbonate ions (Deuser *et al.* 1968). Enriched POM carbon signatures may indicate that the phytoplankton obtain their carbon from bicarbonate ions, as opposed to more depleted CO_2, during periods of low CO_2 availability (Deuser *et al.* 1968). Carbon signatures also provide information relating to phytoplankton growth rates. In this situation, depleted signatures are often associated with low growth rates, while periods with enriched signatures indicate higher growth rates (Gu *et al.* 2006). Carbon signatures can also

be influenced by phytoplankton size through boundary layer effects whereby larger phytoplankton that have larger boundary layers exhibit lower isotopic discrimination while smaller cells have enriched isotope signatures (Korb *et al.* 1996). Similarly, nitrogen isotope signatures reflect the dynamics of the nitrogen pool from which phytoplankton obtain their inorganic nitrogen. The nitrogen available to producers originates either from regenerative processes that utilise ammonia as a nitrogen source resulting in enriched nitrogen isotope signatures, or from direct fixation from dissolved atmospheric nitrogen in the form of nitrates, which leads to isotope signatures being more similar to that of atmospheric nitrogen, or from upwelling which introduces nutrient-rich sub-surface waters to the surface (Dugdale & Goering 1967, Wada & Hattori 1976, O'Reilly *et al.* 2002). In those environments subjected to upwelling events phytoplankton will initially possess relatively depleted nitrogen signatures as there is an abundance of nitrogen sources and discrimination against ^{15}N occurs (O'Reilly *et al.* 2002). As the nitrogen source decreases, discrimination against ^{15}N also decreases resulting in phytoplankton signatures close to those of the nutrient source (O'Reilly *et al.* 2002). Where a significant proportion of the POM is comprised of seaweed detritus, this is evident using stable isotope analysis as seaweeds generally have enriched carbon signatures compared to phytoplankton (Lara *et al.* 2010).

From a South African perspective, Hill (2007) found a fairly consistent trend of offshore depletion of carbon isotope signatures around the coastline of South Africa. This was assumed to reflect the input of nearshore sources of primary production to the POM. This implies that primary production occurring inshore is available to consumers even at some distance from the shore. While the inshore-offshore gradients are assumed to reflect a decreasing input from inshore sources of primary production, this process is poorly understood both spatially and temporally.

Understanding the composition of the POM within Sodwana Bay can provide the necessary information pertaining to those different sources of production, including contributions from macro- and microalgal assemblages and understanding what organic matter sources are available to consumers at different depths and in different habitats. Benthic sedimented organic matter and benthic diatoms were previously discounted as an important source of carbon to consumers, with very little organic matter occurring in the sediments collected from Sodwana Bay (S. Kaehler, RU, pers. comm.). The dynamic nature of the sediment caused by the characteristically strong currents and wave action may prevent the accumulation of benthic organic matter.

This chapter investigated the POM inshore-offshore gradient off Sodwana Bay from a spatio-temporal perspective using both stable isotope and fatty acid analyses to address questions related to POM composition and distribution, in order to verify the assumption that the inshore signature was indicative of a distinct suite of producers, and to determine the degree of temporal variation within the POM. It was hypothesised that there would be a gradient in the type of organic matter, derived from primary producers, moving offshore. It was also hypothesised that there would be distinct inshore and offshore patterns related to the primary producers occurring inshore, including seaweeds, which diminish in abundance further offshore. These patterns are hypothesised to be related to light availability at different depths and its influence on the assemblage of primary producers and their respective rates of primary production.

2.2 Materials and methods

2.2.1 Stable isotope and lipid analysis

POM and oceanographic parameters were sampled monthly between August and December 2010 along a transect running perpendicular to the shore and terminating over the head of a large submarine canyon, Jesser Canyon, which begins approximately 4.6 km offshore. Sampling was commenced a few hours before mid-day. There were six stations – in the intertidal region and at 0.5, 1.0, 2.0, 4.4 and 4.6 km offshore. From February to May 2011 two additional stations were sampled to improve the resolution around the deeper stations, and to also include an oceanic station removed from the shelf (Figure 1.2). The additional stations were 4 and 6.5 km offshore. At each station a CTD cast (YSI 600XLM with conductivity, temperature, depth and oxygen sensors) was conducted and 10 L water samples collected from one metre below the surface and near the bottom, down to a maximum depth of 150 m at the deeper stations. Once ashore, water samples were immediately filtered through precombusted (500 ^{o}C, 6 hours) Whatman GF/F filter papers (glass fibre, 0.4um mesh size) using a vacuum system (vacuum < 4 mmHg) connected to a collection sump. Filter papers were examined and any visible zooplankton removed prior to the filter papers being dried at ~50 ^{o}C overnight in an oven. Dried samples were stored in aluminium foil pouches placed into a container with silica gel desiccant. The surface layers of the filter paper containing POM were then removed and placed into tin capsules and combusted. Stable isotope composition was determined with a Europa Scientific ANCA-SL Elemental Analyser and 20-20 IRMS. Stable isotope signatures were expressed in standard δ notation as $\delta X(‰) = \left(\frac{R_{sample}}{R_{standard}} - 1\right) \times 1000$ where X is the heavier isotope, either ^{13}C or ^{15}N, and R is the ratio of the heavy to the light isotope. Internal standards of casein, ammonium sulphate and beet sugar were used and calibrated against Vienna PeeDee Belemnite and atmospheric nitrogen, for carbon and nitrogen, respectively. Overall precision of stable isotope analyses were 0.12 ‰ for δ^{15}N and 0.10 ‰ for δ^{13}C.

A single transect was conducted to collect POM for lipid analysis. The day prior to the May POM stable isotope sample collection, additional 10 L water samples were collected from the intertidal, and from the surface (1 m deep) and near the bottom, down to a maximum depth of 150 m at the deeper stations, from stations 0.5, 1.0, 4.4, 4.6 and 6.5 km offshore. Water samples from each station were filtered through pre-combusted (500 °C, 6 hours) GF/F filter papers, placed into aluminium foil sleeves and frozen in a standard chest-freezer. Thereafter samples were stored at -80 °C until they were analysed for fatty-acid composition. Before analysis, filter papers for fatty-acid analysis were lypholised. The freeze-dried filters were placed in 2 ml chloroform under nitrogen flow, in lipid-cleaned vials and stored at -20 °C. Total lipids were extracted from each filter paper using a modified Folch procedure (Folch *et al.* 1957). Whole lipids were extracted in 2:1 (v/v) chloroform/methanol, 0.5 ml of chloroform-extracted water was added, and the lipid layers removed and combined after each of 3 chloroform washes. A given quantity of internal standard (19:0) was added to each extract for quantification of fatty acid methyl esters (FAMEs). FAMEs were prepared by heating the extracts suspended in hexane and 14 % boron trifloride-methanol at 80 °C for 1.5 h (adapted from Budge and Parrish (1998)).

Gas chromatographic (GC) analyses of FAMEs were performed with a Hewlett Packard 5890A GC equipped with a bonded and cross-linked 78% cyanopropyl methylpolysiloxane-fused silica capillary column (30 m length, 0.25 mm inner diameter, 0.25 μm film thickness; Quadrex Corporation) with helium as the carrier gas. One microlitre sample aliquots were manually injected, at 250°C, with the oven set at 100°C for the first 3 min. The oven temperature was increased to 150°C at a rate of 5°C .min^{-1}, for 1 min, and raised to 220°C at 3.5°C .min^{-1} (flame ionization detector was set at 260°C). Peaks were integrated using 32 Karat 5.0 software (Beckman Coulter Inc.) and identified using mass spectral (MS) data

derived from a subset of the samples and by comparing retention times with those of external standards (37 component FAMEs and marine PUFA no. 1, Supelco).

Fatty acids that were useful in differentiating between potential sources of primary production were analysed. The average proportions of fatty acids in different groups of primary producers were ascertained from the literature from Carpenter *et al*. (1997), Dalsgaard *et al*. (2003) and Kelly & Scheibling (2012). The fatty acids analysed included 14:0, (average proportion in red seaweeds: 4.45 %, diatoms: 13.51 %), 20:5n-3 (average proportion in dinoflagellates: 10.39 %, brown seaweeds: 11.73 %, diatoms 15.19 %, red seaweeds: 30.34 %), 18:1n-9 (average proportion in vascular plants: 4.5 %, red seaweeds: 7.37 %, dinoflagellates: 13.17 %, brown seaweeds: 14.15 %), 22:6n-3 (average proportion in dinoflagellates: 13.14 %), and the fatty acid ratios 20:5n-3/22:6n-3 (a high ratio indicates a high contribution of diatoms/seaweeds), 16:1n-7/16:0 (a high ratio indicates a high diatom presence), various C^{15} fatty acids (cyanobacteria).

Data on wind patterns in the grid encompassing 27 – 30 °S and 31 – 34 °E, recorded by voluntary observing ships, were supplied by the Southern African Data Centre for Oceanography (SADCO). Values were mean monthly wind speed and monthly modal direction. The mixed layer depth was determined from temperature profiles at the 4.6 km site and was defined as that depth at which a maximum change in temperature occurs, i.e., the thermocline depth, above which the temperature is relatively constant.

2.2.2 Data analysis

Inshore-offshore and depth gradients were investigated using contour plots of the POM variables, $\delta^{13}C$, $\delta^{15}N$, C:N ratios and sample carbon content for all months for both the surface and bottom water samples.

The influence of environmental variables – sampling month, temperature, oxygen concentration, MLD and wind speed – on the POM variables $\delta^{13}C$, $\delta^{15}N$, C:N ratios, and sample carbon content was investigated using Redundancy Analysis (RDA) – a form of linear ordination that indicates those variables that account for the observed patterns within the data matrix. RDA allows for the examination of data collected with a repeated-measures sampling design, such as the monthly data collected in the present study. Salinity data unfortunately could not be used with any confidence due to instrument calibration issues, and were therefore excluded from analyses. Contributions of the variables to the ordination were tested for significance using Monte Carlo simulations (n = 499 permutations and α = 0.05). Ordinations were performed in *CANOCO v4.5*. To balance the data across all months of sampling, the additional stations post December 2010 were excluded from the RDA.

Each fatty acid was expressed as a proportion of the total identified fatty acids (%TFA). Only those fatty acids that were useful in interpreting POM composition were presented. The proportions of these fatty acids at each station were plotted against distance from the shore to investigate any inshore-offshore gradients.

2.3 Results

2.3.1 Environmental variables

The temperature profiles, illustrated in Figure 2.1, partially explain the environmental characteristic patterns observed within the POM. Surface temperatures increased from ~ 22

°C in August to ~ 26 °C in November, decreasing slightly in December, reaching a maximum of ~ 28 °C in March and decreasing thereafter (Figure 2.2 b). Bottom temperatures did not reflect the patterns noted in the surface temperatures (Figure 2.3 c). The lowest recorded bottom temperature was ~ 14 °C in August, at a depth of 124 m. In September, bottom temperature was highest at 19.5 °C, due to the deep mixed layer at the time (Figure 2.3 a), but decreased again in October to ~ 16 °C. There was a gradual increase in the bottom temperature between October and December. Between February and May the bottom temperature was relatively stable, with only minor fluctuations noted. Among bottom samples, the RDA found temperature to be positively correlated with the sample carbon content and the carbon isotope signatures.

Surface dissolved oxygen (DO) concentrations were higher in August (9.88 $mg.l^{-1}$) and fairly consistent among other months (Figure 2.2 b). DO concentration at the bottom samples ranged between 4.38 and 6.83 $mg.L^{-1}$ (Figure 2.2 c). Bottom DO concentrations declined from 6.57 $mg.L^{-1}$ in August to 4.38 $mg.L^{-1}$ in November, and increased in December and the following February. High C:N ratios among bottom POM samples were associated with periods of decreased DO (Figure 2.1 c).

Wind speed between August and November was relatively consistent averaging ~ 10 $m.s^{-1}$. It was lowest, 5.3 $m.s^{-1}$, in February. Wind speed rose from March to values comparable to those pre-December (Figure 2.2 a). The wind most frequently blew from the north-east, being the modal direction in 5 out of 10 months. The C:N ratios of the POM were highest during November and December, following a period of the strongest, northerly winds.

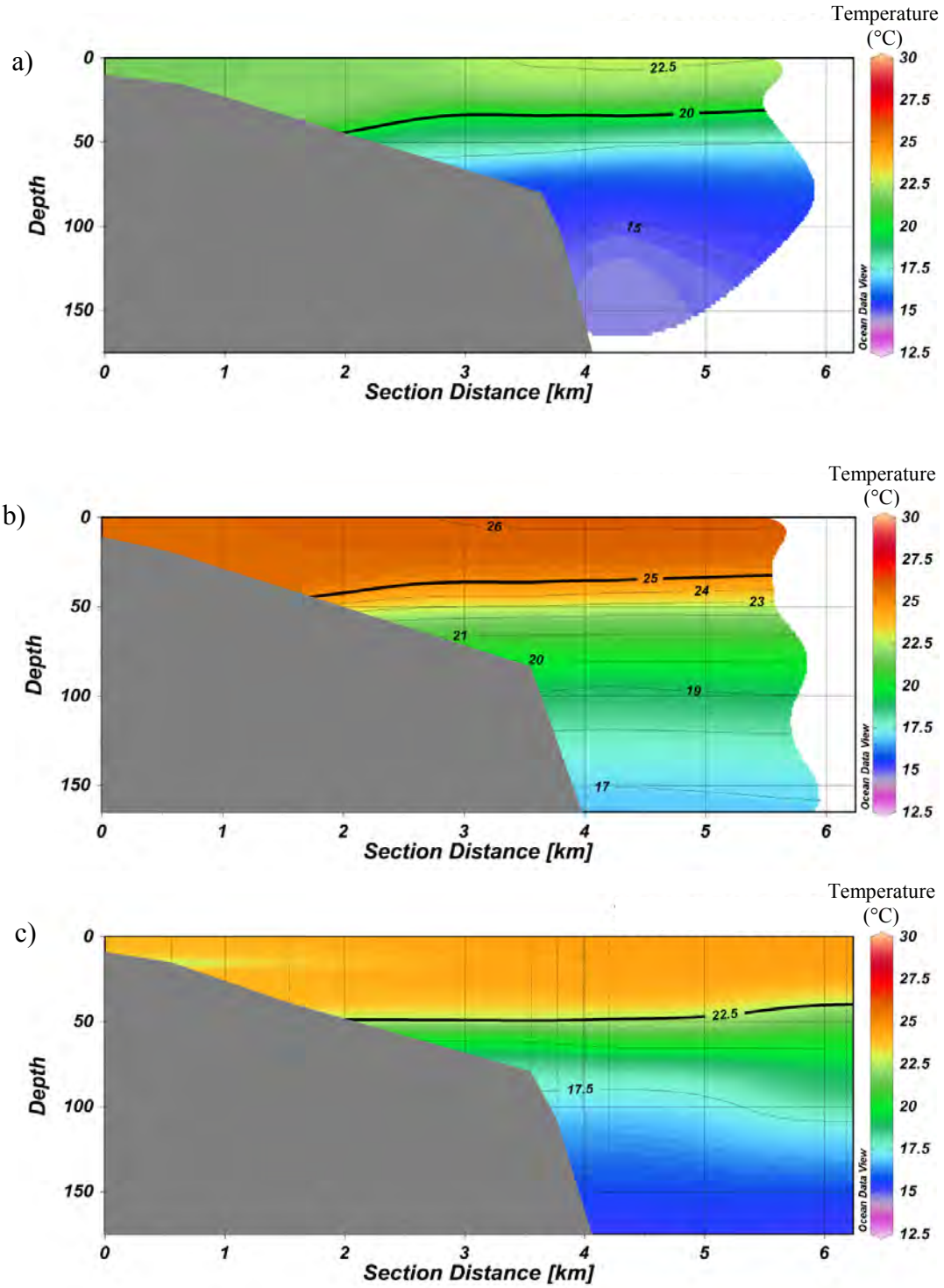


Figure 2.1: Temperature sections for a) August 2010, b) November 2010 and c) May 2011. Mixed layer depth (MLD) represented by bold isotherm. Transects during August and November ended 4.6 km offshore, post-December the transect was extended to 6.5 km offshore.

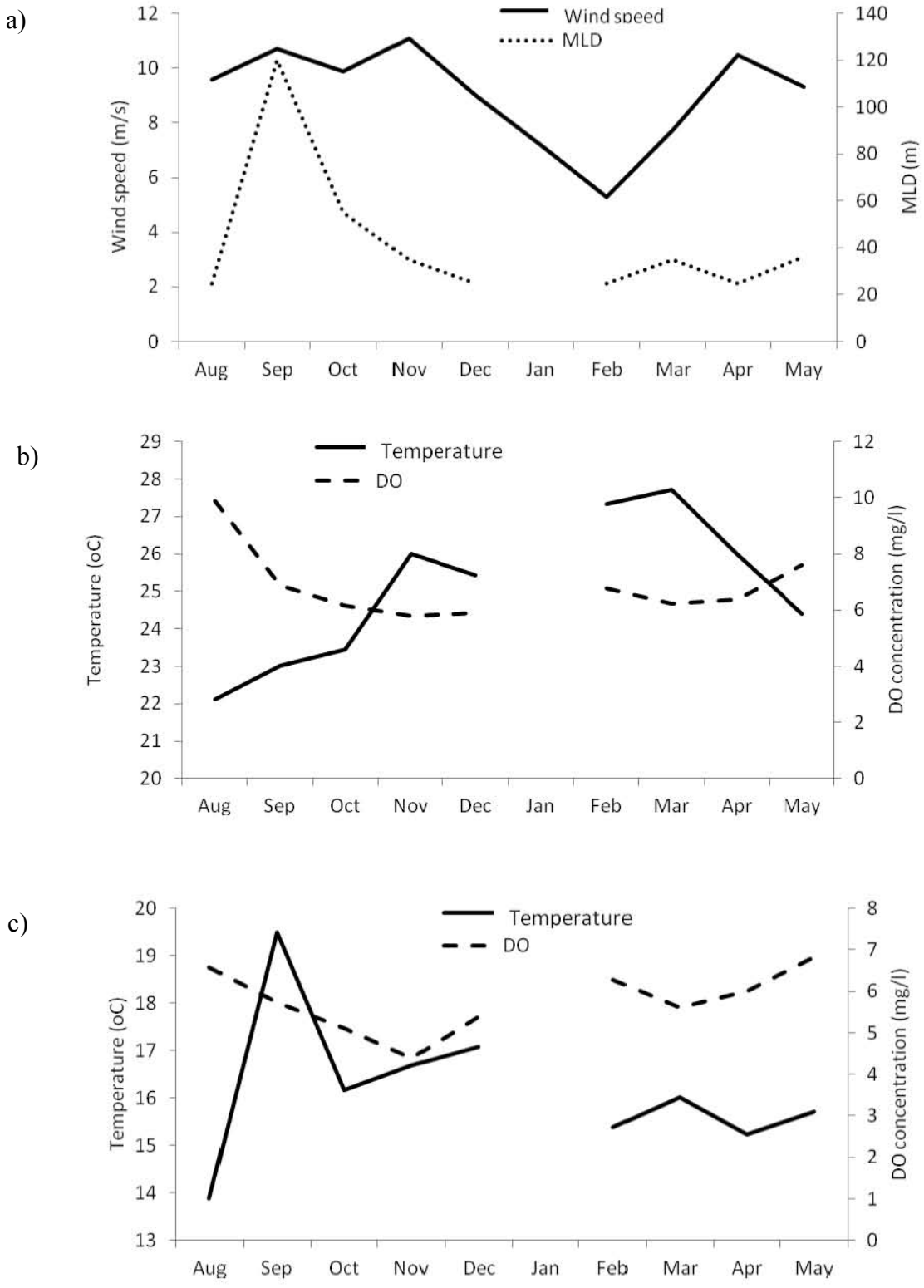


Figure 2.2: Environmental variables for all months sampled. a) Wind speed and mixed layer depth (MLD), and dissolved oxygen (DO) concentration and temperature at sampling depth for b) surface and c) bottom samples, using the 4.6 km station as a representative, with other stations exhibiting similar trends.

MLD increased from 25 m in August to 120 m in September (Figure 2.3 a). In October and November the MLD decreased again, reaching ~ 25 m again in December. From February to May the MLD was between ~ 25 and 35 m. Periods of lower MLD were associated with increased carbon content and enriched carbon signatures in surface POM samples (Figure 2.1 b). Among bottom samples, shallower MLD was associated with higher C:N ratios and low DO concentrations (Figure 2.1 c).

2.3.2 Stable isotopes

The POM variables indicated three distinct periods (Figure 2.3). The first period, between August and October, was characterised by relatively depleted carbon signatures (range: -23.31 to -17.96 ‰) that coincided with relatively enriched nitrogen signatures (range: 4.17 - 9.47 ‰) and intermediate C:N ratios (range: 6.03 to 11.68) (Figure 2.3 a-d, g, h). In the second period, evident in November and December, POM was characterised by extremely high C:N ratios in both surface and bottom samples (11.48 - 16.89) (Figure 2.3 g, h). The third period was between February and May. This period had characteristically enriched POM $\delta^{13}C$ values (-22.92 to -16.83 ‰), with concurrently depleted $\delta^{15}N$ signatures (1.54 - 10.73 ‰), and low C:N ratios (5.19 - 7.70) (Figure 2.3 a-d, g, h).

There was some agreement between the surface and bottom samples. In the surface samples, carbon signatures ranged between -22.83 and -16.83 ‰ (Figure 2.3 a) and were slightly more enriched relative to the bottom samples' signatures that ranged between -23.31 and -18.55 ‰ (Figure 2.3 b). Surface nitrogen ranged between 2.03 and 7.20 ‰ while bottom sample signatures ranged between 1.54 and 10.73 ‰. Surface sample carbon content ranged from 9.19 to 151.08 $\mu gC.L^{-1}$ (Figure 2.3 e). The April and February surface samples possessed the lowest carbon content overall, with the August, December and March samples tending to

have the highest carbon content (Figure 2.3 e). Among the bottom samples, carbon content was relatively high in August, December and February samples while March to May samples had low carbon content (Figure 2.3 f). Offshore bottom samples had lower carbon content, relative to those of the inshore samples, with values ranging between 4.85 and 85.64 $\mu gC.L^{-1}$. Between August and February samples from stations deeper than 40 m tended to have less carbon than shallower, inshore stations (Figure 2.3 f).

While the RDA detected a strong temporal trend in all of the POM variables measured, the POM variability could not be fully explained by the environmental variables suggesting that other, unmeasured physico-chemical variables influenced POM over the study period. This was particularly noticeable in the surface samples. After the temporal aspect was removed from analyses, 18 % of the variation in the combined, surface and bottom sample, analysis was explained by the environmental variables recorded (Table 2.1) and accounted for 57 % of the variability in the surface data and 45 % for the bottom data when analysed separately.

In the combined data RDA, that incorporated both the surface and bottom samples (Figure 2.4 a), the predictor variables significantly explained 18 % of the total variation (Monte Carlo permutations of the first axis, $F = 18.89$, and trace, $F = 3.78$, with $p = 0.03$ for both tests). Almost all the variation (99.9 %) was explained by the first axis that was associated with temperature, DO and the depth (surface/bottom) (Table 2.1). Enriched $\delta^{13}C$ signatures and high loads of particulate carbon were associated with warmer, deeper water. Higher C:N ratios were associated with periods with high wind speed. Because depth was an important predictor variable, the surface and bottom sample data were then analysed separately.

Within the surface sample data, the first two RDA axes (Table 2.1, Figure 2.4 b) significantly accounted for 57 % of the variation (for the first axis $F = 47.00$, $p < 0.01$, and trace $F = 5.88$, $p < 001$). The first axis, which explained most of the variation, was associated with MLD, distance from shore, and the wind directions north and south, while wind speed and DO were associated with the second axis. Particulate carbon content in the samples was negatively correlated with distance from shore and MLD. The C:N ratios were positively correlated with wind speed, particularly northerly winds, with $\delta^{15}N$ signatures exhibiting a similar, yet weaker, correlation.

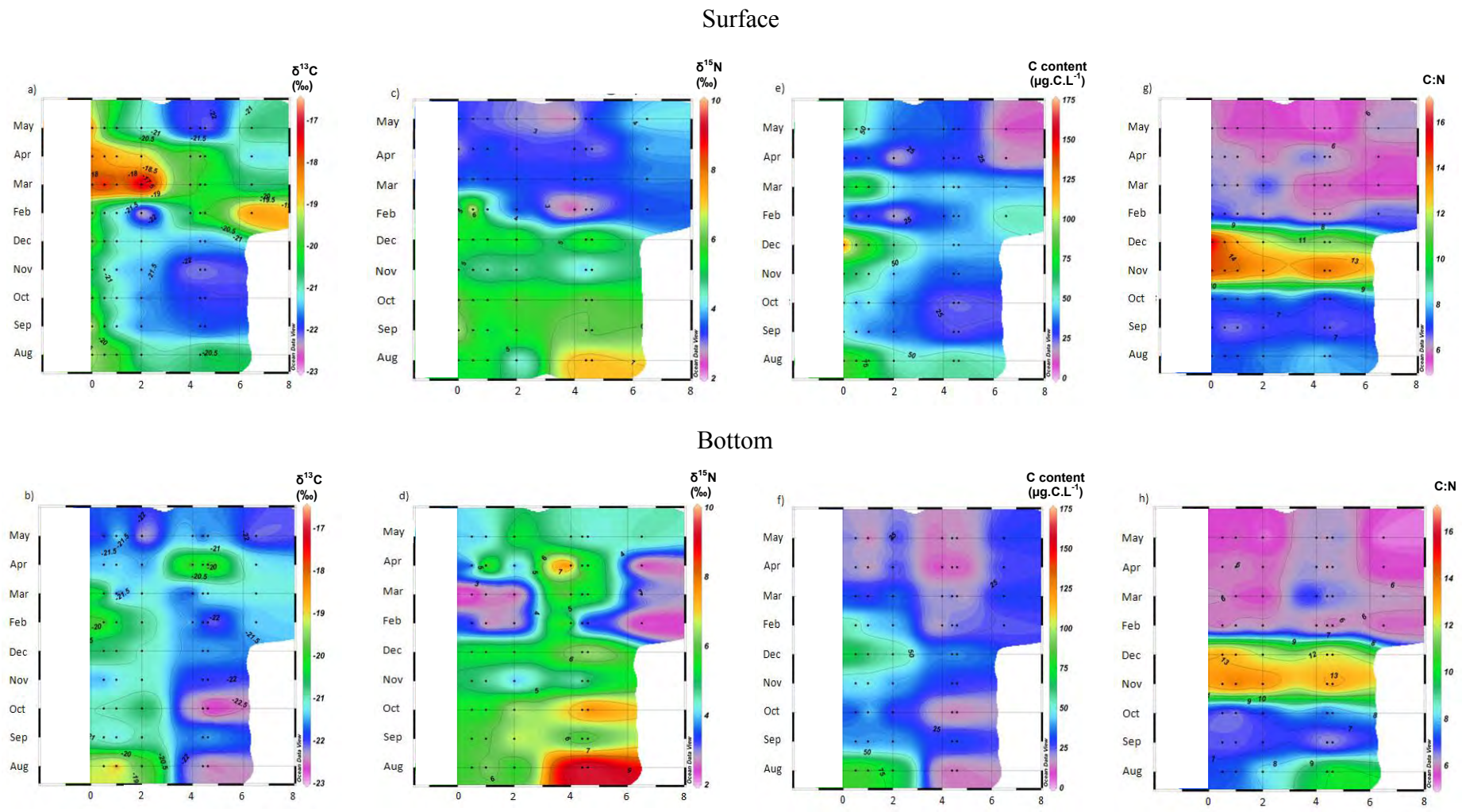


Figure 2.3: Particulate organic matter (POM) δ13C, δ15N, carbon content and C:N ratios for surface and bottom samples. Months in chronological order from bottom to top of plot.

The first two RDA axes explained 45 % of the variation within the bottom sample data (Table 2.1, Figure 2.2 c). The first axis was associated with temperature and distance from shore, while the second axis was associated with DO, wind speed and the wind directions north, south and east. The carbon content within the samples, and the carbon isotope signatures, were positively correlated with temperature. High C:N ratios and enriched nitrogen signatures occurred when strong northerly winds blew and DO concentrations were low.

2.3.3 Fatty acids

The fatty acids, 14:0 and 20:5n-3, both exhibited an overall decline moving offshore (Figure 2.5 a, b). Similarly there was an increasing trend in the proportion of 18:1n-9 and 22:6n-3 (Figure 2.5 c, d). The ratio of the fatty acids 20:5n-3/22:6n-3 declined, among surface samples, moving offshore (Figure 2.5 e). None of the bottom samples possessed 22:6n-3, with 4.6 and 6.5 km stations also containing no 20:5n-3. The ratio of the fatty acids 16:1n-7/16:0 showed no variation due to the consistently high proportion of 16:0 fatty acids in the lipids (Figure 2.5 e). The fatty acid iso 15:0 ranged between 0.41 and 1.61 %, ante 15:0 ranged between 0.18 and 0.54 % and 15:0 ranged between 0.7 and 2.4 % of the total lipid content (Figure 2.5 f).

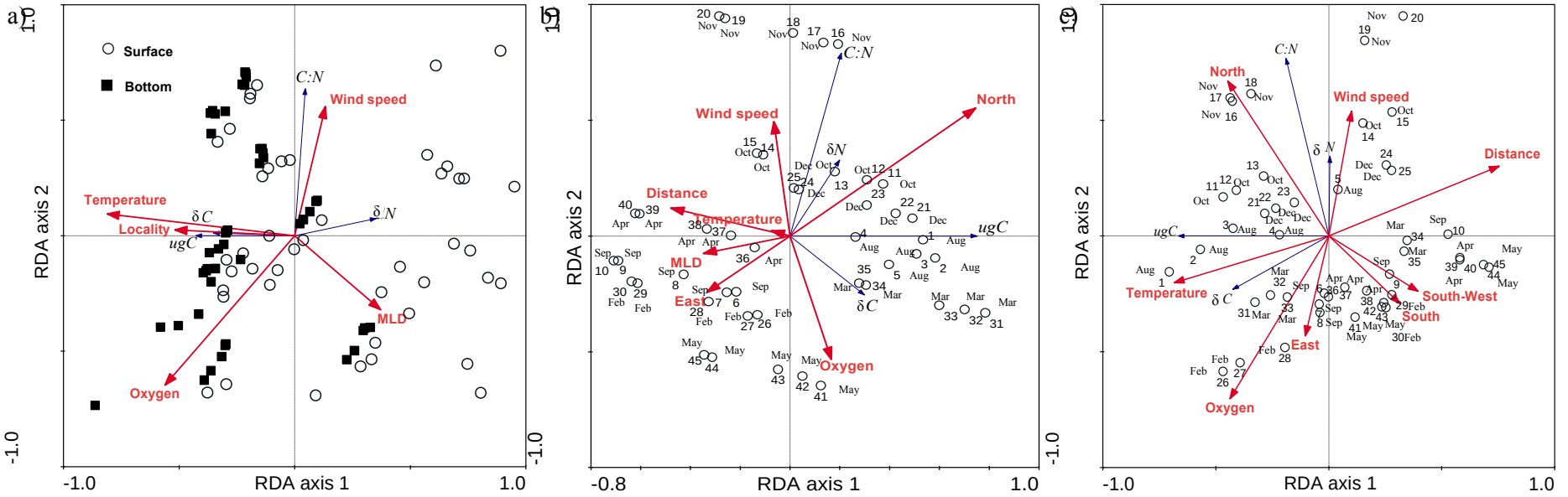


Figure 2.4: RDA ordination plots of particulate organic matter (POM) variables (blue arrows; carbon and nitrogen stable isotope ratios, represented by *δC* and *δN,* respectively, carbon : nitrogen ratio, represented by *C:N*, and the mass of particulate carbon, represented by *ugC*) and environmental variables (red arrows; mixed layer depth, represented by *MLD*, average monthly wind speed and directions, dissolved oxygen concentration and temperature at depth of sample collection, locality (surface or bottom sample), and distance of sample collection from shore. a) combined surface and bottom samples, and separate b) surface and c) bottom samples.

Table 2.1: Summary statistics for redundancy analyses conducted on particulate organic matter (POM) and several environmental variables.

Surface and bottom samples combined	Axis 1	Axis 2		
Eigenvalues	0.18	0.00		
POM-environment correlations	0.43	0.61		
Cumulative percentage variance:				
of POM data	18.4	18.4		
of POM-environment relationship	99.9	100		
Correlations with axes:				
Temperature	-0.35	0.06		
Mixed layer depth (MLD)	0.16	-0.19		
Oxygen	-0.24	-0.39		
Wind speed	0.06	0.34		
Locality	-0.22	0.02		
Sum of all canonical eigenvalues	0.18			
RDA for segregated surface and bottom samples				
	Surface samples		Bottom samples	
	Axis 1	Axis 2	Axis 1	Axis 2
Eigenvalues	0.57	0.00	0.45	0.00
POM-environment correlations	0.75	0.83	0.67	0.81
Cumulative percentage variance				
of POM data	56.6	56.6	44.5	44.5
of POM-environment relationship	100	100	100	100
Correlations with axes				
Temperature	-0.06	0.02	-0.46	-0.16
Wind speed	-0.05	0.41	0.07	0.43
Mixed layer depth (MLD)	-0.26	-0.06	0.07	-0.14
Oxygen	0.12	-0.44	-0.29	-0.57
Distance from shore	-0.36	0.10	0.50	0.24
North	0.56	0.46	-0.30	0.54
South	-0.42	-0.19	0.21	-0.24
East	-0.25	-0.20	-0.07	-0.34
South-west	-0.08	-0.28	0.26	-0.19

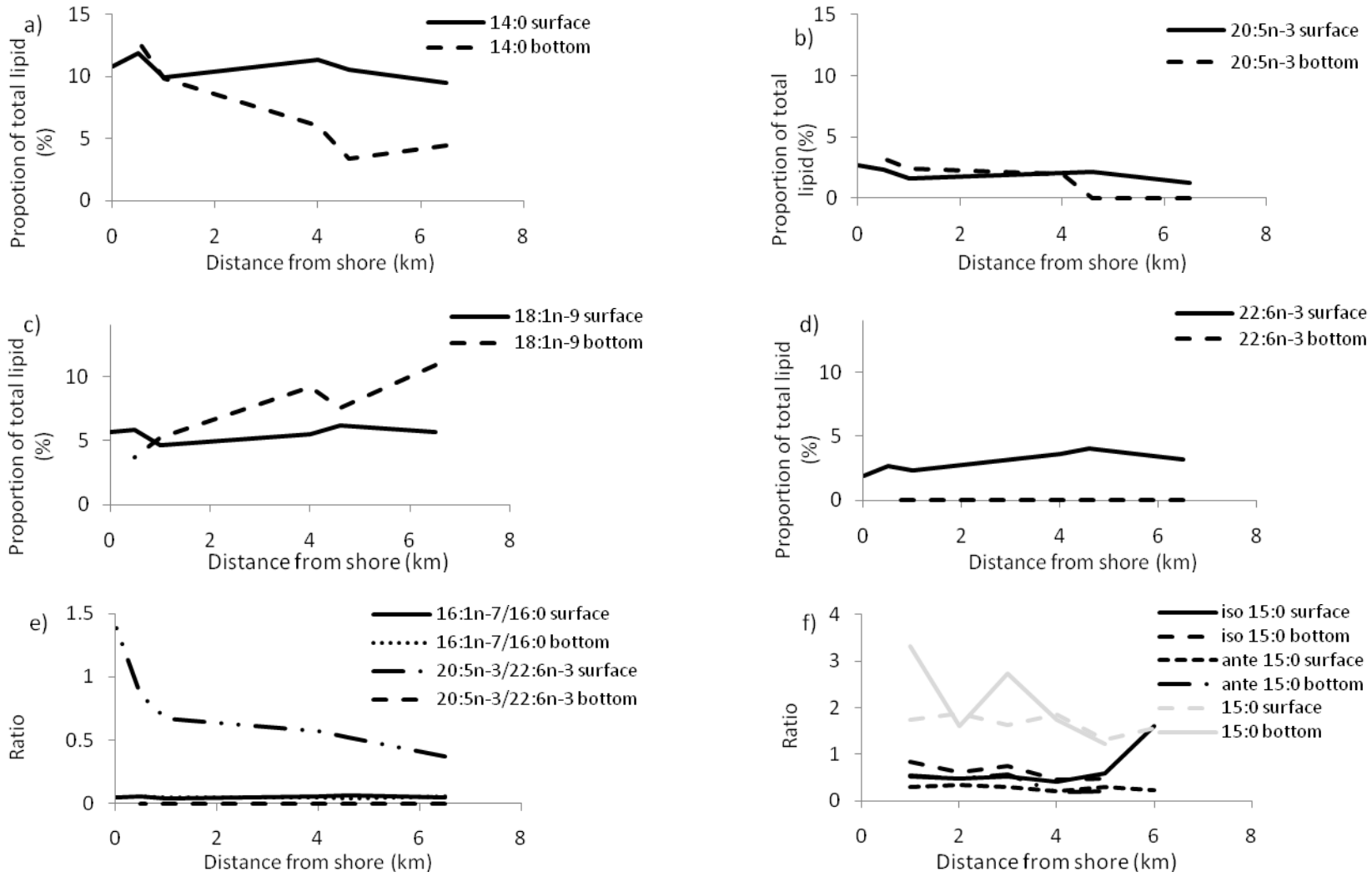


Figure 2.5: Proportions of selected fatty acids in surface and bottom POM samples along an inshore-offshore transect with inferred source and published average proportions in the source in parentheses. a) 14:0 (red seaweeds: 4.45 %, diatoms: 13.51 %), b) 20:5n-3 (dinoflagellates: 10.39 %, brown seaweeds: 11.73 %, diatoms: 15.19 %, red seaweeds: 30.34 %), c) 18:1n-9 (vascular plants: 4.5 %, red seaweeds: 7.37 %, dinoflagellates: 13.17 %, brown seaweeds: 14.15 %), d) 22:6n-3 (dinoflagellates: 13.14 %), e) fatty acid ratios (20:5n-3/22:6n-3: increases with increasing diatom/seaweed contribution; 16:1n-7/16:0: increases with increased diatom presence), f) various C^{15} fatty acids (cyanobacteria). Published proportions from Carpenter *et al*. (1997), Dalsgaard *et al*. (2003) and Kelly & Scheibling (2012).

2.4 Discussion

According to the hypothesis that there would be an inshore-offshore gradient in the type of organic matter, general spatial patterns were observed that included a consistent decline in carbon content together with increasingly depleted isotope signatures in a nearshore to offshore direction. This pattern can probably be ascribed to higher rates of inshore primary production in combination with the input of seaweed detritus into the nearshore POM pool. Carbon content gradients were noted in both the surface and bottom samples. These differences may be explained by higher nutrient concentrations in nearshore water that can support larger concentrations of phytoplankton (Burchall 1968), and a different species assemblage including diatoms. In addition, the greater abundance of seaweeds in shallower waters provides a substantial source of detritus that could increase the amount of organic particulates suspended in the water column and may also account for the relatively enriched inshore POM $\delta^{13}C$ signatures. Seaweeds sampled from the nearshore region had $\delta^{13}C$ signatures ranging between -12 and -20 ‰ except for certain species of red seaweeds that had extremely depleted signatures of less than -30 ‰ (Chapter 3). The most depleted POM $\delta^{13}C$ signatures were found at the farthest sites sampled, while the most enriched samples were consistently from the nearshore/intertidal zones, where seaweed communities colonise shallow wave-swept rocky substrata and continuously shed detrital organic matter into the water. The observed offshore depletion of $\delta^{13}C$ signatures in this study is a common phenomenon in South African waters and was proposed to be linked to a change from a nearshore source of primary production to one of pelagic origin (Hill 2007). The importance of such nearshore, macroalgal production was highlighted in the Aleutian Islands where kelp was conservatively estimated to account for >58 % of the total nearshore primary production (Duggins *et al.* 1989). Together with phytoplankton, seaweeds may contribute to making the

nearshore waters more productive, relative to offshore waters where phytoplankton is assumed to be the sole source of organic primary production,

Lipid analysis indicated further the presence of a nearshore-offshore gradient in the sources of primary production contributing to the POM composition. The proportion of several fatty acids decreased, while others increased, with distance from the shore. The exact sources contributing to inshore and offshore production remain somewhat ambiguous due to a large degree of overlap in fatty acid profiles of primary producers (Kelly & Scheibling 2012). The proportions of fatty acids that are indicative of diatoms (14:0, 20:5n-3) are also indicative of seaweeds (Kelly & Scheibling 2012) and tended to decrease in an offshore direction, especially in bottom water samples. With the reduction in diatoms and/or seaweed detritus in the POM in offshore waters there was an increase in proportions of 18:1n-9 and 22:6n-3 fatty acids, indicating an important dinoflagellate component in the POM of offshore waters, especially at the surface. Furthermore, the declining ratio of 20:5n-3/22:6n-3 clearly indicated a strong gradient from inshore diatom/seaweed organic matter to offshore dinoflagellate dominated organic matter. In a previous study, a ratio of 20:5n-3/22:6n-3 > 1 typically indicated a diatom-dominated phytoplankton community, whereas a ratio < 1 typified a dinoflagellate-dominated community (Budge & Parrish 1998). In addition to the effects of seaweed/diatom input on the POM carbon isotope signatures, isotope ratios of marine phytoplankton, which form an integral part of the POM, may vary with dissolved CO_2 concentrations that are a product of water temperature and pH (Deuser *et al.* 1968). The $\delta^{13}C$ ratios, in the combined-data RDA, were positively correlated with water temperature, and in the surface data negatively correlated with wind speed. In surface waters, $\delta^{13}C$ ratios may therefore partially be a product of increased wind speed and water turbulence that would facilitate the diffusion of CO_2 into the water. Equally, the possibility of temperature effects

on CO_2 concentrations and ultimately $\delta^{13}C$ of the POM may not be discounted. Growth rates in warmer waters, although being high, usually only sustain small phytoplankton populations (Deuser *et al.* 1968). It has previously been found that POM $\delta^{13}C$ was negatively correlated with the concentration of dissolved CO_2 in the water due to the phytoplankton obtaining carbon from bicarbonate ions during CO_2-depleted periods (Deuser *et al.* 1968, Gu *et al.* 2011). Enriched $\delta^{13}C$ values, which were generally observed between February and May, and might therefore be indicative of lower concentrations of dissolved CO_2. Bicarbonate ions in the ocean usually have a $\delta^{13}C$ signature of ~ 0 ‰, whereas dissolved CO_2 has a ratio of between -7 ‰ and -9 ‰ (Deuser *et al.* 1968). Isotopically light phytoplankton, in terms of carbon, should occur in colder waters where the concentration of dissolved CO_2 is high, with the converse true in warmer waters where isotopically heavier bicarbonate and carbonate ions exist in higher proportions and the solubility of CO_2 is reduced (Deuser *et al.* 1968). The observation that $\delta^{13}C$ was more enriched when the water was warmer and the wind calmer, indicates that during these periods the phytoplankton may have been obtaining relatively more of their inorganic carbon from the isotopically heavier bicarbonate and carbonate ions, than during cooler, windier periods when CO_2 availability was likely to be higher.

Temporally, there appeared to be three distinct periods in terms of the POM characteristics. The period from August to November, with depleted $\delta^{13}C$ and enriched $\delta^{15}N$ signatures, the period during November and December when exceptionally high C:N ratios were noted, and the period between February and May, when enriched $\delta^{13}C$ and depleted $\delta^{15}N$ signatures occurred. This suggests that, in terms of the phytoplankton community, different processes may dominate at different times of the year and/or during times of physiological stress due to nutrient availability fluxes. During the first period, enriched $\delta^{15}N$ may have been a product of low nitrogen availability, limited phytoplankton growth and a reliance on isotopically heavier

regenerated nitrogen (Dugdale & Goering 1967). Low growth rates, would also explain the strongly depleted $\delta^{13}C$ values and low overall particulate carbon in the water. During this period, isotopic as well as elemental parameters all pointed towards a strong oceanic influence on the shelf.

During the third period, between February and May, nitrogen isotope ratios were depleted relative to other periods, which points directly to the uptake by primary producers of "new" nitrogen sources. New nitrogen such as upwelled nitrates, or diazotrophically fixed dinitrogen have depleted isotope signatures (~ 2.5 ‰ and 0‰ respectively; Montoya *et al.* (2002)) when compared to regenerated nitrogen. As POC concentrations throughout the water column were extremely low during this period, it is unlikely that a nitrate upwelling event had occurred. Instead, low $\delta^{15}N$ PM signatures were likely a result of increased nitrogen-fixation. This interpretation was supported by the presence of fatty acids occurring in proportions indicative of cyanobacteria. These include iso 15:0 (0.41 – 1.61 %), ante 15:0 (0.18 – 0.54 %) and 15:0 (1.22 – 3.32 %) which have previously been identified in similar proportions in a cyanobacterial study (Carpenter *et al.* 1997). Gu *et al.* (2006) and Montoya *et al.* (2002) furthermore, noted that during a period of high cyanobacterial prevalence, $\delta^{13}C$ signatures were enriched, similar to those found in this study. All lines of evidence, therefore, suggest that the POM during the period between February and May contained an increased abundance of nitrogen-fixing cyanobacteria. The importance of nitrogen fixation in adding to the total nitrogen pool in surface waters has previously been under-emphasised (Capone 2001). In this study nitrogen fixation appeared to be a common occurrence for a substantial period of time. Nonetheless, the new production of POM through nitrogen-fixation (as indicated by C:N close to the Redfield ratio) did not add a substantial amount of organic matter to the POM. Throughout the February to May period, the POC content of the water

was extremely low, when compared to other periods. This indicated that while diazotrophs may have been of increased importance, their actual contribution to the annual carbon budget was limited.

During the second, intermediate period (November and December), both surface and bottom POM samples exhibited extremely high C:N ratios. This probably represented a period of slow growth rate under nutrient limiting conditions (Laws *et al.* 2001). In a laboratory study, the C:N ratio of the diatom *Thalassiosira weissflogii* was shown to be dependent on the growth rate of the culture (Laws *et al.* 2001). At the maximum growth rate the C:N ratio of this species was close to the Redfield ratio of 5.7. When *T. weissflogii* was nutrient-limited the C:N ratio was negatively correlated with growth rate, with the converse occurring under light-limited conditions. Despite the reportedly strong north-easterly winds that blew during this study period, the water column was highly stratified and the surface water temperatures were beginning to peak. Stratification would usually inhibit the entrainment of nutrients into the surface waters and limit growth. High C:N ratios during this period indicate that the proportion of phytoplankton protein was low, while the proportion of carbohydrates was conversely high (Laws *et al.* 2001). Along with reduced growth rates high C:N ratios also often indicate a larger proportion of dead and decaying organic matter in the water column. The correlation between DO concentration and C:N ratios shown in the combined ordination support the theory that there was a high detrital load in the water during the period of high C:N ratios, as the degradation of organic matter is an oxygen demanding process (Tremblay & Gagné 2009). Lara *et al*. (2010) reported elevated C:N ratios, from the southwestern Atlantic. The upper limit of their ratios, however, was much lower at only 8.8, while in the present study a maximum of ~ 15 was noted. Higher C:N ratios were attributed to an increased proportion of detritus or recycled components in the POM (Lara *et al.* 2010), which

supports the observation in the present study. Overall the intermediary "summer" period was characterised by low primary production and an overall high proportion of detrital matter in the water column. While production may have been low, it was during this period that more organic matter was introduced into the water column (especially close to shore) than during any other time (with the exception of August). This may indicate the importance of an inshore organic matter subsidy to the shelf ecosystem in the form of detrital matter.

Vertical trends were also noticeable in this study with more depleted $\delta^{13}C$ values observed in bottom samples and with $\delta^{15}N$ being more variable in bottom samples than surface samples. The depleted carbon isotope signatures may indicate a reliance on a more depleted carbon source. Respired CO_2, produced by benthic organisms, is a likely source of such ^{13}C-depleted inorganic carbon (Raven *et al.* 1995), as opposed to bicarbonates that have a more enriched carbon signature (Deuser *et al.* 1968). The greater observed variability among the nitrogen signatures, especially within months, indicates that the near-benthos POM is subject to a greater variety of influences than the surface POM. A more varied inorganic nitrogen pool available to producers probably exists near the benthos, with a variable input of metabolic nitrogenous waste from benthic consumers likely an important component of this pool, as well as the re-suspension of biogenic sediments and possibly benthic nitrogen-fixation. In a review of oceanic nitrogen fixation by Capone & Carpenter (1982), benthic diazotrophs were estimated to contribute treble the mass of nitrates compared with pelagic diazotrophs. The majority of this benthic fixation was found to occur in coastal waters shallower than 200 m (Capone & Carpenter 1982). A further source of spatial variation in bottom $\delta^{15}N$, is of course light variability at the different bottom depths and the resulting limitations to photosynthesis. The surface POM $\delta^{15}N$ exhibited greater homogeneity within months, likely because all samples were attained from the same depth (1 m).

The mass of carbon in the POM was consistently lower in bottom samples than surface samples, especially at a distance from the shore, indicating there to be a greater amount of surface phytoplankton production than that occurring nearer the bottom. This may be attributed to light attenuation at depth and reduced photosynthesis. Interestingly, between August and February, bottom samples from 40 m depth downwards, had lower carbon content than those taken from shallower sites. This supports the idea of a lower hyper-benthic primary production in depths greater than 30 m. Particulate organic carbon has previously been shown to decrease with depth, linked with surface-water phytoplankton production (Bates *et al.* 2005).

To conclude, in addition to the previously observed offshore depletion in carbon isotope signatures, other spatio-temporal patterns were clearly noticeable. These are suggested to reflect the availability of nutrients, and inorganic carbon, that in turn influence the isotope signatures and population densities of producers, as well as the composition of the POM itself. Evidence from both fatty acid and isotope analyses suggests that inshore particulate organic matter consisted primarily of diatoms and/or seaweed detritus. Moving further offshore, at approximately 2km distance, there was a transition from the inshore profile to one reflecting a largely dinoflagellate dominated POM, with a concurrent reduction in the diatom/seaweed detritus presence. These patterns in the POM are likely to have important consequences on higher trophic level organisms, in terms of the constituents of the organic matter available to consumers at different depth zones and distances from shore. The inshore region is likely to be able to support a larger community of organisms relying on POM, for example filter-feeders, with this zone generally having relatively more organic matter in the water. Also of note, however, was that POM origin and abundance varied greatly in time. Three distinct periods of variable nutrient availability, primary production rates and organic

matter transport were identified during the 10 month study period. Future research involving POM needs to take cognisance of the variability exhibited in this study, both spatially and temporally. Point samples would in them selves not be particularly useful. Sampling in the future should be designed to account for both spatial and temporal variability in POM fluxes such that additional insight into nutrient and organic matter transport as well as the relative contribution of allochthonous and detrital inputs from seaweeds can be understood. This study indicates that while phytoplankton and cyanobacterial production levels at Sodwana Bay are temporally variable their total contribution to the organic carbon pool is limited. Instead during periods of high POM content in the water column, most organic matter seems to be derived from the detrital pool.

Chapter 3

Evidence for high trophic connectivity between different marine habitats along a nearshore-offshore gradient and within submarine canyons off Sodwana Bay, South Africa

3.1 Introduction

A principal goal within ecology is to understand trophic interactions (Rounick & Winterbourn 1986). From a food web perspective, investigations typically focus on the flow of energy through and up trophic levels such that these transfer patterns among communities can be understood (Menge *et al.* 1986, Carpenter *et al.* 1987, Power 1992, Otto *et al.* 2008). Food webs are typically structured through bottom-up (such as nutrients and productivity) and top-down (abundance and distribution of consumers) processes (Power 1992, Menge *et al.* 1997, Shears & Babcock 2002, Frederiksen *et al.* 2006, Baum & Worm 2009). Therefore, within the context of understanding and ultimately managing an ecosystem, information on how the organisms within the community are reliant on other species, and in particular different sources of primary production, is critical. Furthermore, as marine habitats are continuous and interconnected, additional knowledge pertaining to the flow of energy between habitats is required.

Within the marine environment there are a suite of possible sources of primary production available to primary consumers that in turn support higher trophic levels. Autochthonous sources of production include microalgae, both planktonic and benthic, and macrophytes such as seaweeds and seagrasses (Thayer *et al.* 1978, Bustamante & Branch 1996). Allochthonous sources include an import of the latter production into nearby habitats, in the form of detritus (Suchanek *et al.* 1985, Bustamante & Branch 1996), as well as terrestrial and alluvial inputs

(Gordon & Goñi 2003). Phytoplankton is incorporated into food webs by filter-feeders that remove it from the water column and by deposit feeders that consume phytoplankton detritus that flocculates out of the water column (Rudnick 1989, Yahel *et al.* 1998). Phytoplankton is a variable source of production, in terms of species composition and abundance, that is sensitive to environmental influences including temperature, nutrient availability, and light (Eppley 1972, Kromkamp *et al.* 1995). Macrophytes are important directly for grazing or indirectly where up to 90 % of production can become available to consumers through detrital foodwebs (Mann 1973, Stephenson *et al.* 1986). A large proportion of macrophyte production is removed from the tissues when forming detritus, and is therefore not directly available to grazers or filter-feeders (Blum & Mills 1991, Haas *et al.* 2010). Seagrass utilisation by consumers has been shown to be limited, with organisms that utilise seagrass beds as a habitat often consuming seaweed, epiphyte and phytoplankton derived production to a greater extent than the seagrasses, except for specialised seagrass grazers (Stephenson *et al.* 1986). Fish larvae have also been found to utilise seagrass detritus via the planktonic food chain (Thresher *et al.* 1992). Seaweeds may be grazed, by herbivorous fishes for example that may actively defend patches against other herbivores (Vine 1974). Large seaweeds such as kelp are frequently found to be important primary producers and dietary sources, supporting diverse communities and functional groups (Dunton & Schell 1987). Associated with coral reefs are algal turfs that can account for 60 - 80 % of the total macrophyte cover on front- and back-reef crests, while crustose coralline algae may dominate the fore-reef (Klump & McKinnon 1989). Turfs have lower C:N ratios compared with foliose macrophytes possibly due to the large amount of structural carbon, or nutrient limitation, in the foliose species (Miller *et al.* 2009).

Sodwana Bay falls within the iSimangaliso Wetland Park, a World Heritage Site, and is of high conservation significance as it contains both Africa's southern-most coral reefs (Ramsay

& Mason 1990) and the most southerly distributed population of coelacanths *Latimeria chalumnae*, discovered in 2000 (Venter *et al.* 2000), that are classified as critically-endangered by the IUCN (Musick 2000). Owing to the coral reefs it is South Africa's most popular SCUBA diving destination and is therefore important recreationally. Sodwana Bay is important ecologically as it contains a number of habitats along a depth gradient, from the intertidal habitat, which includes sandy beaches and rocky shores, to a habitat of coral reef roughly parallel to the shore followed by deep-reef habitat, and submarine canyon heads offshore (Ramsay 1994). Beyond the relatively narrow shelf, ~3 km offshore, most organisms are pelagic.

Submarine canyons are biologically interesting allowing deep water taxa to move closer inshore as the topography facilitates the intrusion of deeper water (Bosley *et al.* 2004). This provides a mechanism for linkages between inshore production and deep-dwelling organisms to be sustained. Canyons may also perform a concentrating role due to topographic influences on hydrodynamics forming aggregations of food sources for consumers (Harrold *et al.* 1998, Bosley *et al.* 2004). For example, detritivorous sea urchins in canyons offshore of California were found to be in better condition than those living on the continental shelf outside of the canyons. This was ascribed to the observed higher food supply in the canyons compared to the adjacent shelf (Harrold *et al.* 1998).

To date, most biological studies conducted in Sodwana Bay have been of a descriptive nature focusing primarily on species identifications, abundance and distributions with the aim of cataloguing the biodiversity of the area (Anderson *et al.* 2005, Heemstra *et al.* 2006, Sink *et al.* 2006, Samaai *et al.* 2010). Unfortunately little is known about its ecology and the functioning of the system as a whole. This chapter aims, for the first time, to identify trophic interactions between the various Sodwana Bay habitats and to estimate the relative

importance of different sources of primary production to consumers within different habitats along a gradient of depth and distance from shore using stable isotope analysis. It was hypothesised that few consumers would utilise seagrass production, with the major sources of energy and nutrients being derived from seaweeds and phytoplankton. Due to differential distribution of primary producers (inshore seaweeds and offshore phytoplankton) it was furthermore hypothesised that a gradient in the utilisation of inshore production would be evident.

3.2 Materials and methods

3.2.1 General sampling

Samples for stable isotope analysis were collected over a protracted period of time between 2002 and 2011 using a variety of methods and various collectors (Table 3.1).

Samples of fishes, invertebrates and seaweeds were collected by fishing, by hand, snorkelling, SCUBA diving, and with a remotely operated vehicle (ROV) fitted with a manipulator arm. Collections were made from the intertidal/shallow subtidal habitat, < 3 m deep, on the sandy beach and rocky shore, on coral reef to a depth of 30 m, on deep reef (~ 60m), and from the canyon heads (see site description, Chapter 1). The majority of benthic invertebrates and seaweed samples were collected during 2010/2011. Fishes, corals and zooplankton were collected by other researchers. Supplementary samples included green, red and brown seaweeds, seagrass, POM, filter-feeding invertebrates such as mussels, oysters, sponges and sea pens, gastropods, three species of scleractinian coral, and mobile crustaceans such as shrimp and crabs. Data obtained from earlier fieldtrips pertained largely to fish, but also included zooplankton, mussels and sponges. Sample identification, for the latest sample collections, was done using various field guides (King & Fraser 2001, Richmond 2002,

Branch *et al.* 2005, De Clerck *et al.* 2005). Samples from previous collections were identified by the contributors.

Table 3.1: Summary of specimens sampled by various collectors. All specimens were sampled off Sodwana Bay, South Africa between 2002 and 2011.

Sample	Collector	Date	Location
Sand steenbras	Scott *	2006	inshore
Sillago	Scott	2007	inshore
Stone bream	Scott	2007	inshore
Largespot pompano	Scott	2007	inshore
Kosi rockskipper	Scott	2005	inshore
Halfbeak	Kaehler/ McQuaid +	2005	inshore
Blacktail	Scott, Sink *et al* ₸	2007	inshore
Mole crab	Parkinson *et al*.	2010/2011	inshore
Seagrass	Parkinson *et al.*	2010/2011	inshore
Gastropods	Parkinson *et al*.	2010/2011	inshore, coral reef
Crabs	Parkinson *et al*.	2010/2011	inshore, coral reef
Seaweed (red, green, brown)	Parkinson *et al*.	2010/2011	inshore, coral reef, deep reef
Bivalves	Parkinson *et al*.	2010/2011	inshore, coral reef, deep reef, canyon
Bluefin kingfish	Scott	2007	coral reef
Bridle trigger fish	Scott	2007	coral reef
Speckled snapper	Sink *et al*	2002	coral reef
Coral (scleractinians)	Floros *et al* ¥	2011	coral reef
Sponges	Parkinson *et al*.	2010/2011	coral reef
Hermit crabs	Parkinson *et al*.	2010/2011	coral reef
Ascidians	Parkinson *et al*.	2010/2011	coral reef
Cleaner shrimps	Parkinson *et al*.	2010/2011	coral reef
Nudibranchs	Parkinson *et al*.	2010/2011	coral reef, deep reef
Santer	Scott	2007	deep reef
Soldierbream	Scott	2007	deep reef
Smalltooth emperor	Scott	2005	deep reef
Spotcheek emperor	Scott	2005	deep reef
Rosy jobfish	Scott	2005	deep reef
Slinger	Scott	2007	deep reef
Sky emperor	Scott	2005	deep reef
Red tjor tjor	Scott	2005	deep reef
Epaulette soldier	Scott	2005	deep reef
Bluebanded Snapper	Scott	2005	deep reef
Sponges	Parkinson *et al*.	2010/2011	deep reef
Brittle stars	Parkinson *et al*.	2010/2011	deep reef, canyon
Contour rockcod	Sink *et al*	2004	canyon
Tomato rockcod	Sink *et al*	2004	canyon
Blueskin	Sink *et al*	2004	canyon

Coelacanth	Sink *et al*	2004	canyon
Black marlin	Scott	2007	pelagic
Striped marlin	Scott, Kaehler/McQuaid	2007	pelagic
King mackerel	Scott, Kaehler/McQuaid	2007	pelagic
Yellowfin tuna	Scott, Sink *et al*, Kaehler/McQuaid	2007	pelagic
Eastern little tuna	Scott, Kaehler/McQuaid	2005	pelagic
Green jobfish	Scott, Kaehler/McQuaid	2005	pelagic
Tropical yellowtail	Scott	2005	pelagic
Queenfish	Scott, Kaehler/McQuaid	2005	pelagic
Rosy jobfish	Scott	2005	pelagic
Sailfish	Kaehler/McQuaid	2005	pelagic
Bonito	Scott	2007	pelagic
Remora	Scott	2007	pelagic
Dorado	Scott, Sink	2007	pelagic
Zooplankton	Sink *et al*	2002	pelagic
POM	Parkinson *et al*.	2010/2011	

* Lucy Scott; + Sven Kaehler/Christopher McQuaid; ₸ Kerry Sink and colleagues; ¥ Camilla Floros and colleagues.

3.2.2 Sample preparation and analysis

Samples were frozen immediately upon collection. After defrosting, samples were rinsed with distilled water, photographed for identification and a tissue sample was taken for analysis. A database recording sample and photograph number together with the location and date of collection was maintained. Where sufficient quantities were available, muscle tissue was dissected from animal specimens. In cases where invertebrates were too small to allow for muscle extraction, the whole or majority of the organism, excluding hard structures where possible, were used. Seaweed samples were cleaned of epiphytes by scraping the surfaces with a razor blade and a section of the frond removed. Tissue was placed into a labelled micro-centrifuge tube and dried in an oven at ~ 50 °C for 24 to 48 hours. Dried samples were ground using a pestle and mortar and stored in micro-centrifuge tubes placed into a sealed container with a silica gel desiccant until further preparation for stable isotope analysis.

Samples were analysed without acidification (for the removal of carbonates) to eliminate the associated effects on the nitrogen signatures (Mateo *et al.* 2008). Fish carbon isotope ratios were adjusted for lipid content after analysis according to Post *et al*. (2007), whereby $\delta^{13}C$ = -3.32 + 0.99C:N. Where carbonates were present, if the sample reacted to HCl, acidification was conducted for carbon signature determination. In these samples, the tissue was acidified in 15 ml centrifuge tubes by adding 1 ml of a 1 N HCl solution to the sample. If there was a large proportion of carbonate an additional 1 ml of the HCl solution was added. Once CO_2 liberation ceased, ~ 9 ml distilled water was added to each centrifuge tube and they were agitated. Samples were then centrifuged for one to two minutes at approximately 4 000 RPM and the supernatant removed using a pipette being cautious not to remove any of the tissue from the bottom of the tube. Samples were rinsed by adding another ~ 10 ml of distilled water and repeating the centrifuge and draining process. Samples were then dried, ground, weighed into tin capsules and analysed for their post-decarbonation stable isotope ratios. Nitrogen signatures of the unacidified samples were used.

Due to the endangered status of coelacanths, muscle tissue sampling was not a viable option. Scales were collected instead. Scale isotope ratios were adjusted to be comparable with muscle tissue samples of the other fishes that were collected during this study. This was conducted by calculating the average difference between scale and muscle tissue samples from three other fish species – blueskin *Polysteganus coeruleopunctatus*, contour rockcod *Epinephalus poecilinatus* and tomato rockcod *Cephalopholis sonerati*. Scales had, on average, carbon signatures that were 3.06 ‰ more enriched, and nitrogen signatures that were 2.17 ‰ more depleted, than muscle tissue.

Coral tissue was removed from the skeletal structure by spraying the corals with distilled water using an airbrush connected to the first-stage of a SCUBA cylinder into a plastic sandwich bag. The coral tissue from each replicate, excluding any skeletal fragments, was filtered from the water onto a pre-combusted GFF filter (500 °C, 6 hours), dried, and a subsample used for stable isotope analysis.

Tissue samples were then weighed into 8 × 5 mm tin capsules, and combusted and analysed using a Europa Scientific ANCA-SL Elemental Analyser and 20-20 IRMS at the IsoEnvironmental Lab, Grahamstown. Stable isotope ratios were expressed in standard δ notation in parts per thousand as $\delta X(‰) = \left(\frac{R_{sample}}{R_{standard}} - 1\right) \times 1000$ where *X* is the heavy isotope of carbon or nitrogen and *R* is the ratio of heavy:light isotopes (Peterson & Fry 1987). In-house standards of ammonium sulphate, beet sugar and casein were used and calibrated against Vienna PeeDee Belemnite and atmospheric nitrogen, for C and N respectively. Overall precision of stable isotope analyses were 0.12 ‰ for N and 0.10 ‰ for C.

To determine whether the median carbon signatures of the seaweed detritus and offshore POM differed from one another, a non-parametric Mann-Whitney U test was employed, as the assumptions of parametric analyses (such as being normally distributed and homoscedasticity) were violated. Hypothesis tests were conducted at $\alpha = 0.05$. Average offshore POM signatures (from Chapter 2) were used as a proxy for the phytoplankton signature, as it was assumed that little, or no, seaweed or other source of carbon would be present in any offshore sample.

3.2.3 Trophic groups

Biplots of carbon and nitrogen stable isotope signatures were constructed for each habitat defined as either inshore (intertidal to ~ 3m depth), coral reef (10 – 30 m depth), deep reef (~ 60 m depth), submarine canyon (~ 100 m depth) or pelagic. To investigate trophic patterns within each habitat, consumers with similar carbon and nitrogen signatures were grouped. Grouping was somewhat subjective but was necessary to reduce the complexity within the data and to facilitate investigation into broad-scale patterns among consumers. To aid visualisation of the data, a box encompassing the range of average carbon and nitrogen signatures for each group was plotted.

3.2.4 Mixing models

To estimate the relative importance of the inshore and offshore sources of primary production to the consumer groups, a mixing model, *Stable Isotope Analysis in R* (SIAR), was employed (Parnell *et al.* 2010). SIAR uses Bayesian inference and model fitting via Markov chain Monte Carlo simulations, using a Dirichlet distribution, to estimate a credibility interval (the Bayesian analogue of a confidence interval) for the proportion that each source contributes to the consumers' diets. The method accounts for both the variability in consumer and source signatures together with the assumed variability in fractionation between trophic groups.

3.3 Results

3.3.1 Primary producers

It was decided to use the average of the monthly signatures, of surface and bottom samples from the 4.6 km station to represent the offshore production that was devoid of macrophyte detritus. This was chosen because it was the furthest offshore station and was sampled from

August to December 2010, and February to May 2011 (see Chapter 2). The offshore POM signatures were used to represent the offshore production component in the mixing models.

It was found in Chapter 2 that distinct isotope gradients in the POM were present within the data with respect to distance from shore with inshore samples typically having enriched carbon signatures compared with offshore samples. While the contribution of seaweeds to this pattern is likely, as they have enriched carbon signatures relative to phytoplankton, the presence of a distinct inshore phytoplankton community compared with the offshore community was also possible given the patterns in the lipid profiles of the POM. The seaweed detritus carbon signatures were similarly enriched to inshore POM signatures. The enriched signatures of the seaweeds and inshore POM are therefore referred to, collectively, as 'inshore production'. In the mixing model, however, the average net-particle signature was used as a proxy for the inshore production as it was assumed that this would best reflect the isotope signature of particles available to consumers that do not directly graze on seaweeds.

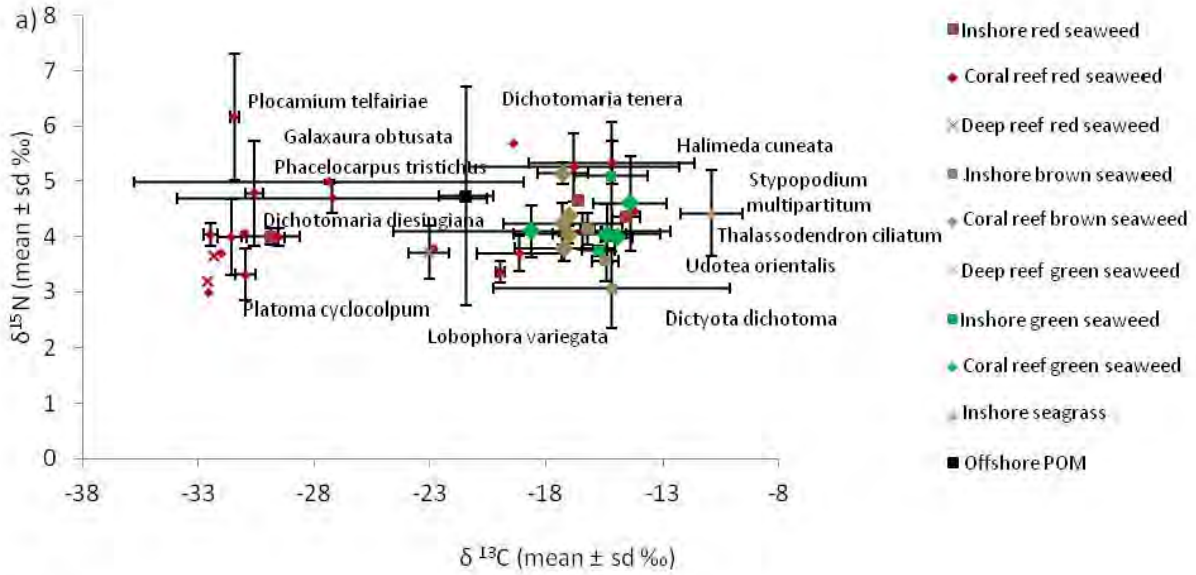
A high degree of variation among the benthic primary producers' carbon isotope signatures with no spatial patterns was evident (Figure 3.1). Some of these signatures were exceptional, such as certain red seaweeds (Table 3.2), with extremely negative carbon signatures (carbon: average ± sd; red seaweed = -29.80 ± 3.46 ‰; nitrogen: 4.17 ± 0.87 ‰), and seagrasses that possessed relatively enriched carbon signatures (carbon: -10.90 ± 1.35 ‰; nitrogen: 4.42 ± 0.78 ‰).

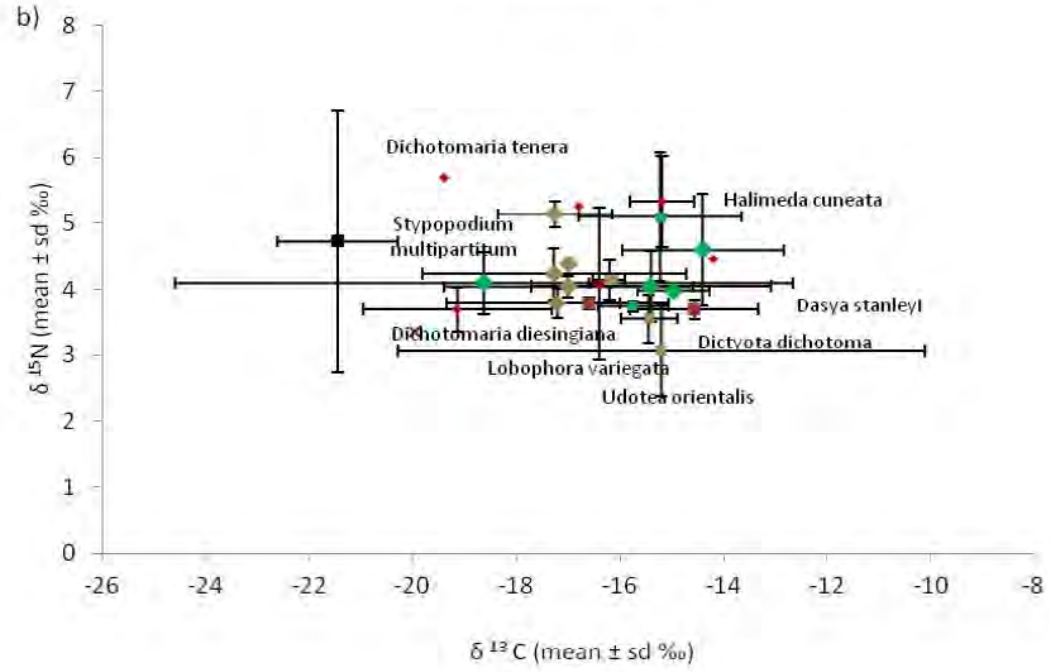
No consumers that were sampled were found to have signatures that reflected an important contribution from the depleted red seaweeds. Similarly, the highly enriched seagrasses did not have any apparent consumers, as the consumers' signatures were far more depleted. Since the depleted red seaweeds and enriched seagrasses were not found to be assimilated into

consumers these two producer sources were removed from the mixing model. As the majority of consumers, except a few grazers (e.g. herbivorous fishes), utilise seaweed production indirectly in the form of detritus, the average seaweed signatures in the detrital chain were determined from net-particles. These were assumed to represent the average composition and signatures of seaweed production available to non-grazers *in situ*. The average of the monthly offshore (4.6 km station; Chapter 2) POM carbon signatures was -21.45 ± 1.17 ‰, and the average nitrogen signature was 5.18 ± 1.18 ‰ (Figure 3.1). The carbon signatures of the seaweed detritus (-15.67 ± 0.51) and offshore POM (-21.45 ± 1.17) were significantly different ($W = 104.00$, $p < 0.01$), facilitating clear interpretation of their assimilation into the foodweb as distinct sources of carbon.

Table 3.2: Red seaweeds sampled off Sodwana Bay, South Africa with depleted carbon signatures.

Species	Location	mean $\delta^{13}C$	mean $\delta^{15}N$
Platoma cyclocolpum	coral reef	-31.56	3.99
Plocamium telfairiae	coral reef	-31.44	6.17
Delesseriaceae cf. *Augophyllum*	coral reef	-32.56	2.99
Balliella crouanioides	coral reef	-31.04	4.06
Unidentified 1	coral reef	-22.86	3.79
Dasya stanleyi	inshore	-29.87	4.00
Meristotheca papulosa	coral reef	-32.03	3.69
Unidentified 2	coral reef	-30.58	4.78
Unidentified 3	coral reef	-25.59	4.30
Nienburgia serrata	coral reef	-27.34	4.99
Phacelocarpus tristichus	coral reef	-32.48	4.04
Laurencia sp.	coral reef	-30.98	3.31
Halymenia durvillei	coral reef	-27.22	4.69
Predaea feldmannii	coral reef	-29.55	3.98
Unidentified 4	deep reef	-23.03	3.71


a)
Plocamium telfairiae
Galaxaura obtusata
Phacelocarpus tristichus
Dichotomaria diesingiana
Platoma cyclocolpum
Dichotomaria tenera
Halimeda cuneata
Stypopodium multipartitum
Thalassodendron ciliatum
Udotea orientalis
Dictyota dichotoma
Lobophora variegata
Inshore red seaweed
Coral reef red seaweed
Deep reef red seaweed
Inshore brown seaweed
Coral reef brown seaweed
Deep reef green seaweed
Inshore green seaweed
Coral reef green seaweed
Inshore seagrass
Offshore POM
δ15N (mean ± sd ‰)
δ 13C (mean ± sd ‰)


b)
Dichotomaria tenera
Halimeda cuneata
Stypopodium multipartitum
Dasya stanleyi
Dichotomaria diesingiana
Dictyota dichotoma
Lobophora variegata
Udotea orientalis
δ 15N (mean ± sd ‰)
δ 13 C (mean ± sd ‰)

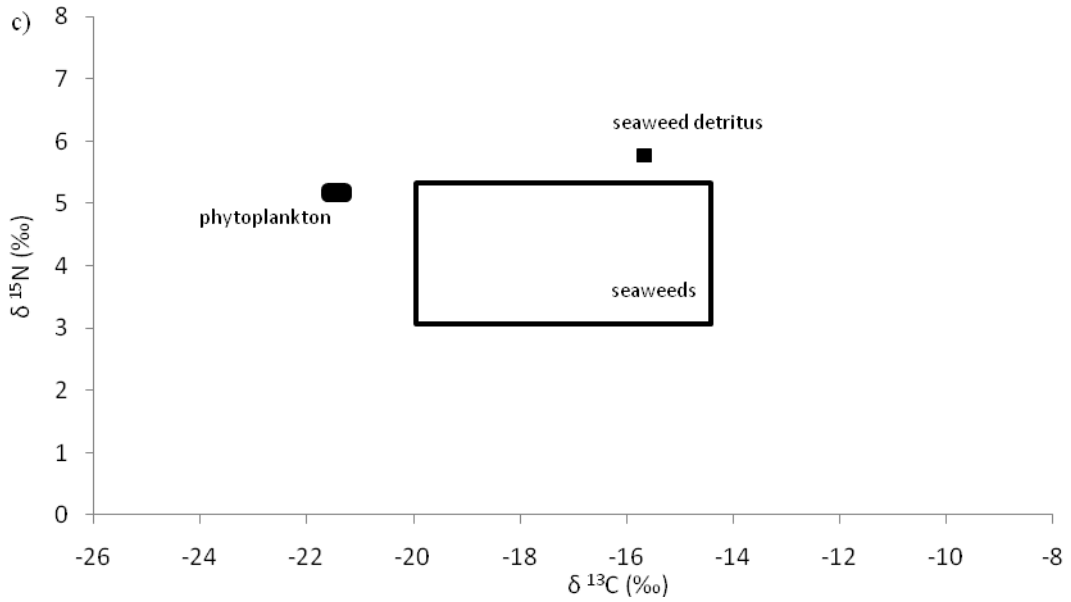


Figure 3.1: Stable isotope signatures of a) all primary producers; b) primary producers, excluding depleted red seaweeds ($\delta^{13}C < -22$ ‰) and seagrass, and c) simplified primary producers with boxes encompassing the range of all average signatures of all species sampled. All samples were collected off Sodwana Bay, South Africa.

3.3.2 Inshore consumers

Carbon signatures ranged from -18.5 ‰ in ghost crabs *Ocypode ryderi*, to -14.8 ‰, for the cone shell *Conus ebraeus* (Figure 3.2 a). The nitrogen signatures ranged between 7 ‰ in black mussels *Perna perna*, and 12 ‰ for both sand steenbras *Lithognathus mormyrus* and blacktail *Diplodus capensis*. Two consumer trophic levels and four consumer groups were evident (Figure 3.2 b, Table 3.3). Group A included two fish species, convict surgeon *Acanthurus triostegus*, kosi rockskipper *Pereulixia kosiensis*, and a cone shell *Conus ebraeus*. Group A consumers had the most enriched carbon signatures, with the mixing model estimating an 80 % contribution of inshore production to this group. Groups B and C, which occupied the highest trophic level, included fishes with nitrogen signatures in the upper range observed (Figure 3.2 b). Most of these fishes are known to prey on marine invertebrates

(Whitfield 2001). Inshore production was estimated to comprise up to 74 and 66 % of the primary production utilised by consumers in groups B and C, respectively (Table 3.3). Group D consisted of a generalist consumer, the pink ghost crab *Ocypode ryderi*, and the mole crab *Emerita austroafricana*. This group of consumers had the most depleted carbon signatures and were estimated to incorporate 51 % of the inshore production into their diets (Table 3.3).

3.3.3 Coral reef consumers

By and large, filter feeders tended to have the most depleted carbon signatures of all consumers on the coral reef while gastropods had predominantly enriched signatures (Figure 3.3 a). The carbon signatures ranged between ~ -22 and -14 ‰. The carbon signatures of the sponges and bivalves, both filter feeders, were the most depleted at ~ -18 ‰, while fishes and cone shells had the highest nitrogen signatures of ~ 13 ‰. Two of the coral species, *Acropora austera* and *Hydnophora* sp., possessed similar signatures to the seaweeds. The third coral species *Platygyra daedalea*, however, was more enriched in ^{13}C. Coral reef consumers spanned two trophic levels (Figure 3.3 b, Table 3.3). The upper trophic levels were occupied by fishes, including bluefin kingfish *Caranx melampygus*, speckled snapper *Lutjanus rivulatus*, several species of predatory cone shells and the cleaner shrimp, *Stenopus hispidus* and *Lysmata amboinensis*. Intermediate consumers included various gastropods. Primary consumers included filter feeding sponges and bivalves, crabs and hermit crabs that all probably feed on detritus. Six groups were distinguished among the coral reef consumers (Figure 3.3 b). Groups A, B and E occurred at an intermediate trophic level and included several gastropods. Group A, with the most enriched carbon signatures, were estimated to incorporate 89 % inshore derived production into their diet (Table 3.3) while groups B and E were more reliant on a pelagic diet. Group D consisted of fishes, a cleaner shrimp and certain species of cone shell (Figure 3.3 b). Group F contained hermit crabs, a cleaner shrimp and

several gastropods, while group C consisted of hermit crabs. Group F had the widest range in carbon signatures, but on average the consumers' diets comprised 67 % offshore phytoplankton-derived primary production. At 82 %, inshore production dominated the contribution to group D, occupying the top trophic level (Table 3.3). There was relatively large overlap between groups E and F and the seaweeds, with group E also overlapping the phytoplankton (Figure 3.3 b). The mixing model estimated that group F that included filter feeders incorporated 67 % offshore phytoplankton-derived primary production in their diet, while the gastropods, in group E, incorporated 60 % inshore production in their diets (Table 3.3).

3.3.4 Deep reef consumers

There was a relatively limited variability in the consumers' signatures from the deep reef and probably partly reflected the limited number of samples obtained from this habitat (Figure 3.4 a). Those organisms sampled, however, showed a clustering of signatures. Sponges possessed the most depleted carbon signatures at ~ -20 ‰, with a cone shell exhibiting the most enriched signature at ~ -16.1 ‰. Three consumer groups, covering two trophic levels, were distinguished (Figure 3.4 b, Table 3.3). Group A consisted of fishes in the top trophic level, including santer *Cheimerius nufar*, slinger *Chrysoblephus puniceus*, smalltooth emperor *Lethrinus microdon*, and soldierbream *Argyrops filamentosus*. These fishes relied predominantly on inshore production, which contributed 70 % to their diets (Table 3.3). Group B included several reef fish species, including red tjor-tjor *Pagellus natalensis*, epaulette soldierfish *Myripristis kuntee,* bluebanded snapper *Lutjanus kasmira*, sky emperor *Lethrinus nebulosus* and rosy jobfish *Pristipomoides filamentosus,* and a cone shell (Figure 3. b). These consumers had a wide range of carbon signatures, ranging from -18.2 to -16.1 ‰, utilising predominantly inshore derived carbon, which contributed 65 % to their diets (Table

3.3). The fishes in groups A and B have been noted to consume various invertebrates, with several of them also including fishes in their diets (van der Elst 1993). Those in group A, with a higher trophic position, were likely including a higher proportion of predatory organisms in their diet possibly including fish. Group C included filter-feeding sea pens and sponges (Figure 3.4 b). These consumers predominantly utilised offshore phytoplankton-derived carbon, with this source contributing 70 % of their dietary carbon. Inshore production was still an important source, with a 30 % contribution, to these consumers (Table 3.3). A tubeworm that was collected possessed signatures overlapping with the seaweed signatures, indicating the direct importance of inshore production in its diet.

3.3.5 Canyon consumers

Few samples were collected from the canyon due to the logistical difficulty in accessing habitats within this depth range. Canyon consumers possessed carbon signatures ranging from -19.62 ‰ for a bivalve, to -14.71 ‰ for the coelacanth *Latimeria chalumnae*, and nitrogen signatures between 7.20 and 15.49 ‰ for a bivalve and contour rockcod *E. poecilinatus*, respectively (Figure 3.5 a). Two distinct groups, from two trophic levels, were noted. Group A consisted of several reef fish species, including blueskin *P. coeruleopunctatus*, tomato rockcod *Cephalopholis sonerati*, contour rockcod, *E. poecilinatus*, and the coelacanth *L. chalumnae* (Figure 3.5 b). Group A was estimated to utilise primarily inshore-derived carbon that contributed 77 % of their diet (Table 3.3). The consumer occupying the highest trophic level was the contour rockcod (Figure 3.5 a). Based on samples from two specimens, the coelacanth's trophic position was similar to other fish species - tomato rockcod and blueskin. Group B consisted of filter-feeding invertebrates, bivalves and a seapen (Figure 3.5 b). Offshore phytoplankton contributed 61 % to the diets of these

consumers. Inshore production was still important, estimated at 39 %, even at this depth and distance from the shore (Table 3.3).

3.3.6 Pelagic consumers

The range of pelagic consumers' carbon signatures was ~ -23 ‰, for the copepod *Gaetanus* sp., to -16 ‰, for striped marlin *Tetrapturus audax*, and the range of nitrogen signatures was between 5.33 and 13.88 ‰ (Figure 3.6 a). The pelagic consumers sampled separated into three groups across two trophic levels (Figure 3.6 b, Table 3.3). Group A comprised the pelagic fishes, occupying the highest trophic level. Among these fishes were yellowfin tuna *Thunnus albacares*, billfishes, such as black marlin *Makaira indica*, striped marlin *Tetrapturus audax,* and sailfish, *Istiophorus platypterus*, and several other pelagic gamefish (Figure 3.6 b). Consumers within this group possessed an enriched carbon source relative to the other groups. Inshore production was the primary source of carbon for group A consumers, estimated at 63 % (Table 3.3). Groups B and C consisted of zooplankton (Figure 3.6 b). Offshore phytoplankton was most important to these consumers, contributing 70 and 73 % to their diets, respectively (Table 3.3).

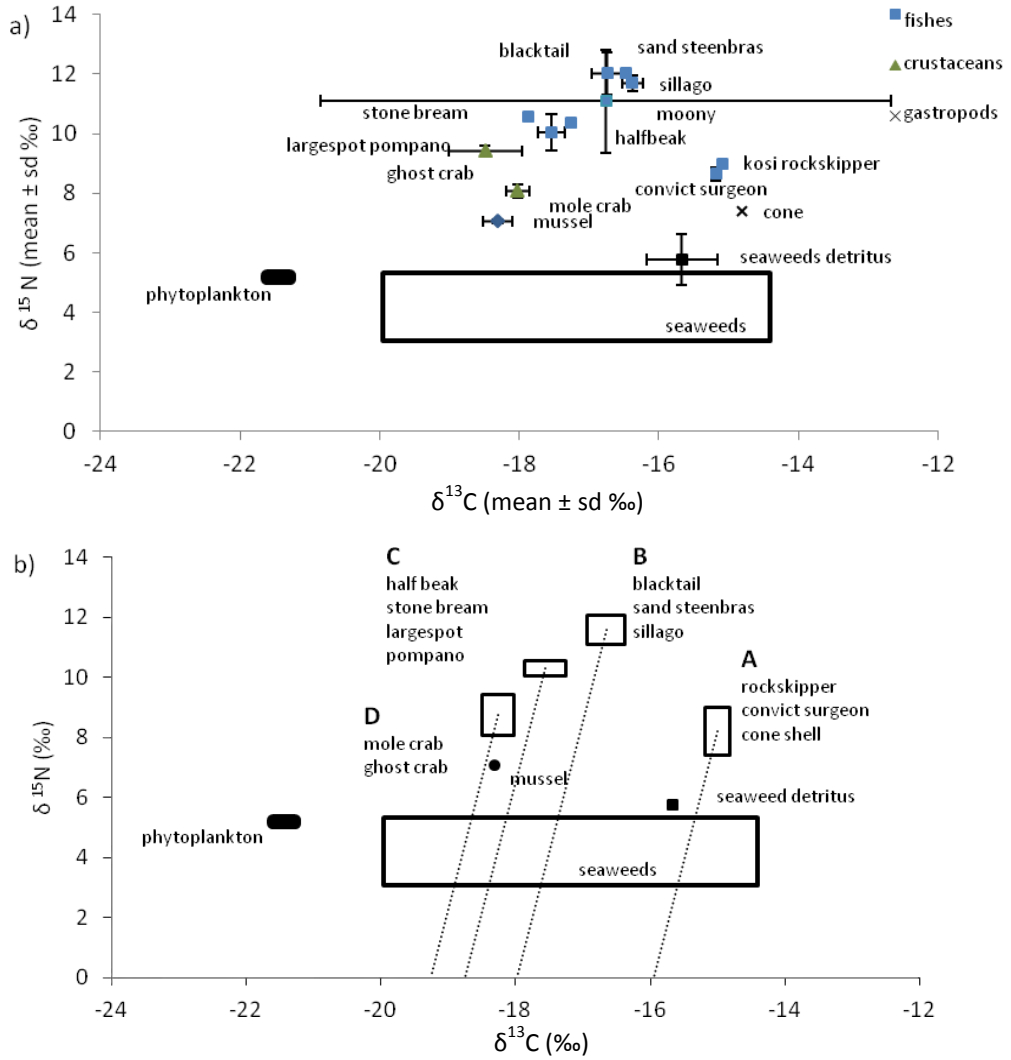
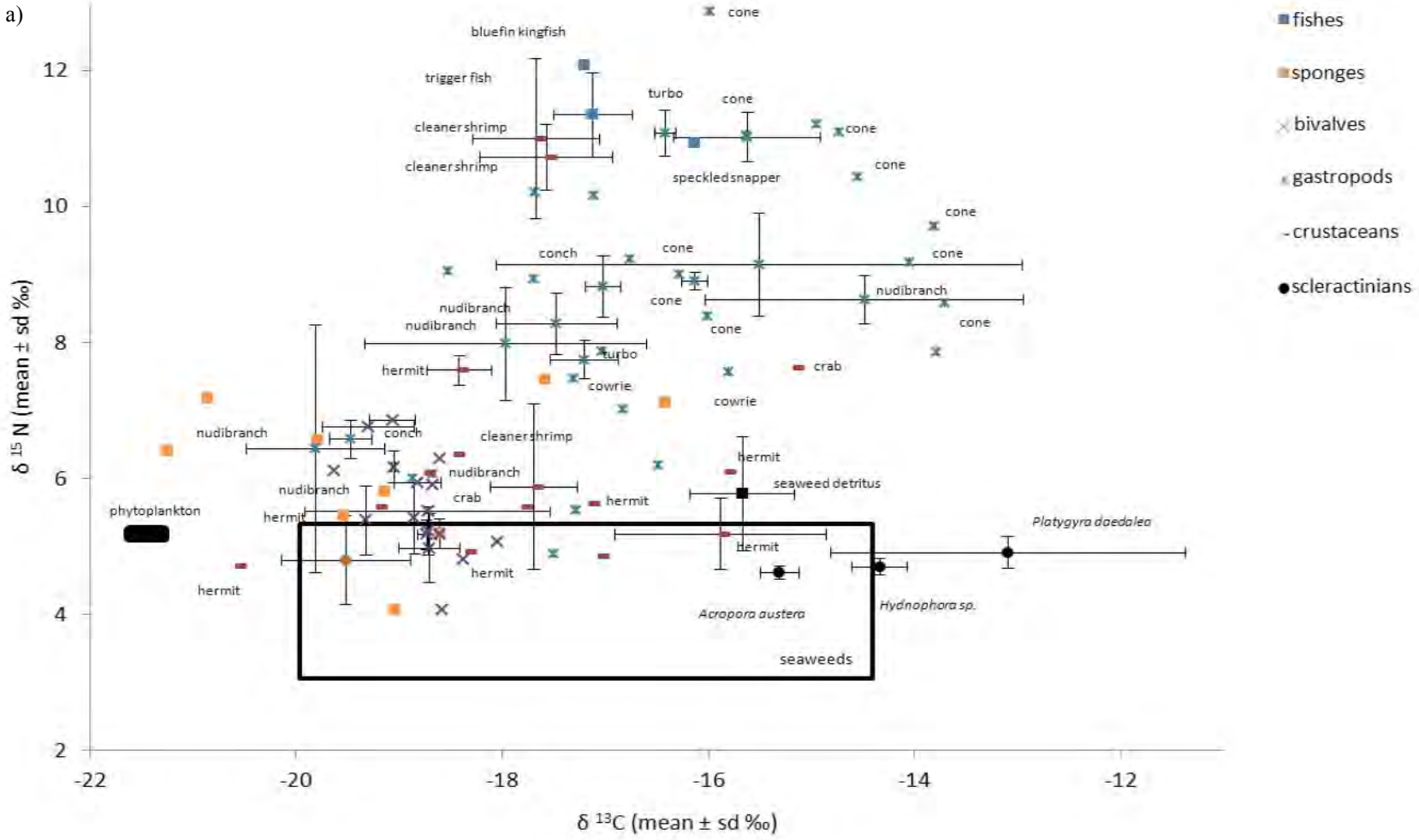


Figure 3.2: a) Carbon and nitrogen stable isotope signatures of consumers sampled from the inshore habitat, b) simplified food web with boxes encompassing the average isotope signatures for each group of consumers. Diagonal lines indicate the trophic enrichment, assuming a 3.4 ‰ enrichment for N and 0.39 ‰ for C, per trophic level (Post 2002).


a)
fishes
sponges
bivalves
gastropods
crustaceans
scleractinians
bluefin kingfish
trigger fish
cleaner shrimp
cleaner shrimp
turbo
cone
speckled snapper
conch
nudibranch
nudibranch
hermit
turbo
cowrie
crab
nudibranch
conch
cleaner shrimp
nudibranch
nudibranch
crab
hermit
phytoplankton
hermit
hermit
hermit
seaweed detritus
Hermit
Platygyra daedalea
Hydnophora sp.
Acropora austera
seaweeds
δ 15 N (mean ± sd ‰)
δ 13C (mean ± sd ‰)
12
10
8
6
4
2
-22
-20
-18
-16
-14
-12

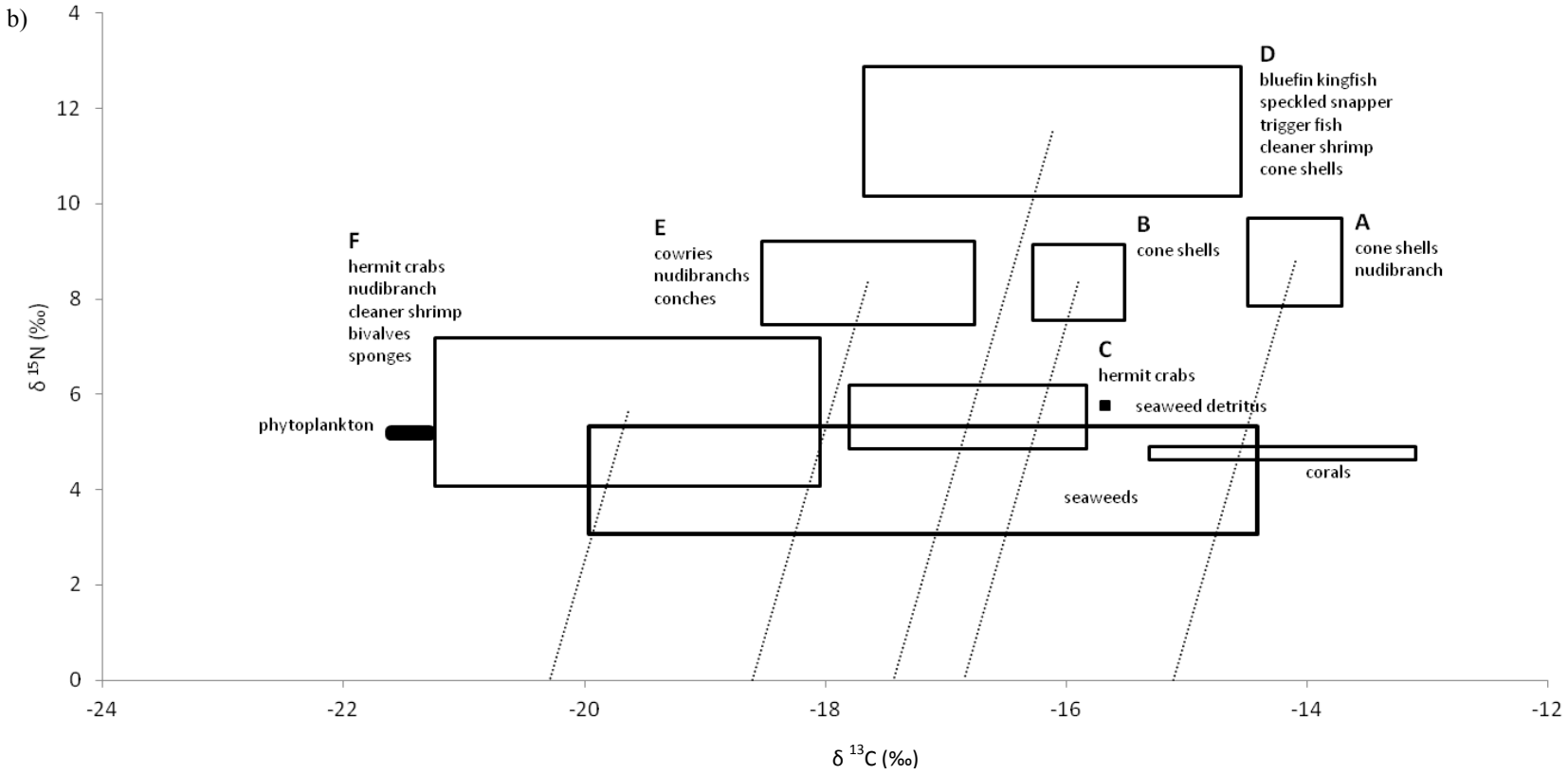


Figure 3.3: a) Carbon and nitrogen stable isotope signatures of consumers sampled from the coral reef habitat, b) simplified food web with boxes encompassing the average isotope signatures for each group of consumers. Diagonal lines indicate the trophic enrichment, assuming a 3.4 ‰ enrichment for N and 0.39 ‰ for C, per trophic level (Post 2002).

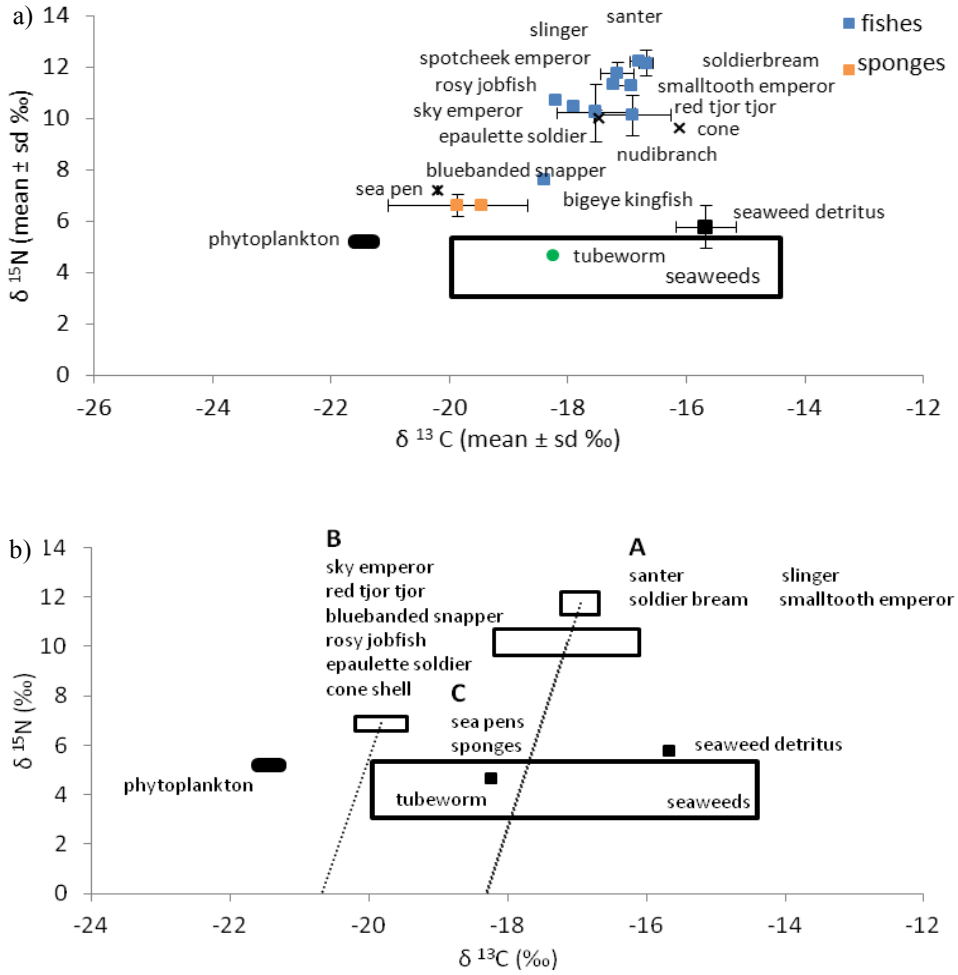


Figure 3.4: a) Carbon and nitrogen stable isotope signatures of consumers sampled from the deep reef habitat, b) simplified food web with boxes encompassing the average isotope signatures for each group of consumers. Diagonal lines indicate the trophic enrichment, assuming a 3.4 ‰ enrichment for N and 0.39 ‰ for C, per trophic level (Post 2002).

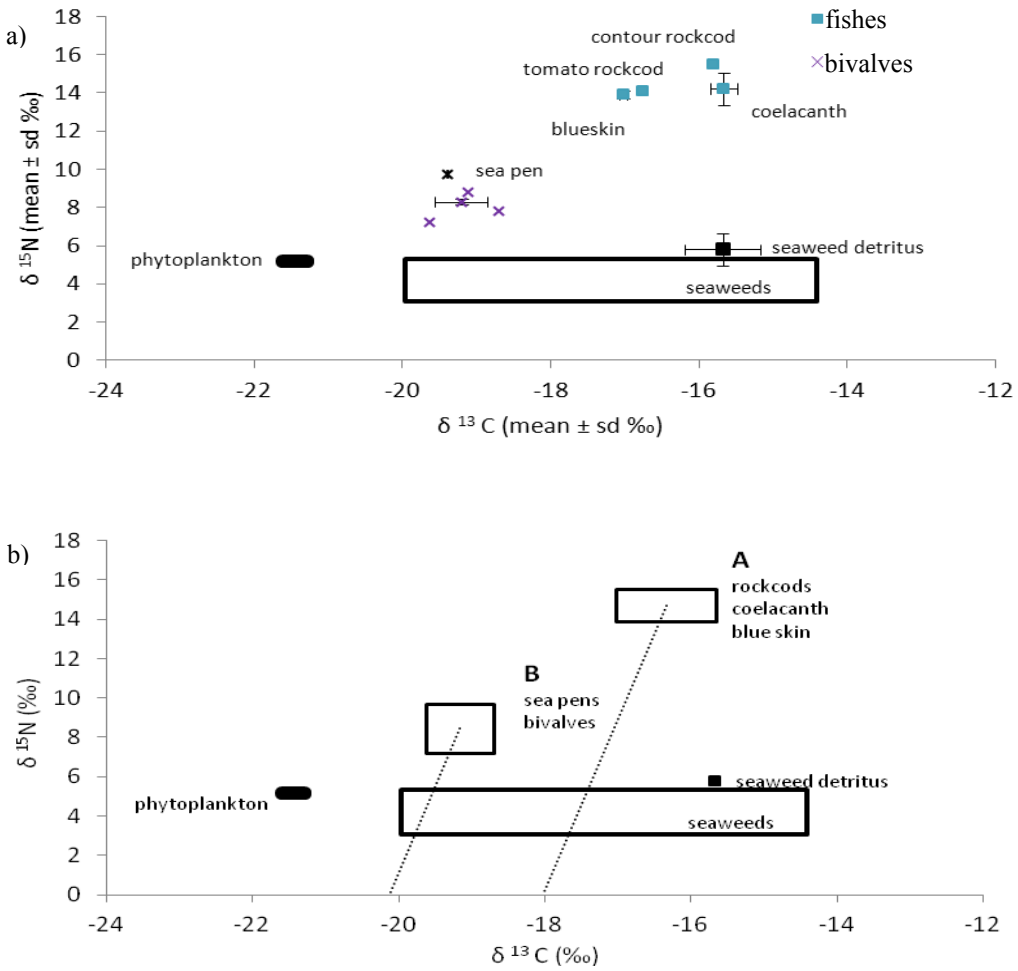


Figure 3.5: a) Carbon and nitrogen stable isotope signatures of consumers sampled from the canyon habitat, b) simplified food web with boxes encompassing the average isotope signatures for each group of consumers. Diagonal lines indicate the trophic enrichment, assuming a 3.4 ‰ enrichment for N and 0.39 ‰ for C, per trophic level (Post 2002).

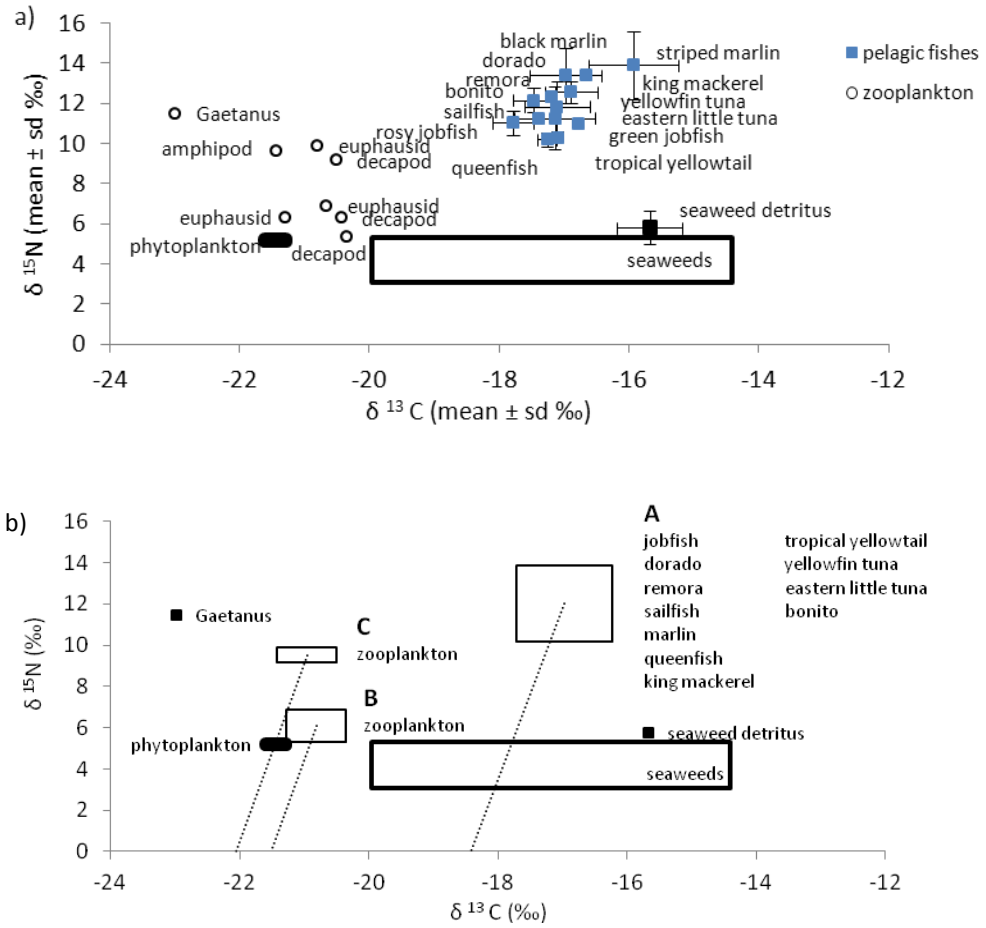


Figure 3.6: a) Carbon and nitrogen stable isotope signatures of consumers sampled from the pelagic habitat, b) simplified food web with boxes encompassing the average isotope signatures for each group of consumers. Diagonal lines indicate the trophic enrichment, assuming a 3.4 ‰ enrichment for N and 0.39 ‰ for C, per trophic level (Post 2002).

Table 3.3: Estimated mean proportion contribution of seaweed (inshore production) and offshore phytoplankton (with 75 % credibility intervals) to various consumer groups in five different habitats off Sodwana Bay, South Africa. Values estimated using *Stable Isotope Analysis in R* (SIAR) (Parnell *et al.* 2010). Estimated trophic level assuming $\delta^{15}N$ fractionation of 3.4 ‰ per trophic level (Post 2002).

	Mean proportion estimated contribution		
Group	Inshore production	Phytoplankton	Trophic level
	Inshore		
A	0.8 (0.72-1)	0.20 (0-0.28)	1
B	0.74 (0.63-0.85)	0.26 (0.15-0.37)	2
C	0.66 (0.52-0.78)	0.34 (0.22-0.48)	1.6
D	0.51 (0.34-0.69)	0.49 (0.31-0.66)	1.2
	Coral reef		
A	0.89 (0.85-1)	0.11 (0-0.15)	1.2
B	0.81 (0.72-0.92)	0.19 (0.08-0.28)	1.1
C	0.68 (0.60-0.76)	0.32 (0.24-0.40)	0.2
D	0.82 (0.75-0.90)	0.18 (0.1-0.25)	2
E	0.60 (0.53-0.66)	0.40 (0.34-0.47)	1.1
F	0.33 (0.29-0.37)	0.67 (0.63-0.71)	0.3
	Deep reef		
A	0.69 (0.58-0.8)	0.31 (0.20-0.42)	2.1
B	0.65 (0.56-0.74)	0.35 (0.26-0.44)	1.6
C	0.30 (0.13-0.45)	0.70 (0.55-0.87)	0.6
	Canyon		
A	0.77 (0.65-0.91)	0.23 (0.09-0.35)	2.9
B	0.39 (0.29-0.5)	0.61 (0.5-0.71)	1.1
	Pelagic		
A	0.69 (0.63-0.75)	0.31 (0.25-0.37)	2.1
B	0.17 (0.02-0.25)	0.83 (0.75-0.98)	0.4
C	0.32 (0.03-0.48)	0.68 (0.52-0.97)	1.4

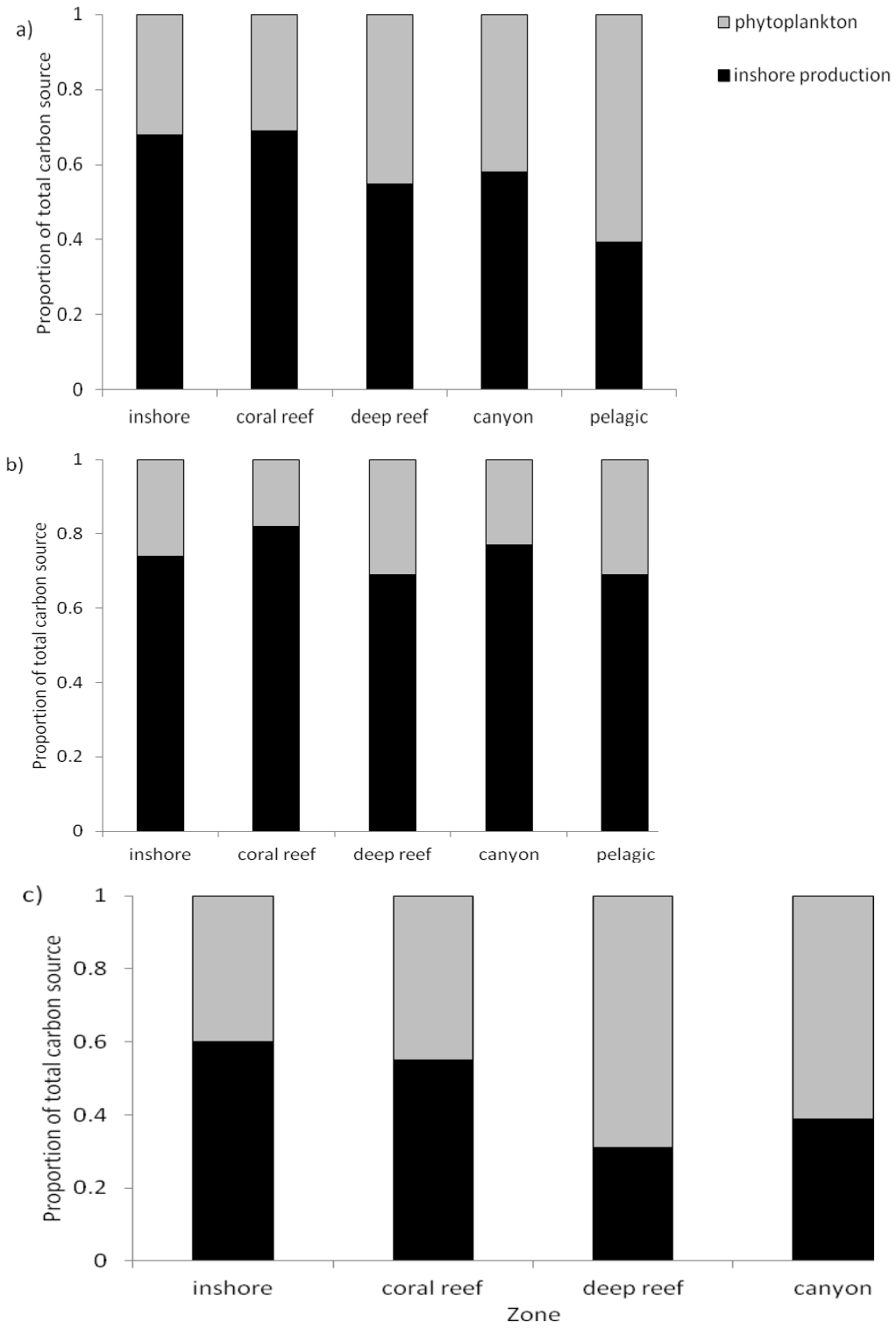


Figure 3.7: Estimated proportional contribution of inshore and offshore phytoplankton production to a) all consumers, b) the top trophic level, and c) benthic invertebrates at five habitats sampled off Sodwana Bay, South Africa.

3.3.7 Inter-habitat comparisons

Estimates of the proportion of each source of primary production utilised by the combined consumer groups from each habitat indicated a slight decline in the utilisation of inshore production from the inshore, at 57%, to the canyon and pelagic consumers, 54 and 46%, respectively (Figure 3.7 a, Table 3.3). The highest trophic level sampled from each habitat did not exhibit a declining trend in the importance of inshore primary production with distance from the shore. Among these consumers, the range in the proportion of inshore production utilised was 69 % for the pelagic fishes, to 82 % for the coral reef fishes (Figure 3.7 b, Table 3.3). Among the benthic invertebrates there was a marked decline in the reliance on inshore production at the deeper sites compared with the coral reef (Figure 3.7 c, Table 3.3), however at 39 % in the canyon consumers, inshore production still contributed significantly to the consumers sampled.

3.4 Discussion

Inshore production was found to be important to all the consumers in all habitats investigated off Sodwana Bay. Among the combined inshore and coral reef consumers, inshore production was the major source of carbon in their diets, at ~ 68 %. The contribution of inshore production to the combined groups of consumers decreased slightly moving offshore, with the offshore phytoplankton contribution becoming increasingly important. The consumers' signatures at all habitats, however, still indicated the importance of inshore production as an important carbon source, estimated to contribute a minimum of ~ 39 %, among pelagic consumers, increasing to 69 % when the zooplankton were excluded. This indicates the importance of inshore production to the entire foodweb of the area, even where seaweed production was low, or non-existent, such as on the deep reef, canyons and pelagic habitats. Macroalgal production has previously been found to be important to inshore consumers

(Dunton & Schell 1987). On the west coast of South Africa, kelp detritus was found to dominate the particulate matter, contributing over 65 % to total POM available to and utilised by mussels (Bustamante & Branch 1996). Kelp were also utilised directly as a food source (Dunton & Schell 1987, Bustamante & Branch 1996). Similar to this study inshore production, in the form of macrophytes, has been shown to make significant contributions to deeper habitats, including submarine canyons (Harrold *et al.* 1998).

The widespread use of inshore production among consumers, despite a lack of seaweeds growing in the canyons evident from ROV surveys (R. Thornycroft, RU, pers. comm.), and the relatively low seaweed biomass in deeper habitats, indicated connectivity of the deep reef and canyons to the inshore habitats where the typically inshore producers are found in greater abundance. This connectivity results in an export of primary production from the shallower inshore regions to the deep reefs, canyon and pelagic habitats. Two possible mechanisms to explain this are suggested. Either there is passive movement of fragments of primary producers to the deeper habitats (Harrold *et al.* 1998) or highly mobile organisms from deeper habitats actively migrate and forage in shallow habitats, thereby importing inshore carbon into deeper habitats. As mobile organisms (fishes) had less variation in their isotopic composition and frequently more enriched carbon isotope signatures than the more sessile benthic organisms, the second mechanism is likely to be of prime importance.

Comparing benthic invertebrates, which have a limited capacity for locomotion and would not be capable of long distance migrations between different habitats, to highly mobile organisms (fish) a trend in the sources of primary production, was noted. Benthic invertebrates exhibited a decrease in the utilisation of inshore primary production with distance from shore. This was probably because the benthic invertebrates, with their limited

ability for locomotion, reflect the locally available organic matter, such as POM, which was shown to exhibit a spatial gradient moving offshore (Chapter 2). POM samples were consistently enriched in $\delta^{13}C$ inshore, with evidence of depletion offshore (Chapter 2). This trend most likely reflects the input of seaweeds to the POM, in the form of detritus, as well as a possibly distinct inshore phytoplankton community including diatoms, which was of significance in the inshore region but declined with distance from the shore. Through this mechanism, benthic invertebrates would be able to utilise seaweed and diatom primary production inshore with the availability of this source further offshore becoming reduced. Non-filter feeding benthic invertebrates tended to possess relatively enriched carbon signatures indicating their reliance on inshore primary production. Suspension feeding invertebrates, in contrast, were repeatedly noted to possess more depleted carbon signatures highlighting their reliance on the more depleted phytoplankton-derived carbon. Due to an insufficient number of suspension feeders from each habitat sampled, the estimated proportions of inshore and offshore production utilised by this functional group were ambiguous. From their relatively depleted carbon signatures, however, a higher contribution of an offshore phytoplankton derived diet appeared likely. Benthic organisms that do not filter-feed should be more likely to access seaweed resources hence their relatively enriched carbon signatures. Filter-feeders, while exhibiting a certain degree of bias in what they ingest, through particle size selectivity (Gili & Coma 1998), are likely to consume suspended matter in the proportion in which they encounter it in the water column as long as their feeding mechanisms allow. Benthic invertebrates have previously been found to have a diet that reflects the availability of carbon sources (Darnaude *et al.* 2004) and it would therefore be expected that the filter-feeders would reflect a phytoplankton-dominated diet, with a variable degree of seaweed, or inshore production, input dependent on the composition of the POM. Previous research has also found phytoplankton production to be the main food source among

filter-feeders. In south-eastern Australia, where terrestrial input is low due to the lack of large riverine input, it was found that seaweeds contributed little, as a carbon source, to the secondary shelf-production (Davenport & Bax 2002). In the latter study, the benthic invertebrate's stable isotope signatures indicated that they assimilated predominantly POM and seagrass into the foodweb. These findings were partially mirrored in this study, whereby the filter-feeders appeared to rely more on POM than the non filter-feeders, however nearshore production, excluding seagrass was still of great importance to the suspension-feeders and other benthic invertebrates. In the Galician upwelling, assimilations by filter-feeders at all sites sampled included at least 40 % from phytoplankton primary production, whereas that from macrophytes was generally < 20 % (Bode *et al.* 2006). The degree to which the filter-feeders utilised phytoplankton production was directly related to its availability in the water column, with a similar pattern exhibited by the deposit feeders (Bode *et al.* 2006). Similarly, in this study, the benthic invertebrates tended to utilise the organic matter relative to its availability to them, with the declining presence of inshore production as determined from the isotope signatures of the POM (Chapter 2) being mirrored in the isotope signatures of the consumers.

Nadon & Himmelman (2006) criticised the interpretation that enriched benthic consumers' signatures indicated the importance of benthic producers, which in this study are included in 'inshore production', due to a lack of decreasing consumer carbon ratios with distance from shore and depth. The authors suggest that such a trend should be evident because of a logical reduction in the occurrence of benthic producers in deeper waters due to limits on photosynthesis. In this study it was found that with increasing distance from shore the benthic consumers did have isotopically lighter carbon signatures. This is supported by the spatial gradient in the isotope signatures and lipid profiles of the POM, moving offshore, to deeper

waters (Chapter 2). The issues raised by Nadon & Himmelman (2006) do not, therefore, apply in this study, and the interpretation that benthic/inshore production is important to benthic consumers, among other groups, is justified.

The three coral species sampled possessed a wide range of carbon signatures, both within (especially *Platygyra daedalea*) and between species. These differences may reflect varying reliance on autochthonous production with more enriched carbon signatures probably indicative of a greater reliance on autochthonous carbon fixation (Ferrier-Pagès *et al.* 2011). These corals occupied a low trophic level similar to bivalves. This indicates their role as low level consumers, probably capturing POM and zooplankton (Lewis & Price 1975, Sebens *et al.* 1996) but also subsidising their carbon through varying levels of autotrophic production from symbiotic zooxanthellae (Odum & Odum 1955). Contributions of these corals to higher trophic levels seemed to be insignificant, among those consumers sampled (Figure 3.3 a, b). Organisms foraging on corals, such as parrotfish, tend to be highly specialised consumers (Rotjan & Lewis 2008) and the method of sample collection, especially hook and line fishing, would have biased the collection to exclude many species that would rely on corals as a source of nutrition. Besides these specialised corallivores, the contribution of corals to the foodweb was apparently low and their function as providers of habitat probably surpasses their role as a food source. Since no corallivores were collected the importance of corals may be underestimated. Further sampling specifically targeting corallivores is required to investigate the importance of this path of production.

In all of the habitats the group in the highest trophic level was dominated by fishes, both among the benthic and pelagic habitats, with these groups exhibiting a similar ultimate carbon source independent of location. This is in sharp contrast to benthic invertebrates that

exhibited isotope signatures that mirrored inshore-offshore isotope gradients in the POM. Among these upper trophic level groups were the canyon-dwelling coelacanth and several species of pelagic gamefishes. These upper trophic level consumer-groups exhibited a similar reliance on inshore production irrespective of where they were caught, with this source comprising between ~ 69 and 82 % of their total carbon source. This indicates that they were assimilating either directly or indirectly inshore food sources and that they may undertake foraging migrations to inshore habitats. Inshore isotope signatures have been found to be fairly consistent in east African coastal waters, with the inshore-offshore gradient found in this study, maintained right up to Kenya (S. Kaehler, RU, pers. comm.). Despite pelagic gamefishes undertaking large migrations and feeding in various coastal waters and not only in Sodwana Bay, their isotope signatures are consistent with an inshore foraging pattern Benthic invertebrates, conversely, must forage on what is in their immediate environment and their signatures reflected this reduction in inshore production available to them with distance from shore.

The degree to which migrations of fishes occur between the deep habitats and the inshore coral reefs is unknown. Stable isotope signatures indicated a major contribution from inshore sources of primary production to the fishes' diets. Daily activities of tropical reef fishes generally revolve around feeding and predator avoidance, with migrations primarily driven by improved access to food at rich feeding grounds (Hobson 1973). The resource rich inshore reefs provide motivation for why fishes from the different habitats should forage inshore. Migratory distances may be relatively short from a few metres (Hobson 1973), up to several hundred metres such as by the striped parrotfish *Scarus croicensis* at Isla Pico Feo on the coast of Panama (Ogden & Buckman 1973) or even upwards of 1 km (Hobson 1973, Gladfelter 1979, Mazeroll & Montgomery 1998). It is therefore possible that deep reef,

canyon and pelagic fishes that were sampled were foraging in shallower habitats driven by inshore production. Alternatively, they were feeding on prey items that foraged inshore but moved between the pelagic or deeper habitats and shallower inshore habitats. It is, however, likely that a combination of the two occur. Heemstra *et al.* (2006), from footage obtained during a submersible expedition, noted an abundance of coral reef fish species in the canyons, forming a link between the inshore region and the deep canyon heads, and concluded that primary production occurring inshore, in shallower waters, sustained much of the canyon fauna. Isotopic signatures of the coelacanth also indicated that they were relying on a relatively enriched carbon source, ultimately derived from inshore primary production. The sighting of a coelacanth by SCUBA divers during a recreational dive at ~ 50 m in Sodwana Bay also supports the idea of inshore foraging (Roberts *et al.* 2006). This sighting coincided with an upwelling event that reduced the temperature of the continental shelf waters, where the water would otherwise have been too warm as they generally favour water cooler than 20 °C (Fricke & Plante 1988) with the shelf waters normally only reaching a minimum of ~ 21 °C during winter. This encroachment of cold water onto the shelf allowed the fish to move to such shallow depths (Roberts *et al.* 2006). It is not certain whether the coelacanth was undertaking a foraging migration but this is possible, considering the abundance of potential prey on the reefs inshore of the canyons. In the Comoros, the coelacanth's temperature limitation is hypothesised to prevent them from exploiting the richer food sources in shallower, warmer water (Fricke & Plante 1988). Upwelling events may provide these fish with a temporary corridor to exploit the shallower resources, otherwise unobtainable under normal temperature regimes. The observations of the reef fishes in the canyon, and the sighting of the coelacanth in unusually shallow depths support the idea of migrations by fishes between deeper habitats further offshore and the inshore habitats, probably driven to some extent by foraging.

In this study, only zooplankton were found to rely almost exclusively on offshore production, upwards of 68 %, possessing similar signatures to the offshore POM. *Gaetanus* sp. was an exception as it had more depleted carbon signatures than the offshore POM. The prey of these copepods were likely to be from selective feeding (Bouillon et al. 2000) on the more depleted, smaller size fractions of the POM (Korb *et al.* 1996). Overall, however, zooplankton were found to rely almost exclusively on offshore production. Despite their swimming capability, the zooplankton's distribution is largely dictated by water currents. Similarly, the food resources available to zooplankton are dictated by currents, and since zooplankton will likely only spend a short time above the continental shelf, due to the influence of the powerful Agulhas Current (Lutjeharms 2006), their signatures reflect a largely offshore phytoplankton-derived diet. While pelagic production was an important energy source to filter-feeders, and in turn to organisms preying on these filter-feeders, it did not appear to contribute much to higher trophic levels such as the predatory fishes. A lack of smaller planktivorous fishes, except those associated with the reefs, may account for this apparent, low utilisation of phytoplankton production by higher trophic levels, especially the pelagic gamefishes.

To conclude, this chapter revealed using stable isotope analysis that the major sources of primary production to consumers in Sodwana Bay were phytoplankton, and those seaweeds that did not have highly depleted carbon signatures. Isotopically depleted red seaweeds, which were sampled at all habitats, as well as seagrass that was restricted to the shallow subtidal habitat, were not found to be of significance to higher trophic levels. There was a high degree of trophic connectivity between the inshore habitats (characterised by enriched primary producer carbon signatures, where seaweeds occur with a relatively high biomass, and diatoms were likely to occur), and deeper habitats offshore, such as the deep reef,

canyons, and pelagic zone (where seaweed biomass was low and a different suite of offshore phytoplankton occurred). When combining all of the consumer groups from each habitat, inshore production was important in all of the habitats, with a minimum contribution of 39 % among the pelagic consumers, when zooplankton were included, increasing to 69 % when the zooplankton were excluded. The trophic connectivity between the inshore habitat and habitats further offshore was most likely achieved predominantly through the movement of organisms between habitats, with a significant level of inshore foraging occurring. Benthic organisms, not able to move large distances between different habitats, reflected the availability of organic matter where they occurred, with a decrease in the typical inshore production signature with distance from shore. Among the benthic invertebrates the estimated contribution of inshore production to their diets decreased from 60 % inshore to 31 % at the deep reef. Fishes, which are more mobile organisms capable of covering large distances, showed a relatively homogeneous utilisation of inshore production ranging between 69 and 82 % regardless of where they were caught. The pelagic and deep benthic fishes, including those from the deep reef and canyon, were likely undertaking foraging migrations to feed in the shallower inshore habitats driven largely by inshore production, and/or were preying on other fishes that foraged inshore but also moved to the deeper habitats. Zooplankton were the only consumers with carbon isotope signatures sufficiently depleted to signify a predominantly phytoplankton derived diet. This suggests strong connectivity between different habitats and dictates that successful management must consider all of the habitats holistically.

Chapter 4

General Discussion

This thesis investigated the trophic dynamics and energy flow in the Sodwana Bay system through the use of soft-tissue stable isotope and lipid analyses. The overall objectives were to investigate and examine the source and utilisation of carbon in order to gain a better understanding of the trophic ecology of the organisms occurring in the various habitats from the intertidal, coral reef, deep reef, canyon and pelagic habitats in the area. Basal trophic level dynamics were investigated both spatially and temporally through monitoring suspended POM. The principal contributors to primary production were found to be both seaweeds (excluding certain red seaweeds with depleted carbon signatures) and phytoplankton. Seagrass, and red seaweeds that possessed depleted carbon signatures, were of no apparent significance as there were no consumers' stable isotope signatures that indicated their consumption in any major proportion. Inshore primary production, contributed by seaweeds and a likely distinct suite of phytoplankton compared to the offshore assemblage, was indicated to be the predominant carbon source to inshore consumers, and was also found to be important for consumers at the furthest habitat sampled, within the canyon head and for pelagic fishes.

An inshore-offshore gradient was apparent in both the sources of production (Chapter 2) and the relative utilisation of the characteristic inshore and offshore production (Chapter 3) in certain consumer groups primarily those including the benthic invertebrates that are either sessile, such as benthic filter feeders, and those that are incapable of long distance movement, such as motile, benthic gastropods. Despite this diminished utilisation of inshore production by benthic invertebrates further offshore, there was still a significant proportion of inshore

production utilised by these organisms, estimated to contribute no less than 31 % to their diets. Among the fishes, which are capable of covering considerable distances during foraging excursions (Hobson 1973, Gladfelter 1979, Mazeroll & Montgomery 1998), there was no inshore-offshore gradient in their utilisation of different sources of production. Fishes exhibited a consistent, utilisation of inshore production regardless of where they were caught. This highlights the importance of the inshore habitats for the integrity of the entire foodweb, and the interconnectedness that exists between habitats. Ensuring the conservation of all the different types of habitats is therefore vital in ensuring ecosystem integrity and function.

Seaweeds and diatoms were probably the most important in their contribution to the foodweb among the assemblage of inshore primary producers. With seaweeds predominantly restricted to the benthos, their depth-related distributions are largely dependent on light (Markager & Sand-Jensen 1992). An increase in water turbidity may therefore negatively impact inshore primary production as well as biomass. Currently, there is little threat of increasing water turbidity through increased sediment load in the area due to the lack of large rivers and therefore minimal sediment input. Eutrophication could also impact on both the seaweed, and phytoplankton communities (Schramm 1999). Increased phytoplankton growth, a characteristic of eutrophication, would increase turbidity and light attenuation thereby reducing the euphotic depth, reduce seaweed production and also alter phytoplankton assemblages (Schramm 1999, Tett *et al.* 2007). With the trend towards increasing development in the Sodwana Bay area, largely restricted thus far to beyond the Park boundaries and alongside the road leading to the entrance to the iSimangaliso Wetland Park (Mograbi & Rogerson 2007), eutrophication may become a threat in the future, if measures to prevent this are not adopted. The most common sources of excess, anthropogenic nutrients include waste water and fertilisers (Tett *et al.* 2007). The latter is not likely to be realised in

the current setting as there are no agricultural practices near to Sodwana Bay, while the latter should be considered if further development neighbouring the beaches in the area occurs.

Recreational activities in Sodwana Bay, which are dominated by SCUBA diving and pelagic game fishing, are generally regarded as non-consumptive. The iSimangaliso Wetland Park has the highest latitude coral reefs in the South West Indian Ocean, and its coral reef biota are the focus of current conservation efforts. While pollution, coastal development and climate change have the potential to impact on any coral reef system, it is fishing that exerts the highest, directed pressure on coral reef and other marine ecosystems (Jennings & Kaiser 1998, Jackson *et al.* 2001). Losses due to fishing, in addition to impacts on the target species, may result in the ultimate removal of a large proportion of the primary production and therefore have broader ecosystem effects (Pauly & Christensen 1995, Pinnegar *et al.* 2000). Some of the apex predators in Sodwana Bay, shown by stable isotope analysis, are the most frequently targeted and caught by recreational fishers in northern KwaZulu-Natal. These species include king mackerel *Scomberomorus commerson*, sailfish *Istiophorus platypterus*, marlin *Tetrapturus audax* and *Makaira* spp., and kingfish. Top-down trophic cascading is therefore likely to result from removing apex pelagic predators from the system. Targeting of coral reef resident species can also potentially lead to trophic cascades, involving the prey of the targeted fishes, if certain species are fished below critical levels (McManus 1997). Larger, apex predators, such as those targeted in Sodwana Bay, have an important role in structuring the fish assemblages on coral reefs (Friedlander & DeMartini 2002). Fishing tends to remove apex predators, and can result in concomitant increases in prey populations, including herbivorous fishes, with broad destructive impacts on the ecosystem likely to follow (Friedlander & DeMartini 2002). If, for example, fishes that prey on invertebrates are removed, herbivorous invertebrates may become more abundant leading to a decline in seaweed stocks, which may in turn increase bioerosion of the reef leading to a reduced

abundance and diversity of fishes through habitat destruction in a top-down trophic cascade (Jennings & Kaiser 1998). Removing herbivorous fishes is also problematic as the competition of invertebrate grazers, such as sea urchins, is reduced leading to population expansions and increased pressure on seaweeds, as urchins are able to survive on lower algal stocks than herbivorous fishes (Jennings & Kaiser 1998). This may lead to the establishment of a new stable system whereby the niche of the main herbivore is now filled by invertebrates (Jennings & Kaiser 1998). Predators of these invertebrates are usually also susceptible to fishing and are also removed from the system, exacerbating the problem (Jennings & Kaiser 1998). Targeting of benthic fish is prohibited in Sodwana Bay, although incidental hooking of benthic fish does occur (personal observations). Given the interconnectivity between habitats that is apparent from this study, trophic cascades that may occur in one habitat would have far-reaching impacts on the ecosystem functioning as a whole. This would especially be true if the suite of inshore primary producers that were shown to be important to consumers in all habitats were altered.

Production by corals was shown to have little importance to those consumers that were sampled. While sampling biases contributed to this observation, it is indisputable that other sources of production are the major drivers of this ecosystem. Corals may thus be seen in the current setting as important providers of habitat to the coral reef biota (Moberg & Folke 1999). Typically, in coral reef communities, there is a grazer-mediated balance between seaweed and coral dominated benthic cover, although other factors such as nutrient loading and physical disturbance levels are also important factors (Littler & Littler 1985). Grazing and other physical disturbances of seaweed tend to facilitate coral dominance (Littler & Littler 1994). Removal of these grazers has been linked with increased seaweed abundance, that smother out the corals (Littler & Littler 1985). Many corals in Sodwana Bay are fragile

and may easily be damaged by divers (Schleyer & Tomalin 2000). Damage caused by algal growth on sites of fishing line fouling was high, while storm-attributed damage was low even after large storm events (Schleyer & Tomalin 2000). Reduction in coral cover favours the propagation of seaweeds, when physical and/or physiological stressors are unsuitable to corals (Littler & Littler 1985). While this may increase primary productivity of the community up to a certain level, eventually the loss of the characteristic coral reef attributes and structure will lead to the ultimate loss of the coral reef community dependent on the associated habitat. The integrity of the grazer community needs to be ensured while the diver- and fishing related damage to corals must be minimised or totally prevented in order to maintain the balance between the seaweeds, which contribute to the inshore production that is important in fuelling the foodweb, and the corals, which are crucial in their provision of habitat to coral reef biota.

Connectivity between the canyon fauna and inshore zones, as indicated through the stable isotope results, only seem plausible through the movement of mobile organisms between the two zones, not from passively transported organic matter. Submarine canyons tend to receive greater amounts of sedimentation and allochthonous production than their immediate surroundings (Houston & Haedrich 1984). Seaweed detritus is hypothesised to reach canyons by lateral transport along the seafloor, as opposed to sinking in from the water column above the canyon (Vetter & Dayton 1998). Currents may follow the course of canyons thereby facilitating the passage of organic matter in the form of detritus from shallower areas to great depths (Harrold *et al.* 1998). Some canyons, however, due to flow dynamics, are continually flushed out thus preventing sedimentation of imported production (Houston & Haedrich 1984). Carson Canyon, on the Grand Banks to the east of Newfoundland, Canada is an example of a canyon where organic matter imports are low, with pelagic production

identified as the major source of organic matter for canyon-resident fishes (Houston & Haedrich 1984). In Carmel Canyon in Monterey Bay, California, kelp *Macrocystis pyrifera* was exported as large aggregations produced in nearby kelp forests to the canyons where they were a major source of organic carbon to the consumers, constituting an estimated 20 – 83 % of the total carbon imported into the canyon. This high percentage was partly due to the unusually close proximity of the canyon to a kelp forest (Harrold *et al.* 1998). Large proportions of seaweed detritus may be consumed directly, with this supplementary food source noted to confer advantages to the consumers living in the canyons, relative to conspecifics living outside the canyon (Harrold *et al.* 1998, Vetter & Dayton 1998). It would seem unlikely that transport of macroalgal detritus into the canyons of Sodwana Bay was of any significance, with currents at depths of 100 – 140 m measured to be between 20 and 80 $cm.s^{-1}$ parallel to the coast, in a north to south direction (Roberts *et al.* 2006), and large beds of seaweeds comparable to kelp beds not occurring in the area, thereby hindering any drift of detritus from the inshore zones to the canyons. Further evidence for the potential connectivity between different depth zones lies in the behaviour of coelacanths. Coelacanths feed opportunistically on co-inhabiting cave fishes at a depth of 164 – 243 m in the Comorian Archipelago (Fricke & Hissmann 1994, 2000). Additionally coelacanths were recorded undertaking nocturnal foraging trips with some individuals descending to depths of around 700 m (Fricke & Hissmann 1994). In addition to the likely predation on reef fishes, the closest distance between a canyon and the coral reef being only 140 m (Heemstra *et al.* 2006), movements between shallower reefs and the deeper canyons, by coelacanths may therefore be possible. Heemstra *et al*. (2006) suggest that foraging trips by Sodwana coelacanths to deeper waters is unlikely due to the abundance of potential prey in their immediate surrounds, and the observed decreasing fish biomass in greater depths. Even if the coelacanths in Sodwana Bay do not undertake deeper excursions, other more mobile fish

species occurring in their depth range may (Heemstra *et al.* 2006). The canyons then are likely to be linked with both shallower and deeper habitats through the movement of mobile organisms, and are not an end-point to the energy received from inshore zones.

To conclude, inshore production is an important source of primary production, with this source contributing significantly to the entire foodweb. The consumptive, pelagic game fishing, as well as the supposedly non-consumptive diving practices occurring in Sodwana Bay need to be monitored and regulated so as not to have undesirable effects on the coral reef community, and through the observed inter-connectedness, the entire system that could cause the loss of abundance and biodiversity of organisms. Cognisance of the inter-connectedness of all of the habitats needs to be maintained, when considering any of the habitats and their respective communities.

References

Anderson RJ, McKune C, Bolton JJ, DeClerck O, Tronchin E. 2005. Patterns in subtidal seaweed communities on coral-dominated reefs at Sodwana Bay on the KwaZulu-Natal coast, South Africa. *African Journal of Marine Science* **27**: 529-537.

Bates NR, Hansell DA, Moran SB, Codispoti LA. 2005. Seasonal and spatial distribution of particulate organic matter (POM) in the Chukchi and Beaufort Seas. *Deep-Sea Research II* **52**: 3324-3343.

Baum JK, Worm B. 2009. Cascading top-down effects of changing oceanic predator abundances. *Journal of Animal Ecology* **78**: 699-714.

Blum LK, Mills AL. 1991. Microbial growth and activity during the initial stages of seagrass decomposition. *Marine Ecology Progress Series* **70**: 73-82.

Bode A, Alvarez-Ossorio MT, Varela M. 2006. Phytoplankton and macrophyte contributions to littoral food webs in the Galician upwelling estimated from stable isotopes. *Marine Ecology Progress Series* **318**: 89-102.

Bosley KL, Lavelle JW, Brodeur RD, Wakefield WW, Emmett RL, Baker ET, Rehmke KM. 2004. Biological and physical processes in and around Astoria submarine Canyon, Oregon, USA. *Journal of Marine Systems* **50**: 21-37.

Bouillon S, Chandra Mohan N, Sreenivas N, Dehairs F. 2000. Sources of suspended organic matter and selective feeding by zooplankton in an estuarine mangrove ecosystem as traced by stable isotopes. *Marine Ecology Progress Series* **208**: 79-92.

Branch GM, Griffiths CL, Branch ML, Beckley LE. 2005. *Two Oceans: A guide to the marine life of southern Africa*. Cape Town: David Philip Publishers. 360 pp

Budge SM, Parrish CC. 1998. Lipid biogeochemistry of plankton, settling matter and sediments in Trinity Bay, Newfoundland. II. Fatty acids. *Organic Geochemistry* **29**: 1547-1559.

Burchall J. 1968. Primary production studies in the Agulhas Current region off Natal - June, 1965. 18 pp. Oceanographic Research Institute.

Bustamante RH, Branch GM. 1996. The dependence of intertidal consumers on kelp-derived organic matter on the west coast of South Africa. *Journal of Experimental Marine Biology and Ecology* **196**: 1-28.

Bustamante RH, Branch GM, Eekhout S, Robertson B, Zoutendyk P, Schleyer M, Dye A, Hanekom N, Keats D, Jurd M, McQuaid C. 1995. Gradients of intertidal primary productivity around the coast of South Africa and their relationships with consumer biomass. *Oecologia* **102**: 189-201.

Capone DG. 2001. Marine nitrogen fixation: what's the fuss? *Current Opinion in Microbiology* **4**: 341-348.

Capone DG, Carpenter EJ. 1982. Nitrogen fixation in the marine environment. *Science* **217**: 1140-1142.

Carlier A, Le Guilloux E, Olu K, Sarrazin J, Mastrototaro F, Taviani M, Clavier J. 2009. Trophic relationships in a deep Mediterranean cold-water coral bank (Santa Maria di Leuca, Ionian Sea). *Marine Ecology Progress Series* **397**: 125-137.

Carpenter EJ, Harvey HR, Fry B, Capone DG. 1997. Biogeochemical tracers of the marine cyanobacterium *Trichodesmium*. *Deep-Sea Research I* **44**: 27-38.

Carpenter SR, Kitchell JF, Hodgson JR, Cochran PA, Elser JJ, Elser MM, Lodge DM, Kretchmer D, He X, von Ende CN. 1987. Regulation of lake primary productivity by food web structure. *Ecology* **68**: 1863-1876.

Celliers L, Schleyer MH. 2002. Coral bleaching on high-latitude marginal reefs at Sodwana Bay, South Africa. *Marine Pollution Bulletin* **44**: 1380-1387.

Celliers L, Schleyer MH. 2008. Coral community structure and risk assessment of high-latitude reefs at Sodwana Bay, South Africa. *Biodiversity and Conservation* **17**: 3097-3117.

Clements KD, Choat JH. 1993. Influence of season, ontogeny and tide on the diet of the temperate marine herbivorous fish *Odax pullus* (Odacidae). *Marine Biology* **117**: 213-220.

Corbisier TN, Soares LSH, Petti MAV, Muto EY, Silva MHC, McClelland J, Valiela I. 2006. Use of isotopic signatures to assess the food web in a tropical shallow marine ecosystem of Southeastern Brazil. *Aquatic Ecology* **40**.

Dalsgaard J, St. John M, Kattner G, Müller-Navarra D, Hagen W. 2003. Fatty acid trophic markers in the pelagic marine environment. *Advances in Marine Biology* **46**: 225-340.

Darnaude AM, Salen-Picard C, Harmelin-Vivien ML. 2004. Depth variation in terrestrial particulate organic matter exploitation by marine coastal benthic communities off the Rhone River delta (NW Mediterranean). *Marine Ecology Progress Series* **275**: 57-57.

Davenport SR, Bax NJ. 2002. A trophic study of a marine ecosystem off southeastern Australia using stable isotopes of carbon and nitrogen. *Canadian Journal of Fisheries & Aquatic Sciences* **59**: 514.

De Clerck O, Bolton JJ, Anderson RJ, Coppejans E. 2005. *Guide to the seaweeds of KwaZulu-Natal*. Belgium: National Botanic Garden of Belgium. 294 pp

DeNiro MJ, Epstein S. 1978. Influence of diet on the distribution of carbon isotopes in animals. *Geochimica et Cosmochimica Acta* **42**: 495-506.

DeNiro MJ, Epstein S. 1981. Influence of diet on the distribution of nitrogen isotopes in animals. *Geochimica et Cosmochimica Acta* **45**: 341-351.

Deuser W, Degens E, Guillard R. 1968. Carbon isotope relationships between plankton and sea water. *Geochimica et Cosmochimica Acta* **32**: 657-660.

Duffy DC, Jackson S. 1986. Diet studies of seabirds: a review of methods. *Colonial Waterbirds* **9**: 1-17.

Dugdale RC, Goering JJ. 1967. Uptake of new and regenerated forms of nitrogen in primary productivity. *Limnology and Oceanography* **12**: 196-206.

Duggins DO, Simenstad CA, Estes JA. 1989. Magnification of secondary production by kelp detritus in coastal marine ecosystems. *Science* **245**: 170-173.

Duineveld GCA, Lavaleye MSS, Berghuis EM. 2004. Particle flux and food supply to a seamount cold-water coral community (Galicia Bank, NW Spain). *Marine Ecology Progress Series* **277**: 13-23.

Dunton KH, Schell DM. 1987. Dependence of consumers on macroalgal (*Laminaria solidungula*) carbon in an arctic kelp community: ^{13}C evidence. *Marine Biology* **93**: 615-625.

Ehleringer JR, Rundel PW, Nagy KA. 1986. Stable isotopes in physiological ecology and food web research. *Trends in Ecology & Evolution* **1**: 42-45.

Eppley RW. 1972. Temperature and phytoplankton growth in the sea. *Fishery Bulletin* **70**: 1063-1085.

Ferrier-Pagès C, Peirano A, Abbate M, Cocito S, Negri A, Rottier C, Riera P, Rodolfo-Metalpa R, Reynaud S. 2011. Summer autotrophy and winter heterotrophy in the temperate symbiotic coral *Cladocora caespitosa*. *Limnology and Oceanography* **56**: 1429-1438.

Flemming BW. 1981. Factors controlling shelf sediment dispersal along the southeast African continental margin. *Marine Geology* **42**: 259-277.

Floros CD, Samways MJ, Armstrong B. 2004. Taxonomic patterns of bleaching within a South African coral assemblage. *Biodiversity and Conservation* **13**: 1175-1194.

Folch J, Lees M, Sloane Stanley GH. 1957. A simple method for the isolation and purification of total lipides from animal tissues. *The Journal of Biological Chemistry* **226**: 497-509.

Forget M-H, Platt T, Sathyendranath S, Fanning P. 2011. Phytoplankton size structure, distribution, and primary production as the basis for trophic analysis of Caribbean ecosystems. *ICES Journal of Marine Science: Journal du Conseil* **68**: 751-765.

Frederiksen M, Edwards M, Richardson AJ, Halliday NC, Wanless S. 2006. From plankton to top predators: bottom-up control of a marine food web across four trophic levels. *Journal of Animal Ecology* **75**: 1259-1268.

Fricke H, Hissmann K. 1994. Home range and migrations of the living coelacanth *Latimeria chalumnae*. *Marine Biology* **120**: 171-180.

Fricke H, Hissmann K. 2000. Feeding ecology and evolutionary survival of the living coelacanth *Latimeria chalumnae*. *Marine Biology* **136**: 379-386.

Fricke H, Plante R. 1988. Habitat requirements of the living coelacanth, *Latimeria chalumnae*, at Grande Comore, Indian Ocean. *Naturwissenschaften* **75**: 149-151.

Friedlander AM, DeMartini EE. 2002. Contrasts in density, size, and biomass of reef fishes between the northwestern and the main Hawaiian islands: the effects of fishing down apex predators. *Marine Ecology Progress Series* **230**: 253-264.

Fry B, Wainright SC. 1991. Diatom sources of ^{13}C-rich carbon in marine food webs. *Marine Ecology Progress Series* **76**: 149-157.

Gaston TF, Schlacher TA, Connolly RM. 2006. Flood discharges of a small river into open coastal waters: plume traits and material fate. *Estuarine, Coastal and Shelf Science* **69**: 4-9.

Gili J-M, Coma R. 1998. Benthic suspension feeders: their paramount role in littoral marine food webs. *Trends in Ecology & Evolution* **13**: 316-321.

Gladfelter WB. 1979. Twilight migrations and foraging activities of the copper sweeper *Pempheris schomburgki* (Teleostei: Pempheridae). *Marine Biology* **50**: 109-119.

Glassom D, Celliers L, Schleyer MH. 2006. Coral recruitment patterns at Sodwana Bay, South Africa. *Coral Reefs* **25**: 485-492.

Gordon ES, Goñi MA. 2003. Sources and distribution of terrigenous organic matter delivered by the Atchafalaya River to sediments in the northern Gulf of Mexico. *Geochimica et Cosmochimica Acta* **67**: 2359-2375.

Green AN, Goff JA, Uken R. 2007. Geomorphological evidence for upslope canyon-forming processes on the KwaZulu-Natal shelf, SW Indian Ocean, South Africa. *Geo-Marine Letters* **27**: 399-409.

Green AN, Uken R. 2005. First observations of sea-level indicators related to glacial maxima at Sodwana Bay, northern KwaZulu-Natal. *South African Journal of Science* **101**: 236-238.

Gu B, Chapman AD, Schelske CL. 2006. Factors controlling seasonal variations in stable isotope composition of particulate organic matter in a soft water eutrophic lake. *Limnology and Oceanography* **51**: 2837-2848.

Gu B, Schelske C, Waters M. 2011. Patterns and controls of seasonal variability of carbon stable isotopes of particulate organic matter in lakes. *Oecologia* **165**: 1083-1094.

Haas AF, Naumann MS, Struck U, Mayr C, el-Zibdah M, Wild C. 2010. Organic matter release by coral reef associated benthic algae in the Northern Red Sea. *Journal of Experimental Marine Biology and Ecology* **389**: 53-60.

Harrold C, Light K, Lisin S. 1998. Organic enrichment of submarine-canyon and continental-shelf benthic communities by macroalgal drift imported from nearshore kelp forests. *Limnology and Oceanography* **43**: 669-678.

Heemstra PC, Fricke H, Hissmann K, Schauer J, Smale M, Sink KJ. 2006. Interactions of fishes with particular reference to coelacanths in the canyons at Sodwana Bay and the St Lucia Marine Protected Area of South Africa. *South African Journal of Science* **102**: 461-465.

Hill JM. 2007. A stable isotope approach to trophic ecology: resolving foodwebs in intertidal ecosystems. PhD thesis, Rhodes University, South Africa, 159pp.

Hissmann K, Fricke H, Schauer J, Ribbink AJ, Roberts MJ, Sink KJ, Heemstra PC. 2006. The South African coelacanths - an account of what is known after three submersible expeditions. *South African Journal of Science* **102**: 491-500.

Hobson ES. 1973. Diel feeding migrations in tropical reef fishes. *Helgoländer Wissenschaftliche Meeresuntersuchungen* **24**: 361-370.

Houston KA, Haedrich RL. 1984. Abundance and biomass of macrobenthos in the vicinity of Carson Submarine Canyon, northwest Atlantic Ocean. *Marine Biology* **82**: 301-305.

Howarth RW, Marino R, Lane J, Cole JJ. 1988. Nitrogen fixation in freshwater, estuarine, and marine ecosystems. 1. Rates and importance. *Limnology and Oceanography* **33**: 669-687.

Jackson JB, Kirby MX, Berger WH, Bjorndal KA, Botsford LW, Bourque BJ, Bradbury RH, Cooke R, Erlandson J, Estes JA, Hughes TP, Kidwell S, Lange CB, Lenihan HS, Pandolfi JM, Peterson CH, Steneck RS, Tegner MJ, Warner RR. 2001. Historical overfishing and the recent collapse of coastal ecosystems. *Science* **293**: 629-637.

Jennings S, Kaiser MJ. 1998. The effects of fishing on marine ecosystems. *Advances in Marine Biology* **34**: 201-352.

Kadye W, Booth AJ. 2012. Integrating stomach content and stable isotope analyses to elucidate the feeding habits of non-native sharptooth catfish *Clarias gariepinus*. *Biological Invasions* 779-795.

Kelly JR, Scheibling RE. 2012. Fatty acids as dietary tracers in benthic food webs. *Marine Ecology Progress Series* **446**: 1-22.

King D, Fraser V. 2001. *More Reef Fishes and Nudibranchs: East and South Coast of Southern Africa*. Cape Town: Struik Publishers. pp

Kling GW, Fry B, O'Brien WJ. 1992. Stable isotopes and planktonic trophic structure in arctic lakes. *Ecology* **73**: 561-566.

Klump DW, McKinnon AS. 1989. Temporal and spatial patterns in primary production of a coral-reef epilithic algal community. *Journal of Experimental Marine Biology and Ecology* **131**: 1-22.

Korb RE, Raven JA, Johnston AM, Leftley JW. 1996. Effects of cell size and specific growth rate on stable carbon isotope discrimination by two species of marine diatom. *Marine Ecology Progress Series* **143**: 283-288.

Kromkamp J, Peene J, van Rijswijk P, Sandee A, Goosens N. 1995. Nutrients, light and primary production by phytoplankton and microphytobenthos in the eutrophic, turbid Westerschelde estuary (The Netherlands). *Hydrobiologia* **311**: 9-19.

Lara RJ, Alder V, Franzosi CA, Kattner G. 2010. Characteristics of suspended particulate organic matter in the southwestern Atlantic: influence of temperature, nutrient and phytoplankton features on the stable isotope signature. *Journal of Marine Systems* **79**: 199-209.

Laws EA, Popp BN, Bidigare RR, Riebesell U, Burkhardt S, Wakeham SG. 2001. Controls on the molecular distribution and carbon isotopic composition of alkenones in certain haptophyte algae. *Geochemistry Geophysics Geosystems* **2**: 1-31.

Layman CA, Araujo MS, Boucek R, Hammerschlag-Peyer CM, Harrison E, Jud ZR, Matich P, Rosenblatt AE, Vaudo JJ, Yeager LA, Post DM, Bearhop S. 2011. Applying stable isotopes to examine food-web structure: an overview of analytical tools. *Biological Reviews* no-no.

Lewis JB, Price WS. 1975. Feeding mechanisms and feeding strategies of Atlantic reef corals. *Journal of Zoology* **176**: 527-544.

Littler MM, Littler DS. 1985. Factors controlling relative dominance of primary producers on biotic reefs. In *Proceedings of the Fifth International Coral Reef Congress*, 35-39. Tahiti.

Littler MM, Littler DS. 1994. Tropical reefs as complex habitats for diverse macroalgae. In *Seaweed ecology and physiology,* eds. C. S. Lobban & P. J. Harrison, 72-75. New York: Cambridge University Press.

Lutjeharms JRE. 2006. The coastal oceans of south-eastern Africa. In *The Sea,* eds. A. R. Robinson & K. H. Brink, 781-832. Cambridge, MA: Harvard University Press.

Mann KH. 1973. Seaweeds: their productivity and strategy for growth. *Science* **182**: 975-981.

Mann KH. 1988. Production and use of detritus in various freshwater, estuarine, and coastal marine ecosystems. *Limnology and Oceanography* **33**: 910-930.

Markager S, Sand-Jensen K. 1992. Light requirements and depth zonation of marine macroalgae. *Marine Ecology Progress Series* **88**: 83-92.

Mateo MA, Serrano O, Serrano L, Michener RH. 2008. Effects of sample preparation on stable isotope ratios of carbon and nitrogen in marine invertebrates: implications for food web studies using stable isotopes. *Oecologia* **157**: 105-115.

Mazeroll AI, Montgomery WL. 1998. Daily migrations of a coral reef fish in the Red Sea (Gulf of Aqaba, Israel): initiation and orientation. *Copeia* **1998**: 893-905.

McManus JW. 1997. Tropical marine fisheries and the future of coral reefs: a brief review with emphasis on Southeast Asia. *Coral Reefs* **16, Suppl**: S121-S127.

Menge BA, Daley BA, Wheeler PA, Dahlhoff E, Sanford E, Strub PT. 1997. Benthic-pelagic links and rocky intertidal communities: bottom-up effects on top-down control? *Proceedings of the National Academy of Sciences* **94**: 14530-14535.

Menge BA, Lubchenco J, Gaines SD, Ashkens LR. 1986. A test of the Menge-Sutherland model of community organisation in a tropical rocky intertidal food web. *Oecologia* **71**: 75-89.

Miller RJ, Reed DC, Brzenski MA. 2009. Community structure and productivity of subtidal turf and foliose algal assemblages. *Marine Ecology Progress Series* **388**: 1-11.

Minagawa M, Wada E. 1984. Stepwise enrichment of ^{15}N along food chains: Further evidence and the relation between $\delta^{15}N$ and animal age. *Geochimica et Cosmochimica Acta* **48**: 1135-1140.

Moberg F, Folke C. 1999. Ecological goods and services of coral reef ecosystems. *Ecological Economics* **29**: 215-233.

Mograbi J, Rogerson CM. 2007. Maximising the local pro-poor impacts of dive tourism: Sodwana Bay, South Africa. *Urban Forum* **18**: 85-104.

Montoya JP, Carpenter EJ, Capone DG. 2002. Nitrogen fixation and nitrogen isotope abundances in zooplankton of the oligotrophic North Atlantic. *Limnology and Oceanography* **47**: 1617-1628.

Muñoz AA, Ojeda FP. 1997. Feeding guild structure of a rocky intertidal fish assemblage in central Chile. *Environmental Biology of Fishes* **49**: 471-479.

Müren U, Berglund J, Samuelsson K, Andersson A. 2005. Potential effects of elevated sea-water temperature on pelagic food webs. *Hydrobiologia* **545**: 153-166.

Musick JA. 2000. *Latimeria chalumnae*. In: IUCN 2011. IUCN Red List of Threatened Species. Version 2011.2. www.iucnredlist.org. Downloaded on 08 March 2012.

Nadon M-O, Himmelman JH. 2006. Stable isotopes in subtidal food webs: have enriched carbon ratios in benthic consumers been misinterpreted? *Limnology and Oceanography* **51**: 2828-2836.

O'Reilly CM, Hecky RE, Cohen AS, Plisnier P-D. 2002. Interpreting stable isotopes in food webs: recognising the role of time averaging at different trophic levels. *Limnology and Oceanography* **47**: 306-309.

Odum HT, Odum EP. 1955. Trophic structure and productivity of a windward coral reef community on Eniwetok Atoll. *Ecological Monographs* **25**: 291-320.

Ogden JC, Buckman NS. 1973. Movements, foraging groups, and diurnal migrations of the striped parrotfish *Scarus croicensis* Bloch (Scaridae). *Ecology* **54**: 589-596.

Otto SB, Berlow EL, Rank NE, Smiley J, Brose U. 2008. Predator diversity and identity drive interaction strength and trophic cascades in a food web. *Ecology* **89**: 134-144.

Parnell AC, Inger R, Bearhop S, Jackson AL. 2010. Source partitioning using stable isotopes: coping with too much variation. *PLoS One* **5**: 1-5.

Pauly D, Christensen V. 1995. Primary production required to sustain global fisheries. *Nature* **374**: 255-257.

Peterson BJ, Fry B. 1987. Stable isotopes in ecosystem studies. *Annual Review of Ecology and Systematics* **18**: 293-320.

Pinnegar JK, Polunin NVC, Francour P, Badalamenti F, Chemellow R, Harmelin-Vivien M-L, Hereu B, Milazzo M, Zabala M, D'Anna G, Pipitone C. 2000. Trophic cascades in benthic marine ecosystems: lessons for fisheries and protected-area management. *Environmental Conservation* **27**: 179-200.

Post DM. 2002. Using stable isotopes to estimate trophic position: models, methods, and assumptions. *Ecology* **83**: 703-718.

Post DM, Layman CAA, D A, Takimoto G, Quattrochi J, Montanã CG. 2007. Getting to the fat of the matter: models, methods and assumptions for dealing with lipids in stable isotope analyses. *Oecologia* **152**: 179-189.

Power ME. 1992. Top-down and bottom-up forces in food webs: do plants have primacy? *Ecology* **73**: 733-746.

Ramsay PJ. 1994. Marine geology of the Sodwana Bay shelf, southeast Africa. *Marine Geology* **120**: 225-247.

Ramsay PJ, Mason TR. 1990. Development of a type zoning model for Zululand coral reefs, Sodwana Bay, South Africa. *Journal of Coastal Research* **6**: 829-852.

Ramsay PJ, Miller WR. 2006. Marine geophysical technology used to define coelacanth habitats on the KwaZulu-Natal shelf, South Africa. *South African Journal of Science* **102**: 427-434.

Rau GH, Mearns AJ, Young DR, Olson RJ, Schafer HA, Kaplan IR. 1983. Animal $^{13}C/^{12}C$ correlates with trophic level in pelagic food webs. *Ecology* **64**: 1314-1318.

Raven JA, Walker DI, Johnston AM, Handley LL, Kübler JE. 1995. Implications of ^{13}C natural abundance measurements for photosynthetic performance by marine macrophytes in their natural environment. *Marine Ecology Progress Series* **123**: 193-205.

Richmond MD (ed.). 2002. A Field Guide to the Seashores of Eastern Africa and the Western Indian Ocean Islands. Milan. Sida/SAREC 461pp

Ridderinkhof H, Lutjeharms JRE, de Ruijter WPM. 2001. A research cruise to investigate the Mozambique Current. *South African Journal of Science* **97**: 461-464.

Riegl B, Piller WE. 2003. Possible refugia for reefs in times of environmental stress. *International Journal of Earth Sciences* **92**: 520-531.

Riegl B, Schleyer MH, Cook PJ, Branch GM. 1995. Structure of Africa's southernmost coral communities. *Bulletin of Marine Science* **56**: 676-691.

Roberts MJ, Ribbink AJ, Morris T, van den Berg MA, Engelbrecht DC, Harding RT. 2006. Oceanographic environment of the Sodwana Bay coelacanths (*Latimeria chalumnae*), South Africa. *South African Journal of Science* **102**: 435-443.

Rotjan RD, Lewis SM. 2008. Impact of coral predators on tropical reefs. *Marine Ecology Progress Series* **367**: 73-91.

Rounick JS, Winterbourn MJ. 1986. Stable carbon isotopes and carbon flow in ecosystems. *BioScience* **36**: 171-177.

Rudnick DT. 1989. Time lags between the deposition and meiobenthic assimilation of phytodetritus. *Marine Ecology Progress Series* **50**: 231-240.

Sala E, Ballesteros E. 1997. Partitioning of space and food resources by three fish of the genus *Diplodus* (Sparidae) in a Mediterranean rocky infralittoral ecosystem. *Marine Ecology Progress Series* **152**: 273-283.

Samaai T, Gibbons M, Kerwath S, Yemane D, Sink K. 2010. Sponge richness along a bathymetric gradient within the iSimangaliso Wetland Park, South Africa. *Marine Biodiversity* **40**: 205-217.

Schleyer MH, Celliers L. 2003a. Biodiversity on the marginal coral reefs of South Africa: What does the future hold? *Zoologische Verhandelingen Leiden* **345**: 387-400.

Schleyer MH, Celliers L. 2003b. Coral dominance at the reef-sediment interface in marginal coral communities at Sodwana Bay, South Africa. *Marine and Freshwater Research* **54**: 967-972.

Schleyer MH, Celliers L. 2005. Modelling reef zonation in the Greater St Lucia Wetland Park, South Africa. *Estuarine, Coastal and Shelf Science* **63**: 373-384.

Schleyer MH, Tomalin BJ. 2000. Damage on South African coral reefs and an assessment of their sustainable diving capacity using a fisheries approach. *Bulletin of Marine Science* **67**: 1025-1042.

Schramm W. 1999. Factors influencing seaweed responses to eutrophication: some results from EU-project EUMAC. *Journal of Applied Phycology* **11**: 69-78.

Schumann EH. 1988. Physical oceanography off Natal. In *Coastal ocean studies off Natal, South Africa,* ed. E. H. Schumann, 271. New York: Springer-Verlag.

Sebens KP, Vandersall KS, Savina LA, Graham KR. 1996. Zooplankton capture by two scleractinian corals, *Madracis mirabilis* and *Montastrea cavernosa*, in a field enclosure. *Marine Biology* **127**: 303-317.

Shears NT, Babcock RC. 2002. Marine reserves demonstrate top-down control of community structure on temperate reefs. *Oecologia* **132**: 131-142.

Sink KJ, Boshoff W, Samaai T, Timm PG, Kerwath SE. 2006. Observations of the habitats and biodiversity of the submarine canyons at Sodwana Bay. *South African Journal of Science* **102**: 466-474.

Sink KJ, Branch GM, Harris JM. 2005. Biogeographic patterns in rocky intertidal communities in KwaZulu-Natal, South Africa. *African Journal of Marine Science* **27**: 81-96.

Stephenson RL, Tan FC, Mann KH. 1986. Use of stable carbon isotope ratios to compare plant material and potential consumers in a seagrass bed and a kelp bed in Nova Scotia, Canada. *Marine Ecology Progress Series* **30**: 1-7.

Suchanek TH, Williams SL, Ogden JC, Hubbard DK, Gill IP. 1985. Utilization of shallow-water seagrass detritus by Caribbean deep-sea macrofauna: $\delta^{13}C$ evidence. *Deep Sea Research Part A. Oceanographic Research Papers* **32**: 201-214.

Suring E, Wing SR. 2009. Isotopic turnover rate and fractionation in multiple tissues of red rock lobster (*Jasus edwardsii*) and blue cod (*Parapercis colias*): consequences for ecological studies. *Journal of Experimental Marine Biology and Ecology* **370**: 56-63.

Tamelander T, Renaud PE, Hop H, Carrol ML, Ambrose WJ, Hobson KA. 2006. Trophic relationships and pelagic-benthic couppling during summer in the Barents Sea Marginal Ice

Zone, revealed by stable carbon and nitrogen isotope measurements. *Marine Ecology Progress Series* **310**: 33-46.

Tett P, Gowen R, Mills D, Fernandes T, Gilpin L, Huxham M, Kennington K, Read P, Service M, Wilkinson M, Malcolm S. 2007. Defining and detecting undesirable disturbance in the context of marine eutrophication. *Marine Pollution Bulletin* **55**: 282-297.

Thayer GW, Parker PL, LaCroix MW, Fry B. 1978. The stable carbon isotope ratio of some components of an eelgrass, *Zostera marina*, bed. *Oecologia* **35**: 1-12.

Thresher RE, Nichols PD, Gunn JS, Bruce BD, Furlani DM. 1992. Seagrass detritus as the basis of a coastal planktonic food chain. *Limnology and Oceanography* **37**: 1754-1758.

Tremblay L, Gagné J-P. 2009. Organic matter distribution and reactivity in the waters of a large estuarine system. *Marine Chemistry* **116**.

Uchima M. 1988. Gut content analysis of neritic copepods *Acartia omorii* and *Oithona davisae* by a new method. *Marine Ecology Progress Series* **48**: 93-97.

Uken R, Green AN. 2006. Coelacanth (*Latimeria chalumnae*) resting traces in a cave floor, Chaka Canyon, South Africa. *South African Journal of Science* **102**: 474-475.

UNEP. 2005. Greater St Lucia Wetland Park KwaZulu-Natal, South Africa. 9 pp.

van der Elst R. 1993. *A Guide to the Common Sea Fishes of Southern Africa* Cape Town: Struik Publishers. 398 pp

Vander Zanden MJ, Rasmussen JB. 2001. Variation in $\delta^{15}N$ and $\delta^{13}C$ trophic fractionation: implications for aquatic food web studies. *Limnology and Oceanography* **46**: 2061-2066.

Venter P, Timm PG, Gunn G, le Roux E, Serfontein C, Smith E, Bensch M, Harding D, Heemstra PC. 2000. Discovery of a viable population of coelacanths (*Latimeria chalumnae* Smith, 1939) at Sodwana Bay, South Africa. *South African Journal of Science* **96**: 567-568.

Vetter EW, Dayton PK. 1998. Macrofaunal communities within and adjacent to a detritus-rich submarine canyon system. *Deep Sea Research Part II: Topical Studies in Oceanography* **45**: 25-54.

Viana AR, Faugères JC, Stow DAV. 1998. Bottom-current-controlled sand deposits - a review of modern shallow- to deep-water environments. *Sedimentary Geology* **115**: 53-80.

Vine PJ. 1974. Effects of algal grazing and aggressive behaviour of the fishes *Pomacentrus lividus* and *Acanthurus sohal* on coral-reef ecology. *Marine Biology* **24**: 131-136.

Wada E, Hattori A. 1976. Natural abundance of ^{15}N in particulate organic matter in the North Pacific Ocean. *Geochimica et Cosmochimica Acta* **40**: 249-251.

West JB, Bowen GJ, Cerling TE, Ehleringer JR. 2006. Stable isotopes as one of nature's ecological recorders. *Trends in Ecology and Evolution* **21**: 408-414.

Yahel G, Anton FP, Fabricius K, Marie D, Vaulot D, Genin A. 1998. Phytoplankton distribution and grazing near coral reefs. *Limnology and Oceanography* **43**: 551-563.