11(2):064-070 (2017)

Journal of FisheriesSciences.com

E-ISSN 1307-234X

Research Article

Searching Relationships between Tissue Elemental Concentrations and Geographical Distribution of Bigeye Tuna (*Thunnus Obesus)* from the South Atlantic Ocean

Joana Raimundo[1,2], Miguel Caetano[1*], Carlos Vale[2], Rui Coelho[1], Mário Mil-Homens[1,2], Miguel Neves Dos Santos[1,3]

[1]IPMA-Portuguese Institute for the Sea and Atmosphere, Rua Alfredo Magalhães Ramalho, Lisbon, Portugal

[2]CIIMAR, Marine and Environmental Research Center, Rua dos Bragas, Porto, Portugal

[3]ICCAT-International Commission for the Conservation of Atlantic Tunas, Corazón de María, Madrid, Spain

Received: 17.03.2017 / Accepted: 18.04.2017 / Published online: 20.04.2017

Abstract: Tunas are at the top of the food chains and have high metabolism and feeding rates, leading to increased metal accumulation in their tissues. Several tools using biological, biochemical and chemical measurements have been used to identify populations, geographic distributions and migrations of fish species in different environments. Elemental composition (V, Cr, Co, Ni, Cu, Zn, As, Se, Cd and Pb) was determined in muscle and liver tissues of bigeye tuna (*Thunnus obesus*) caught in three areas of the south Atlantic Ocean. Concentrations in liver (V, Co, Cu, Zn, As, Cd and Pb) point to increased values in specimens captured near the South Africa and South America relatively to the open-ocean areas. The obtained results showed that tissue elemental concentrations in *T. obesus* from the south Atlantic areas can be used as a natural tag. Due to its role, liver seems to be the best indicator tissue.

Keywords: *Thunnus obesus;* Trace elements; Natural tag; Atlantic tuna populations; Liver

---

[*]**Correspondence to:** **Joana Raimundo,** Miguel Caetano, IPMA-Portuguese Institute for the Sea and Atmosphere, Rua Alfredo Magalhães Ramalho, Lisbon, Portugal, Tel: +351.21.3027070; E-mail: mcaetano@ipma.pt

Introduction

Marine contamination has been considered an important issue during the last decades. The number of contaminants entering the ecosystems increased dramatically, and the synergism (and antagonism) among them is poorly supported. Consequently, the evaluation of the influence of contaminants in marine life is a scientific challenge. Accumulation of chemicals in marine resources, habitat loss and decrease of biodiversity are often related to environmental contamination. Metals are persistent in the environment and often displaying concentrations considered toxic (Ikem and Egiebor, 2005; Khansari *et al.*, 2005). The mechanisms of contaminant accumulation in remote areas far from point sources is insufficiently documented and a challenge for the scientific community.

Pelagic top predators are important in oceanic ecosystems (Madigan *et al.*, 2014). They are particularly sensitive for anthropogenic impacts due to their specific life-history traits, like long life-span, late maturation and low reproductive rates (Heithaus *et al.*, 2008). Marine predators can be used as indicators of the marine environmental health (Ramos and González-Solís, 2012). These apical species are subject to high levels of pollutants at the top of the trophic webs and many of them are characterized by large breeding ranges, allowing direct comparison of pollutant levels among remote areas (Aguilar *et al.*, 2002; Hobson *et al.*, 2004; Ramos and González-Solís, 2012). Tunas are migratory predators that often carry out long-distance migrations, both horizontally and vertically, to track food resources and reproduce at remote spawning grounds (Patterson *et al.*, 2008; Block *et al.,* 2001, 2011; Macdonald *et al.*, 2013). They are found at the top of the food chains, considered to have high performance, metabolism and feeding rates favoring enhanced accumulation of metals in their tissues (Engman and Jorhem, 1998; Storelli *et al.*, 2005). Bigeye tuna, *Thunnus obesus* (Lowe, 1839) is a marine pelagic fish species marked by large populations and global distributions in the Atlantic, Indian and Pacific oceans (Gonzalez *et al.*, 2008). The Atlantic population is assessed as a single stock, low genetic heterogeneity (e.g. Chow *et al.,* 2000; Gonzalez *et al.,* 2008); despite several uncertainties exist mainly due to limited information on their biology, fishery statistics and abundance indices (Anon 2005; Arrizabalaga *et al.*, 2008). Research studies that allow a better comprehension of spatial distributions of this data-poor stock are relevant.

The aims of the present study were to search whether elemental composition in muscle and liver of *T. obesus* captured in the central-south Atlantic Ocean and in two areas closer to the South America and South Africa continents differ, reflecting anthropogenic sources.

Material and Methods

Bigeye tuna (*Thunnus obesus*) specimens were caught in 2009 by Portuguese longliners targeting swordfish in the Central- and South Atlantic Ocean. Only the individuals captured off South American (AM, n=6) and African (AF, n=8) coastal areas, and in the open ocean at the Central-South Atlantic Ocean (OO, n=9) were considered **(Figure 1)**. Fork length (FL, cm) of the 23 tunas

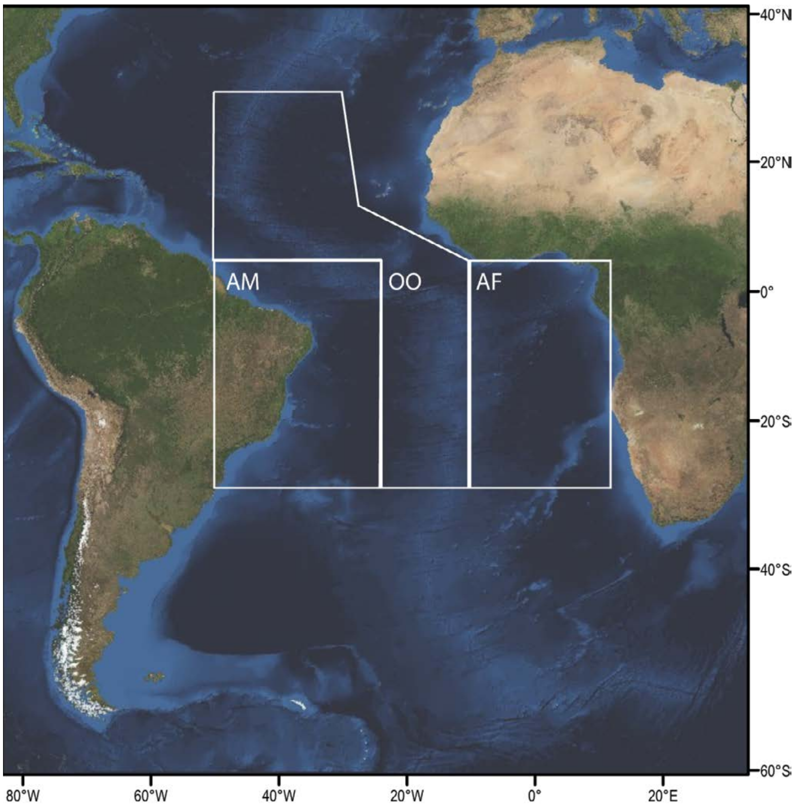

Figure 1: Capture areas of bigeye tuna (*Thunnus obesus*): South American coast (AM), South African coast (AF) and open ocean areas (OO).

were registered and dorsal muscle and liver samples collected. Individual age was estimated applying the growth equation (1) accepted by ICCAT for the Atlantic tuna (Hallier *et al.*, 2005),

$$L_t=217.3(1-e^{-0.18})^{(t+0.709)} \quad (1)$$

where L_t is the tuna length at an age *t*. Tissue samples were immediately stored at -20ºC in individual plastic bags. All labware was cleaned with HNO_3 (20%) and rinsed with Milli-Q water (18.2 M.cm) to minimize potential contamination. Vanadium (V), Cr, Co, Ni, Cu, Zn, As, Se, Cd and Pb concentrations in individual muscle and liver tissues were determined in freeze dry, ground and homogenised samples after digestion with HNO_3 (bidistilled, 65% v/v) and H_2O_2 (sp, 30% v/v) as detailed in Raimundo *et al.* (2013). Procedural blanks were prepared using the same analytical procedure and reagents, accounting for less than 1% of the total metal in samples. Metal concentrations were quantified using a quadropole ICP-MS (Thermo Elemental, X-Series). Analytical methods accuracy was assessed by the analysis of international certificate reference material (DORM-2 – dogfish muscle; DORM-3 – fish protein; DOLT-4 – fish liver). No significant (p>0.05, Mann-Whitney) differences were observed between obtained results and certified values. Metal concentrations were tested for normality of distribution and homogeneity of variances, the no agreement with parametric tests assumptions led to the application of the non-parametric tests Kruskal-Wallis H (KW-H) and Mann-Whitney (U). Tests were used to assess differences among element concentrations in tissues and among capture areas. Principal Component Analysis (PCA) was applied to the three capture areas (AM, AF and OO) for each tissue to search for element and area associations. Correlations between elemental concentrations and tuna biological parameters were calculated using Spearman correlation. The significance for statistical analyses used was always p<0.05. The statistical analyses were performed using STATISTICA (Statsoft).

Results and Discussion

Median sizes of the analyzed specimens varied from 140 cm FL in OO to 157 cm FL in AM and AF. No significant differences of size were observed (p>0.05) between areas. Individuals captured in the three areas presented a median age of 6 years, ranging from 5-8 years in AM, 3-8 years in AF and 4-8 years in OO. At these ages all tuna specimens are considered adults.

Median elemental concentrations (including all areas and both tissues) varied over broad ranges from 171 mg Kg^{-1} for Zn to 0.043 mg Kg^{-1} for Co, with the remaining elements varying according to the following order: As, Se > Cu > Cr > Cd > V > Ni > Pb. No correlations between concentrations and fish size were obtained (p>0.05), suggesting that within the studied range of ages, this biological parameter had a minor effect on metal variation in both tissues. The lack of relationships is in line with findings of Besada *et al.* (2006) for a broader range of sizes of *T. obesus*, *T. alalunga* and *T. albacores* captured in the Atlantic Ocean. Previous studies with other fish species from the Adriatic and Mediterranean also conclude that size has no effect on trace element accumulation in muscle and liver (Canli and Atli, 2003; Juresa and Blanusa, 2003). **Figure 2** showed that partitioning among tissues varied with the studied elements. Liver presented enhanced median concentrations of Co (0.47 mg Kg^{-1}), As (31 mg Kg^{-1}), Se (118 mg Kg^{-1}), Cu (70 mg Kg^{-1}), Zn (277 mg Kg^{-1}) and Cd (110 mg Kg^{-1}) compared with muscle (0.009, 7.7, 3.4, 1.2, 16, and 0.19 mg Kg^{-1}, respectively). All differences were statistically significant (p<0.05). Muscle and liver showed comparable median levels of Pb (0.10 mg Kg^{-1} and 0.051 mg Kg^{-1}, respectively), Ni (0.12 and 0.15 mg Kg^{-1}, respectively), V (0.18 and 0.17 mg Kg^{-1}, respectively) and Cr (0.67 and 0.60 mg Kg^{-1}, respectively). Data on organotropism exhibits distinct elemental composition among tissues varying with species (e.g. Mackay *et al.*, 1975; Canli and Atli, 2003; Deheyn *et al.*, 2005; Licata *et al.*, 2005). The organotropism is most likely related to tissue function and element affinity to certain proteins such as metallothioneins (MTs), besides the availability of elements in water and food (Hamilton and Mehrle, 1986; Roesijadi, 1992; De Smet *et al.*, 2001; Atli and Canli, 2003). Liver has an important capacity for storage, redistribution, detoxification or transformation of contaminants and is also considered as an active site of pathological effects induced by contaminants (Evans *et al.*, 1993). In general, elemental turnover rates are higher in liver than in muscle, whose elemental composition reflects intake on a longer time scale. Accordingly, liver/muscle ratios are used as a tool to evaluate recent element accumulation. Although Cd and Pb are considered elements with no biological role, ratios obtained for Cd (213-720) were up to three orders of magnitude higher than for Pb (0.28-2.0) **(Figure 3)**. The high ratios found for Cd, followed by Se, Cu and Co (three essential elements), support the hypothesis of the central role of liver in the accumulation and functioning as elemental storage. However, this hypothesis is not applicable to Pb, since median and maximum concentrations **(Figure 2)** in the muscle were above levels registered in the liver. Thus, ratios lower than the unit for Pb suggest increased affinity of this element to the muscle tissues as previously observed in tunas and other fish

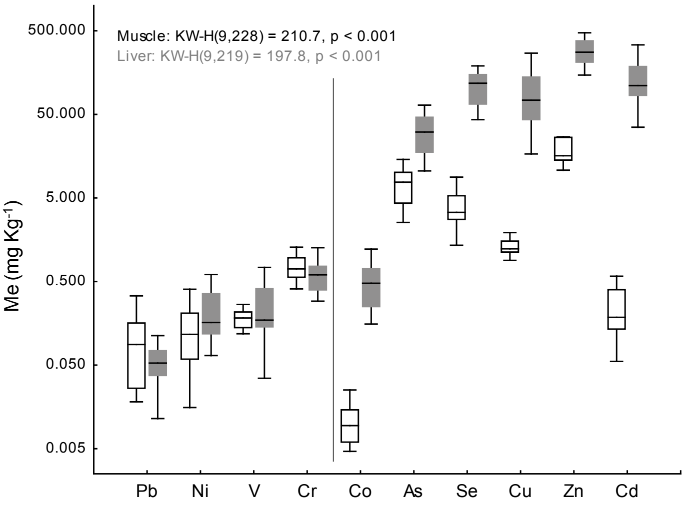

Figure 2: Median, 25 and 75 percentiles, minimum and maximum concentrations of V, Cr, Co, Ni, Cu, Zn, As, Se, Cd and Pb (mg Kg^{-1}, dw) in the muscle (white boxplots) and liver (grey boxplots) of *Thunnus obesus* captured in South American coast (AM), South African coast (AF), and in the open ocean areas (OO). The y-axis is in the log10 scale.

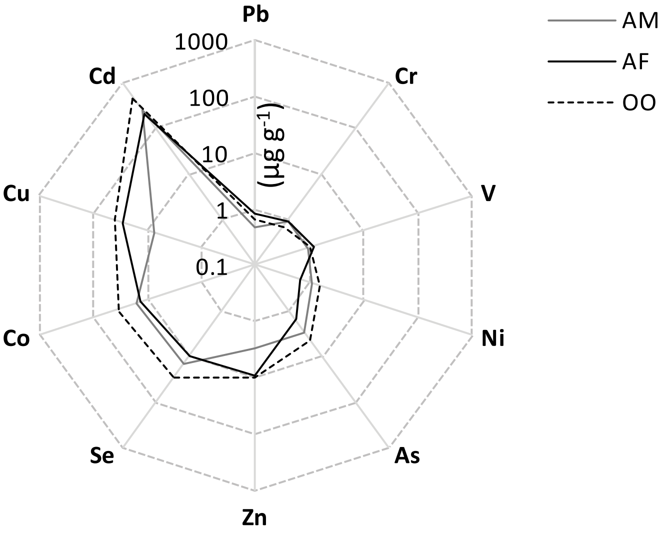

Figure 3: Liver/muscle ratios for V, Cr, Co, Ni, cu, z, As, Se, Cd and Pb in *Thunnus obesus* captured in South American coast (AM), South African coast (AF), and in the open ocean areas (OO). The axis is in the log10 scale.

species (e.g., Atli and Canli, 2003; Vizzini *et al.*, 2010). For Cd, Cu, Co, Se, Zn, As and Ni, ratios were comparable in the three sampling areas or slightly higher in specimens from the central Atlantic area (OO). The tissue elemental regulation, as a possible detoxification mechanism, may be a common feature of tuna species. The effectiveness of the mechanism preventing toxicity effects depends on the element. In fact, similarly high liver/muscle Cd ratios (110-726) can be considered for albacore tuna (*Thunnus alalunga*) captured in the Mediterranean Sea (Storelli and Marcotrigiano, 2004).

In general, obtained values in liver and muscle were comparable or above the ones reported for various tuna species (*T. obesus*, *T. thynnus*, *T. alalunga*, *T. albacores*) from the Mediterranean Sea (Hernández-Hernández *et al.*, 1990; Storelli and Marcotrigiano, 2004; Percin *et al.*, 2011), Australia (Padula *et al.*, 2008) and Atlantic Ocean (Besada *et al.*, 2006). Levels of contaminants in the muscle have been determined in several tuna species to evaluate its importance as a dietary source for humans (e.g. Storelli and Marcotrigiano, 2004; Besada *et al.*, 2006; Percin *et al.*, 2011). Despite most studies only points Hg as a problematic metal for human consumption (e.g. Storelli *et al.*, 2002; Besada *et al.*, 2006; Vizzini *et al.*, 2010), 26% of the studied specimens in our study presented levels of Cd (0.10-0.12 mg Kg^{-1}, ww) close to the maximum allowed for human consumption (0.10 mg Kg^{-1}, ww; European Directive 1259/2011). For Pb, all samples presented concentrations up to two orders of magnitude below (0.004-0.086 mg Kg^{-1}, ww) the admissible value (0.40 mg Kg^{-1}, ww). The same tendency was found for Cu and Zn considering the limits proposed by Australia (Cu: 10 mg Kg^{-1}; Zn: 150 mg Kg^{-1}) and Canada (Cu: 100 mg Kg^{-1}; Zn: 100 mg Kg^{-1}) (Vizzini *et al.*, 2010).

To evaluate the spatial distribution of elemental composition a principal component analysis (PCA) was done for liver and muscle separately **(Figure 4)**. Geographic separation was not clearly observed in muscle, with no association of trace elements with a specific area. Muscle appears to integrate the element accumulation during tuna life time. On the other hand, ordination based on the liver elemental composition **(Figure 4A)**, separates specimens captured closer to the coast (AM and AF) from the open ocean (OO) (accounting for 23% of the variability in the element concentrations). Most elements (V, Co, Cu, Zn, As, Cd and Pb) showed enhanced values in AM and AF regions. Moreover, a separation between specimens from the AM and AF coasts can also be discerned (accounting for 32% of the variability in the element concentrations). Geographic variation may be described by changes in the species' dietary and/or trophic position among areas. Tunas from the AF coast presented enhanced levels of V, Co, Cu, Zn, Cd and Pb compared to tunas from AM. These geographical differences found in the South Atlantic tuna population are in line with data reported in the environment. Studies carried out in the Atlantic waters showed higher Cd and Co concentrations in the Guinea Dome and Angola Dome (Bowie *et al.*, 2002; Pohl *et al.*, 2011). Enhanced levels of Cu were also reported in the African coast and linked to sources existing both in the continent and in the shelves (Loeff *et al.*, 1997). Despite limited information is published on trace element concentration in other fish species from this remote area (Gnandi *et al.*, 2013; Bandowe *et al.*, 2014), increased concentration of Cd, Co and Cu in water may influence the levels found in tuna's prey. Being the increase of concentrations found in the liver suggests that food is the accumulation pathway. Despite that tunas have long range migratory movements across the Atlantic, the enhanced accumulation of Cd, Co and Cu in specimens from AF, as in the environment, allows to geolocate them in that caught area. Therefore, the concentration of these elements in liver of tunas can be used as a natural tag to distinguish *T. obesus* from different areas. The similar concentrations of Ni in water from south America (AM) and African (AF) coast (Bowie *et al.*, 2002) was reflected in liver of analysed tunas with similar levels between the two areas. For As, tunas captured in the AM area presented higher concentrations in liver. According to Cutter *et al.* (2001), concentrations of this metalloid in water are, slightly higher in the western and equatorial Atlantic than in the eastern Atlantic. The increased concentrations were found in the Brazil Current, which is within the caught area of tunas (AM) that showed higher levels of As. Despite arsenic biomagnification in the marine organisms is not consensual, several studies reveal the accumulation of high concentrations in marine organism tissues and through the food chain (Rahman *et al.*, 2012).

Conclusions

Although this specie have a long range migratory movements across the Atlantic, the obtained results showed that sub-populations of tuna *T. obesus* from the central-south Atlantic areas can be identified using tissue elemental concentrations as a natural tag. During migration, feeding habits change associated with prey availability and consumption, presumably ingesting food with different contamination levels. Due to its role, liver seems to be the best indicator tissue for distinguishing T. *obesus* sub-populations.

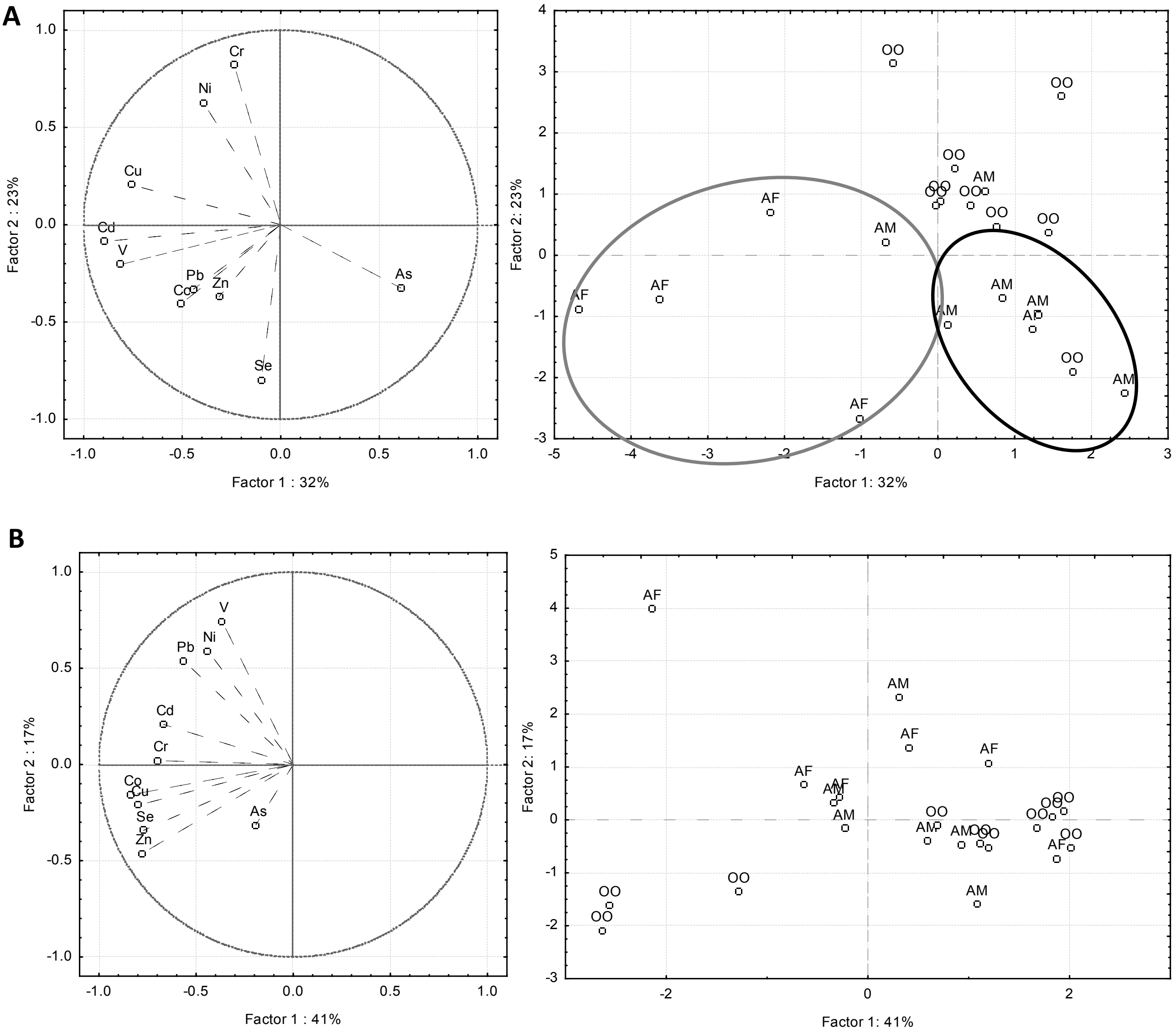

Figure 4: Principal component analysis (PCA) of V, Cr, Co, Ni, Cu, Zn, As, Se, Cd and Pb concentrations (mg Kg^{-1}, dw) in liver (A) and muscle (B) of *Thunnus obesus* captured in the South American coast (AM), South African coast (AF), and in the open ocean areas (OO).

Acknowledgments

The samples for this study were collected during experimental fishing conducted within the scope of the SELECT-PAL project. Thanks are due to João Táta Regala and Sérgio Amorim, for collecting the samples. Joana Raimundo acknowledges the pos-doctoral grant by FCT (SFRH/BPD/91498/2012). Rui Coelho is supported by an Investigador-FCT contract from FCT (IF/00253/2014), supported by the EU European Social Fund and the Programa Operacional Potencial Humano.

References

Aguilar, A., Borrel, A., Reijnders, P.J.H. (2002) Geographical and temporal variation in levels of organochlorine contaminants in marine mammals. Mar Environ Res **53,** 425-452.

Anon (2005) 2004 ICCAT bigeye tuna year program symposium. Collect Vol Sci Pap- ICCAT **57,** 3-60.

Arrizabala, H., Pereira, J.G., Royer, F., Galuardi, B., Goñi, N, et al. (2008) Bigeye tuna (Thunnus obesus) vertical movements in the Azores Islands determined with pop-up satellite archival tags. Fish Oceanog **17,** 74-83.

Atli, G., Canli, M. (2003) Natural occurrence of metallothionein-like proteins in the liver of fish Oreochromis niloticus and effects of cadmium, lead, copper, zinc, and iron exposures on their profiles. Bull Environ Contam Toxicol **70,** 618-627.

Bandowe, B., Bigalke, M., Boamah, L., Nyarko, E., Saalia, F, et al. (2014) Polycyclic aromatic compounds (PAHs and oxygenated PAHs) and trace metals in fish species from Ghana (West Africa): Bioaccumulation and health risk assessment. Environ Int **65,** 135-146

Besada, V., González, J.J., Schultze, F. (2006) Mercury, cadmium, lead, arsenic, copper and zinc concentrations in albacore, yellowfin tuna and bigeye tuna from the Atlantic Ocean. Ciencias Marinas **32,** 439-445.

Block, B.A., Dewar, H., Blackwell, S.B., Williams, T.D., Prince, E.D., et al. (2001) Migratory movements, depth preferences, and thermal biology of Atlantic bluefin tuna. Science **293,** 1310-1314.

Block, B.A., Jonsen, I.D., Jorgensen, S.J., Winship, A.J., Shaffer, S.A., et al. (2011). Tracking apex marine predator movements in a dynamic ocean. Nature **475,** 86-90.

Canli, M., Atli, G. (2003) The relationships between heavy metal (Cd, Cr, Cu, Fe, Pb, Zn) levels and the size of six Mediterranean fish species. Environ Pollut **121,** 129-136.

Chow, S., Okamoto, H., Miyabe, N., Hiramatsu, K., Barut, N. (2000). Genetic divergence between Atlantic and Indo-Pacific stocks of bigeye tuna (Thunnus obesus) and admixture around South Africa. Mol Ecol **9,** 221-227.

De Smet, H., De Wachter, B., Lobinski, R., Blust, R. (2001) Dynamics of (Cd, Zn)-metallothionein in gills, liver, and kidney of common carp Cyrpinus carpio during cadmium exposure. Aquat Toxicol **52,** 269-281.

Deheyn, D.D., Gendreau, P., Baldwin, R.J., Latz, M.I. (2005) Evidence for enhanced bioavailability of trace elements in the marine ecosystem of Deception Island, a volcano in Antarctica. Mar Environ Res **60,** 1-33.

Engman, J., Jorhem, L. (1998) Toxic and essential elements in fish from Nordic waters, with the results seen from the perspective of analytical quality assurance. Food Addit Contam **15,** 884-892.

Evans, D.W., Doo, D.K.Do., Hanson, P. (1993) Trace element concentration in fish livers: implication of variations with fish size in pollution monitoring. Mar Pollut Bull **26,** 329-354.

Gnandi, K., Bandowe, B., Deheyn, D., Porrachia, M., Kerstend, M., et al. (2013) Polycyclic aromatic hydrocarbons and trace metal contamination of coastal sediment and biota from Togo. J Environ Monit **13,** 2033.

Hamilton, S.J., Mehrle, P.M. (1986) Metallothionein in fish: Review of its importance in assessing stress from metal contaminants. T Am Fish Soc **115,** 596-609.

Heithaus, M.R., Frid, A., Wirsing, A.J., Worm, B. (2008) Predicting ecological consequences of marine top predator declines. Trends Ecol Evol **23,** 202-210.

Hernández-Hernández, F., Medina, J., Ansuátegui, J., Conesa, M. (1990) Heavy metal concentrations in some marine organisms from the Mediterranean Sea (Castellón, Spain): Metal accumulation in different tissues. Scientias Marinas **54,** 113-129.

Hobson, K.A., Riget, F.F., Outridge, P.M., Dietz, R., Born, E. (2004). Baleen as a biomonitor of mercury content and dietary history of North Atlantic Minke Whales (Balaenopetra acutorostrata): combining elemental and stable isotope approaches. Sci Total Environ **331,** 69-82.

Ikem, A., Egiebor, N.O. (2005) Assessment of trace elements in canned fishes (mackerel, tuna, salmon, sardines and herrings) marketed in Georgia and Alabama (United States of America). J Food Comp Anal **18,** 771-787.

Juresa, D., Blanusa, M. (2003) Mercury, arsenic, lead, and cadmium in fish and shellfish from the Adriatic Sea. Food Addit Contam **20,** 241-246.

Khansari, F.E., Ghazi-Khansari, M., Abdollahi, M. (2005). Heavy metals content of canned tuna fish. Food Chem **93,** 293-296.

Korsmeyer, K.E., Dewar, H. (2001) Tuna metabolism and energetics. In: Block, Stevens (Eds.), Tuna Physiology, Ecology, and Evolution. Academic Press, San Diego, California pp: 36-78.

Lehodey, P., Hampton, J., Leroy, B. (1999) Preliminary results on age and growth of bigeye tuna (Thunnus obesus) from the western and central pacific ocean as indicated by daily growth increments and tagging data. Oceanic Fisheries Programme, Secretariat of the Pacific Community Noumea, New Caledonia.

Licata, P., Trombetta, D., Cristani, M.T., Naccari, C., Martino, D., et al. (2005) Heavy metals in liver and muscle of bluefin tuna (Thunnus thynnus) caught in the Straits of Messina. Environ Monit Assess **107,** 239-248.

Macdonald, J.I., Farley, J.H., Clear, N.P., Williams, A.J., Carter, T.I. et al. (2013) Insights into mixing and movement of South Pacific albacore Thunnus alalunga derived from trace elements in otoliths. Fish Res **148,** 56-63.

Mackay, N.J., Kazacos, M.N., Williams, R.J., Leedow, M.I. (1975) Selenium and heavy metals in Black Marlin Mar Pollut Bull **6,** 57-61.

Madigan, D.J., Baumann, Z., Carlisle, A.B., Hoen, D.K., Popp, B.N., et al. (2014) Reconstructing transoceanic migration pattern of Pacific bluefin tuna using a chemical tracer toolbox. Ecology **95,** 1674-1683.

Padula, D.J., Daughtry, B.J., Nowak, B.F. (2008) Dioxins, PCBs, metals, metalloids, pesticides and antimicrobial residues in wild and farmed Australian southern bluefin tuna (Thunnus maccoyii). Chemosphere **72,** 34-44.

Patterson, T.A., Evans, K., Carter, T.I., Gunn, J.S. (2008) Movement and behavior of large southern bluefin tuna (Thunnus maccoyii) in the Australian region determined using pop-up satellite archival tags. Fish Oceanography **17,** 352-367.

Percin, F., Sogut, O., Altınelataman, C., Soylak, M. (2011) Some trace elements in front and rear dorsal ordinary muscles of wild and farmed bluefin tuna (Thunnus thynnus L. 1758) in the Turkish part of the eastern Mediterranean Sea. Food Chem Toxicol **49,** 1006-1010.

Rahman M., Hasegawa, H., Lim, R. (2012) Bioaccumulation, biotransformation and trophic transfer of arsenic in the aquatic food chain. Environ Res **116,** 118-135.

Raimundo, J., Vale, C., Caetano, M., Anes, Giacomello, E., Menezes, G. (2013) Trace-element concentrations in muscle and liver of 11 commercial fish species from Condor Seamount, Azores Archipelago (Portugal). Deep-Sea Res **98,** 137-147.

Ramos, R., González-Solís, J. (2012) Trace me if you can: the use of intrisic biogeochemical markers in marine top predators. Front Ecol Environ **10,** 258-266.

Roesijadi, G. (1992) Metallothioneins in metal regulation and toxicity in aquatic animals. Aquat Toxicol 22, 81-114.

Storelli, M.M., Marcotrigiano, G.O. (2004) Content of mercury and cadmium in fish (Thunnus alalunga) and cephalopods (Eledone moschata) from the south-eastern Mediterranean Sea. Food Addit Contam **21,** 1051-1056.

Storelli, M.M., Giacominelli, R., Storelli, A., Marcotrigiano, G.O. (2005) Accumulation of mercury, cadmium, lead and arsenic in Swordfish and Bluefin tuna from the Mediterranean Sea: a comparative study. Mar Pollut Bull **44,** 281-288.

Sun, C.L., Huang, C.L. , Yeh, S.Z. (2001) Age and growth of the bigeye tuna, Thunnus obesus, in the western Pacific Ocean. Fish Bull **99,** 502-509.

Vizzini, S., Tramati, C., Mazzola, A. (2010) Comparison of stable isotope composition and inorganic and organic contaminant levels in wild and farmed bluefin tuna, Thunnus thynnus, in the Mediterranean Sea. Chemosphere **78,** 1236-1243.