## Microbial Ecology and Biogeography

OF THE

## Southern Ocean

David Wilkins

September 22, 2012

## **Contents**

List of Figures	iii
List of Tables	v
List of Acronyms	vii
Acknowledgements	ix
Abstract	xi
Introduction	1
Microbial ecology of the Southern Ocean	1
Oceanography of the Southern Ocean	1
Water masses and fronts	1
Effect of climate change	1
Role of the Polar Front in biogeography	1
Project questions and hypotheses	1
The Polar Front as a major biogeographic boundary in the Southern Ocean	3
Summary	3
Introduction	3
Methods	3
Sampling and metagenomic sequencing	3
Phylogenetic analysis of metagenomic data	6
Functional analysis of metagenomic data	6
Results	6
Metagenomic sequencing	6
Phylogenetic analysis of metagenomic data	6
Functional analysis of metagenomic data	6
Discussion	6
Conclusions	6
Meso-scale biogeographic drivers of planktonic diversity	7
Conclusions	9

## **List of Figures**

1	Map showing site	es of seawater samples u	sed in the Polar Front study	4
---	------------------	--------------------------	------------------------------	---

## **List of Tables**

1	Details of samples used in Polar Front study	7.																			5
---	--	----	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	---

## Acronyms

AZ Antarctic Zone.

**CEAMARC/CASO** Collaborative East Antarctic Marine Census/Climate of Southern Ocean.

CTD Conductivity, Temperature and Depth.

NZ North Zone.

**OTU** Operational Taxonomic Unit.

**PF** Polar Front.

**PFZ** Polar Frontal Zone.

SZ South Zone.

**UFO** Unidentified Flying Object.



## Acknowledgements



## **Abstract**

## Introduction

This is a test of the acronyms: I saw a Unidentified Flying Object (UFO). It was not the first UFO I'd ever seen. In fact, I've seen 100 UFOs.

Here is some greek: µg.

Microbial ecology of the Southern Ocean

Oceanography of the Southern Ocean

Water masses and fronts

Effect of climate change

Role of the Polar Front in biogeography

Project questions and hypotheses

# The Polar Front as a major biogeographic boundary in the Southern Ocean

Sections of this chapter have been previously published in Wilkins D., Lauro F. M., Williams T. J., Demaere M. Z., Brown M. V., Hoffman J. M., Andrews-Pfannkoch C., Mcquaid J. B., Riddle M. J., Rintoul S. R., and Cavicchioli R. Biogeographic partitioning of Southern Ocean picoplankton revealed by metagenomics. *Molecular Ecology*, 2012.

#### Summary

#### Introduction

#### Methods

#### Sampling and metagenomic sequencing

Sampling<sup>1</sup> was conducted on board the RSV *Aurora Australis* during cruise V3 Collaborative East Antarctic Marine Census/Climate of Southern Ocean (CEAMARC/CASO) from 13 December 2007 – 26 January 2008. This cruise occupied the SR3 latitudinal transect from Hobart, Australia (44° S) to the Mertz Glacier, Antarctica (67° S) within a longitudinal range of 140–150° E. Nineteen samples (16 surface, 3 deep) were obtained along almost the entire latitudinal range (Figure 1).

A range of data were recorded by integrated instruments on the RSV *Aurora Australis* including location, water column depth, water temperature, salinity, fluorescence and meterological data (Table 1). These data were used to locate the Polar Frontal Zone (PFZ) based on a surface temperature gradient of  $\sim 1.35$  °C across a distance of 45–65 km, placing the Polar Front (PF) at approximately  $-59.70^{\circ}$  of latitude, consistant with previous descriptions (Moore *et al.*, 1999; Sokolov and Rintoul, 2002). Samples were accordingly grouped into "North" and "South" zones, while the three deep samples composed a "Deep" zone (Table 1). The North Zone (NZ) represents waters from the Subtropical, Subantarctic and PFZ regions, while the South Zone (SZ) represents the Antarctic Zone (AZ).

At each station,  $\sim$  250–560 L of seawater was pumped from  $\sim$  1.5–2.5 m below the sea surface into drums stored at ambient temperature on deck. In the case of deep samples,  $\sim$  225–230 L of seawater was collected from Niskin bottles attached to a CTD (SeaBird, Bellevue, USA). Seawater samples were

<sup>&</sup>lt;sup>1</sup>Sampling was performed by Jeffrey M. Hoffman and Jeffrey B. Mcquaid

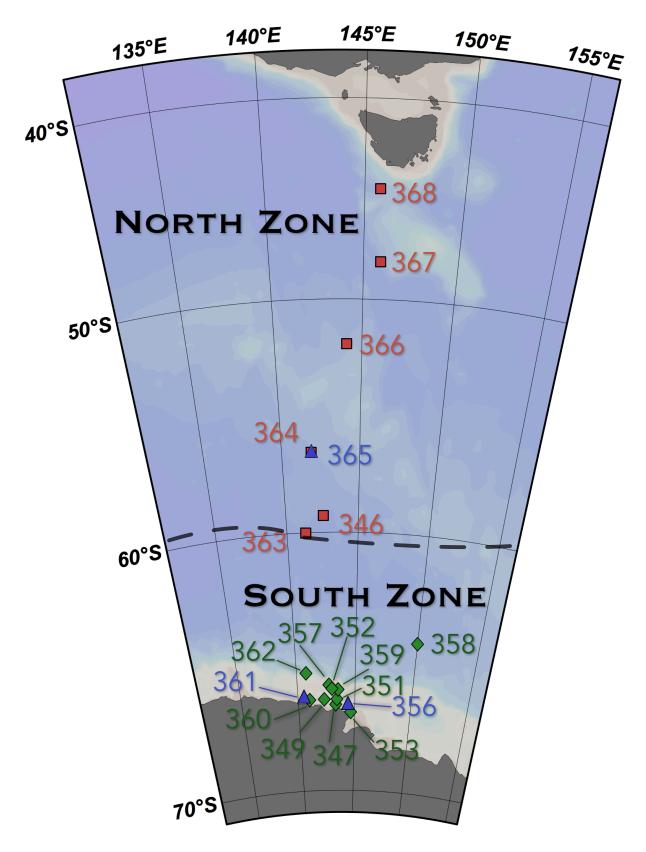


Figure 1: Sites of seawater samples used in this study. Red squares indicate surface samples from the North Zone; green diamonds samples from the South Zone; and blue triangles indicate deep samples. The dashed line gives the approximate location of the Polar Front.

Table 1: Sampling time, location and physiochemical properties of samples used in this study. All data were retrieved from underway instruments aboard the RSV Aurora Australis, with the exception of temperature, salinity and fluorescence data for the three deep samples, which was obtained from

Sample	Zone	Local Time	Latitude	Longitude	Water	Sample	Water	Salinity (PSU)	Fluorescence $(\mu g/1)$	Volume
ı				)	Column	Depth (m)	Tempera-	•		seawater
					Depth (m)		ture (°C)			filtered (L)
346	North	20/12/07	-59.312	142.5949	4294	2	2.9	33.75	0.25	500
347	South	23/12/07	-66.0213	142.738	450	2	9.0	34.2	4	250
349	South	27/12/07	-66.5662	142.3169	370	1.5	-1.3	34.4	2.3	250
351	South	28/12/07	-66.5587	143.4303	823	1.5	-0.55	34.3	1.3	200
352	South	29/12/07	-66.765	143.324	164	2.5	-0.75	34.3	3.1	200
353	South	30/12/07	-67.0521	144.6786	180	2	-1.8	34.4	0.3	200
356	Deep	03/01/08	-66.7617	144.4138	920	920	-1.89	34.69	0.1	230
357	South	05/01/08	-66.1719	143.0193	580	2	-0.4	34.15	2.5	200
358	South	09/01/08	-64.3001	150.0306	3550	2	0	33.55	0.45	200
359	South	12/01/08	-66.1903	143.5292	540	2	-0.24	34.21	2.5	200
360	South	13/01/08	-66.5817	141.0211	316	2	-0.65	34.04	6.19	200
361	Deep	14/01/08	-66.4727	140.5572	1203	1170	-1.77	34.56	0.1	225
362	South	19/01/08	-65.5367	140.8287	1064	2	0.7	32.2	0.5	200
363	North	22/01/08	-60.0001	141.3094	4473	2	3.3	33.77	0.1	500
364	North	23/01/08	-56.6953	141.878	3693	2	4	33.7	0.5	200
365	Deep	23/01/08	-56.6967	141.9125	3693	3693	0.48	34.69	0.1	230
366	North	24/01/08	-52.0233	144.1362	3180	2	7.6	33.84	0.25	200
367	North	25/01/08	-48.2487	145.9025	3490	2	11	34.43	0.2	200
368	North	26/01/08	-44.718	145.7775	3201	2	14.75	34.96	1.25	260
								1		

prefiltered through a 20  $\mu$ m plankton net, then filtrate was captured on sequential 3.0  $\mu$ m, 0.8  $\mu$ m and 0.1  $\mu$ m polyethersulfone membrane filters (Port Washington, USA), and immediately stored at -20 °C (Rusch *et al.*, 2007; Ng *et al.*, 2010).

DNA extraction<sup>2</sup> was performed at the J. Craig Venter Institute (Rockville, USA) as described in Rusch *et al.* (2007). Pyrosequencing was performed on a GS20 FLX Titanium instrument (Roche, Branford, USA) also at the J. Craig Venter Institute as described in Lauro *et al.* (2011). Duplicate reads and reads with many pyrosequencing errors were removed as described in Lauro *et al.* (2011).

#### Phylogenetic analysis of metagenomic data

A subset of the RefSeq microbial (bacterial and archaeal) genome database (release 41, retrieved May 31 2012 from ftp://ftp.ncbi.nih.gov/refseq/release/) was prepared by excluding sequences with the words "shotgun", "contig", "partial", "end" or "part" in their headers (Angly et al., 2009). Because this database was not expected to contain representative genomes for every species present, Operational Taxonomic Units (OTUs) in this study are defined by the best species match to this database, and may for example represent congeners.

The metagenomic reads from each sample were compared against this database using TBLASTX, with default parameters except for: E-value threshold  $1.0 \times 10^{-3}$ , cost to open gap 11, cost to extend gap 1, masking of query sequence by SEG masking with lookup table only.

TODO HERE

#### Functional analysis of metagenomic data

#### Results

Metagenomic sequencing

Phylogenetic analysis of metagenomic data

Functional analysis of metagenomic data

Discussion

**Conclusions** 

<sup>&</sup>lt;sup>2</sup>DNA extraction was performed by Cynthia Andrews-Pfannkoch and others at the J. Craig Venter Institute

## Meso-scale biogeographic drivers of planktonic diversity

## **Conclusions**

## References

- Angly F. E., Willner D., Prieto-Davó A., Edwards R. A., Schmieder R., Vega-Thurber R., Antonopoulos D. A., Barott K., Cottrell M. T., Desnues C., Dinsdale E. A., Furlan M., Haynes M., Henn M. R., Hu Y., Kirchman D. L., McDole T., McPherson J. D., Meyer F., Miller R. M., Mundt E., Naviaux R. K., Rodriguez-Mueller B., Stevens R., Wegley L., Zhang L., Zhu B., and Rohwer F. The GAAS Metagenomic Tool and Its Estimations of Viral and Microbial Average Genome Size in Four Major Biomes. *PLoS Computational Biology*, 5(12):e1000593, 2009.
- Lauro F. M., Demaere M. Z., Yau S., Brown M. V., Ng C., Wilkins D., Raftery M. J., Gibson J. A., Andrews-Pfannkoch C., Lewis M., Hoffman J. M., Thomas T., and Cavicchioli R. An integrative study of a meromictic lake ecosystem in Antarctica. *The ISME journal*, 5(5):879–895, 2011.
- Moore J. K., Abbott M. R., and Richman J. G. Location and dynamics of the Antarctic Polar Front from satellite sea surface temperature data. *Journal of Geophysical Research*, 104:3052–3073, 1999.
- Ng C., Demaere M. Z., Williams T. J., Lauro F. M., Raftery M., Gibson J. A., Andrews-Pfannkoch C., Lewis M., Hoffman J. M., Thomas T., and Cavicchioli R. Metaproteogenomic analysis of a dominant green sulfur bacterium from Ace Lake, Antarctica. *The ISME journal*, 4(8):1002–1019, 2010.
- Rusch D. B., Halpern A. L., Sutton G., Heidelberg K. B., Williamson S., Yooseph S., Wu D., Eisen J. A., Hoffman J. M., Remington K., Beeson K., Tran B., Smith H., Baden-Tillson H., Stewart C., Thorpe J., Freeman J., Andrews-Pfannkoch C., Venter J. E., Li K., Kravitz S., Heidelberg J. F., Utterback T., Rogers Y.-H., Falcón L. I., Souza V., Bonilla-Rosso G., Eguiarte L. E., Karl D. M., Sathyendranath S., Platt T., Bermingham E., Gallardo V., Tamayo-Castillo G., Ferrari M. R., Strausberg R. L., Nealson K., Friedman R., Frazier M., and Venter J. C. The Sorcerer II Global Ocean Sampling expedition: northwest Atlantic through eastern tropical Pacific. *PLoS Biology*, 5(3):e77–e77, 2007.
- Sokolov S. and Rintoul S. R. Structure of Southern Ocean fronts at 140°E. *Journal of Marine Systems*, 37 (1):151–184, 2002.
- Wilkins D., Lauro F. M., Williams T. J., Demaere M. Z., Brown M. V., Hoffman J. M., Andrews-Pfannkoch C., Mcquaid J. B., Riddle M. J., Rintoul S. R., and Cavicchioli R. Biogeographic partitioning of Southern Ocean picoplankton revealed by metagenomics. *Molecular Ecology*, 2012.