

THE POTENTIAL EFFECT OF NITROGEN REMOVAL PROCESSES ON THE $\delta^{15}\text{N}$ FROM DIFFERENT TAXA IN THE MEXICAN SUBTROPICAL NORTH EASTERN PACIFIC

Camalich, J.¹, A. Sánchez¹, S. Aguíñiga¹ & E. F. Balart²

¹ Centro Interdisciplinario de Ciencias Marinas - Instituto Politécnico Nacional. Av. Instituto Politécnico Nacional s/n Col. Playa Palo de Santa Rita Apdo. Postal 592. C.P. 23090, La Paz, B.C.S., México. ² Centro de Investigaciones Biológicas del Noroeste. Instituto Politécnico Nacional 195, Playa Palo de Santa Rita Sur; La Paz, B.C.S. México. C.P. 23096 * Corresponding author. Present address: IMARES Wageningen UR, Zuiderhaaks 5 1797 SH 't Horntje, Texel, Netherlands.

ABSTRACT. The sub-tropical north eastern Pacific is one of the major zones in the ocean where nitrogen is removed by bacterial processes which are enhanced by low oxygen concentrations commonly found in the water column along the Pacific coast upwelling areas. It is well established that the nitrogen isotopic signal ($\delta^{15}\text{N}$) increases in relation to trophic levels but little is known about the transfer of this $\delta^{15}\text{N}$ signal from the dissolved fraction to higher trophic levels in oceanic regions with low oxygen. The objectives of this study are: 1) to report $\delta^{15}\text{N}$ values from different abiotic and biotic components collected in the low-oxygen oceanic region in front of Bahía Magdalena (Mexican subtropical north-eastern Pacific); 2) to compare the $\delta^{15}\text{N}$ of different trophic levels with analogous organisms in regions where nitrogen fixation is the dominating process, which will allow us to evaluate the actual transfer of $\delta^{15}\text{N}$ enriched in ^{15}N through the trophic web up to top predators. The $\delta^{15}\text{N}$ was higher in both abiotic and biological compared to those reported from zones where N fixation is the dominating process. Oxygen concentrations in the oceanic area in front of Bahía Magdalena are low (< 2ml/l) at shallow water depths (< 100m) but not anoxic. Despite this we found that the $\delta^{15}\text{N}$ signal reflects denitrification and this signal is transferred up through the food web.

Keywords: Subtropical north eastern Pacific, nitrogen cycle, $\delta^{15}\text{N}$, oxygen minimum zone.

Efecto potencial del proceso de remoción de nitrógeno sobre el $\delta^{15}\text{N}$ de distintos taxa en el Pacífico noreste mexicano subtropical

RESUMEN. El Pacífico subtropical noroeste es una de las zonas más importantes del océano en las cuales el nitrógeno es utilizado por procesos bacterianos que se intensifican bajo condiciones bajas de oxígeno como las que se encuentran comúnmente en las zonas de surgencia a lo largo de las costas del Pacífico. El incremento en la señal isotópica de N con respecto al nivel trófico ($\delta^{15}\text{N}$) es bien conocido, sin embargo su transferencia desde la fracción disuelta hasta niveles tróficos altos no ha sido estudiada a profundidad en zonas del océano en las cuales las concentraciones de oxígeno son bajas. Los objetivos de este estudio son: 1) reportar valores de $\delta^{15}\text{N}$ de diferentes compartimentos (abióticos y bióticos) recolectados en la zona oceánica de baja concentración de oxígeno frente a Bahía Magdalena (Pacífico subtropical noreste Mexicano); 2) comparar $\delta^{15}\text{N}$ de diferentes niveles tróficos con organismos análogos de regiones en las cuales la fijación de nitrógeno es el proceso dominante; esto nos permitirá evaluar la transferencia real de $\delta^{15}\text{N}$ enriquecido en ^{15}N a través de la red trófica hasta depredadores tope. El $\delta^{15}\text{N}$ de los componentes abióticos y bióticos fue más alto que los reportados en regiones con una alta tasa de fijación de N. Las concentraciones de oxígeno en la zona de estudio son bajas (< 2ml/l) a profundidades superficiales (< 100m) aunque no anóxicas. No obstante, la señal de $\delta^{15}\text{N}$ refleja desnitrificación y esta señal es transferida a lo largo de la cadena trófica.

Palabras clave: Pacífico nororiental subtropical, ciclo del nitrógeno, $\delta^{15}\text{N}$, zona de mínimo oxígeno.

Camalich, J., A. Sánchez, S. Aguíñiga & E. F. Balart. 2012. The potential effect of nitrogen removal processes on the $\delta^{15}\text{N}$ from different taxa in the mexican subtropical north eastern Pacific. *CICIMAR Oceánides*, 27(2): 27-35.

INTRODUCTION

Upwelling areas, where nutrient rich water is transferred from the deep ocean to the productive surface layers, have an important role in coastal fisheries around the world as they enhance primary productivity, which in turn allows for a higher secondary production. However, in some areas a combination of high productivity and poor water exchange can create large areas in the water column with very low oxygen concentrations (0.5 ml/l) called oxygen minimum zones (OMZ, Levin *et al.*, 2002). Within these OMZ, processes take place which transfer nitrogen, one of the most important nutrients in the ocean, to a form which cannot be used by phytoplankton and thus result in a loss of biologically available nitrogen. The major ni-

trogen removal processes behind this are denitrification, in which heterotrophic bacteria convert nitrate (NO_3^-) to dinitrogen gas (N_2), and anaerobic ammonium oxidation (anammox), where microbes use ammonium (NH_4^+) and nitrite (NO_2^-) to produce N_2 (Lam *et al.*, 2009, Fig. 1).

Although OMZ represent less than the 1% of the ocean volume worldwide, it has been estimated that as much as 20 - 40% (or approximately 200×10^6 ton/year) of the oceanic nitrogen is lost in the Arabian Sea and the North and South Eastern Tropical Pacific (Codispoti *et al.*, 2001; Devol, 2008). With the alleged warming of the oceans due to climate change, a substantial expansion of OMZ is predicted to occur (Stramma *et al.*, 2008). Another emerging pro-

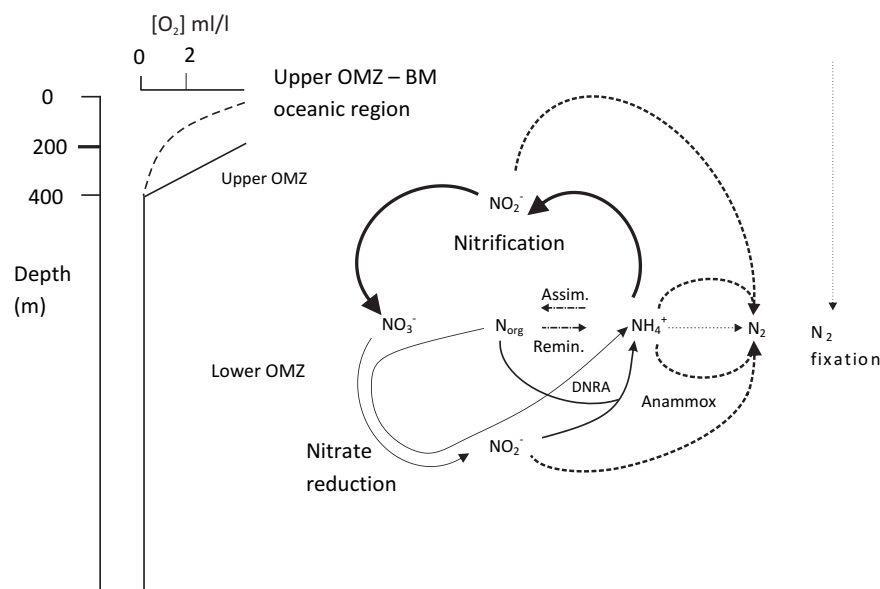


Figure 1. The N cycle at the OMZ. The processes nitrate reduction (denitrification) and anammox can potentially increase the isotopic signal at the dissolved phase. Nitrate reduction and anammox are considered as loss of N (modified from Lam *et al.*, 2009).

blem is the increase of coastal areas not connected to OMZ with low oxygen conditions (hypoxia), which is considered to be one of the major emerging anthropogenically induced problems (Vaquer-Sunyer & Duarte, 2008). These expansions will lead to an increase in nitrogen removal processes (Stramma *et al.*, 2008) and could therefore cause a significant imbalance in the global budget of this important nutrient (Zehr & Ward, 2002; Zehr, 2009). Furthermore, oxygen minimum zones are inhospitable to many species and therefore serve as biogeographic barriers (Helly & Levin, 2004; Rogers, 2000) possibly causing ecological modification as some species are displaced or removed.

Nitrogen is found in two stable isotopic forms in nature, the lighter ^{14}N (99.6%) and the heavier ^{15}N (<0.4%). Due to their difference in mass the heavier isotope reacts at a slightly slower rate compared to ^{14}N , thereby causing chemical fractionation (Devol, 2008). In addition there are many biological reactions that can alter the ratio of heavy-to-light isotopes (Peterson & Fry, 1987), as ^{15}N is selectively discriminated and therefore accumulates in the residual nitrogen pool.

Several studies have used the stable isotopic composition of nitrogen ($\delta^{15}\text{N}$) to trace nitrogen removal processes. Water column denitrification in the OMZ and the incorporation of NO_3^- by phytoplankton in the ocean surface leads to an increase in nitrate $\delta^{15}\text{N}$. For

example, Brandes *et al.* (1998) showed that as a result of intense denitrification within the water column in the OMZ of the Arabian Sea and the eastern tropical North Pacific Ocean (ETNP), there is a marked increase in the $\delta^{15}\text{N}$ signal of NO_3^- from the deep water to the surface. In addition, a significant difference between the $\delta^{15}\text{N}$ of nitrate measured in the North Eastern Pacific (15 ‰) and the average open ocean (5 ‰) has been detected due to the N removal processes in OMZ waters (Altabet *et al.*, 1999; Brandes *et al.*, 1998; Cline & Kaplan, 1975). Although this process is commonly known, the transference of this relatively high $\delta^{15}\text{N}$ signal from the dissolved fraction up through the trophic web has not previously been reported in this region. Our objectives with this study are: 1) to report the $\delta^{15}\text{N}$ from different abiotic (sediment and NO_3^-) and biological components (phytoplankton, zooplankton, cephalopods: *Dosidicus gigas*, benthic-pelagic crustaceans: *Pleuoncodes planipes*, demersal fishes, sea lions: *Zalophus californianus* and dolphins: *Tursiops truncatus*) from the low-oxygen oceanic region in front of Bahía Magdalena, Mexico (Fig 2); 2) to highlight the potential effect of nitrogen removal process in the $\delta^{15}\text{N}$ values from the base of the food web through top predators.

Low water column oxygen concentrations favor bacterial nitrogen removal processes leaving a pool of nitrate high on $\delta^{15}\text{N}$. Since this signal is transferred through the food web, our hypothesis is that higher trophic levels including

top predators are considerably more enriched in ^{15}N than similar taxa living in regions where the $\delta^{15}\text{N}$ at the base of the food web is lower.

MATERIAL AND METHODS

Samples of water, sediments, phytoplankton, zooplankton, benthic-pelagic crustaceans and demersal fishes were collected on board the research vessel BIP XII during four campaigns (March and November, 2006 and 2007) at the oceanic region in front of the Bahía Magdalena-Almejas lagoon complex (Fig. 2). Phytoplankton and zooplankton were collected simultaneously using a bongo net towed from the surface (61 cm diameter, 200 and 500 μm mesh respectively), stored in conical tubes and kept cold (4°C) for later analysis. Sediment samples were collected at different depths (from 40 to 400 m) using a Smith-McIntyre grab and stored in clean plastic bags at -20° . Water samples were collected at 50 m and 200 m (only during November 2007) using a Niskin bottle, filtered through pre-combusted GF/F filters (0.45 μm) and stored in cleaned Nalgene bottles. The demersal fishes and the red crab (*P. planipes*) were collected using a bottom-trawl net with a head rope of 34 m and a 50-mm mesh size and preserved by freezing for later analysis (no *P. planipes* samples were collected in November 2006). Portions of muscle from stranded sea mammals were acquired by continuous patrolling along Isla Margarita (Fig 2).

Oxygen profiles

The world ocean atlas is the result of a global collection of samples supported by different programs including the World Ocean Data Base (WOD) and the Global Oceanographic Data Archaeology and Rescue (GODAR). In the case of oxygen concentrations, values have in most cases been obtained by instruments mounted on oceanographic rosettes and in some cases obtained from a modified Winkler titration (ocean discrete samples in the WOD) (García *et al.*, 2010). The data was downloaded from <http://www.nodc.noaa.gov/OC5/WOA09/wao9data.html> and the profiles constructed using Ocean Data View (Schlitzer, 2011). The selection of profiles from WOA09 correspond to those closest available for our sampling stations.

Stable isotope analysis

Nitrogen stable isotopes from water samples were analyzed following the ammonia diffusion method from by Sigman *et al.* (1997) at the Facultad de Ciencias del Mar y Limnología (UNAM-Mazatlán). Sediments were dried and approximately 20 mg were weighed and packed into tin capsules and sent to the University of Davis for the isotopic analysis. The phytoplankton and zooplankton samples were freeze-dried and set under acid environment (1N HCl) using a glass desiccator during 24 h following Lorrain *et al.* (2003). After

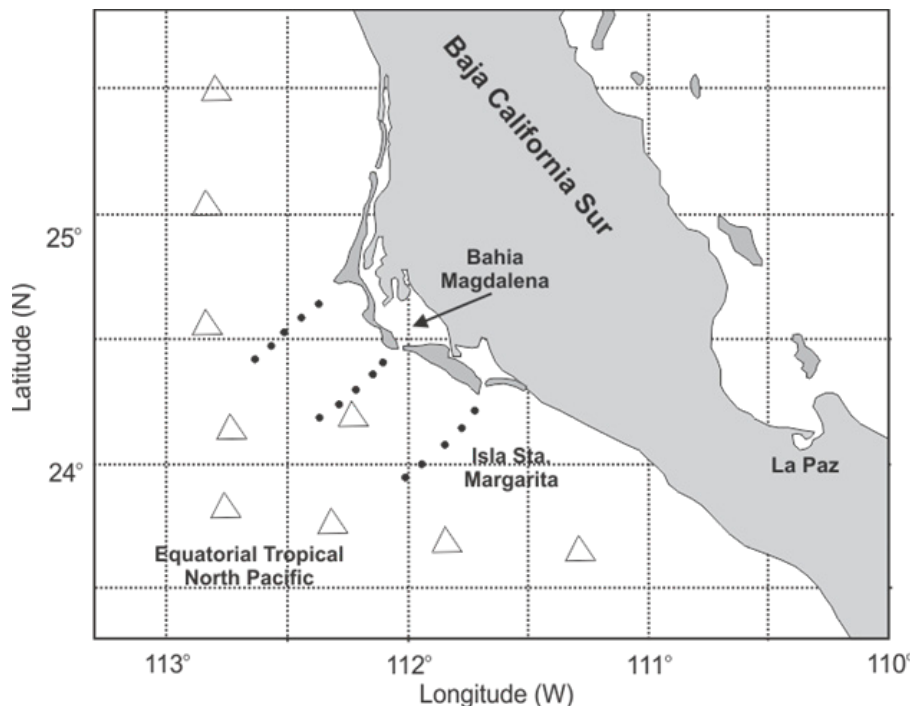


Figure 2. Map of sampling sites (•) and points selected (Δ) offshore Bahía Magdalena for the O₂ profiles using WOA09 data

decalcification, approximately 1 mg of sample was weighed and packed into tin capsules for analysis. Samples from the giant squid (*Dosidicus gigas*) mantle were freeze-dried and 1 mg packed into tin capsules. Samples of muscle of benthic-pelagic (22 – 32 mm) red crab were scraped from the exoskeleton, freeze-dried and 1 mg packed into tin capsules for analysis. For demersal fishes a portion of dorsal muscle was freeze dried and 1mg packed into tin capsules. In the case of marine mammals the biopsies were dissected to remove the adipose tissue and rinsed in methanol before being freeze-dried and packed into tin capsules. The analysis reproducibility was 0.1‰ for $\delta^{15}\text{N}$ ($n = 19$, UC Davis internal standard). The highest standard deviation from duplicates was 0.3‰.

Literature data comparison

We used the concept of N^* proposed by Gruber and Sarmiento (1997) to distinguish between regions in the ocean with contrasting N processes such as removal (denitrification) and fixation. The N^* concept and its mathematical development are based on a large collection of samples which describes the stoichiometry behind the Redfield ratio. Based on Gruber (2008) we thus selected regions of the Atlantic ocean with positive N^* value (Caribbean, West coast of the Iberian Peninsula, Brazilian coast, Bay of Biscay France, coast of Island, coast of Virginia U.S., North Sea U.K) as a criterion for N fixation and searched the literature for species of similar taxa to those collected at the northeastern subtropical Pacific to use in a comparison of $\delta^{15}\text{N}$ transfer in N-fixing and N-removing environments.

The intense denitrification around the studied area is evidenced by N^* value of -4 (at 300 m) (Gruber, 2008). On the other hand, values of N^* from Alaska and the Atlantic Ocean (-1 to 4 respectively, Table 1), were large cyanobacteria blooms constantly occur (e.g. Zehr & Ward, 2002) reveal N fixation (Carpenter & Capone, 2008)

RESULTS

Oxygen profiles

The oxygen profiles constructed for the studied area show a fast reduction in O_2 -concentration in the first two hundred meters at all stations (Fig. 3). Hypoxic conditions ($[\text{O}_2] < 2.1$ ml/l) starts between 50-150 m for all stations.

N stable isotopes

The analysis of $\delta^{15}\text{N-NO}_3^-$ ranged from 6‰ to 7.6‰ at 50 m and from 11‰ to 13.4‰ at 300 m. The sediment values ranged from 6.3‰ to 9.3‰. The phytoplankton values ranged from

8‰ to 9‰ and zooplankton values from 12.9‰ to 13.9‰. The cephalopod values ranged from 15.1‰ to 17.7‰, red crab values from 12.6‰ to 16.6‰, demersal fish from 13.8‰ to 18‰, dolphins from 16.8‰ to 17.8‰ and sea lion values from 18.4‰ to 20.7‰.

Literature data comparison

Nitrogen stable isotope data ($\delta^{15}\text{N}$) from taxa similar to those found in front of Bahía Magdalena were obtained from ten different studies from regions in the Atlantic Ocean with positive N^* values, with the exception of Hobson *et al.* (2002) which is from Alaska and has a negative N^* (Table 1). Aguilar *et al.* (2008) used stable isotopes in marine fish from the Atlantic Ocean and Caribbean to compare different levels of human impacts. The study of Bode *et al.* (2007) was conducted on the Iberian Atlantic shelf and described the temporal variations of the pelagic food web using stable isotopes. Corbisier *et al.* (2006) used stable isotopes to determine food sources and reconstruct the food web in a coastal area in Brazil. Hobson *et al.* (2002) used stable isotopes to model the Arctic food web from particulate organic matter (POM) to top predators. Le Loc'h *et al.* (2008) described the benthic food web using stable isotopes from the continental shelf in the north eastern Atlantic. Although the article of Logan and Lutcavage (2008) was not focused on ecological descriptions, important values from fish species were found for this comparison. Petursdottir *et al.* (2008) described trophic routes from benthic and pelagic crustaceans to mesopelagic fish species using stable isotopes and fatty acids. Stowasser *et al.* (2009) analyzed stable isotopes and fatty acids from deep sea fishes collected in the North Atlantic. Sigman *et al.* (1997) described the method for $\delta^{15}\text{NO}_3^-$ analysis using samples from the Sargasso Sea, a region of the ocean commonly known for intense N_2 fixation. The mean and standard deviations of each component are summarized in Table 1.

DISCUSSION

In the present study our purpose was to highlight the utility of $\delta^{15}\text{N}$ as a tracer of N removal processes along the food web in front of Bahía Magdalena. Therefore the results presented are averaged values of four season samplings. Details regarding seasonal changes and specific species can be found in Camalich (2011). The data comparison of analogous biotic components from the studied region and the Atlantic Ocean shows the transfer of dissolved nitrogen, and its progressive enrichment, along the food web. The $\delta^{15}\text{N-NO}_3^-$ found in this study were higher compared to those reported as av-

Table 1. Average values and standard deviations of $\delta^{15}\text{N}$ from different components in areas where nitrogen fixation is the dominating process.

Compartment	N*		Citation
	Gruber 2008	d15N (‰) Mean \pm S.D.	
NO_3^-	3	5	Sigman <i>et al.</i> , 1997
POM	3 - 4	5.6 \pm 1.7	Bode <i>et al.</i> , 2007; Corbisier <i>et al.</i> , 2006
Crustaceans*	3	10 \pm 0.5	Corbisier <i>et al.</i> , 2006; Petursdottir <i>et al.</i> , 2008
Cephalopods*	2 - 3	11 \pm 1.3	Bode <i>et al.</i> , 2007; Corbisier <i>et al.</i> , 2006
Demersal species*	1 - 2	12.5 \pm 0.1	Aguilar <i>et al.</i> , 2008; Bode <i>et al.</i> , 2007; Corbisier <i>et al.</i> , 2006; Le Loc'h <i>et al.</i> , 2008; Logan & Lutcavage, 2008; Logan & Lutcavage, 2010; Petursdottir <i>et al.</i> , 2008; Stowasser <i>et al.</i> , 2009
<i>Delphinus delphis</i>	3	13.1	Bode <i>et al.</i> , 2007
<i>Pusa hispida</i>	-1	17.5	Hobson <i>et al.</i> , 2002

*A detailed description of the species and values used for this comparison can be found in Camalich (2011).

erage for open oceans where N fixation is the dominant process (Table 1), and are consistent with the range of values reported by Liu and Kaplan (1989), Brandes *et al.* (1998) and Voss *et al.* (Voss *et al.*, 2001) from the Eastern tropical north Pacific OMZ (8‰ to 16‰). In addition we found that the $\delta^{15}\text{N}$ of the sedimentary organic matter in the study area were higher compared to areas in the Atlantic Ocean. The sediment underlying oxygen minimum zones are a good register of the $\delta^{15}\text{N}$ sinking particles since the low oxygen enhance the preservation of surface organic matter (Altabet *et al.*, 1999). Although the O_2 -concentration in the water column in the area is potentially not low enough (~ 2 ml/l) for denitrification, the record of ^{15}N -enriched particles in the sediments appear to confirm the presence of nitrogen removal processes in the overlying water column.

Both phytoplankton and zooplankton in the study area were enriched in ^{15}N compared to the Atlantic regions (Fig. 4). Zooplankton $\delta^{15}\text{N}$ in the Bahía Magdalena oceanic region fit into the values measured at the southern end of the Baja California peninsula reported by Lopez-Ibarra (2008) and Olson *et al.* (2010). In addition, the giant squid and the red crab had higher values compared to the Atlantic cephalopods and crustaceans (Fig. 4). Following the same trend, marine mammals (dolphins and sea lions) off Bahía Magdalena were enriched in ^{15}N even though they maintained similar diets as those found for the Atlantic (Table 1). Both the jumbo squid and the red crab were sampled at depths from 50 to 400m. Gilly (2006) showed that jumbo squid migrate vertically in the Gulf of

California and the north eastern Pacific, probably hunting one of its preferred prey species, *Pleuroncodes planipes*.

In the case of higher trophic levels the average $\delta^{15}\text{N}$ values showed a ^{15}N enrichment compared to those reported in the literature living on the Atlantic (Table 1). Some demersal fishes and marine mammals of the region have been suggested as a good monitor of biogeochemical processes since they observed a strong spatial and temporal fidelity (Camalich, 2011). In the study of Ménard *et al.* (2007) conducted at the Arabian Sea, migratory tuna (*Thunnus albacares*) and sword fish (*Xiphias gladius*) recorded a change in the $\delta^{15}\text{N}$ signal corresponding to a change in N dynamics. As they found the highest $\delta^{15}\text{N}$ values in fishes in areas with high N removal rates, their results suggest that top predators can be used as monitors of water column denitrification. Our finding of a cascading effect in the $\delta^{15}\text{N}$ -signal from the base of the food web to top predators in the area supports that conclusion. However, it is not clear if the enhanced signal is caused by denitrification as the O_2 -concentration in the more shallow parts of the study area is too high (~ 2 ml/l) to cause extensive denitrification (Gruber, 2008; Knowles, 1982). It is possible that the high $\delta^{15}\text{N}$ signal has been transferred into the area from deeper waters or carried northward from the more O_2 -depleted regions such as the Mazatlán area (Brandes *et al.*, 1998) or the Gulf of Tehuantepec (Kienast *et al.*, 2002). Another possible explanation is that the high $\delta^{15}\text{N}$ is due to anammox in the water column. Anammox is triggered at O_2 concentration of ~ 2 ml/l as in the

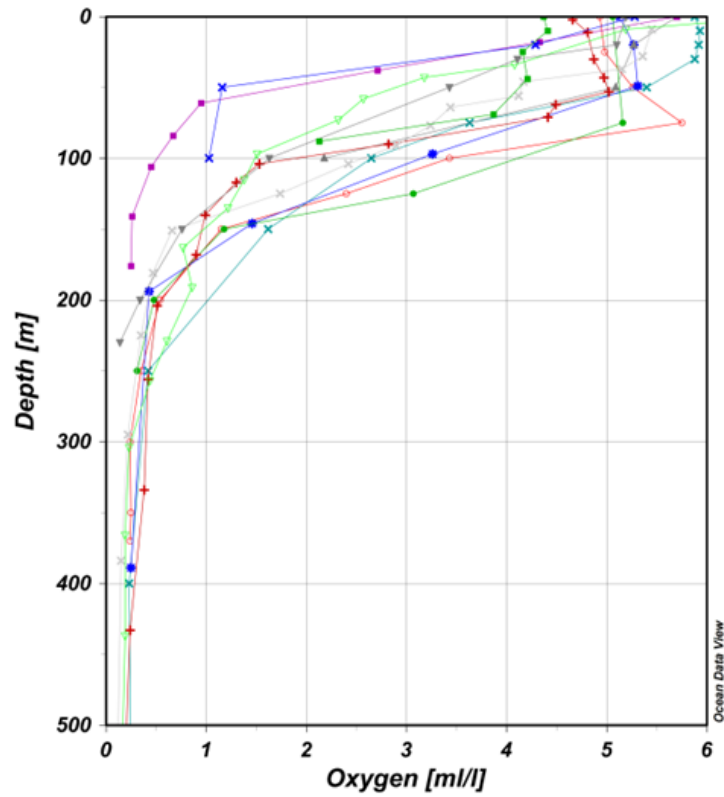


Figure 3. Oxygen concentration (ml/l) profiles at the oceanic region in front of Bahía Magdalena (data from WOA09).

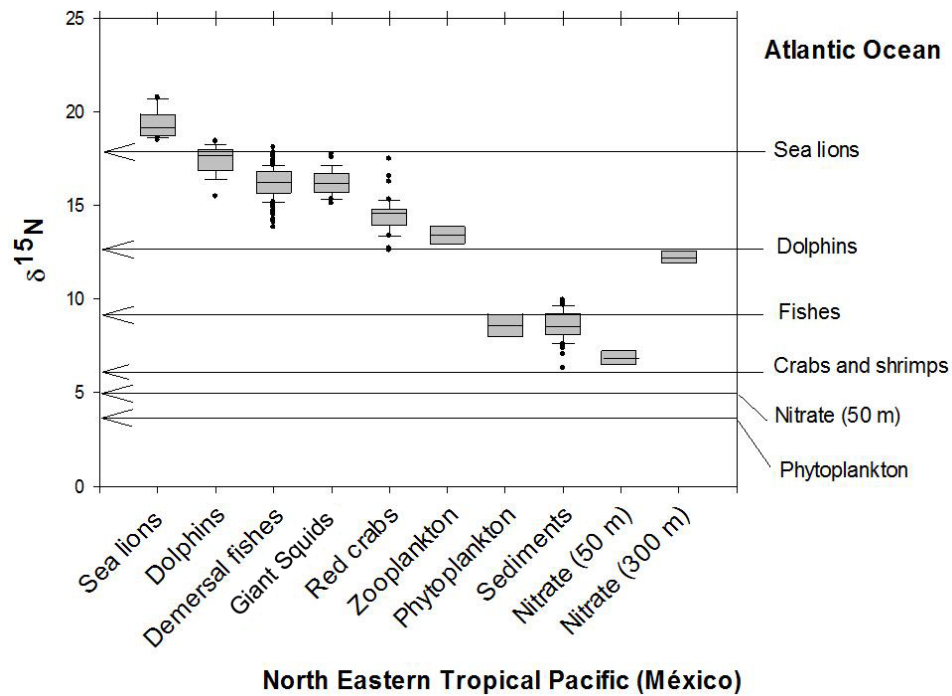


Figure 4. Comparison of $\delta^{15}\text{N}$ from abiotic (nitrate and sediments) and biological samples from the northern ETP (Camalich, 2011) compared to the Atlantic Ocean (Bode *et al.*, 2007; Corbisier *et al.*, 2006; Gruber & Sarmiento, 1997; Hobson *et al.*, 2002; Knapp *et al.*, 2008; Le Loc'h *et al.*, 2008; Stowasser *et al.*, 2009).

shallower parts of the study area and in recent years it has become clear that in addition to denitrification, anammox may be an important nitrogen removal process (Kuypers *et al.*, 2003; Lam *et al.*, 2009). Just as for denitrification the signal of this process could thus be printed on the water column nitrates and transferred to the base of the food web (Holtappels *et al.*, 2010; Song & Tobias, 2011). Elucidating the contribution of denitrification and anammox to the enrichment of ^{15}N in low oxygen areas and determining the potential transfer of the $\delta^{15}\text{N}$ signal from anammox processes throughout the food web is important in future studies.

ACKNOWLEDGEMENTS

This study was supported by the grants SEP-CONACYT (project C01-46806, 2005-2008), SAGARPA-CONACYT (project 2003-02-019) and project EP2 of CIBNOR. We are grateful to the crew of the R/V BIP XII and Arturo Tecuapetla for their assistance during the field work. The first author is grateful to CONACYT and COFAA for the scholarships provided during his doctoral work. Thanks to Elisabeth Svensson for her help in improving the final manuscript.

REFERENCES

- Aguilar, C., G. González-Sansón, I. Faloh & R.A. Curry. 2008. Spatial variation in stable isotopes ($\delta^{13}\text{C}$ and $\delta^{15}\text{N}$) in marine fish along the coast of Havana City: Evidence of human impacts from harbor and river waters. *J. Coast. Res.*, 24(5): 1281-1288.
- Altabet, M.A., C. Pilskaln, R. Thunell, C. Pride, D. Sigman, F. Chavez, & R. Francois. 1999. The nitrogen isotope biogeochemistry of sinking particles from the margin of the Eastern North Pacific. *Deep Sea Res. Part I*, 46(4): 655-679.
- Bode, A., M.T. Alvarez-Ossorio, M.E. Cunha, S. Garrido, J.B. Peleteiro, C. Porteiro, L. Valdés & M. Varela. 2007. Stable nitrogen isotope studies of the pelagic food web on the Atlantic shelf of the Iberian Peninsula. *Prog. Oceanogr.*, 74(2-3): 115-131.
- Brandes, J.A., A.H. Devol, T. Yoshinari, D.A. Jayakumar & S.W.A. Naqvi. 1998. Isotopic composition of nitrate in the central Arabian Sea and eastern tropical North Pacific: A tracer for mixing and nitrogen cycles. *Limnol. Oceanogr.*, 43(7): 1680-1689.
- Camalich, J. 2011. *The register of oceanographic variability on demersal fishes and top predators at the oceanic front off Bahía Magdalena México*, PhD. Thesis. Centro Interdisciplinario de Ciencias Marinas, Instituto Politécnico Nacional, La Paz, BCS, 187 p.
- Carpenter, E.J. & D.G. Capone. 2008. Pelagic nitrogen fixation, 141-198. In: Capone, D.G., D.A. Bronk, M.R. Mulholland, E.J. Carpenter (Eds.), *The Marine Nitrogen Cycle: Overview and Challenges*. Academic Press, San Diego..
- Cline, J.D. & I.R. Kaplan. 1975. Isotopic fractionation of dissolved nitrate during denitrification in the eastern tropical north Pacific Ocean. *Mar. Chem.*, 3(4): 271-299.
- Codispoti, L.A., J.A. Brandes, J.P. Christensen, A.H. Devol, S.W.A. Naqvi, H.W. Paerl & T. Yoshinari. 2001. The oceanic fixed nitrogen and nitrous oxide budgets: Moving targets as we enter the anthropocene? *Sci. Mar.*, 65(SUPPLEMENT 2): 85-105.
- Corbisier, T.N., L.S.H. Soares, M.A.V. Petti, E.Y. Muto, M.H.C. Silva, J. McClelland & I. Valiela. 2006. Use of isotopic signatures to assess the food web in a tropical shallow marine ecosystem of Southeastern Brazil. *Aquat. Ecol.*, 40(3): 381-390.
- Devol, A.H. 2008. Denitrification including Anammox, 263-302. In: Capone, D.G., D.A. Bronk, M.R. Mulholland & E.J. Carpenter (Eds.), *Nitrogen in the Marine Environment* (2nd Ed.). Academic Press, San Diego..
- Garcia, H.E., R.A. Locarnini, T.P. Boyer, J.I. Antonov, O.K. Baranova, M.M. Zweng & D.R. Johnson. 2010. Dissolved Oxygen, Apparent Oxygen Utilization, and Oxygen Saturation. In: Levitus, S. (Ed.), *World Ocean Atlas 2009*. NOAA Atlas NESDIS 70, U.S. Government Printing Office, Washington, D.C., 344 pp.
- Gilly, W.F. 2006. Horizontal and vertical migration of *Dosidiscus gigas* in the Gulf of California revealed by electronic tagging, 3-7. In: Olson, R.J., & J.W. Young (Eds.), *The role of squid in open ocean ecosystems*. Global ocean ecosystem dynamics, Honolulu, Hawaii. 116 p.
- Gruber, N. & J.L. Sarmiento. 1997. Global patterns of marine nitrogen fixation and denitrification. *Global Biogeochem. Cycles*, 11(2): 235-266.
- Gruber, N. 2008. The Marine Nitrogen Cycle: Overview and Challenges, 1-50. In: Capone, D.G., D.A. Bronk, M.R. Mulholland, E.J. Carpenter (Eds.), *Nitrogen in the Ma-*

- rine Environment* (2nd Edition). Academic Press, San Diego.
- Helly, J.J. & L.A. Levin. 2004. Global distribution of naturally occurring marine hypoxia on continental margins. *Deep Sea Res. Part I*, 51(9): 1159-1168.
- Hobson, K.A., A. Fisk, N. Karnovsky, M. Holst, J.M. Gagnon & M. Fortier. 2002. A stable isotope ($d^{13}C$, $d^{15}N$) model for the North Water food web: Implications for evaluating trophodynamics and the flow of energy and contaminants. *Deep Sea Res. Part II*, 49(22-23): 5131-5150.
- Holtappels, M., G. Lavik, M.M. Jensen & M.M.M. Kuypers. 2010. ^{15}N -labeling experiments to dissect the contributions of heterotrophic denitrification and anammox to nitrogen removal in the OMZ waters of the ocean. *Methods Enzymol.*, 486: 223-251.
- Kienast, S.S., S.E. Calvert & T.F. Pedersen. 2002. Nitrogen isotope and productivity variations along the northeast Pacific margin over the last 120 kyr: Surface and subsurface paleoceanography. *Paleoceanography*, 17(4): 7.1-7.17.
- Knowles, R. 1982. Denitrification. *Microbiol Rev.*, 46(1): 43-70.
- Kuypers, M.M.M., A.O. Silekers, G. Lavik, M. Schmid, B.B. Jørgensen, J.G. Kuenen, J.S. Sinninghe Damsté, M. Strous & M.S.M. Jetten. 2003. Anaerobic ammonium oxidation by anammox bacteria in the Black Sea. *Nature*, 422(6932): 608-611.
- Lam, P., G. Lavik, M.M. Jensen, J. van de Vossenberg, M. Schmid, D. Woebken, G. Dimitri, R. Amann, M.S.M. Jetten & M.M.M. Kuypers. 2009. Revising the nitrogen cycle in the Peruvian oxygen minimum zone. *Proc. Nat. Acad. Sci. U.S.A.*, 106(12): 4752-4757.
- Le Loc'h, F., C. Hily & J. Grall. 2008. Benthic community and food web structure on the continental shelf of the Bay of Biscay (North Eastern Atlantic) revealed by stable isotopes analysis. *J. Mar. Syst.*, 72(1-4): 17-34.
- Levin, L., D. Gutiérrez, A. Rathburn, C. Neira, J. Sellanes, P. Muñoz, V. Gallardo & M. Salamanca. 2002. Benthic processes on the Peru margin: A transect across the oxygen minimum zone during the 1997-98 El Niño. *Prog. Oceanogr.*, 53(1): 1-27.
- Liu, K.-K. & I.R. Kaplan. 1989. The Eastern Tropical Pacific as a source of $d^{15}N$ -enriched nitrate in seawater off southern California. *Limnol Oceanogr.*, 34(5): 820-830.
- Logan, J.M. & M.E. Lutcavage. 2008. A comparison of carbon and nitrogen stable isotope ratios of fish tissues following lipid extractions with non-polar and traditional chloroform/methanol solvent systems. *Rapid Commun. Mass Spectrom.*, 22(7): 1081-1086.
- Lopez-Ibarra, G. 2008. *Estructura trófica de los copépodos pelágicos en el Océano Pacífico Oriental Tropical*. Ph D Thesis. Centro Interdisciplinario de Ciencias Marinas, Instituto Politécnico Nacional, La Paz, B.C.S., México. 107 p.
- Lorrain, A., N. Savoye, L. Chauvaud, Y.M. Paulet & N. Naulet. 2003. Decarbonation and preservation method for the analysis of organic C and N contents and stable isotope ratios of low-carbonated suspended particulate material. *Anal. Chim. Acta*, 491(2): 125-133.
- Ménard, F., A. Lorrain, M. Potier & F. Marsac. 2007. Isotopic evidence of distinct feeding ecologies and movement patterns in two migratory predators (yellowfin tuna and swordfish) of the western Indian Ocean. *Mar. Biol.*, 153(2): 141-152.
- Olson, R.J., B.N. Popp, B.S. Graham, G.A. López-Ibarra, F. Galván-Magaña, C.E. Lennert-Cody, N. Bocanegra-Castillo, N.J. Wallsgrove, E. Gier, V. Alatorre-Ramírez, L.T. Ballance & B. Fry. 2010. Food-web inferences of stable isotope spatial patterns in copepods and yellowfin tuna in the pelagic eastern Pacific Ocean. *Prog. Oceanogr.*, 86: 124-138.
- Peterson, B.J. & B. Fry. 1987. Stable isotopes in ecosystem studies. *Ann. Rev. Ecol. Syst.*, 18: 293-320..
- Petursdottir, H., A. Gislason, S. Falk-Petersen, H. Hop & J. Svavarsson. 2008. Trophic interactions of the pelagic ecosystem over the Reykjanes Ridge as evaluated by fatty acid and stable isotope analyses. *Deep Sea Res. Part II*, 55(1-2): 83-93.
- Rogers, A.D. 2000. The role of the oceanic oxygen minima in generating biodiversity in the deep sea. *Deep Sea Res. Part II*, 47(1-2): 119-148.

- Schlitzer, R. 2011. Ocean Data View, <http://odv.awi.de>.
- Sigman, D.M., M.A. Altabet, R. Michener, D.C. McCorkle, B. Fry & R.M. Holmes. 1997. Natural abundance-level measurement of the nitrogen isotopic composition of oceanic nitrate: an adaptation of the ammonia diffusion method. *Mar. Chem.*, 57(3-4): 227-242.
- Song, B. & C.R. Tobias. 2011. Molecular and stable isotope methods to detect and measure anaerobic ammonium oxidation (anammox) in aquatic ecosystems. *Methods Enzymol.*, 496: 63-89.
- Stowasser, G., R. McAllen, G.J. Pierce, M.A. Collins, C.F. Moffat, I.G. Priede & D.W. Pond. 2009. Trophic position of deep-sea fish-Assessment through fatty acid and stable isotope analyses. *Deep Sea Res. Part I*. 56, 812-826.
- Stramma, L., G.C. Johnson, J. Sprintall & V. Mohrholz. 2008. Expanding oxygen-minimum zones in the tropical oceans. *Science*, 320(5876): 655-658.
- Vaquer-Sunyer, R. & C.M. Duarte. 2008. Thresholds of hypoxia for marine biodiversity. *Proc. Nat. Acad. Sci. U.S.A.*, 105(40): 15452-15457.
- Voss, M., J.W. Dippner & J.P. Montoya. 2001. Nitrogen isotope patterns in the oxygen-deficient waters of the Eastern Tropical North Pacific Ocean. *Deep-Sea Res. Pt. I*, 48(8): 1905-1921.
- Zehr, J.P. & B.B. Ward. 2002. Nitrogen cycling in the ocean: New perspectives on processes and paradigms. *Appl. Environ. Microbiol.*, 68(3): 1015-1024.
- Zehr, J.P. 2009. New twist on nitrogen cycling in oceanic oxygen minimum zones. *Proc. Nat. Acad. Sci. U.S.A.*, 106(12): 4575-4576.

