

Isoscapes Development ($\delta^{13}\text{C}$ and $\delta^{15}\text{N}$) in the Golfo Nuevo and Golfo San José: A Baseline for the Study of the Marine Pelagic Trophic Web



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Index

Index	2
Introduction	3
Objective	4
Material and Methods	5
Field sampling	5
Laboratory processing.....	5
Stable isotopes analysis	5
Results and Discussion.....	6
Acknowledgments	7
Bibliography.....	7

Introduction

Stable isotopes ecology is a modern tool to, for example, understand trophic food ecology involving diet, trophic relationships, competition, niche, habitat and resource partitioning and migration patterns (Fry 2006). However, abiotic factors such as temperature, salinity, dissolve CO₂, nutrients available in the ocean realm, among others, can produce effects on the stable isotopes fractionation (Fry 2006), besides the physiological approach (Schoeller 1999). Isoscapes or *isotopic landscapes* are a type of maps made from stable isotopes distribution in certain time and space (Bowen 2010; West et al. 2010). The first application of isoscapes were to understand the migration of birds and monarch butterflies in North America through $\delta^2\text{H}$ and $\delta^{18}\text{O}$ in rain and inert tissue, feathers and wings respectively (Hobson 1999). Every day isoscapes based on $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ are being published, showing local variations and seasonality. $\delta^{13}\text{C}$ is commonly known to be used as a determinant of organic matter origin, which in the case of the marine environment tells about inshore/benthic or offshore/pelagic origin (Rubenstein and Hobson 2004). On the other hand, $\delta^{15}\text{N}$ gives an approximation of trophic position (TP) in a marine trophic food web (trophic level in a marine food chain), being arbitrary determined TP = 1 of phytoplankton as a starter for the baseline (Post 2002; Caut et al. 2009).

It is well known that the organisms that are part of the pelagic baseline (i.e.: particulate organic matter, phytoplankton, zooplankton) can affect directly through prey items to top predators (i.e.: dolphins, marine birds, sea lions, seals) or to their own predators (i.e.: baleen whales) as stable isotopes can be tracked through the trophic food with the corresponding trophic discrimination/enrichment (TDF/TEF) factor applied (Post 2002).

The Golfo Nuevo (GN) and the Golfo San José (GSJ), both located in the Chubut province, Argentina (Fig. 1) constitute a great case study to see the differences on how $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ isotopes vary in space and time. Both are enclosed basins with a mouth to open ocean but, Golfo Nuevo shows an anticyclonic circulation starting from the left southern coast (Rivas and Ripa 1989; Tonini et al. 2022); Golfo San José is divided in 2 dominions: West and East, the first one showing cyclonic gyres with plumes injections from the Golfo San Matías in the north while the other dominion shows an anticyclonic gyres with steady waters. There are variations in

their salinity and surface sea water around the year (Rivas 1990; Amoroso and Gagliardini 2010; Tonini et al. 2022).

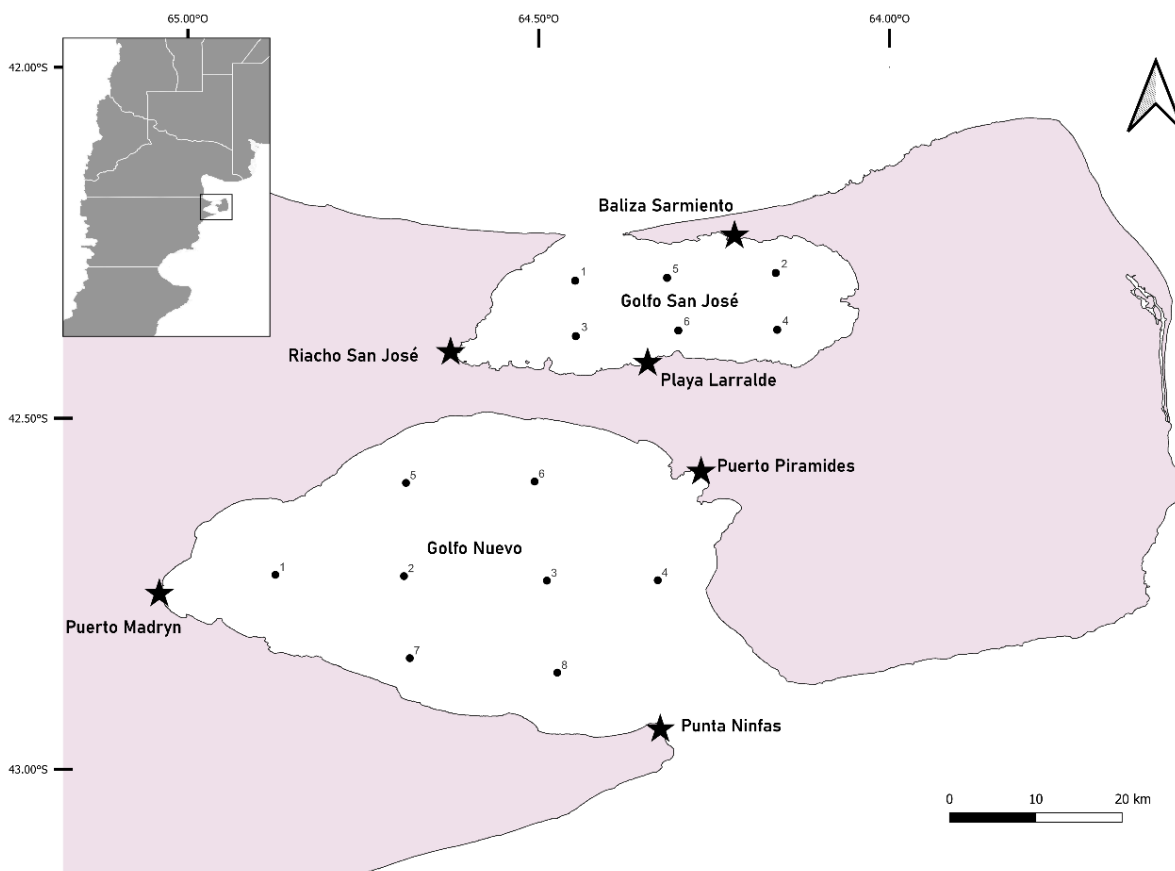


Figure 1: Golfo Nuevo and Golfo San José sampling stations as showed by black dots. Stars show places of important location places.

Objective

Use phyto- and zooplankton and particulate organic matter (POM) stable isotopes ($\delta^{13}\text{C}$ and $\delta^{15}\text{N}$) to characterize two marine-coastal ecosystems with different oceanographic features where different human activities are taking place. These areas are the GN and GSJ located in Argentina. Understanding in detail the matter and energy flow through both gulfs trophic webs, registering animals' movement to identify important areas for species conservation and management, will lead us in a proper way to take better care of the environment.

Material and Methods

Field sampling

1. Boat surveys were made through a hole year (Fall, Winter and Spring 2022; Summer 2023) in the GN and GSJ.
2. For POM, water samples using a Niskin Bottle were taken in a 5L plastic container.
3. For phytoplankton, a special net of 20 μm mesh was towed for 7 minutes at 2 knots at the subsurface horizontally twice: one for stable isotopes and other one to be stored in formaldehyde to characterize the previous one.
4. For zooplankton, a special net of 333 μm mesh was towed for 7 minutes at 2 knots at 30mts of depth obliquely.

Laboratory processing

1. POM samples were filtered through pre-combusted 47 mm diameter filters of 0.7 μm of pore.
2. Phytoplankton samples were filtered through pre-combusted 47 mm diameter filters of 0.7 μm of pore. Fixed samples were characterized using a Leica microscope at a group taxonomic level.
3. Zooplankton samples were filtered through pre-combusted 47 mm diameter filters of 0.7 μm of pore.

Stable isotopes analysis

1. Lipid extraction was carried out to all the zooplankton samples with several rinses of a 2:1 mixture of chloroform and methanol every 24hs during 3 days. Phytoplankton and POM samples were not treated.
2. Finally, samples were freeze dried at -40°C for 24hs.
3. Nitrogen and carbon ratios were measured with approximately 1.0-1.2 mg of zooplankton and 6.0-6.2 mg of phytoplankton and POM using a continuous-flow isotope ratio mass spectrometer connected to an elemental analyzer (EA-IRMS) at the University of New Mexico (UNM), Center for Stable Isotopes (CSI).

4. Results are reported in δ notation in per mil units (‰) based on PeeDee Belemnite and atmospheric N₂ as internationally accepted standards for $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$, respectively:

$$\delta X = (R_{\text{sample}} / R_{\text{standard}} - 1) \times 1000$$

where R represents the relationship between heavy, and light isotope ($^{13}\text{C}/^{12}\text{C}$ or $^{15}\text{N}/^{14}\text{N}$) for samples and standards, and X is ^{13}C or ^{15}N . International isotope secondary standards given by the International Atomic Energy Agency were used to calibrate estimates of nitrogen and carbon at a precision of 0.30 and 0.20‰, respectively.

Results and Discussion

Table 1: max, min and mean values of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ per season in each gulf for the different samples collected.

Season	Gulf	Sample	Max $\delta^{13}\text{C}$	Min $\delta^{13}\text{C}$	Mean $\delta^{13}\text{C}$	Max $\delta^{15}\text{N}$	Min $\delta^{15}\text{N}$	Mean $\delta^{15}\text{N}$
Fall 2022	GN	POM	-14.5	-24.0	-20.7	17.0	7.4	10.7
		Phytoplankton	-13.7	-20.2	-16.9	21.4	13.0	17.0
		Zooplankton	-18.4	-19.6	-19.1	15.7	14.4	14.8
	GSJ	POM	-19.8	-23.6	-21.3	10.1	8.2	9.1
		Phytoplankton	-14.9	-24.3	-18.1	18.1	6.3	15.2
		Zooplankton	-18.1	-18.7	-18.3	13.3	3.3	12.9
Winter 2022	GN	POM	-22.2	-23.5	-23.1	9.1	7.8	8.1
		Phytoplankton	-14.9	-17.9	-16.7	7.8	6.5	7.3
		Zooplankton	-18.5	-19.5	-18.9	11.9	9.6	10.2
	GSJ	POM	-21.4	-22.9	-21.9	9.3	8.3	8.7
		Phytoplankton	-16.1	-18.9	-16.8	12.7	8.2	9.2
		Zooplankton	-17.8	-18.1	-17.9	12.2	10.6	11.2
Spring 2022	GN	POM	-18.0	-23.5	-21.4	11.3	8.4	9.8
		Phytoplankton	-17.1	-20.0	-18.1	13.1	12.7	13.0
		Zooplankton	-16.7	-17.2	-17.0	14.9	12.8	13.8
	GSJ	POM	-20.4	-22.7	-21.8	11.4	8.3	9.4
		Phytoplankton	-16.3	-19.9	-18.1	13.3	10.1	11.1
		Zooplankton	-18.3	-19.0	-18.7	14.0	12.0	12.6
Summer 2023	GN	POM	-20.4	-23.7	-22.1	10.1	8.0	8.6
		Phytoplankton	-16.9	-20.3	-18.6	10.9	9.5	10.2
		Zooplankton	-17.8	-21.7	-19.5	12.9	11.6	12.2
	GSJ	POM	-20.0	-22.2	-21.3	10.2	7.9	9.1
		Phytoplankton	-17.6	-20.6	-18.9	11.2	10.0	10.5
		Zooplankton	-17.4	-18.7	-18.1	14.2	13.5	13.9

Table 1 shows the isotopic values for the different samples collected during the seasons of 2022 and summer 2023 (a hole year). In all cases, phytoplankton samples were represented by the taxonomic group of diatoms. On the other hand, most of the zooplankton samples were represented by small copepods species, if it wasn't the case, the other group were cladocerans.

One thing to be highlighted is that, mean $\delta^{15}\text{N}$ values for both gulfs during fall 2022 of phytoplankton and zooplankton are not as expected (Table 1). The first ones are enriched compared to the second one, an unusual trend given the fact that zooplankton feeds on phytoplankton or particulate organic matter. $\delta^{15}\text{N}$ value of nitrate increases as nitrate consumption increases due to an isotopic effect ranging from 5‰ to 8‰ during nitrate assimilation by phytoplankton (Yoshikawa et al. 2024). The spatial patterns of nitrate values in the euphotic zone are conserved in phytoplankton and are subsequently transferred to organisms with higher trophic positions (zooplankton) with ^{15}N enrichment of ~3‰ per trophic position (Minagawa and Wada 1984). Given this, the proposed hypothesis is that during the phytoplankton growth during this particular season and year, zooplankton was trying to “catch up” and was at the early stages of grazing reflecting lower $\delta^{15}\text{N}$ values.

Besides isotopic values, chlorophyll “a”, nutrients (nitrate, phosphate and silicate), sea surface temperature and salinity are being measured as well to be explanatory variables for the distribution of the isotopic values. In addition, a hole year was already sampled and ready to be analyzed at the UNM so comparisons between 2 years can be achieved. Isoscapes are being developed with the free software Ocean Data View (ODV) (Schlitzer 2002; Beckers et al. 2014) and it is expected to propose General Additive Models (GAMs) to tag along them.

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