Scientific Works. Series C. Veterinary Medicine. Vol. LXVIII (2), 2022
ISSN 2065-1295; ISSN 2343-9394 (CD-ROM); ISSN 2067-3663 (Online); ISSN-L 2065-1295

FRESHNESS INDICATORS, PHOSPHORUS CONTENT AND HUMIDITY CORRELATIONS IN ANGLERFISH AND MONKFISH SAMPLES *(Lophius spp.)* FROM THE NORTH SEA AND ATLANTIC OCEAN

Cătălina Nicoleta BOIŢEANU[1], Florin NEACSU[2]

[1]University of Agronomic Sciences and Veterinary Medicine of Bucharest, Faculty of Veterinary Medicine, 105 Splaiul Independenţei, District 5, Bucharest, Romania
[2]Baylor University, Department of Chemistry and Biochemistry, Waco, TX 76798, USA

Corresponding author email: catalina.boitzeanu@yahoo.com

Abstract

The paper aimed to present the correlations between the freshness indicators (pH, TVB-N) and water content, phosphorus content and humidity, protein content and TVB-N (mg %) in wild Anglerfish/ Monkfish captured from the North Sea and Atlantic Ocean during naval expeditions aboard the research ship Walther Herwig III^rd^. It is based on the research data obtained at Max Rubner Institute Department of Safety and Quality of Milk and Fish Products Hamburg between 2013 and 2014. The processed data were the following indicators: pH, TVB-N, phosphates content, percent of water, salt, protein, fat and ashes. Correlations were made using chi square test. Angler- and Monkfish (Lophius spp.) samples analysed (n = 77), presented a reasonable positive correlation (R^2 = 0.44) between the pH values and the percentage of water, a very weak positive correlation (R^2 = 0.1) between TVB-N and protein, a high negative correlation (R^2 = - 0.7018) between the phosphate and water content and a strong negative correlation (R^2 = - 0.7961) between the protein and the water content.

Key words*: Lophius spp., freshness indicators, proximate analysis, phosphorus content, humidity.*

INTRODUCTION

Anglerfish (*Lophius piscatorius*) and Monkfish (*L. americanus*) are belonging to the *Lophiidae* Family. They have massive heads, that are broad, flat and depressed, making the rest of the body (normally called the tail) seem like an appendage.

The Anglerfish is a demersal fish, well adapted to its environment, living in coastal regions and at high depths. In northern latitudes, the Anglerfish spawns between April and June mainly north, W and S of the British Isles at a depth beyond 400 meters.

Seven species of *Lophius* are known worldwide, six within the Atlantic Ocean and only one within the Northwest Pacific. The genus supports valuable fisheries (except for *Lophius vaillanti*), most for an extended time, though the exploitation of *Lophius gastrophysus* along the coast of Brazil is comparatively recent (Fariña *et al.*, 2008).

The primary goal of this study was to research the composition of *Lophius* spp. tail muscle which is the main part of the body that is usually destined to human consumption.

MATERIALS AND METHODS

Lophius spp. live in many seas. The foremost important areas of distribution include the N Atlantic and the Mediterranean. They include the American waters from Newfoundland to Brazil, and also the Gulf of Guinea to the New Zealand zone. The fish genus *Lophius* is exploited worldwide. *Lophius* spp. is caught by demersal trawlers with trawls or long lines, mostly gill nets. They are usually prepared for sale on board where the head is removed, the fish is skinned and gutted.

This species is widely found in coastal waters of the N-E Atlantic, from the Barents Sea to the Strait of Gibraltar, the Mediterranean Sea and the Black Sea. The greatest capture volumes within the last decade are reported by France, UK, Ireland and Spain (International Council for the Exploration of the Sea, 2021; Norwegian Institute of Marine Research, 2021).

According to International Council for the Exploration of the Sea (ICES) the main fisheries areas for Black-bellied Anglerfish *(L. budegassa*) and White Anglerfish (*L. piscatorius*) are divisions 7b.-k, 8.a-b and 8d (W and S-W Ireland, Bay of Biscay), 8c and 9a (Cantabrian Sea, Atlantic Iberian waters); sub-areas 1 and 2 (N-E Arctic); sub-areas 4 and 6, and in Division 3.a (North Sea, Rockall and W of Scotland, Skagerrak and Kattegat) (International Council for the Exploration of the Sea, 2018).

L. piscatorius and L. budegassa are mainly caught by Spanish and Portuguese bottom-trawlers and net fisheries (gill net and trammelnets). As with *L. piscatorius, L. budegassa* is a crucial target species for the artisanal fleets and a by-catch for the trawl fleets targeting hake or crustaceans. Since 2010, Spanish landings of *L. piscatorius* were on the average 83% of total landings of the stock. Reported values for *L. budegassa* represented < 1% (on average) of the full landings of the stock. The total allowable catch advice for the Celtic Seas, West of Scotland, Bay of Biscay and Iberian Sea is 34,275 t in 2022, while the most recent estimated stocks were around 45,651 t (International Council for the Exploration of the Sea, 2021).

The stocks assessed are also distributed over a large area of the Northeast Atlantic shelf from ICES Division IIIa to IXa: here are currently 4 stocks of Anglerfish (*Lophius piscatorius* and *L. budegassa* in Divisions VIIb-k and VIIIa,b,d and *L. piscatorius* and *L. budegassa* in Divisions VIIIc and IXa)(International Council for the Exploration of the Sea, 2021).

The *Lophius* spp. samples we analysed in Germany come from the N-E Atlantic (North Sea) and were mainly cached during the fishing expeditions 366 and 372 respectively of Walther Herwig IIIrd the main research and exploration ship of Thünen Institute Hamburg (Figure 1). Fish weight was between 857 g and 6.34 kg. The medium size for commercial landings from this area is 1 m or 20 kg (ICES, 2020). Three species of Anglerfish dominate in the fishery. On the German market may be found the White Anglerfish (*Lophius piscatorius*), which might reach a length of up to 2 meters and a weight of up to 40 kilograms. The commercial weight of obtainable tails is sometimes between 1 and 3 kg.

Figure 1. Walther Herwig IIIrd research ship (Thünen Institute of Sea Fisheries)

Black-bellied Anglerfish (*Lophius budegassa*), grows up to 70 centimeters long.

Mainly on the American Atlantic Coast is located the home of the American Anglerfish (*Lophius americanus*), which might reach up to 1.2 meters and is known as the Monkfish (Figure 2).

Figure 2. Monkfish (*Lophius americanus*) specimen

European landings are limited in volume (between 9,000 and 10,000 metric tons catch weight per year). The market share in Germany in 2013 was around 0.5%. Due to of the meat quality, which is extremely valued by connoisseurs, good prices may be achieved for *Lophius* spp.

Some are sold with head on but mainly without it, also skinned, because the appearance of the Anglerfish sometimes it is not commercially offered as an entire fish (Figure 3). They are mainly sold as fresh (whole with head on/ tail skinless fillets/ whole skin-on tails) or deep frozen as tail skinless fillets/ whole skin-on tails (Figure 4).

The marketed meat, hosts a fine aroma and it is ideal for roasting with a fine inherent flavour of the lean, white meat, which is nearly boneless (Figure 5).

Figure 3. Whole Anglerfish (*Lophius piscatorius*)

Figure 4. Anglerfish (*Lophius piscatorius*) edible portion skinning

Figure 5. Economically valuable part of *Lophius* spp.

Both lipid quantity and quality of fish are subject to changes because of their age and size and environmental conditions like season and feeding (Karl *et al.*, 2018). Moreover, both fat content and the fatty acid composition are species specific (Borresen, 1992; Hamre *et al.,* 2003; Iverson *et al.,* 2002; Lenas *et al.,* 2011). *Lophius* spp. belong to the semi-fatty (3–6% fat) fish species (Karl *et al.*, 2013). They store their lipids mainly within the muscle tissue, but only little information is accessible on the distribution within the edible part. However, the flesh quality is correlated to the fat, water and protein content and to its distribution within the muscle tissue. The lipid distribution is additionally a very important factor for the values of contaminants eventually present within edible parts with fat content. Determination in parts of the fillets and/or in parts of entire fish can yield different contamination levels, depending on the respective lipid content (Karl & Lahrssen-Wiederholt, 2013). This work provides information on the amount of fat, water and protein within the muscle tissue of *Lophius* spp. from the North Sea fishing areas. Additionally, we establish a relationship between the lipid and water content, the share of TVB-N and protein content, the water content and pH, the protein and the water content respectively (Figures 7 to 10) using available muscle flesh thawed samples (n=77).

Previous chemical analysis

Proximate composition, fatty acids profiles and other nutritional values were evaluated for fillets of *Lophius* spp. Crude protein (N × 6.25) and total lipid content were determined on the edible portions of fish by the methods descrybed within the AOAC manual (AOAC, 1975).
Moisture content was determined by drying the samples in an air circulation oven for 8 h at 100°C. Samples for ash determination were heated in a muffle furnace at 550°C for 6 h to constant weight. (Jhaveri *et al.*, 1984).
Moisture, total lipid and protein contents ranged from 828 to 845, 3.0 to 3.7 and 135 to 170 g/ kg muscle, respectively (Prego *et al.*, 2012). Other previous research that associated the chemical composition and nutritional value of Anglerfish is scarce (Table 1) or 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).

MATERIALS AND METHODS

Fish samples were collected from the N-E Atlantic (correspondent German Bight of the North Sea) and were cached from a bottom trawl catch during the fishing expeditions no.366 and no.372 of Walther Herwig III[rd] (Figure 6).

Table 1. Nutritional values and energy of 100g Anglerfish and Monkfish (edible portions) medium values compared to lobster (Fish Information Center, 2021; Pehrsson *et al.*, 2016; Strobel *et al.*, 2012).

Proximate composition	Anglerfish	Monkfish	Lobster
Fat	4.6 g	1.5 g	2.0 g
Protein	20.3 g	14.9 g	16.0 g
Sodium	180 mg	180 mg	270 mg
Calories	88 kcal	76 kcal	89 kcal
Iodine in raw liver	96 µg	100 µg	185 µg
Omega-3 fatty acids	0.1 g	0.13 g	79 mg

Between 26.07.2013-20.08.2013 and 23.01.2014 - 23.02.2014 respectively and previously 77 Angler- and Monkfish (*Lophius* spp.) of various size were collected by the above mentioned German fishing research vessel (German Oceanographic Data Center, 2013; 2014).

Figure 6. Anglerfish (*Lophius piscatorius*) caught in the North Sea

Individuals were weighed, lengths were measured and then were manually filleted. The skin was removed and muscles were weighed (FAO, 1995). Tail muscle meat of *Lophius* spp. was minced and homogenised. Corresponding left and right tail parts of every individual were combined and deep frozen. Samples were stored at –20ºC until further analysis.

Proximate composition (water, ash, protein and lipid content in %) of every muscle part homogenate was measured. Total sum, as percentage, was in the range of 98-102%.

Water content was indirectly determined after drying an aliquot of the homogenised tail muscle flesh for 12 h at 105ºC.

Protein nitrogen was measured by Dumas method (Miller *et al.,* 2007) using a LECO TruSpecN (LECO Instruments GmbH, Germany).

Percent of protein was calculated by multiplying % N by 6.25 (AOAC, 2005).

Ash content was determined according to Antonacopoulos (1973).

Lipid content was determined by modifying the Smedes method (1999) (according to Karl *et al.,* 2012). After double extraction from 5 g sample with cyclohexane and isopropanol, the fat was transferred within the cyclohexane phase, after adding water and stirring by means of an Ultra-Turrax instrument (IKA-Werke, Germany). Centrifugation and phases separation was followed by gravimetric measurement of fat after separation and concentration from the cyclohexane phase. Organic phase was separated from the aqueous layer and evaporated. Lipid content was determined after drying the remained residue for 1 h at 105ºC.

Total phosphorus-determination

The total phosphorus content was determined photometrically from the nitric acid extract of the ash in line with a modified official German method § 64 LFBG to detect and measure phosphorus in meat (German Food & Feed Code, 2008).

Total volatile basic nitrogen (TVB-N)

A perchloric acid extract was prepared using 20 g minced and homogenised fish tail muscle flesh with 180 mL 6% (w /w) perchloric acid. The filtered acid extract was used for the determination of total volatile basic nitrogen (TVB-N)

Determination of pH values

After mincing and homogenizing, the sample was diluted 1:1 with deionised water, stirred and the pH was determined by means of a pH-electrode (Oehlenschläger *et al.*, 2002).

Quality assurance and statistical analysis

For the analytical internal quality control the matrix meat MUVA Reference material food supplements no.752 (MUVA GmbH, Kempten, Germany) was used. Certified values were given for ash, moisture, lipid and nitrogen. All analysed components' values showed excellent agreement with the certified values.

Statistical analysis according to MUVA Kempten Statistical Protocol was conducted (https://www.muva.de/fileadmin//user_upload/ Statistisches-Protokoll.pdf). Pairs of variables were subjected to t tests ($p < 0.05$).

RESULTS AND DISCUSSIONS

Proximate composition of the tail muscle flesh of *Lophius* spp. (Monkfish and Anglerfish samples analysed) is compiled into Tables 2 and 3. The sizes of the fish are typical for bottom trawl catches of *Lophiidae* in this area. The *Lophius* spp. samples covered a larger length and weight range of 32–76 cm and 857g–6.34 kg, respectively.

Table 2. Results of proximal analysis in the tail muscle samples from Monkfish (*Lophius* spp.) caught in the Atlantic Ocean (n=13)

Component	I st catch (n=3)	II nd catch (n=10)	Min.	Max.
Moisture %	83 ± 0.7	84.8 ± 0.9	81.84	86.8
Protein%	16 ± 0.5	15.32 ± 0.8	14.2	15.9
Ash %	1.3 ± 0.3	1.05 ± 0.1	0.9	1.66
Lipids %	0.5	0.4	0.4	0.5
pH	6.5 ± 0.12	6.8 ± 0.13	6.4	6.9
NaCl%	0.5± 0.4	0.4 ± 0.1	0.3	1
TVB-N <mg/100g>	11.3 ± 2.7	10.9 ± 1.6	7.1	14.3
Phosphate g P_2O_5/kg	4 ± 0.7	3.2 ± 0.3	3	4.5

Table 3. Proximate composition of Anglerfish, caught in the North Sea (n=64)

Catch nr.	Water %	Protein %	Ash %	Lipids %	Phosphate g P_2O_5/ kg	NaCl %
1 (n=10)	84.56± 0.74	15.40±0.64	1.01± 0.04	0.34	3.23± 0.13	0.30± 0.08
2 (n=10)	89.57± 0.78	8.88± 0.78	1.20± 0.17	0.25	1.16± 0.28	0.48± 0.16
3 (n=10)	88.92± 0.89	10.41±0.72	0.69± 0.07	0.38	2.23± 0.24	0.09± 0.02
4 (n=10)	90.22± 0.81	9.23± 0.46	0.92± 0.05	0.21	1.53± 0.21	0.14± 0.02
5 (n=10)	86.73± 1.62	12.47±1.21	1.62± 0.04	0.39	1.92± 0.4	1.20± 0.07
6 (n=4)	84.87± 0.11	14.08	1.02± 0.03	0.42	3.6± 0.08	0.19± 0.01
7 (n=10)	88.74± 1.75	12.89±1.61	1.00± 0.03	0.24	2.35± 0.19	0.39± 0.10
Min.	83.49	7.88	0.78	0.21	0.86	0.11
Max.	89.87	16.24	1.58	0.42	3.69	1.28

Lophius spp. samples were collected from fishing grounds of the Atlantic Ocean and North Sea to review the composition of the muscle tissue. Little information is available on the composition and nutritional values of *Lophius* spp.

Overall lipid, water, protein and ash contents were calculated as arithmetic means (averages) ± standard deviations (SD) of the respective values analysed from the edible tail muscle parts. Proximate composition of the *Lophius* spp. edible part analysed during the present study (Tables 2 and 3) was generally within the range previously reported. A number of 30 analysed Anglerfish samples presented a higher percentage in moisture (88.92 to 90.22%), thus all other components of dry matter had smaller amounts (protein from 9 to 10%; lipids from 0.21 to 0.38%; ash from 0.69 to 1.20%). The high water percent could not be correlated with an increased salt or phosphorus values.

Lowest mean lipid content of 0.21% was correlated with high water percentage and low protein percentage respectively. The higher lipid content corresponded to the lowest water content.

Protein content of both species stayed within the range of 9-16%. Lowest protein value was equal to 7.88%. Conversely, the highest protein content was 16.5%, while the phosphate ranged from 1.23 to 4.5 g P_2O_5/ kg.

Ash content ranged between 0.55 and 1.69%.

Prego *et al.* (2012) analysed the proximate composition of Anglerfish (n=5): according with his results the values found were: moisture content varied from 82.8 to 84.5%, total lipids from 0.3 to 0.37% and protein from 13.5 to 16.1%, in agreement with other results (Jhaveri *et al.*, 1984; Barros-Velászquez *et al.*, 2008; García-Soto *et al.*, 2011).

Previously, Jahveri *et al.* (1985) had reported in edible portions of Monkfish: 83.29 ± 0.63% moisture, 15.85 ± 0.54% protein, 1.21 ± 0.04% ash and 0.53 ± 0.09% lipids.

Composition differences could not be associated with the dimensions of the fish, although in other fish species, the maturation stage may affect the muscle structure (Karl *et al.,* 2013).

Water and lipid content of the muscle tissue

For both species, a negative correlation between the water and lipid content of the muscle tissue was found ($r = -0.7071$, see

Figure 10). Previously, Karl *et al.* (2018) have reported the identical tendency in other fish species.

The average composition (fat, ash, protein and water) of the thawed *Lophius* spp. samples, the pH of the muscle and also the phosphorus content, given as P_2O_5, are found in Tables 2 and 3.

Crude protein content of tail muscle flesh ranged between 8.8 and 16%, results being similar compared to the protein content reported in other studies (which ranged between 13.5 and 16%).

Water content of the sampled fillets ranged between 81.8 and 91.5% compared to available literature data which showed the proportion of water in fillets at around 83-84%.

Fat contents were comparable with those from other studies, varying between 0.2 and 0.5% (Jhaveri *et al.*, 1984; Barros-Velászquez *et al.*, 2008; García-Soto *et al.*, 2011).

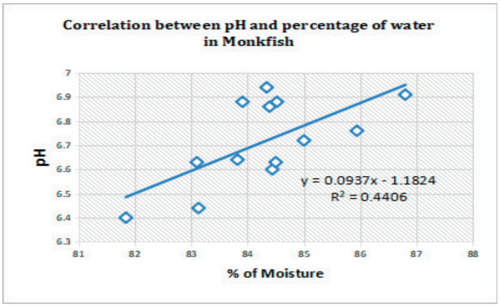

Figure 7. Scatter plot showing a reasonable positive correlation (R^2 = 0.4406) between the pH-values (Y) and the water content (X) in Monkfish, caught in Atlantic Ocean (n = 13)

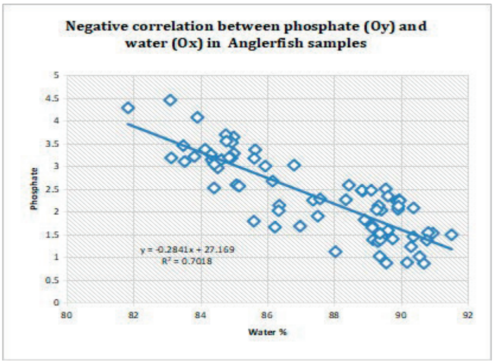

Figure 8. Scatter plot showing a high negative correlation (R^2 = - 0.7018) between the the phosphate (g P_2O_5/ kg) content and water percent in Anglerfish, caught in the North Sea (n = 64)

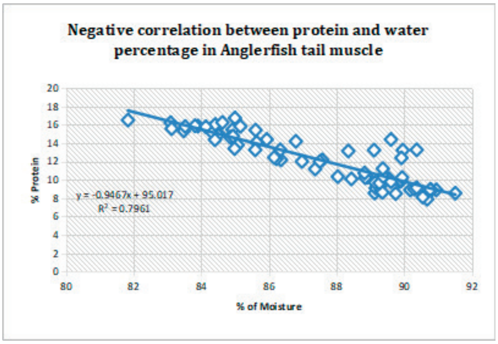

Figure 9. Scatter plot showing a strong negative correlation (R^2 = - 0.7961) between the protein (Y) and the water content (X) in Anglerfish, caught in the North Sea, (n = 64)

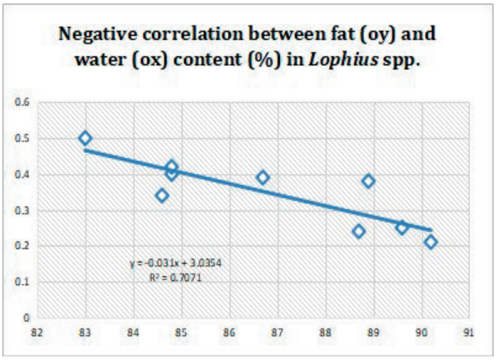

Figure 10. Scatter plot showing a negative correlation (R^2 = - 0.7071) between the fat (Y) and the water content (X) medium values in Anglerfish, caught in the North Sea (n = 77)

Most analysed samples had low pH-values <7.0 (pH=6.4–6.7). Fresh fish muscle has pH values <6.5, and the critical pH is 6.8-7.0 (Lin *et al.*, 2015).

The low pH value of analysed *Lophius* spp. samples corresponds well to literature data reported for fresh *Lophiidae* in which liver samples of Anglerfish had a variation of pH from 6.33 to 6.78 when stored in slurry ice one week.

Composition of *Lophius* spp. tail muscle flesh should be compared to other demersal fish species, therefore further research is required. Feed composition and season of capture may influence the composition of fish (Hamre *et al.*, 2003; Yildiz *et al.*, 2008).

The total volatile basic nitrogen (TVB-N) is a widely used instrument to assess the degree of freshness of marine fish species and legal limits are set for unprocessed fishery products. In freshly caught fish TVB-N equals to 4–5 mg N/100 g values which may be experimentally increased up to over 200 mg N / 100 g or even 500 mg N/ 100g (Ruiz-Capillas and Horner, 1999, Azam *et al,* 2005).

All analysed thawed samples during this study had low TVB-N contents ranging between 7.1 and 14.3 mg N / 100 g (Table 2). Differences between the TVB-N values measured in this study were not significant.

CONCLUSIONS

The results indicate a good freshness of the fish and proper maintenance of the cold chain.

Differences in muscle composition between the individuals were noticeable. The obtained data confirmed a large variation within the muscle composition of the various *Lophius* spp. samples analysed (Table 3).

Consequently, when analysing control and/or monitoring samples, large differences in the water, protein and lipid content of the edible parts of *Lophius* spp. can be expected even in specimens fished simultaneously.

In 30 of analysed samples low protein and high water content along with elevated pH value were detected while the literature offers little information on the water content, protein and pH of the edible muscle meat of *Lophiidae*.

Due to high negative correlation between the amount of phosphate and the percentage of water in analysed samples (Figure 8) we conclude that differences in tail muscle structure of *Lophius* spp. could be induced by different diet composition which the fish encountered in the wild.

ACKNOWLEDGEMENTS

We would like to thank German Academic Exchange Service (DAAD) and Max Rubner Institute (MRI). Special gratitude to Mr. Marcel Balaci (Josef Möbius Bau AG), Professor Claudia Sala and Associate Professor Laurențiu Tudor.

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