Comparative chemical composition of different muscle zones in angler (*Lophius piscatorius*)

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ABSTRACT (ojo 206)

The present study focuses on the lipid composition of angler (*Lophius piscatorius*) muscle. Distribution of lipid classes and groups and fatty acid composition of neutral and polar lipid fractions corresponding to two edible sites (head and tail) were investigated. Proximate composition and essential and toxic mineral contents were also analysed. Moisture, total lipid and protein contents ranged from 828-845, 3.0-3.7 and 135-170 g kg^{-1} muscle, respectively. Phospholipids were found to be the most abundant lipid group (690-720 g kg^{-1} total lipids) and triacylglycerol presence was very low (3.2-7.7 g kg^{-1} total lipids). The most abundant fatty acid group comprised polyunsaturated fats. In both neutral and polar lipid fractions, 22:6 n-3 was found to be the major fatty acid, followed by 16:0 and 18:1 n-9. Nutritionally valuable high concentrations of Zn and Mn were observed, and As was the most abundant toxic element. Higher protein and trimethylamine oxide values were observed in the tail site, but lower in moisture, sterol and triacylglycerol contents; however, an inhomogeneous muscle distribution could not be demonstrated with regards to total lipids, phospholipids, free fatty acids or α-tocopherol contents. Mean concentrations of essential and toxic elements were higher in the head site in most cases, especially for Zn, Cd, Cr and V.

Running Title: Chemical composition of angler muscle

Keywords: food analysis; food composition; *Lophius piscatorius*; muscle; head; tail; proximate composition; lipid fractions; fatty acids; minerals

ABBREVIATIONS LIST

DHA: docosahexaenoic acid
EPA: eicosapentaenoic acid
FAME: fatty acid methyl esters
FFA: free fatty acids
MUFA: monounsaturated fatty acids
NL: neutral lipids
PL: phospholipids
POL: polar lipids
PUFA: polyunsaturated fatty acids
SFA: saturated fatty acids
STS: sterols
TAG: triacylglycerols
TMAO: trimethylamine oxide

1. INTRODUCTION

Seafood products are known to provide significant amounts of different beneficial nutrients such as nutritional and digestible proteins, lipid-soluble vitamins, essential minerals and highly unsaturated fatty acids (Aitken et al., 1982; Simopoulos, 1997). Most of these constituents have been shown to play important roles in human diet. Among them, the lipid fraction is now the subject of a great deal of attention due to its high content of n-3 polyunsaturated fatty acids (n-3 PUFA), which have exhibited a positive effect in the prevention of certain human diseases (Ackman & Ratnayake, 1990; Weber, 1992).

Chemical constituents of marine species have been proven to be inhomogeneously distributed throughout the body, most likely as a result of physiological and anatomical factors (Pearson et al., 1977). The lipid matter has been recognised as the most strongly affected of these constituents, with wide differences in lipid content among tissues in both fatty (Piclet, 1987; Gallardo et al., 1989) and lean (Aubourg et al., 1999; Aubourg et al., 2007) fish. Additionally, previous studies have shown that mineral concentrations of fish muscle zones may be influenced by different biological factors (Lal, 1995; Noël et al., 2011).

Angler (*Lophius piscatorius*) is a monkfish belonging to the *Lophiidae* family. It has a very large head, which is broad, flat and depressed, making the rest of the body (normally called the tail) appear to be a mere appendage. This species is widely found in coastal waters of the northeast Atlantic, from the Barents Sea to the Strait of Gibraltar, the Mediterranean Sea and the Black Sea. This high-value commercial species has long attracted a great deal of interest because of its firm and flavourful flesh, and the countries that have reported the greatest capture volumes in the last decade are France, UK, Denmark, Spain and Norway (FAO, 2007).

Previous research related to angler has focused on the assessment of contaminant components, such as organochlorine compounds (Bordajandi et al., 2006) and toxic minerals (Mormede & Davis, 2001; Afonso et al., 2008; Anacleto et al., 2009). Preliminary research into quality changes in this species during storage or technological processing has assessed microbial presence assessment (Mediel et al., 2000), volatile amine formation (Ruiz-Capillas & Horner, 1999), nucleotide degradation (Mendes et al., 2001) and employment of advanced chilling strategies (Barros-Velázquez et al., 2008; García-Soto et al., 2011). Previous research related to chemical composition and nutritional value of angler, is scarce and has focused on proximate and amino acid composition (Jhaveri et al., 1984), fatty acid composition (Sirot et al., 2008), and assessment of α-tocopherol and essential minerals (Jhaveri et al., 1984; Carvalho et al., 2005).

The primary goal of the present study was to investigate the lipid composition of angler muscle. For this purpose, the distribution of lipid classes and groups and the fatty acid composition of neutral and polar lipid fractions corresponding to two edible sites (head and tail) were investigated. The work also focused on the proximate composition and on essential and toxic mineral assessments.

2. MATERIALS AND METHODS

2.1. Fish material and sampling

Angler individuals (n = 20; 300-450 g weight; 24-30 cm length) were captured in the North Atlantic Fishing Bank of the Grand Sole, unloaded in Vigo Harbour and transported to our laboratory. From the time of capture until arrival at the laboratory, the fish were stored in ice. In all cases, gonads of individual fishes were at the $5^{th}/6^{th}$ stage of Maier's scale of gonad maturity.

The fish were distributed into five groups (four individuals per group), which were considered independently in the statistical analysis (n = 5). Within each fish group, the white muscle of two different zones (head and tail) was considered and carefully separated. The absence of bones, blood and dark muscle was verified. Chemical analyses were carried out separately on each of the selected zones of each fish group. Solvents and chemical reagents used in this study were of reagent grade (Merck, Darmstadt, Germany); in the case of tocopherol analysis, solvents used were liquid chromatographic grade.

2.2. General composition analysis

Moisture content was determined as the weight difference in the homogenised muscle (1-2 g) before and after 4 h at 105ºC (AOAC, 1990). Results were calculated as g water kg^{-1} muscle.

Protein content was measured using the Kjeldahl method (AOAC, 1990) with a conversion factor of 6.25. Results were calculated as g protein kg^{-1} muscle.

The total lipid fraction was extracted using the Bligh and Dyer (1959) method and was quantified as g total lipid kg^{-1} muscle. Throughout the study, total lipid extracts were kept at –40ºC in a nitrogen atmosphere.

Trimethylamine oxide (TMAO) content was determined using a previous reduction of a trichloracetic acid extract of the muscle with titanium (III) chloride (Parkin & Hultin, 1982); the trimethylamine content was assessed using the spectrophotometric method described by Tozawa et al. (1971). Results were expressed as mg TMAO-nitrogen (TMAO-N) kg^{-1} muscle.

2.3. Lipid classes and groups analysis

Phospholipids (PL) were quantified by measuring the organic phosphorus in the total lipid extracts according to the Raheja et al. (1973) method, which is based on a complex formation with ammonium molybdate. Results were calculated as g PL kg^{-1} muscle and as g PL kg^{-1} total lipids.

Sterols (STS) were determined on total lipid extracts by the method of Huang et al. (1961) based on the Liebermann-Buchardt reaction. Results were calculated as g STS kg^{-1} muscle and as g STS kg^{-1} total lipids.

To measure the triacylglycerol (TAG) content, the total lipid extracts were first purified on 20 x 20 cm thin-layer chromatography plates coated with a 0.5 mm-layer of silica gel G from Merck (Darmstadt, Germany) using a mixture of hexane-ethyl ether-acetic acid (90-10-1, v-v-v; two developments) as eluent (Aubourg et al., 1991). Once the TAG fraction was purified, the method of Vioque and Holman (1962) was used to measure the ester linkage content according to the conversion of the esters into hydroxamic acids and subsequent complexion with Fe (III). Results were calculated as g TAG kg^{-1} muscle and as g TAG kg^{-1} total lipids.

Free fatty acid (FFA) content of the total lipid extracts was determined following the Lowry and Tinsley (1976) method, which is based on the formation of a complex with cupric acetate-pyridine. In this study, benzene was replaced by toluene as organic solvent. Results were calculated as g FFA kg^{-1} muscle and as g FFA kg^{-1} total lipids.

Tocopherol isomers were analysed according to the Cabrini et al. (1992) method. In this analysis, tocopherols were extracted from the muscle with hexane, dried under nitrogen flux, dissolved in isopropanol and injected into an HPLC system (ODS column, 15 cm x 0.46 cm i.d.); detection was performed at 280 nm. The presence of different tocopherol isomers (α, β, γ and δ) was assessed. Only the α-tocopherol isomer

was detected, and its content was calculated with a calibration curve prepared from commercial α-tocopherol. Results were expressed as mg α-tocopherol kg^{-1} muscle and as g α-tocopherol kg^{-1} total lipids.

2.4. Fatty acid analysis

Total lipid extracts from both muscle sites were fractionated on pre-packed Sep-Pak® SEP amino propyl cartridges (Waters, Dublin, Ireland). For this analysis, cartridges were first activated by successive elutions of CH_3OH (3 mL), $CHCl_3$ (5 mL) and hexane (5 mL). Then, 1 mg of total lipid extracts was dissolved in 1 mL $CHCl_3$ and applied to the cartridge. Neutral lipids (NL) were eluted with 3 mL $CHCl_3$, whereas polar lipids (POL) were eluted after addition of 2 mL CH_3OH. Throughout the study, NL and POL fractions were kept at –40ºC in a nitrogen atmosphere.

NL and POL extracts were converted into fatty acid methyl esters (FAME) by using acetylchloride and were then analysed by gas-liquid chromatography (Perkin-Elmer 8700 chromatograph, Madrid, Spain) according to an established procedure (Aubourg et al., 1991). For it, a fused silica capillary column SP-2330 (0.25 mm i.d. x 30 m, Supelco, Inc., Bellefonte, PA, USA) was employed and the temperature program was as follows: increased from 145 to 190ºC at 1.0ºC/min and from 190ºC to 210ºC at 5.0ºC/min; held for 13.5 min at 210ºC. The carrier gas was nitrogen at 10 psig and detection was performed with a flame ionisation detector at 250ºC. A programmed temperature vaporiser injector was employed in the split mode (150:1) and was heated from 45 to 275ºC at 15ºC/min. Peaks corresponding to FAME were identified by comparing their retention times with those of standard mixtures (Qualmix Fish, Larodan, Malmo, Sweden; FAME Mix, Supelco, Inc.). Peak areas were automatically integrated; 19:0 fatty acid was used as internal standard for quantitative purposes. Content of each fatty acid was expressed as mg kg^{-1} muscle.

2.5. Analysis of essential and toxic minerals analysis

Two groups of elements were analysed. The first group included five essential elements (Co, Cu, Fe, Mn and Zn) that are present in all animal tissues (Johnson & Fischer, 1994; Lal, 1995). The second group comprised seven elements (As, Cd, Cr, Hg, Ni, Pb and V), which are referred to as toxic and thought to reflect exogenous influences related to environmental pollution (Engman & Jorhem, 1998; Noël et al., 2011).

Samples corresponding to head and tail sites were dried in a stove at 50ºC to constant weight and subsequently ground in an agate mortar. A 325±20 mg fraction of each sample was weighed in a 55 mL Teflon vessel and a mixture of 6 mL of 69% HNO_3, 2 mL of 30% H_2O_2 and 4 mL of Milli-Q water was added prior to the microwave digestion. Then, samples of fish muscle were introduced into a Mars-X-Press CEM (Matthews, NC, USA) microwave oven with three blanks and three samples of certified reference material (DORM-2 from the National Research Council of Canada; Table 1), which were included to verify the success of the analytical procedure. Samples were completely digested and diluted to a volume of 50 mL; sample handling was accomplished inside of a clean cabinet (Cruma 670 FL, Barcelona, Spain).

In the digested samples, the mineral concentrations of As, Cd, Co, Cr, Cu, Mn, Ni, Pb, V and Zn were determined using a quadrupole ICP-MS (Thermo Elemental, X-Series, Bremen, Germany) equipped with a Peltier Impact bead spray chamber and a concentric Meinhard nebulizer. The experimental parameters were as follows: forward power of 1400 W; peak jumping mode; 150 sweeps per replicate; dwell time of 10 ms, and dead time of 30 ns. Two procedural blanks were prepared using the analytical procedures and reagents described above and were included in each batch of 20

samples. Quantification was achieved by single mode analysis, by applying the corrections provided by the equipment software for polyatomic and isobaric interferences.

Fe was determined by flame atomic absorption spectrometry (Varian 220 FS, Victoria, Australia) with a nitrous oxide-acetylene flame. Hg was determined by atomic absorption spectrometry using a silicon UV diode detector Leco AMA-254 (Plzen, Czech Republic) after pyrolysis of each sample in a combustion tube at 750°C under an oxygen-rich atmosphere and collection on a gold amalgamator (Costley et al., 2000). In all cases, procedural blanks accounted for less than 1% of element concentrations in the samples. The precision and accuracy of the analytical procedures was controlled through repeated analyses of certified reference materials containing the studied elements (Table 1). Quantification results are expressed as mg kg^{-1} muscle.

2.6. Statistical analysis

Data (n = 5) obtained from the different chemical analyses were subjected to the ANOVA method ($p<0.05$) to investigate differences between body zones, i.e., the head and tail comparison (Statsoft Inc., *Statistica*, version 6.0, 2001; incluir city, state y country ??). Comparison of means was performed using a least-squares difference (LSD) method.

3. RESULTS AND DISCUSSION

3.1. General composition analysis

Moisture and total lipid contents ranged from 828-845 and 3.0-3.7 g kg^{-1} muscle (Table 2), respectively, in good agreement with the results reported in previous research carried out on angler muscle (Jhaveri et al., 1984; Barros-Velázquez et al., 2008; García-Soto et al., 2011). Total lipid range can be considered low compared to the lipid ranges of other lean fish species such as hake (*Merluccius merluccius*) (Aubourg et al., 1999) and turbot (*Psetta maxima*) (Aubourg et al., 2007). Additionally, moisture content was relatively high, which corresponds the known inverse ratio between water and lipid constituents in fish (Pearson et al., 1977; García-Arias et al., 1994). Comparison of the two muscle sites indicated a higher moisture content in the head location than the tail location; consequently, and according to the above-mentioned inverse ratio with lipid matter, a lower mean value was observed for the total lipid content in the head muscle zone.

Protein content range from 135-161 g kg^{-1} muscle (Table 2), in agreement with the contents reported by previous research on proximate composition of angler muscle (Jhaveri et al., 1984). Compared to most lean and fatty fish species, this range of protein contents can be considered relatively low, particularly for the head zone (Aitken et al., 1982; Piclet, 1987). However, digestibility value of angler muscle has been reported as 91.3% (Jhaveri et al., 1984), which places it in the digestibility range of teleostean fish species (88-96%; Piclet, 1987). An inhomogeneous distribution of protein content was observed in the present work, with a higher protein level observed in the tail zone.

The TMAO molecule has been recognised as ubiquitous constituent of marine species that plays an important role in osmoregulation during the life of the fish (Aitken et al., 1982). After the death of the fish and during the processing and storage steps, TMAO can induce different types of degradation that lead to the formation of different molecular spoilage indicators, such as trimethylamine, dimethylamine and formaldehyde. As a result of the formation of such spoilage and breakdown molecules, an important loss of quality is sustained (Finne, 1992; Huss, 1995).

In the present work, TMAO-N assessment revealed an inhomogeneous distribution of this molecule between the two sites under study (Table 2), with the highest level observed in the tail zone. A higher TMAO concentration in tail muscle compared to the dorsal and ventral muscle zones has already been reported in farmed and wild blackspot seabream (*Pagellus bogaraveo*) (Álvarez et al., 2009). This difference in TMAO distribution can be explained on the basis of a greater interchange surface with the environment in the tail zone. Consistent with the role of the TMAO molecule in osmoregulation, a higher content of this molecule would be necessary in the tail zone to prevent dehydration of the fish body.

TMAO content has also been shown to be dependent on species, area of capture, and size and condition of fish (Aitken et al., 1982). Thus, a higher content has been recorded in wild individuals than in their farmed counterparts in species such as cod (*Gadus morhua*) (Herland et al., 2007) and blackspot seabream (*P. bogaraveo*) (Álvarez et al., 2009). Previous research has shown that the highest TMAO-N contents can be found in elasmobranches, squids and gadoid fish (750-2500 mg kg^{-1} muscle). The present results indicated that the TMAO-N content in angler muscle can be considered relatively low, being similar to values reported for flat and pelagic fish species (Finne, 1992; Huss, 1995).

3.2. Total lipid composition analysis

Total lipid composition was investigated by analysing lipid classes and groups and determining the fatty acid composition of NL and POL fractions.

3.2.1. Lipid classes and groups

Table 3 shows the contents of different lipid classes and groups observed in the two muscle locations; results are expressed on both muscle and total lipid basis.

PL have been shown to represent the most abundant total lipid group (Table 3). Their concentration of PL did not differ between the two locations. This lipid group has been reported to be an important constituent of cell membranes and supports a structural role in living bodies, so its presence in muscle is hardly affected by internal factors such as anatomical and physiological characteristics (Pearson et al., 1977). This lack of content differences between the muscle sites is in agreement with previous research on albacore (*Thunnus alalnuga*) (Gallardo et al., 1989) and on lean fish species such as turbot (*P. maxima*) (Aubourg et al., 2007) and blackspot seabream (*P. bogaraveo*) (Álvarez et al., 2009).

The range of values found for STS (Table 3) is higher than that previously reported by Jhaveri et al. (1984) for the same species (0.186 g kg^{-1} muscle). Additionally, an inhomogeneous distribution between the two zones was observed for the STS group in this investigation; in terms of both muscle and total lipid contents, a higher concentration of this lipid group was observed in the head zone. STS are reported to contribute to functional properties and play structural roles in living bodies (Pearson et al., 1977; Aubourg et al., 1991); accordingly, no differences in the STS distribution were observed in different zones of blackspot seabream (*P. bogaraveo*) (Álvarez et al., 2009) or in albacore tuna (*T. alalunga*) (Gallardo et al., 1989). However, previous research conducted on wild and farmed turbot (*P. maxima*) showed that a marked STS accumulation was achieved in the edge zone, especially in the case of farmed fish (Aubourg et al., 2007).

TAG values were generally lower than those obtained for STS and PL (Table 3). Values range obtained for TAG is in agreement with the lipid class distribution of a lean fish species (Pearson et al., 1977; Álvarez et al., 2009) but is markedly different from

the corresponding profile of a fatty fish species (Gallardo et al., 1989). Higher TAG mean contents were obtained from the head muscle than from the tail (Table 3); when expressed in terms of the total lipid content, this difference was significant.

Value ranges found in the present study for the presence of FFA correspond to very low development of lipid hydrolysis in a lean fish species (Aubourg et al., 1999; Aubourg et al., 2007). Because the analysis was carried out after the catching/slaughtering steps, values obtained in the present research should correspond to the in vivo metabolic action of lipases and phospholipases on high-molecular-weight lipids such as TAG and PL, respectively. No difference in the hydrolytic enzyme activity of the different muscle zones under study was observed, although higher mean values were detected in the head site.

The α-tocopherol contents obtained for fish muscle are expressed in Table 3. Values obtained were higher than those reported for the same species by Afonso et al. (2008) (1.1±1.4 mg kg^{-1} muscle). Table 3 shows that a higher mean value was found in the tail zone than in the head zone, although differences were not found significant. However, an inhomogeneous distribution of α-tocopherol in muscle has been observed in other wild fish, such as turbot (*P. maxima*) (Aubourg et al., 2007) and Atlantic halibut (*Hippoglossus hippoglossus*) (Ruff et al., 2004); in such experiments, higher α-tocopherol contents were observed in edible zones that had higher total lipid contents. In the current experiment, a higher mean total lipid content in the tail zone was accompanied by a higher mean α-tocopherol presence in the same site (Tables 2-3).

Tocopherols are known lipid-soluble chain-breaking antioxidants, and the main role of these antioxidants is to protect the unsaturated fatty acids from oxidation. Different isomers (α-, β-, γ- and δ-) have been identified in plants and all have been found in most seaweeds and unicellular algae. However, α-tocopherol has been reported to be the only tocopherol that accumulates in higher marine animals from natural diets (Sigurgisladóttir et al., 2003). When farmed fish are considered, deposition of different tocopherol isomers (primarily α- and γ-isomers) as a result of diet has often been observed (Parazo et al., 1998).

3.2.2. Fatty acids

Fatty acid composition of NL and POL fractions was studied in the two different zones of angler muscle (Table 4). In the neutral fraction, 22:6 n-3 fatty acid was the most abundant, followed by 16:0, 18:1 n-9 and 18:0 fatty acids. A similar profile was observed for the polar fraction; thus, the following order was determined for the most abundant fatty acids in terms of decreasing content: 22:6 n-3 > 16:0 > 18:1 n-9 > 20:5 n-3 > 20:4 n-6. In a previous study, the fatty acid composition of angler captured along the French coast was analyzed (Sirot et al., 2008); in that study, no muscle site differentiation was carried out and the following decreasing sequence was reported: 22:6 n-3 > 16:0 > 20:5 n-3 > 18:1 n-9 > 18:0. Present results and discussion concerning differences in distribution between sites and types of lipid fractions will now be focused on fatty acid group contents and fatty acid ratio values.

In both muscle zones and in both lipid fractions, PUFA was the most abundant group, in agreement to a previous study on angler muscle in which no muscle site differentiation was achieved (Sirot et al., 2008). In the present work, contents of all groups (saturated fatty acids, SFA; monounsaturated fatty acids, MUFA; PUFA) were shown to be higher in the polar fraction than in the neutral fraction in both the tail and the head. This result can be explained on the basis that PL was found to be the most abundant total lipid group (Table 3) and that similar total lipid contents were observed in both muscle zones (Table 2).

Comparison of NL between muscle sites showed higher mean contents for the three acid groups in the head zone than in the tail zone; these differences were found to be significant for MUFA and PUFA. It was concluded that the head zone has a higher neutral lipid content. With regards to the POL fraction, a definite trend related to the distribution of fatty acid groups between muscle zones was not obtained. This conclusion is in agreement to the above-mentioned structural role of PL (main component of the polar fraction), which is thought to render it robust against internal and external factors (Pearson et al., 1977; Gallardo et al., 1989).

A majority of the Western population does not consume adequate levels of n-3 fatty acids through natural dietary sources, such as fish. As a result, great interest has been recently accorded to the n-3/ n-6 ratio of foods included in the human diet. The value of this ratio has shown to have a great effect on the development of certain health problems (Ackman & Ratnayake, 1990; Weber, 1992), and the recommended ratio for the whole diet is near 1/6 (n-3/ n-6) (Simopoulos, 1994). In the present work (Table 4), although higher mean values of the n-3/ n-6 ratio were obtained in the polar fraction in both the head and tail, these differences were not found to be significant. However, it should be noted that both edible zones exhibited n-3/ n-6 ratios that would be beneficial for maintaining the recommended values in the whole human diet.

In response to the recent interest in n-3 fatty acids, great attention has been accorded to the two most abundant components (22:6 n-3 or docosahexaenoic acid, DHA; 20:5 n-3 or eicosapentaenoic acid, EPA) of the n-3 fatty acid series. As a result, EPA has been recognised as being beneficial for human health as it reduces the risk of cardiovascular diseases (Hall et al., 2008) and DHA has been reported to contribute to the development of certain functions related to the nervous system and visual functions in human beings (Linko & Hayakawa, 1996). Because of this interest, the DHA/EPA ratio distribution in the different sites and lipid fractions of angler muscle were analysed in the present work (Table 4). In all cases, a value greater than 4 was obtained for this ratio. Higher mean values were obtained in the NL fraction, and such differences were significant in the tail zone. Comparison of muscle zones did not indicate a definite trend. Previous research on turbot (*P. maxima*) (Aubourg et al., 2007) and blackspot seabream (*P. bogaraveo*) (Álvarez et al., 2009) did not identify differences in this fatty acid ratio between different muscle zones.

3.3. Mineral composition analysis

Marine organisms absorb minerals from their diet and the surrounding water and deposit them in their skeletal tissues and organs. Many such elements are widely known to be present in enzyme active centers that are responsible for the development of important functions in all animals; thus, marine-derived foods can serve as a good source of essential elements (Johnson & Fischer, 1994; Lal, 1995). Additionally, fish and shellfish are good bioindicators of trace element contamination in the marine environment because they occupy different trophic levels and can exhibit large bioaccumulation factors; by studying fish and shellfish, the harmful effects of certain metals and metalloids to marine environment and to human health have been recognized (Engman & Jorhem, 1998; Noël et al., 2011). For both the essential and toxic groups, elements corresponding to the transition and electronegative groups of the periodic table have been reported to be strongly bound to other constituents, whereas elements corresponding to electropositive groups generally remain dissolved in the ionic state in the cell medium (Piclet, 1987). Preliminary studies have indicated that the concentration of essential and toxic minerals in fish is influenced by a number of factors such as seasonal and biological differences (species, size, dark/white muscle, age, sex and

sexual maturity), food source and environment (water chemistry, salinity, temperature and contaminants) (Lal, 1995; Noël et al., 2011).

In the present research, the presence of different essential (Co, Cu, Fe, Mn and Zn) and toxic (As, Cd, Cr, Hg, Ni, Pb and V) elements was assessed in two angler muscle zones (Table 5).

Among the essential elements analysed, Zn and Fe were found to be the most abundant, whereas Co was present at very low levels. Higher mean values were obtained for all essential elements in the head zone compared with the tail zone; such differences were significant in the case of Zn. The Cu and Fe contents found in the present study agree with the mean values reported for the muscle of most fish species; however, Zn and Mn concentrations can be considered to fall above this mean value, whereas Co content measured here is below the mean value (Piclet, 1987; Lal, 1995; Engman & Jorhem, 1998).

Previous research has reported on the essential mineral content in angler muscle, although a muscle zone study had not been conducted. In an earlier study (Jhaveri et al., 1984), angler captured along the coast of southern New England (USA) coast were analysed; results demonstrated lower values than those in the present study with respect to Cu, Fe and Zn (0.28, 3.20 and 4.10 mg kg^{-1} muscle, respectively). In a second study (Carvalho et al., 2005), angler captured along the Portuguese coast were analysed; again, lower values than those in the present study were obtained for Cu, Fe, Mn and Zn (0.17, 1.36, 0.24 and 3.57 mg kg^{-1} muscle, respectively).

With regards to the toxic elements assessment, As was found to be the most abundant, whereas the abundances of Cd and Pb were very low. In most cases (except for As and Hg), higher mean values were observed in the head zone than in the tail zone; these differences were significant in the cases of Cd, Cr and V.

Previous works have assessed toxic elements in angler muscle; in such studies, no muscle zone analysis was included. In angler caught along the coast of southern New England (USA) (Jhaveri et al., 1984), similar values for Cd (0.011 mg kg^{-1} muscle) but lower values for Ni (0.03 mg kg^{-1} muscle) were obtained compared with those determined in the present study. In angler caught along the Portuguese coast (Carvalho et al., 2005), lower values were obtained for Cd, Cr, Hg, Ni and Pb (0.00, 0.19, 0.22, 0.03 and 0.01 mg kg^{-1} muscle, respectively) when compared with those determined in the present work. In another study (Afonso et al., 2008), a different sample of angler from the Portuguese coast was reported to have a similar Hg content (0.43 mg kg^{-1} muscle) to that determined in the present research.

With regards to the assessment of As in angler muscle, lower values have been reported in previous investigations compared to values determined in this study. Anacleto et al. (2009) reported a values range of 4.4-20.3 mg kg^{-1} muscle for angler captured along the Portuguese coast. Similarly, Mormede and Davis (2001) repòrted a 2.70-21.47 mg kg^{-1} muscle range for angler captured in the Rockall Basin, northwest of Scotland and Ireland.

It has been reported that intrinsic factors such as growth (size, weight), age, sex, sexual maturity, physiology and stress influence the accumulation of trace metals in marine organisms (Phillips and Rainbow, 1993). Additionally, geographical variations and differences in feeding habitats and availability have also been reported to induce differences in metal levels (Engman & Jorhem, 1998). Differences between present results and previously reported values for the same species and for related fish species can be explained on the basis of these intrinsic and extrinsic factors.

4. CONCLUSIONS

The present research has provided new data concerning the lipid composition and the essential and toxic element contents of angler captured in the Grand Sole Fishing Bank. Angler muscle contains low total lipid and protein contents, which are accompanied by a relatively high moisture level. The lipid classes and groups analysis showed PL to be the most abundant group, whereas TAG presence was demonstrated to be very low. In both the NL and POL fractions, the contents of the PUFA group was generally higher than the contents of the SFA and MUFA groups; 22:6 n-3 was the major fatty acid. A healthy and beneficial n-3/ n-6 ratio was observed in all cases. Naturally valuable muscle contents were also observed regarding essential elements such as Cu, Fe, Zn and Mn; the contents were especially high for the latter two elements. Among toxic elements, As was found to be the most abundant.

A difference in the distribution of chemical components was observed by comparing the two muscle locations under study. Higher protein and TMAO-N concentrations but lower for moisture, STS and TAG contents were observed in the tail site. However, an inhomogeneous muscle distribution could not be demonstrated with regards to total lipids, PL, FFA and α-tocopherol contents. The mean concentrations of essential elements were higher in the head site than in the tail site for all elements considered; this difference was significant in the case of Zn. For toxic elements, higher mean values were also obtained for the head site, with the exceptions of As and Hg; differences were found to be significant for Cd, Cr and V.

Conflict of interest

The authors declare that there are no conflicts of interest.

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REFERENCES

Ackman, R., & Ratnayake, W. (1990). Chemical and analytical aspects of assuring an effective supply of omega-3 fatty acids to the consumer. In R. Lees, & M. Karel (Eds.), *Omega-3 fatty acids in health and disease* (pp. 215-233). New York, NY, USA and Basel, Switzerland: Marcel Dekker, Inc.

Afonso, C., Lourenço, H., Pereira, C., Martins, M., Carvalho, M., Castro, M., Nunes, Mª L. (2008). Total and organic mercury, selenium and α-tocopherol in some deep-water fish species. *Journal of the Science of Food and Agriculture, 88*, 2543-2550.

Aitken, A., Mackie, I., Merrit, J., & Windsor, M. (1982). In *Fish: Handling and processing* (pp. 2-19). Edinburgh, Scotland, UK: Ministry of Agriculture, Fisheries and Food, Torry Research Station.

Álvarez, V., Medina, I., Prego, R., Aubourg, S. (2009). Lipid and mineral distribution in different zones of farmed and wild blackspot seabream (*Pagellus bogaraveo*). *European Journal of Lipid Science and Technology, 111*, 957-966.

Anacleto, P., Lourenço, H., Ferraría, V., Afonso, C., Carvalho, L., Martins, F., Nunes, Mª L. (2009). Total arsenic cointent in seafood consumed in Portugal. *Journal of Aquatic Food Product Technology, 18*, 32-45.

AOAC (1990) *Official methods for analysis of the Association of Analytical Chemistry* (15th ed.) (pp. 931-937). Arlington, VA, USA: Association of Official Chemists, Inc.

Aubourg, S., Gallardo, J. M., Sotelo, C. (1991). Distribution of triglycerides, phospholipids and polyunsaturated fatty acids in different sites in raw albacore (*Thunnus alalunga*) muscle: Changes after cooking. *Canadian Institute of Food Science and Technology Journal, 24*, 287-291.

531 Aubourg, S., Losada, V., Prego, R. (2007). Distribution of lipids and trace minerals in
532 different muscle sites of farmed and wild turbot (*Psetta maxima*). *International*
533 *Journal of Food Science and Technology*, *42*, 1456-1464.
534 Aubourg, S., Rey-Mansilla, M., Sotelo, C. (1999). Differential lipid damage in various
535 muscle zones of frozen hake (*Merluccius merluccius*). *Zeitschrift für*
536 *Lebensmittel Untersuchung und Forschung*, *208*, 189-193.
537 Barros-Velázquez, J., Gallardo, J., Calo, P., Aubourg, S. (2008). Enhanced quality and
538 safety during on-board chilled storage of fish species captured in the Grand Sole
539 North Atlantic fishing bank. *Food Chemistry, 106*, 493-500.
540 Bligh, E., Dyer, W. (1959). A rapid method of total extraction and purification.
541 *Canadian Journal of Biochemistry and Physiology, 37*, 911-917.
542 Bordahandi, L., Martín, I., Abad, E., Rivera, J., González, M. (2006). Organochlorine
543 compounds (PCBs, PCDDs and PCDFs) in seafish and seafood from the Spanish
544 Atlantic Southwest Coast. *Chemosphere, 64*, 1450-1457.
545 Cabrini, L., Landi, L., Stefanelli, C., Barzanti, V., Sechi, A. (1992). Extraction of lipid
546 and lipophilic antioxidants from fish tissues: A comparison among different
547 methods. *Comparative Biochemistry and Physiology. Biochemistry and*
548 *Molecular Biology*, *101*, 383-386.
549 Carvalho, M., Santiago, S., Nunes, Mª L. (2005). Assessment of the essential element
550 and heavy metal content of edible fish muscle. *Analytical and Bioanalytical*
551 *Chemistry*, *382*, 426-432.
552 Costley, C., Mossop, K., Dean, J., Garden, L., Marshall, J., Carroll, J. (2000).
553 Determination of mercury in environmental and biological samples using
554 pyrolysis atomic absorption spectrometry with gold amalgamation. *Analytica*
555 *Chimica Acta, 405*, 179–183.

556 Engman, J., Jorhem, L. (1998). Toxic and essential elements in fish from Nordic
557 waters, with the results seen from the perspective of analytical quality
558 assurance. *Food Additives and Contaminants*, *15*, 884-892.
559 FAO (2007). Fishery statistics. Capture production. In *Food and Agriculture*
560 *Organization of the United Nations*, *Vol. 100/1* (pp. 197-198). Rome, Italy:
561 Yearbook 2005.
562 Finne, G. (1992). Non-protein nitrogen compounds in fish and shellfish. In G. Flick, &
563 R. Martin (Eds.), *Advances in Seafood Biochemistry* (pp. 393-399). Lancaster,
564 PA, USA: Technomic Publishing.
565 Gallardo, J. M., Aubourg, S., Pérez-Martín, R. (1989). Lipid classes and their fatty acids
566 at different loci of albacore (*Thunnus alalunga*): Effects of the pre-cooking.
567 *Journal of Agricultural and Food Chemistry, 37*, 1060-1064.
568 García-Arias, T., Sánchez-Muniz, F., Castrillón, A., Navarro, P. (1994). White tuna
569 canning, total fat, and fatty acid changes during processing and storage. *Journal*
570 *of Food Composition and Analysis, 7,* 119-130.
571 García-Soto, B., Sanjuás, M., Barros-Velázquez, J., Fuertes-Gamundi, R., Aubourg, S.
572 (2011). Preservative effect of an organic acid-icing system on chilled fish lipids.
573 *European Journal of Lipid Science and Technology, 113*, 487-496.
574 Hall, W., Sanders, K., Sanders, T., Chowienczyk, P. (2008). A high-fat meal enriched
575 with eicosapentaenoic acid reduces postprandial arterial stiffness measured by
576 digital volume pulse analysis in healthy men. *Journal of Nutrition*, *138*, 287-291.
577 Herland, H., Esaiassen, M., Olsen, R. (2007). Muscle quality and storage stability of
578 farmed cod (*Gadus morhua* L.) compared to wild cod. *Journal of Aquatic Food*
579 *Product Technology*, *16*, 55-66.

580 Huang, T., Chen, C., Wefler, V., Raftery, A. (1961). A stable reagent for the
581 Liebermann-Buchardt reaction. *Analytical Chemistry*, *33*, 1405-1407.
582 Huss, H. (1995). Chemical composition (principle constituents, lipids, proteins, N-
583 containing extractives and vitamins and minerals). In H. Huss (Ed.), *Quality and*
584 *quality changes in fresh fish* (pp. 20-34). Rome, Italy: FAO Fisheries Technical
585 Paper No. 348.
586 Jhaveri, S., Karakoltsidis, P., Montecalvo, J., Constantinides, S. (1984). Chemical
587 composition and protein quality of some Southern New England marine species.
588 *Journal of Food Science, 49*, 110-113.
589 Johnson, M., Fischer, J. (1994). Role of minerals in protection against free radicals.
590 *Food Technology, 48(5)*, 112-120.
591 Lal, S. (1995). Macro and trace elements in fish and shellfish. In A. Ruiter (Ed.), *Fish*
592 *and fishery products: composition, nutritive properties and stability* (pp. 187-
593 214). Wallingford, CN, USA: CAB International.
594 Linko, Y., Hayakawa, K. (1996). Docosahexaenoic acid: A valuable nutraceutical ?
595 *Trends in Food Science and Technology*, *7*, 59-63.
596 Lowry, R., Tinsley, I. (1976). Rapid colorimetric determination of free fatty acids.
597 *Journal of the American Oil Chemists' Society*, *53*, 470-472.
598 Mediel, M., Rodríguez, V., Codina, G., Martín-Casanova, N. (2000). Isolation of
599 mycobacteria from frozen fish destined for human consumption. *Applied and*
600 *Environmental Microbiology, 66*, 3637-3638.
601 Mendes, R., Quinta, R., Nunes, Mª L. (2001). Changes in baseline levels of nucleotides
602 during ice storage of fish and crustaceans from the Portuguese coast. *European*
603 *Food Research and Technology, 212*, 141-146.

604 Mormede, S., Davies, I. (2001). Heavy metal concentrations in commercial deep-sea
605 fish from the Rockall Trough. *Continental Shelf Research, 21*, 899-916.
606 Noël, L., Chafey, C., Testu, C., Pinte, J., Velge, P., Guérin, T. (2011). Contamination
607 levels of lead, cadmium and mercury in imported and domestic lobsters and
608 large crab species consumed in France: Differences between white and brown
609 meat. *Journal of Food Composition, 24*, 368-375.
610 Parazo, M., Lall, S., Castell, J., Ackman, R. (1998). Distribution of α- and γ-
611 tocopherols in Atlantic salmon (*Salmo salar*) tissues. *Lipids*, *33*, 697-704.
612 Parkin, K., Hultin, H. (1982). Some facts influencing the production of dimethylamine
613 and formaldehyde in minced and intact red hake muscle. *Journal of Food*
614 *Processing and Preservation*, *6*, 73-97.
615 Pearson, A., Love, J., Shorland, F. (1977). "Warmed-over" flavor in meat, poultry and
616 fish. *Advances in Food Research*, *23*, 2-61.
617 Phillips, D., & Rainbow, P. (1993). *Biomonitoring of trace aquatic contaminants* (pp.
618 1-371). London, UK: Elsevier.
619 Piclet, G. (1987). Le poisson aliment. Composition – intérêt nutritionnel. *Cahiers de*
620 *Nutrition et Diététique*, *XXII*, 317-335.
621 Raheja, R., Kaur, C., Singh, A., Bhatia, A. (1973). New colorimetric method for the
622 quantitative determination of phospholipids without acid digestion. *Journal of*
623 *Lipid Research*, *14*, 695-697.
624 Ruff, N., Fitzgerald, R., Cross, T., Lynch, A., Kerry, J. (2004). Distribution of alpha-
625 tocopherol in fillets of turbot (*Scophthalmus maximus*) and Atlantic halibut
626 (*Hippoglossus hippoglossus*), following dietary alpha-tocopheryl acetate
627 supplementation. *Aquaculture Nutrition*, *10*, 75-81.

628 Ruiz-Capillas, C., Horner, W. (1999). Determination of trimethylamine and total
629 volatile basic nitrogen in fresh fish by flow injection analysis. *Journal of the*
630 *Science of Food and Agriculture, 79*, 1982-1986.
631 Sigurgisladóttir, S., Ackman, R., O'Keefe, S. (2003). Selective deposition of alpha-
632 tocopherol in lipids of blue mussels (*Mytilus edulis*). *Journal of Food Lipids*, *1*,
633 97-109.
634 Simopoulos, A. (1994). Fatty acids. In I. Goldberg (Ed.), *Functional foods, designer*
635 *foods, pharmafoods, nutraceuticals* (pp. 355-392). New York, NY, USA:
636 Chapman and Hall.
637 Simopoulos, A. (1997). Nutritional aspects of fish. In J. Luten, T. Börrensen, & J.
638 Oehlenschläger (Eds.), *Seafood from producer to consumer, Integrated*
639 *approach to quality* (pp. 589-607). London, UK: Elsevier Science.
640 Sirot, V., Oseredczuc, M., Bemrah-Aouachria, N., Volatire, J.L., Leblanc, J.C. (2008).
641 Lipid and fatty acid composition of fish and seafood consumed in France:
642 CALIPSO study. *Journal of Food Composition and Analysis, 21*, 8-16.
643 Tozawa, H., Erokibara, K., & Amano, K. (1971). Proposed modification of Dyer's
644 method for trimethylamine determination in codfish. In R. Kreuzer (Ed.), *Fish*
645 *Inspection and Quality Control* (pp. 187-190). London, UK: Fishing News
646 Books Ltd.
647 Vioque, E., Holman, R. (1962). Quantitative estimation of esters by thin-layer
648 chromatography. *Journal of the American Oil Chemists' Society*, *39*, 63-66.
649 Weber, P. (1992). Dietary fatty acids and eicosanoid Biochemistry. In G. Flick, & R.
650 Martin (Eds.), *Advances in Seafood Biochemistry* (pp. 181-184). Lancaster, PA,
651 USA: Technomic Publishing.
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TABLE 1

Analysis results* of certified reference material (CRM)**

Essential/toxic element	Measured	Certified
Co	0.176 ± 0.028	0.182 ± 0.031
Cu	2.25 ± 0.13	2.34 ± 0.16
Fe	138 ± 12	142 ± 10
Mn	3.80 ± 0.27	3.66 ± 0.34
Zn	23.0 ± 3.9	25.6 ± 2.3
As	17.6 ± 0.2	18.0 ± 1.1
Cd	0.040 ± 0.001	0.043 ± 0.008
Cr	34.3 ± 2.6	34.7 ± 5.5
Hg	4.51 ± 0.15	4.64 ± 0.26
Ni	17.8 ± 1.6	19.4 ± 3.1
Pb	0.054 ± 0.006	0.065 ± 0.007
V	0.47 ± 0.03	NC***

* Results (n=3; three replicates) are expressed as mg kg^{-1} muscle.

** CRM consisted on fish muscle material (DORM-2) from the National Research Council of Canada.

*** NC, not certified.

TABLE 2

General composition* parameters in different muscle zones of angler**

Parameter	Muscle zone	
	Head	Tail
Moisture	840.9 ± 4.4 b	830.0 ± 1.7 a
Total lipids	3.22 ± 0.29	3.42 ± 0.25
Proteins	140.2 ± 5.1 a	157.7 ± 3.3 b
Trimethylamine oxide-nitrogen (TMAO-N)	238.0 ± 78.6 a	407.5 ± 43.6 b

* Values expressed as g kg^{-1} muscle, except for TMAO-N (mg kg^{-1} muscle).

** Mean values of five independent determinations (n = 5) ± standard deviations. For each parameter, values followed by different letters (a, b) express significant ($p<0.05$) differences between head and tail zones, respectively. No letters are included in case of no significant differences ($p>0.05$).

TABLE 3

Content* on lipid classes and groups in different muscle zones of angler muscle**

Lipid parameter	Muscle zone	
	Head	**Tail**
Phospholipids	2.288 ± 0.176 (715.1 ± 6.5)	2.370 ± 0.255 (696.9 ± 8.1)
Sterols	0.355 ± 0.040 b (111.0 ± 1.4) b	0.288 ± 0.019 a (84.7 ± 0.7) a
Triacylglycerols	0.024 ± 0.008 (7.43 ± 0.26) b	0.011 ± 0.010 (3.27 ± 0.03) a
Free fatty acids	0.119 ± 0.055 (37.30 ± 2.08)	0.073 ± 0.038 (21.54 ± 1.23)
α-tocopherol	1.990 ± 0.212 (0.618 ± 0.066)	2.335 ± 0.251 (0.683 ± 0.073)

* Values expressed as g kg^{-1} muscle, except for α-tocopherol (mg kg^{-1} muscle). Values in brackets correspond to g kg^{-1} total lipids.

** Mean values of five independent determinations (n = 5) ± standard deviations. For each lipid component, values followed by different letters (a, b) express significant ($p<0.05$) differences between head and tail zones, respectively. No letters are included in case of no significant differences ($p>0.05$).

TABLE 4

Fatty acid content (mg kg^{-1} muscle) and fatty acid ratio assessment* in neutral and polar lipid fractions of different zones of angler muscle**

Fatty acid	Neutral fraction		Polar fraction	
	Head	Tail	Head	Tail
14:0	1.79 ± 0.77	1.36 ± 0.37	11.07 ± 1.66	10.27 ± 1.43
15:0	0.23 ± 0.12	0.32 ± 0.09	4.43 ± 0.45	5.28 ± 0.73
16:0	15.04 ± 3.30	11.05 ± 2.62	196.26 ± 3.33	193.29 ± 7.55
16:1 n-7	5.92 ± 2.08	3.54 ± 0.99	31.10 ± 6.18	30.53 ± 6.80
17:0	0.42 ± 0.19	0.51 ± 0.14	11.40 ± 1.08	12.08 ± 0.94
18:0	9.98 ± 0.70	11.02 ± 1.80	28.13 ± 2.24	22.71 ± 5.37
18:1 n-9	13.22 ± 3.26	9.96 ± 2.52	111.26 ± 3.78	110.81 ± 12.95
18:1 n-7	3.87 ± 0.96	2.79 ± 0.71	22.04 ± 2.08	20.67 ± 2.04
18:2 n-6	1.22 ± 0.26	0.85 ± 0.14	9.57 ± 1.31	9.25 ± 1.16
18:3 n-3	3.99 ± 1.08	2.10 ± 0.96	2.03 ± 0.98	3.93 ± 1.21
20:1 n-9	2.94 ± 1.22	2.21 ± 0.61	7.84 ± 1.33	5.78 ± 2.55
18:4 n-3	1.66 ± 0.57	0.69 ± 0.46	2.99 ± 0.74	2.08 ± 0.39
20:4 n-6	5.69 ± 0.78	3.84 ± 1.02	37.82 ± 5.73	40.90 ± 4.28
20:5 n-3	5.92 ± 1.41	3.60 ± 1.16	56.93 ± 6.46	66.44 ± 6.05
24:1 n-9	1.78 ± 0.54	1.02 ± 0.39	15.92 ± 1.32	15.77 ± 1.07
22:5 n-3	2.75 ± 1.09	1.77 ± 0.54	20.45 ± 5.63	21.25 ± 5.07
22:6 n-3	35.01 ± 5.31	25.06 ± 6.05	290.65 ± 23.36	294.62 ± 23.49
SFA	z 27.46 ± 3.78	z 24.26 ± 2.88	y 251.29 ± 3.87	y 243.63 ± 8.74
MUFA	z 27.73 ± 4.45 b	z 19.52 ± 3.23 a	y 188.16 ± 5.63	y 183.56 ± 13.41
PUFA	z 56.24 ± 6.52 b	z 37.91 ± 6.85 a	y 420.44 ± 31.74	y 438.47 ± 26.02
n-3/ n-6 ratio	7.14 ± 1.34	7.08 ± 1.14	7.87 ± 1.01	7.74 ± 0.69
DHA/EPA ratio	5.91 ± 0.65	y 6.96 ± 0.60	5.11 ± 0.46	z 4.43 ± 0.31

* Mean values of five independent determinations (n = 5) ± standard deviations. For each fatty acid group or ratio, values followed by different letters (a, b) express significant ($p<0.05$) differences between head and tail zones in each lipid fraction (neutral and polar). For each fatty acid group or ratio, values preceded by different letters (z, y) express significant ($p<0.05$) differences between neutral and polar fractions in each muscle zone (head and tail). No letters are included in case of no significant differences ($p>0.05$).

** Abbreviations: SFA (saturated fatty acids), MUFA (monounsaturated fatty acids), PUFA (polyunsaturated fatty acids), DHA (docosahexaenoic acid; 22:6 n-3) and EPA (eicosapentaenoic acid; 20:5 n-3).

TABLE 5

Element contents* in different zones of angler muscle**

Essential/toxic element	Muscle zone	
	Head	**Tail**
Co	0.079 ± 0.005	0.077 ± 0.007
Cu	2.34 ± 0.73	1.45 ± 0.69
Fe	17.5 ± 7.4	8.9 ± 7.3
Mn	0.92 ± 0.24	0.60 ± 0.14
Zn	29.5 ± 6.0 b	15.5 ± 1.1 a
As	27.3 ± 4.7	31.1 ± 5.9
Cd	0.011 ± 0.003 b	0.005 ± 0.001 a
Cr	0.75 ± 0.11 b	0.54 ± 0.07 a
Hg	0.425 ± 0.030	0.471 ± 0.041
Ni	0.164 ± 0.033	0.151 ± 0.046
Pb	0.080 ± 0.048	0.051 ± 0.041
V	1.86 ± 0.18 b	0.50 ± 0.08 a

* Values expressed as mg kg^{-1} muscle.

** Mean values of five independent determinations (n = 5) ± standard deviations. For each element, values followed by different letters (a, b) express significant ($p<0.05$) differences between head and tail zones, respectively. No letters are included in case of no significant differences ($p>0.05$).