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Publications to date

Muhling, B.A. and L.E. Beckley (submitted). Seasonal variation in horizontal and vertical structure of larval fish assemblages off south-western Australia, with implications for larval transport. (Submitted to *Journal of Plankton Research*)

Muhling, B.A, L.E. Beckley, J.A. Koslow and A.F. Pearce (in press). Larval fish assemblages and water mass structure off the oligotrophic south-western Australian coast. *Fisheries Oceanography.*

Muhling, B.A., L.E. Beckley and M.P. Olivar (submitted). Ichthyoplankton assemblage structure in two meso-scale Leeuwin Current eddies. (Resubmitted after review to *Deep Sea Research II* - special issue).

Waite, A.M., P.A. Thompson, S. Pesant, M. Feng, L.E. Beckley, C. Domingues, D. Gaughan, C. Hanson, C. Holl, J.A. Koslow, M. Meuleners, J. Montoya, T. Moore, B.A. Muhling, H. Paterson, S. Rennie, J. Strzelecki and L. Twomey (submitted). The Leeuwin Current and its Eddies: An Introductory Overview. (Resubmitted after review to *Deep Sea Research II* - special issue)

Waite, A.M., B.M. Muhling, C. Holl, L.E. Beckley, J. Montoya, J. Strzelecki, P.A. Thompson and S. Pesant (submitted). Food web structure in two counter-rotating eddies based on d ^{15}N and d ^{13}C isotopic analyses. (Resubmitted after review to *Deep Sea Research II* - special issue)

2.2.9 Microzooplankton off south west Western Australia: Their spatial and temporal variations and impact of phytoplankton

Investigator / Institution

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Executive Summary

Seasonal and spatial variability in the microzooplankton assemblage off south western Western Australia was evident. The microzooplankton formed an important component of the planktonic food web in this region, consuming ~60% of the primary productivity. Their biomass was greatest in winter, responding to increases in prey availability. Their diversity was limited inshore, possibly due to the highly variable environment encountered. Diversity increased with distance offshore as physical and chemical properties in addition to prey availability and predation become more stable. Strictly heterotrophic cells were evenly distributed between inshore and offshore. Those organisms harbouring endosymbionts had a higher biomass offshore where basic metabolic costs are covered by photosynthesis during periods of low prey availability. Cells that are primarily photosynthetic, that may phagocytize particles were favoured inshore where they survived low nutrient environments more efficiently than do strictly autotrophic cells.

The grazing impact exerted by microzooplankton on phytoplankton was generally balanced in winter; however in summer phytoplankton growth rates tended to exceed grazing rates, effectively decoupling these rates. Inshore, the sporadic availability of diatoms caused this decoupling, as these phytoplankton are often too large to be consumed by microzooplankton. The mechanisms involved offshore in decoupling between phytoplankton growth and grazing losses during summer were more complex. Microzooplankton appeared to graze at reduced rates, effectively maintaining the low phytoplankton biomass just above threshold feeding levels.

Introduction

Microzooplankton has been studied since the mid 1980s when their important function in the planktonic food chain was identified. Since this time they have been studied extensively in the Atlantic, Pacific and Antarctic regions, as well as in other, well-studied water bodies. Microzooplankton repackage carbon from small pico (<2 µm) sized phytoplankton cells to micro (20 – 200 µm) sized particles which are available to larger consumers. Unravelling the microbial food web is fundamental to understanding how the system works.

The region off the west coast of Australia represents the only pole-wards flowing eastern boundary current in the world, resulting in an oligotrophic region with comparatively low productivity and a different food web compared to other eastern boundary currents (Pearce 1991). Studying microzooplankton in this region, allows clarification of the role of microzooplankton in a temperate, oligotrophic waters in a coastal environment.

Aims and objectives

The objectives of this study were to:

1 Characterize the microzooplankton assemblage off south western WA and their temporal and spatial variability.

2 Quantitatively assess the herbivory of microzooplankton on phytoplankton, and its temporal and spatial variability.

3 Examine the distribution of three functionally different groups of micro-zooplankton on the Two Rocks transect.

Methods

Microzooplankton biomass, abundance and species richness were analysed from 190 samples of ~850 mL of seawater fixed with Lugol's iodine. These were routinely collected from the Two Rocks transect (defined below), using the microscopy method of Utermöhl (1958). Cells were identified using the methods described in Marshall (1969), Thomas (1997) and Boltovskoy (1999). Each cell was classified into three functional groups, HETERO for strictly heterotrophic cells, ENDO for cells with endosymbionts and MIXO for primarily autotrophic cells that may phagocytize particles. The biomass for each group was compared between inshore (station A and B) and offshore (stations C, D and E). The grazing impact of microzooplankton on phytoplankton was evaluated using the dilution method (Landry and Hassett 1982) with changes in chlorophyll *a* and picoplankton measured by flow-cytometry to quantify changes to the phytoplankton community. The computer packages SPSS and PRIMER were used to analyse the data.

Study site

During this study, five sites along a transect running from 4 km south west of Two Rocks, Western Australia, to 85 km offshore (See Fig. 2.32), were sampled from February 2002 – December 2004 (31.5 - 31.8˚S, 115.6 – 114.9˚E). The sites represented the coastal lagoon (A, water depth 15 m), inner shelf (B, 40 m), outer shelf (C, 100 m), shelf break (D, 300 m) and offshore (E, 1000 m) habitats. Two sampling strategies were followed: inshore stations A, B and C were sampled monthly, weather permitting, using the 8 m *RV Mesocat*; stations A – E were sampled quarterly from larger vessels, *RV Naturaliste*, *RV Maritime Image* or the *RV Southern Surveyor*. Samples for microzooplankton were routinely collected from the surface and deep chlorophyll *a* maximum (DCM) using 10 L Niskin bottles. Water was collected from the surface only from station A, C and E during quarterly cruises for microzooplankton grazing experiments.

Results

There were 157 morphotypes of microzooplankton discriminated from 190 samples. The major groups were ciliates and heterotrophic dinoflagellates, although nauplii, acantharians, radiolarians and foraminiferans were also found. Of the ciliates, *Strombidium* sp. and *Strobilidium* sp. were the most dominant forms and 19 genera of tintinnids were found. *Protoperidinium* sp. and *Gyrodinium*/*Gymnodinium* sp. were the most abundant dinoflagellates, although *Ceratium furca* was an important contributor to dinoflagellate blooms inshore. The average abundance of microzooplankton in the surface was 2400 cells L^{-1} with a biomass of 2.9 μg C L^{-1}, and in the DCM the average abundance was 2100 cells L^{-1} and the average biomass was 2.1 μg C L^{-1} (Fig. 2.36). There was a significant increase in microzooplankton biomass in winter (p=0.002), but the peak biomass of 15 μg C L^{-1} was in autumn, and caused by a *C. furca* bloom.

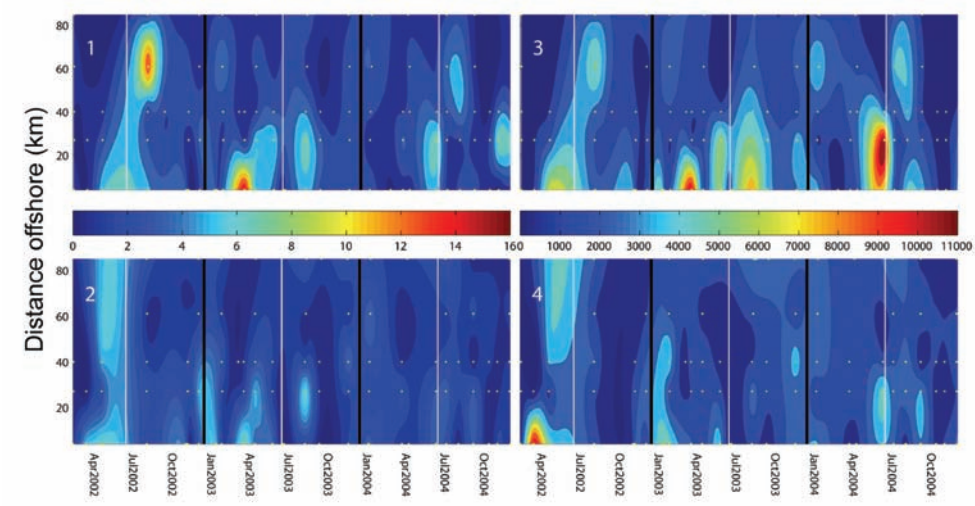

Figure 2.36: Biomass and abundance of microzooplankton from the Two Rocks (WA) transect, 2002-04. Panels 1 and 2 represent microzooplankton carbon μg C L^{-1}; surface (1), deep chlorophyll *a* maximum (2). Panels 3 and 4 represent microzooplankton cell abundance L^{-1}; surface (3), deep chlorophyll *a* maximum (4).

Species richness increased significantly with distance offshore ($F_{(4, 173)}$ =23, p=0.000), winter was significantly richer ($F_{(3, 173)}$ = 4 p=0.009) than in other seasons, across the transect; and samples from the DCM were richer than those from the surface ($F_{(1, 173)}$ =5.1 p=0.024). The variability between samples, as measured by the Multivariate Dispersion Index, for each site, decreased with distance offshore ($F_{(1, 4)}$ = 19, p=0.02) (Fig. 2.37A).

The three functional groups differed in their distribution between inshore and offshore. The HETERO group had no difference in distribution. The ENDO group had a significantly greater biomass offshore ($t_{(1, 173)}$ = -1.9, p=0.056), while the MIXO group had a significantly greater biomass inshore ($t_{(1,173)}$ =3.9, p<0.01).

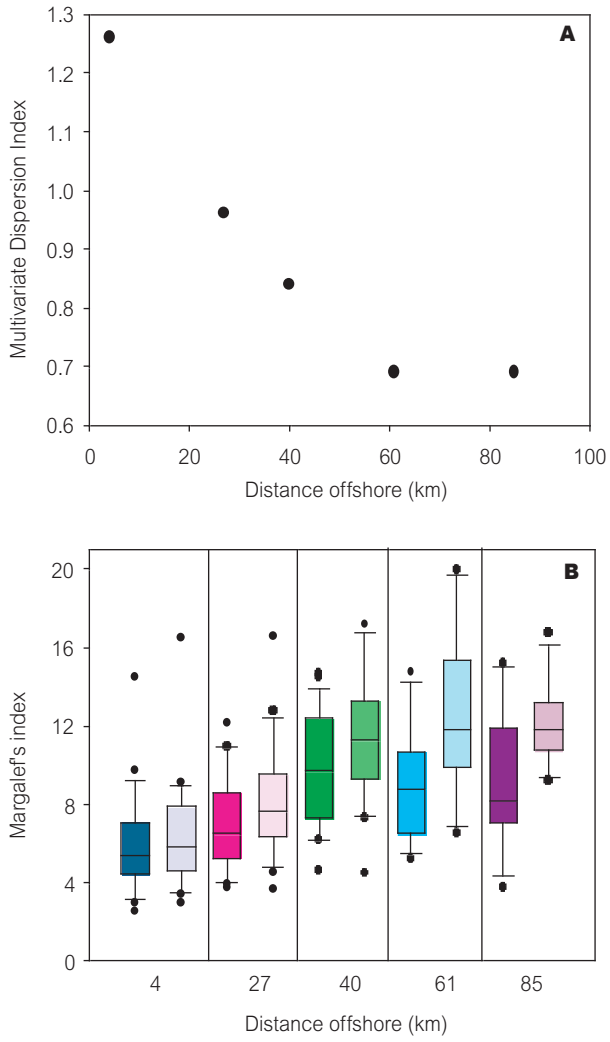

Figure 2.37: Species diversity: Panel A, Multivariate dispersion Index for microzooplankton, calculated from scatter of points from Non-metric multi dimension scaling plot of square root transformed microzooplankton biomass data. Panel B, Distribution of Margalef's index of species richness for each station, top and bottom of box represent the 75th and 25th percentiles, wiskers represent the 90th and 10th percentiles. Dark colour surface data, light colour deep chlorophyll *a* maxima data. From April 2003 to December 2004, from the Two Rocks transect Western Australia.

The relationship between phytoplankton growth and microzooplankton grazing differs between summer and other seasons (Fig. 2.38). During the summer the phytoplankton biomass was low, although its apparent growth rate was high. At the offshore station the grazing rate was also high during summer, effectively maintaining the low phytoplankton biomass. In the lagoon and outer shelf the grazing rate did not rise in accordance with the phytoplankton growth rate, resulting in decoupling. Most results showed linear grazing responses, however saturation feeding was seen in results from the outer shelf and offshore during summer, and threshold feeding was evident in some experiments during the year, mostly in the lagoon and outer shelf. There is also evidence of saturated feeding and a threshold response by microzooplankton throughout the year. Microzooplankton grazing on total phytoplankton production and standing stocks in the lagoon ranged from 33-55 and 69-89% in summer and 68-96% and 48-56% in winter, respectively, and at the two offshore stations 29 – 91% and 60 – 82% in summer and 26-85% and 39-55% in winter, respectively. Picoplankton growth was generally balanced by microzooplankton grazing (Fig. 2.38). Heterotrophic bacteria and *Prochlorococcus* spp. had the highest growth and grazing rates (2 – 2.5 d^{-1}) and cryptophyte 1 the lowest (0.5 – 1 d^{-1}). Rates on *Synechococcus* sp. and the pico-eukaryotes were varied, with grazing occasionally exceeding growth.

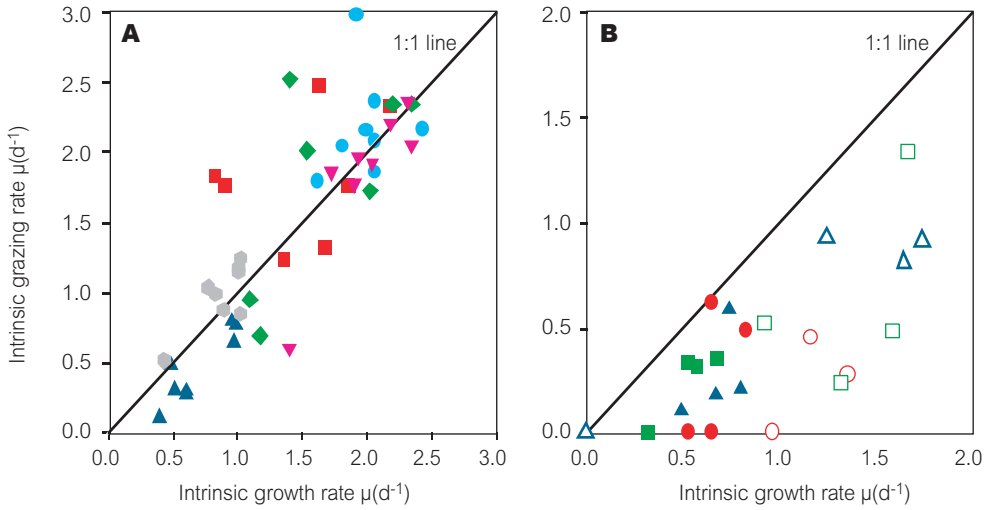

Figure 2.38: Relationship between intrinsic growth rates and mortality due to microzooplankton grazing. Line is the 1 to 1 relationship between growth and grazing.

Panel A: growth and grazing for six pico-phytoplankton populations;

- Prochlorococcus spp.
- Synechococcus spp.
- Cryptophyte 1
- Cryptophyte 2
- Pico-eukaryotes
- Heterotrophic bacteria

Panel B: growth and grazing determined from total chlorophyll *a*;

- Lagoon
- outer shelf
- offshore

Open symbols summer (December – February), closed symbols all other seasons (March – November).

Discussion

The Two Rocks transect may be divided into 3 zones based on the abundance and biomass of microzooplankton data. The lagoon was influenced by near-shore processes; the inner shelf was influenced (to some degree) by near shore processes, the summer Capes Current and also periodically by the Leeuwin Current; and the outer shelf, shelf break and 1000 m stations were dominated by the Leeuwin Current, with some influence from the Capes Current during summer at the outer shelf; the offshore station was occasionally influenced by oceanic water. Increases in microzooplankton biomass during winter presumably represent responses to increases in phytoplankton biomass and productivity that result from new nutrient being introduced into the photic zone during autumn and winter when the mixed layer deepens.

The physical and chemical differences between the lagoon and offshore station explain some of the differences between these regions. In the lagoon, bottom-up forces of temperature and salinity changes, combined with episodic nutrient inputs, resulted in low species richness and highly variable abundance and biomass, and in rapid turnover of the dominant microzooplankton species. The offshore stations provide a comparatively stable environment where nutrient limitation caused consistent microzooplankton abundance and biomass, and where prey availability, niche diversification and selective grazing by predators resulted in a relatively diverse community.

The distribution of the three functional groups may be explained by the advantages gained by the different modes. The strictly heterotrophic organism gained no advantage inshore or offshore demonstrating their viability and diversity. The ENDO group may cover their metabolic costs by harvesting photosynthetic products directly from their endosymbionts. This gives them a competitive advantage during periods of low prey availability. The MIXO group are similar to strict autotrophs, hence their higher biomass inshore, however they have a greater advantage during periods of low nutrient availability as they may consume particles and assimilate nutrients directly.

The grazing impact of microzooplankton on total phytoplankton off south western Western Australia differs between summer and the rest of the year. High phytoplankton growth rates in the lagoon during summer were not coupled with grazing. Large diatoms, known to bloom sporadically in the lagoon during summer, may have been the cause as they are not preferentially grazed on by microzooplankton. Offshore, the likelihood of saturation feeding occurring during summer, when phytoplankton biomass is low, suggests that microzooplankton are capable of reducing feeding rates, which presumable prevent the phytoplankton biomass being grazed to threshold levels where microzooplankton cease to feed.

The impact of microzooplankton grazing on specific picoplankton populations is generally balanced between seasons. However, while growth and grazing of picoplankton is balanced, growth and grazing on total phytoplankton is not balanced. The larger phytoplankton cells that contribute to this imbalance are likely to be grazed by mesozooplankton, which are equipped to consume phytoplankton cell.

Microzooplankton constitute an important component of the planktonic assemblage off south western, Western Australia. Their impact on phytoplankton varies seasonally, and their diversity increases with distance offshore.

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References

Boltovskoy, D. (1999). South Atlantic Zooplankton. Backhuys Publishers, Leiden.

Landry, M. R, Hassett, R. P. (1982). Estimating the Grazing Impact of Marine Micro-zooplankton. *Marine Biology* 67: 283-288.

Marshall, S. M., (1969) Tintinnida. *Conseil International pour l'exploration de la mer.*

Pearce, A. F. (1991) Eastern boundary currents of the southern hemisphere. Journal of the Royal Society of Western Australia 74: 34-45.

Thomas, C. R. (1997). Identifying marine phytoplankton. Academic Press. San Diego, California.

Utermöhl, H. (1958). Zur Vervollkommnung der quantitativen phytoplankton-methodik. Internationale Vereinigung fuer Theoretische und Angewandte Limnologie. Mitteilungen 9:1-38.

Conference attendance and presentations

Award

SRFME Student Symposium 2003, Best abstract
SRFME Student Symposium 2004, 2nd presentation

Symposium

Third International Symposium on Zooplankton, Spain, May 2003
SRFME Student Symposium 2003
SRFME Student Symposium 2004
AMSA 2004
SRFME Student Symposium 2005
CMM seminar series 2005

PhD awarded in December 2006

Publications

The Nanostructural Network Analysis Organisation (NANO) Major National Research Facilty Annual Report. p12.

Paterson, H. L. and B. Knott (in prep.). The role of mixotrophy/autotrophy ratio in contrasting inshore/offshore environments: in the Indian Ocean off south Western Australia. (Intended for *Journal of Plankton Research*)

Paterson, H. L., B. Knott and A. Waite (submitted). Microzooplankton community structure, and herbivory on phytoplankton, in an eddy pair in the Indian Ocean off Western Australia. (Resubmitted after review to *Deep-Sea Research II*)

Paterson, H. L. and J.A. Koslow (in prep.). Microzooplankton: Biomass, abundance and composition covering lagoon, shelf and shelf break (1000 m deep) waters of temperate south western Australia, 2002 to 2004. (Intended for *Journal of Plankton Research*)

Paterson, H. L., S. Pesant, P. Clode, B. Knott and A. Waite (submitted). Systematics of a rare radiolarian - *Coelodiceras spinosum* Haecker (Sarcodina: Actinopoda: Phaeodaria: Coelodendridae). (Resubmitted after review to *Deep-Sea Research II*)

2.2.10 Diurnal variations in physical processes & phytoplankton response

Investigator / Institution

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Supervisors

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Executive Summary

Oligotrophy, high irradiance and consistent diurnal cycles in water column stratification and mixing in the coastal waters offshore Perth, Western Australia presented an opportunity to study natural phytoplankton community responses to an interesting combination of physical processes. This research aimed to determine the phytoplankton response for varying strengths of water column stratification and irradiance at sites with different exposures. We estimated the phytoplankton community response through examination of vertical chlorophyll *a* concentration variability and distribution on an hourly timescale, in vivo fluorescence fluctuations, *in situ* primary production experiments and measurements of photosystem II efficiency (yield). A combination of air–sea heat fluxes and wind stress control the vertical stratification strength, measured through the potential energy anomaly. In general, the water column is well mixed in the morning because of night-time cooling and convection and stratifies during the day in response to solar heating. Our field study showed that chlorophyll *a* concentration and in vivo fluorescence (which were 48 % correlated) were both lowest in the surface (1-5 m) during prolonged (hours) periods of stratification when the irradiance experienced was highest and phytoplankton were retained in the surface layer due to an absence of vertical mixing (generally around midday). Short-term *in situ* primary production rates (measured through 1-hour carbon uptake incubations and normalised to chlorophyll *a*