

Seasonal Variation of the Chemical Composition, Fatty Acid Profiles and Mineral Elements of *Diplodus Annularis* (Linnaeus, 1758) Caught in the Tunisian Coastal Water

Ilhem Ketata Khitouni¹, Nourhène Boudhrioua Mihoubi², Abderrahmen Bouain¹, Faouzi Ben Rebah^{3,*}

¹UR Biodiversité et Ecosystèmes Aquatiques, FSS, Sfax, Tunisia

²UR Ecophysiologie et Procédés Agroalimentaires, ISB-Sidi Thabet, Tunisia

³Laboratoire de Biochimie et de Génie Enzymatique des Lipases, ENIS, Sfax-Tunisia

*Corresponding author: benrebahf@yahoo.fr

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Abstract The present study investigated the seasonal variation of the chemical composition and fatty acid profiles of Annular sea bream *Diplodus annularis* from the Gulf of Gabes (Tunisia). Moisture, protein, fat and ash contents of the fish muscle were examined as a function of season and sex. Protein, fat and ash average contents were 18.65, 6.47 and 2.20 g/100 g, respectively. The major fatty acids in *D. annularis* lipids were palmitic acid, oleic acid, eicosapentaenoic acid, docosahexaenoic acid and myristic acid. Palmitic acid was the most abundant saturated fatty acid ranging from 26.38 to 32.79 % of the total fatty acids. Oleic acid was the main monounsaturated fatty acid ranging from 26.28 to 35.76 %. The high amounts of saturated and monounsaturated fatty acids in the investigated species are almost in agreement with other studies. Interestingly, omega-3 fatty acids were present at high level in spring (18.04 %). Moreover, the fish muscle contained appreciable concentrations of essential elements (Ca, K, Na and Mg). The present work suggests that this fish could be used as a source of healthy diet for humans.

Keywords: fish chemical composition, fatty acids, fish muscle, *diplodus annularis*, coastal catch fish

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1. Introduction

Seafood products are known to provide significant amounts of different beneficial nutrients such as nutritional and digestible proteins, lipid, essential minerals and highly unsaturated fatty acids [1,2]. Most of these constituents are economically important for human nutrition because of their high nutritional quality. Fish is a major source of protein and it also contains nutritionally valuable lipids. The lipid fraction is now the subject of a great deal of attention due to its high content of ω -3 polyunsaturated fatty acids (ω -3 PUFA), which have exhibited a positive effect in the prevention of certain human diseases (hypertension, inflammation, psoriasis, aggression, depression and cancer) [3,4]. Moreover, PUFA are essential for normal growth, development, and reproduction in all vertebrates, including fish and humans because vertebrates are not able to synthesise ω -3 or ω -6 fatty acids and they must be supplied through diet. Generally, the fish chemical composition varies not only between species, but also between individuals depending on sexual cycle, age, feed, stage of maturity, environment,

season, organs and also muscle location [5,6]. For example, great variations in fatty acid composition of fish lipid were reported [7,8]. Also, mineral concentrations of fish muscle zones may be influenced by different biological factors [9,10]. Consequently, is very important to determine the seasonal biochemical composition of fish in order to ensure the nutritional value as well as eating quality of fish.

Tunisia is a major producer and exporter of seafood and the fishing sector is still of great economic, social and environmental importances. Tunisian seafood products include: cuttlefish and squid, shellfish and mussels, octopus, tuna fish, shrimp, sardines, anchovy, sea bass and annular sea bream, etc. The country's pelagic sources have been evaluated around 100,451 tons/year [11]. Among the pelagic fish, annular sea bream represent the important proportion and is one of the highly consumed fish species in Tunisia. According to [12], the reproduction period of this fish is from February to September and varies with water temperature. Its first sexual maturity was reached for 8-10 cm size approximately. It consumes worms, crustaceans, molluscs, echinoderms, hydrozoans and algae. Although the Annular sea bream constitute a large portion of catches in Tunisia, no detailed seasonal chemical

composition studies of this fish are available. Thus, this study aimed to determine the seasonal variation of the chemical composition and fatty acid profiles of *Diplodus annularis* caught in Tunisian coastal water.

2. Materials and Methods

Diplodus annularis specie was caught from Gabes gulf area (Tunisia). Females and males were analyzed separately because the species shows sexual dimorphism. Both sexes are well distinguished but there are some cases of protandric hermaphroditism. However, the sex of the fish can be determined based on the color and the appearance of the gonads: yellowish and granular form for females and white and smooth for males. Samples were obtained monthly over the year. Fishes were rapidly transported on ice to the laboratory for preparation to chemical analyses. The length (16-21 cm) and weight (50-100 g) of the whole fish were measured in order to select homogenous samples (superior to sexual maturity size). Only muscle was used to determine the chemical composition. Ten fish male and ten fish females were sampled each month. The status of sexual maturation and reproductive period of fish samples were determined macroscopically. The gonadosomatic index GSI was calculated as follows:

$$GSI = \frac{W_g}{W_{ev}} \times 100; \quad (1)$$

$\left\{ \begin{array}{l} \text{where } W_g : \text{gonad weight;} \\ W_{ev} : \text{eviscerated fish weight} \end{array} \right\}$

Moisture content was repeated 10 times (for males and females) after dehydration in an oven at 105°C to a constant weight (for 48 h). For the next analysis ten dry samples were crushed by a Moulinex® blender. The fish dry powder of fish muscles was divided in 3 parts and used to determine the protein, fat and ash contents. Fat content was quantified by Soxhlet extraction, proteins by Kjeldahl procedure, and ash by incineration in a muffle furnace at 550°C [13]. Analyses were repeated three times and performed over the year.

Fatty acid composition was determined using Bligh and Dyer method (1959) [14]. A homogenized fresh sample (25 g) was extracted using chloroform/methanol/water mixture (5V/10V/5V). Fat extracts were converted into fatty acid methyl esters (FAME) by using acetylchloride and were then analyzed by gas-liquid chromatography (Perkin-Elmer 8700 chromatograph, Madrid, Spain) [15]. A fused silica capillary column SP-2330 (0.25 mm i.d. × 50 m, Supelco, Inc., Bellefonte, PA, USA) was employed and the temperature program was as follows: increased from 145 to 190°C at 1.0°C/min and from 190°C to 210°C at 5.0°C/min; held for 13.5 min at 210°C. The carrier gas was nitrogen at 10 psig and detection was performed with a flame ionization detector at 250°C. A programmed temperature vaporizer injector was employed in the split mode (150:1) and was heated from 45 to 275°C at 15°C/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.

The mineral element contents (Ca, Na, Mg, Fe, Zn and K) were determined by an inductively coupled plasma optical emission spectrophotometer (ICP-OES) (Perkin-Elmer, Model 4300 DV, Norwalk, CT) [13].

The energetic value (calorific value) of fish muscle was estimated by multiplying the percentages of protein (PC) and fat (FC) contents with their respective standard factors of 4 and 9 kcal/100 g of fish sample [16] using the following equation :

$$\text{Energetic value} = (4PC + 9FC) \text{ kcal/100 g weight } (2)$$

Data were subjected to Statistical analysis using SPSS software® version 11.0 (Statistical Package for Social Sciences). Values are expressed as mean ± error deviation. To determine the pertinent factors affecting the chemical contents, variance analysis was performed for moisture, protein, fat and ash contents measured in the muscle according to fish sex and size. Correlation matrixes were established between the measured variables (moisture, protein, fat and ash). Honestly significant difference (HSD) with ANOVA one factor was performed for establishing the index of significance for histograms plate. Every factor presenting $p < 0.05$ was considered significant.

3. Results

The seasonal variations of gonadosomatic (GSI) of *D. annularis* species is presented in Table 1. The gonadosomatic indexes present four stages of development: the stage before maturation from December to March, the maturation stage from March to Mai, the spawning period from Mai to August and the stage after ponte from September to November. The gonado-somatic indexes of *Diplodus annularis* (Table 1) varies between $0.46 \pm 0.10\%$ (in August) to $6.61 \pm 0.13\%$ (in May) for males and between $0.43 \pm 0.04\%$ (in August) to $7.70 \pm 0.68\%$ (in May) for females. The maturation of the sexual products begins in March and achieved in August.

Table 1. Seasonal variations of the gonadosomatic index (GSI) of *D. annularis*; mean values ± SD

| | Male | Female |
|------|-----------|-----------|
| Dec | 1.09±0.34 | 1.65±0.25 |
| Jan | 0.84±0.09 | 1.76±0.27 |
| Feb | 0.48±0.06 | 1.01±0.15 |
| Mar | 0.53±0.14 | 1.04±0.18 |
| Apr | 4.14±0.11 | 5.07±0.75 |
| May | 6.61±0.13 | 7.70±0.68 |
| June | 3.01±0.67 | 6.18±0.17 |
| July | 0.82±0.30 | 0.48±0.09 |
| Aug | 0.46±0.10 | 0.43±0.04 |
| Sept | 0.59±0.17 | 0.65±0.10 |
| Oct | 0.52±0.16 | 1.42±0.13 |
| Nov | 0.86±0.13 | 1.17±0.17 |

Table 2. Variations of moisture, ash, total fat and crude protein contents (in g/100 g of fresh fish: mean Values \pm SD.) and energetic values (in kcal/100 g fresh fish \pm SD.) of *D. annularis*; Values followed by different small letters (a, b, c, d, e, f and g) showed significant differences at $p < 0.05$

| Season | Month | Sex | Moisture | Ash | Fat | Protein | Energetic value |
|--------|-----------|--------|--------------------|----------------------|-------------------|---------------------|---------------------|
| Winter | December | Female | 71.74 \pm 0.18c | 1.72 \pm 0.08a | 6.83 \pm 0.17e | 18.26 \pm 0.53bcd | 134.58 \pm 3.65e |
| | | Male | 71.67 \pm 0.09d | 2.35 \pm 0.18c | 7.65 \pm 0.04fg | 18.24 \pm 0.06c | 141.81 \pm 0.62d |
| | January | Female | 70.92 \pm 0.52bc | 1.79 \pm 0.16ab | 9.00 \pm 0.17g | 19.06 \pm 0.13de | 157.30 \pm 1.98g |
| | | Male | 67.68 \pm 0.12ab | 1.85 \pm 0.16ab | 10.26 \pm 0.04j | 18.23 \pm 0.20c | 165.30 \pm 1.13g |
| | February | Female | 69.76 \pm 0.19b | 1.96 \pm 0.19abcd | 8.15 \pm 0.03f | 18.33 \pm 0.05bcd | 146.75 \pm 0.50f |
| | | Male | 71.53 \pm 0.65d | 1.99 \pm 0.02abc | 7.87 \pm 0.02gh | 17.93 \pm 0.09c | 142.55 \pm 0.62de |
| Spring | March | Female | 69.81 \pm 0.27b | 2.38 \pm 0.05f | 2.86 \pm 0.09b | 21.56 \pm 0.02g | 112.02 \pm 0.97b |
| | | Male | 76.07 \pm 0.53ef | 2.07 \pm 0.01abc | 1.60 \pm 0.02b | 18.34 \pm 0.16c | 87.84 \pm 0.83a |
| | April | Female | 75.67 \pm 0.11d | 2.17 \pm 0.04cdef | 2.33 \pm 0.06a | 20.16 \pm 0.23f | 101.68 \pm 1.50a |
| | | Male | 77.88 \pm 0.58g | 1.70 \pm 0.04a | 2.65 \pm 0.04c | 16.48 \pm 0.17a | 89.85 \pm 0.88a |
| | May | Female | 78.15 \pm 0.39e | 2.23 \pm 0.10def | 3.01 \pm 0.08b | 17.92 \pm 0.27bc | 98.8 \pm 1.81a |
| | | Male | 74.87 \pm 0.74e | 2.31 \pm 0.04c | 3.70 \pm 0.07d | 19.17 \pm 0.07d | 110.06 \pm 0.93b |
| Summer | June | Female | 77.69 \pm 0.63e | 2.42 \pm 0.04f | 3.65 \pm 0.26c | 17.39 \pm 0.13ab | 102.42 \pm 1.93a |
| | | Male | 76.38 \pm 0.69ef | 2.35 \pm 0.08c | 1.04 \pm 0.19a | 20.49 \pm 0.07e | 91.33 \pm 2.07a |
| | July | Female | 75.54 \pm 0.25d | 1.98 \pm 0.08abcde | 5.92 \pm 0.05d | 16.39 \pm 0.09a | 118.87 \pm 0.86c |
| | | Male | 76.68 \pm 0.41ef | 1.81 \pm 0.01ab | 6.03 \pm 0.12e | 17.45 \pm 0.13b | 124.12 \pm 1.64c |
| | August | Female | 66.00 \pm 0.21a | 2.08 \pm 0.02bcdef | 10.85 \pm 0.10h | 17.34 \pm 0.08ab | 167.03 \pm 1.25h |
| | | Male | 71.94 \pm 0.57d | 1.95 \pm 0.01abc | 9.06 \pm 0.04i | 17.03 \pm 0.12b | 149.73 \pm 0.82f |
| Autumn | September | Female | 64.88 \pm 0.80a | 1.84 \pm 0.06abc | 11.96 \pm 0.10i | 19.58 \pm 0.05ef | 185.96 \pm 0.92i |
| | | Male | 66.56 \pm 0.41a | 2.08 \pm 0.06abc | 10.73 \pm 0.20k | 18.03 \pm 0.06c | 168.69 \pm 1.69g |
| | October | Female | 66.41 \pm 0.60a | 3.92 \pm 0.09g | 5.98 \pm 0.23d | 18.55 \pm 0.55cde | 128.12 \pm 1.24d |
| | | Male | 69.18 \pm 0.61bc | 4.18 \pm 0.12d | 8.20 \pm 0.08h | 18.19 \pm 0.05c | 146.64 \pm 1.02ef |
| | November | Female | 71.55 \pm 0.35c | 2.32 \pm 0.05ef | 8.57 \pm 0.09fg | 23.20 \pm 0.44h | 170.02 \pm 2.31h |
| | | Male | 70.73 \pm 0.19cd | 2.15 \pm 0.27bc | 7.43 \pm 0.05f | 20.11 \pm 0.07e | 147.35 \pm 0.78f |

Table 3. Variance analysis of chemical contents and energetic value of *D. annularis* according to sexes and season

| Source of variation | Parameters | F (Fisher number) | P (p-value) |
|---------------------|-----------------|-------------------|-------------------|
| Sex | Moisture | 2.47 | 0.12 |
| | Protein | 0.001 | 0.97 |
| | Fat | 0.24 | 0.62 |
| | Ash | 5.00 | 0.03 |
| | Energetic value | 1.27 | 0.26 |
| Season | Moisture | 22.38 | <10 ⁻³ |
| | Protein | 5.63 | <10 ⁻³ |
| | Fat | 31.87 | <10 ⁻³ |
| | Ash | 7.91 | <10 ⁻³ |
| | Energetic value | 35.55 | <10 ⁻³ |
| Sex \times Season | Moisture | 0.64 | 0.60 |
| | Protein | 5.87 | 0.01 |
| | Fat | 0.74 | 0.53 |
| | Ash | 0.74 | 0.53 |
| | Energetic value | 0.44 | 0.72 |

The moisture, ash, total fat and protein contents of *D. annularis* species for both fish sexes are illustrated in Table 2. Moisture content ranged from 66.56 (in September) to 77.88% (in April) for males and from 64.88 (in September) to 78.15% (in May) for females with significant differences ($p < 0.05$). The highest levels, for both sexes, were found in summer. In the same time, lowest values of the fat content were observed ($1.04\% \pm 0.19$ and $2.33\% \pm 0.06$, for males and females, respectively). In addition, this fish species contained low fat for period from March to June. However, high values were found in September ($10.73\% \pm 0.20$ and $11.96\% \pm 0.10$, for males and females, respectively) with significant differences while compared to those obtained over the year. The protein content varied from 16.39 ± 0.55 (in July) to 23.20 ± 0.44 (in November) for females and from 16.48 ± 0.17 (in April) to 20.49 ± 0.07 (in June) for males. Ash content varied also over the year; however it remained under a maximum of $4.18\% \pm 0.12$ obtained in October for males with significant differences ($p < 0.05$). The period of the gonadic maturation being spread out between March and June corresponding to fish muscle with medium nutritional values (between 87.84 ± 0.83 and 124.12 ± 1.64 Kcal/100 g fresh fish) (Table 1). However,

before and after the gonadic maturation, the fish species have high nutritional values (168.69 ± 1.69 and 185.96 ± 0.92 Kcal/100 g fresh fish for males and females, respectively). The variance analysis of moisture, protein, fat and ash contents according to fish sex and season factors and the fish sex \times season interaction is presented in the Table 3. Season has a highly significant effect on moisture, protein, fat and ash contents ($p < 10^{-3}$) while the fish sex has a significant effect on ash content ($p = 0.03$). In addition, the sex-season interaction has a significant effect on protein content ($p = 0.01$).

Fatty acids profiles in *D. annularis* are presented in Table 4. The fatty acids are almost 90% of the total oil. Higher concentrations of palmitic acid (C16:0), oleic acid (C18:1), eicosapentaenoic acid (C20:5), docosahexaenoic acid (C22:6) and myristic acid (C14:0) were observed. The main fatty acids having high concentration over the year were palmitic (C16:0) acid with value varied from $26.38\% \pm 0.1$ to $32.79\% \pm 0.44$ (for males in spring and summer, respectively) and oleic (C18:1) acid with value varied from $26.28\% \pm 0.42$ to $35.76\% \pm 0.16$ (for females in winter and spring, respectively). The fatty acids with the highest percentage after these main fatty acids were C22:6, C18:0, C14:0 and C20:5 with values varied from $6.68\% \pm 0.01$ (for males in spring) to $10.67\% \pm 0.1$ (for females in winter), from 6.20 ± 0.1 to $9.88\% \pm 0.18$ (for females and males in spring, respectively), from $3.94\% \pm 0.01$ (for males in spring) to $6.15\% \pm 0.13$ (for females and in winter) and $1.13\% \pm 0.01$ to $11.03\% \pm 0.01$ (for males in summer and spring, respectively), respectively. Interestingly, in the Tunisian *D. annularis*, the fatty acid profiles of muscle exhibited a dominating saturated fatty acids (SFAs) and monounsaturated fatty acids (MUFAs). Seasonal variation of SFAs in muscle varies from 41.28% (in spring) to 47.44% (in summer) for female, and between 42.43% (in spring) and 50.34% (in winter) for males. The total MUFAs varied between 34.58% (in winter) and 37.68% (in autumn) for females, and from 34.73% (in winter) to 37.58% (in summer) for males. The

total polyunsaturated fatty acids (PUFAs) varied between 16.55% (in autumn) and 21.54% (in spring) for females, and from 14.32% (in summer) to 22.24% (in spring) for males. The ω -3 fatty acids content varied between 13.27% (in summer) and 17.61% (in spring) for females and between 10.25% (in summer) and 18.04% (in spring) for males. However, the highest and the lowest values of ω -3

fatty acid content were observed in spring and in summer, respectively. In addition, high value of $\Sigma\omega3/\Sigma\omega6$ ratio was observed essentially in spring (4.29 for males and 4.48 for females). No significant effect of fish sexes was noted on individual fatty acid variations, while the season has a significant effect on palmitic acid (C16:0) content ($p < 0.05$).

Table 4. Fatty acid profiles of *D. annularis* (% of total fatty acids); mean values \pm SD

| Nature | Fatty acids | Winter (January) | | Spring (April) | | Summer (July) | | Autumn (October) | |
|--------|------------------------------|------------------|------------------|------------------|------------------|------------------|------------------|------------------|------------------|
| | | Male | Female | Male | Female | Male | Female | Male | Female |
| SFA | C12:0 | 0.16 \pm 0.01 | 0.19 \pm 0.02 | 0.16 \pm 0.02 | 0.27 \pm 0.05 | 0.31 \pm 0.01 | 0.39 \pm 0.02 | 0.29 \pm 0.01 | 0.21 \pm 0.01 |
| | C14:0 | 5.27 \pm 0.11 | 6.15 \pm 0.13 | 3.94 \pm 0.01 | 4.63 \pm 0.16 | 4.87 \pm 0.01 | 5.36 \pm 0.04 | 4.66 \pm 0.07 | 4.46 \pm 0.1 |
| | C15:0 | 2.25 \pm 0.03 | 0.94 \pm 0.01 | 1.30 \pm 0.01 | 0.93 \pm 0.08 | 0.62 \pm 0.12 | 0.56 \pm 0.01 | 0.96 \pm 0.01 | 1.14 \pm 0.02 |
| | C16:0 | 31.66 \pm 0.42 | 29.14 \pm 0.26 | 26.38 \pm 0.1 | 28.39 \pm 0.5 | 32.79 \pm 0.44 | 32.36 \pm 0.01 | 30.23 \pm 0.41 | 30.21 \pm 0.34 |
| | C17:0 | 2.64 \pm 0.01 | 0.94 \pm 0.01 | 0.75 \pm 0.04 | 0.85 \pm 0.01 | 0.53 \pm 0.01 | 0.41 \pm 0.04 | 1.30 \pm 0.01 | 1.68 \pm 0.02 |
| | C18:0 | 8.35 \pm 0.05 | 7.08 \pm 0.02 | 9.88 \pm 0.18 | 6.20 \pm 0.1 | 8.96 \pm 0.01 | 8.33 \pm 0.14 | 8.01 \pm 0.07 | 8.03 \pm 0.07 |
| MUFA | C14:1 | 0.45 \pm 0.01 | 0.42 \pm 0.01 | 0.46 \pm 0.01 | 0.45 \pm 0.01 | 0.56 \pm 0.01 | 0.49 \pm 0.01 | 0.49 \pm 0.01 | 0.50 \pm 0.03 |
| | C15:1 | 1.78 \pm 0.02 | 1.18 \pm 0.05 | 2.29 \pm 0.1 | 0.19 \pm 0.01 | 0.29 \pm 0.01 | 0.23 \pm 0.01 | 0.32 \pm 0.01 | 0.44 \pm <0.01 |
| | C16:1 | 3.80 \pm 0.01 | 6.25 \pm 0.22 | 4.20 \pm 0.09 | 0.43 \pm 0.01 | 0.78 \pm 0.01 | 0.92 \pm 0.01 | 0.62 \pm 0.02 | 1.46 \pm 0.01 |
| | C17:1 | 1.07 \pm 0.01 | 0.44 \pm 0.01 | 0.21 \pm 0.01 | 0.32 \pm 0.02 | 0.34 \pm 0.01 | 0.32 \pm 0.01 | 0.42 \pm 0.01 | 0.33 \pm 0.01 |
| | C18:1 | 27.62 \pm 0.2 | 26.28 \pm 0.42 | 28.15 \pm 0.31 | 35.76 \pm 0.16 | 35.59 \pm 0.22 | 33.38 \pm 0.31 | 35.59 \pm 0.61 | 34.93 \pm 0.59 |
| PUFA | C18:2 | 1.88 \pm 0.05 | 1.95 \pm 0.01 | 0.77 \pm 0.01 | 2.36 \pm 0.01 | 2.51 \pm 0.01 | 2.75 \pm 0.05 | 1.46 \pm 0.01 | 1.70 \pm 0.02 |
| | C18:3 | 0.98 \pm 0.01 | 0.91 \pm 0.01 | 2.08 \pm 0.02 | 1.14 \pm 0.01 | 0.75 \pm 0.01 | 0.79 \pm 0.01 | 1.42 \pm 0.02 | 0.37 \pm 0.01 |
| | C20:4 | 0.96 \pm 0.01 | 1.27 \pm 0.01 | 1.34 \pm 0.01 | 0.42 \pm 0.09 | 0.80 \pm 0.01 | 0.39 \pm 0.01 | 1.21 \pm 0.012 | 0.74 \pm 0.01 |
| | C20:5 | 1.90 \pm 0.02 | 5.64 \pm 0.05 | 11.03 \pm 0.01 | 6.74 \pm 0.01 | 1.13 \pm 0.01 | 4.73 \pm 0.02 | 4.07 \pm 0.01 | 5.15 \pm 0.23 |
| | C22:5 | 0.34 \pm 0.01 | 0.49 \pm 0.01 | 0.32 \pm 0.01 | 0.44 \pm 0.01 | 0.42 \pm 0.01 | 0.40 \pm 0.01 | 0.40 \pm 0.01 | 0.41 \pm 0.01 |
| | C22:6 | 8.84 \pm 0.09 | 10.67 \pm 0.1 | 6.68 \pm 0.01 | 10.42 \pm 0.09 | 8.70 \pm 0.03 | 8.13 \pm 0.09 | 8.44 \pm 0.11 | 8.16 \pm 0.08 |
| | ω 3 | 11.08 | 16.80 | 18.04 | 17.61 | 10.25 | 13.27 | 12.92 | 13.73 |
| | ω 6 | 3.83 | 4.14 | 4.20 | 3.93 | 4.06 | 3.93 | 4.11 | 2.82 |
| | ω 3/ ω 6 | 2.89 | 4.05 | 4.29 | 4.48 | 2.52 | 3.37 | 3.14 | 4.86 |
| | Σ SFA | 50.34 | 44.46 | 42.43 | 41.28 | 48.10 | 47.44 | 45.48 | 45.75 |
| | Σ MUFA | 34.73 | 34.58 | 35.32 | 37.17 | 37.58 | 35.35 | 37.47 | 37.68 |
| | Σ PUFA | 14.92 | 20.94 | 22.24 | 21.54 | 14.32 | 17.20 | 17.04 | 16.55 |
| | Σ MUFA/ Σ PUFA | 2.33 | 1.65 | 1.58 | 1.72 | 2.62 | 2.05 | 2.20 | 2.27 |

SFA: saturated fatty acid; MUFA: monounsaturated fatty acid; PUFA: polyunsaturated fatty acid, ω -3 FA: omega-3 fatty acid; DHA: docosahexanoic; EPA: Eicosapentaenoic.

Table 5. Mineral contents (Fe, Zn, K, Mg, Na and Ca) in muscle of *D. annularis* (in mg/100 g of fresh fish); means \pm SD

| Mineral element | Winter (January) | | Spring (April) | | Summer (July) | | Autumn (October) | |
|-----------------|------------------|------------------|------------------|------------------|------------------|------------------|------------------|------------------|
| | Male | Female | Male | Female | Male | Female | Male | Female |
| Fe | 0.11 \pm 0.01 | 0.11 \pm 0.01 | 0.09 \pm 0.01 | 0.11 \pm 0.01 | 0.11 \pm 0.01 | 0.10 \pm 0.01 | 0.33 \pm 0.01 | 0.32 \pm 0.01 |
| Zn | 0.24 \pm 0.01 | 0.02 \pm 0.01 | 0.18 \pm 0.01 | 0.01 \pm 0.01 | 0.31 \pm 0.01 | 0.28 \pm 0.01 | 0.10 \pm 0.01 | 0.08 \pm 0.01 |
| Ca | 21.61 \pm 0.01 | 18.04 \pm 0.01 | 50.49 \pm 0.01 | 58.42 \pm 0.01 | 57.68 \pm 0.01 | 31.23 \pm 0.01 | 60.02 \pm 0.01 | 42.91 \pm 0.01 |
| K | 51.19 \pm 0.01 | 42.99 \pm 0.01 | 35.04 \pm 0.01 | 46.84 \pm 0.01 | 22.59 \pm 0.01 | 23.48 \pm 0.01 | 33.63 \pm 0.01 | 26.12 \pm 0.01 |
| Mg | 12.49 \pm 0.01 | 10.17 \pm 0.01 | 7.06 \pm 0.01 | 7.77 \pm 0.01 | 11.56 \pm 0.01 | 8.72 \pm 0.01 | 4.08 \pm 0.01 | 4.52 \pm 0.01 |
| Na | 25.44 \pm 0.01 | 30.38 \pm 0.01 | 19.47 \pm 0.01 | 24.00 \pm 0.01 | 15.83 \pm 0.01 | 13.38 \pm 0.01 | 14.49 \pm 0.01 | 14.57 \pm 0.01 |

The mineral contents of of *D. annularis* muscle were shown in Table 5. Values decreased from Ca to Zn as follows: Ca > K > Na > Mg > Fe > Zn, respectively. *D. annularis* is rich in potassium and calcium in all seasons. The K content varied between 22.59 \pm 0.01 mg/ 100 g of fresh fish in summer for males to 51.19 \pm 0.01 mg/ 100 g of fresh fish in winter for males. The Ca content showed the highest value in summer for males (57.68 \pm 0.01 mg/ 100 g of fresh fish) and in spring for females (58.42 \pm 0.01 mg/ 100 g of fresh fish). For Mg content, the lowest values of 4.08 \pm 0.01 and 4.52 \pm 0.01 mg/ 100 g of fresh fish respectively for males and females were observed in autumn. For Na, winter showed the highest values with 12.49 \pm 0.01 and 10.17 \pm 0.01 mg/ 100 g of fresh fish for males and females, respectively. The Fe content varied from 0.09 \pm 0.01 (in spring) to 0.33 \pm 0.01 mg/ 100 g of fresh fish (in autumn) for males. Similar values were observed for females. Generally, the season has a significant effect on the variation of Fe, Zn, Na, and Ca (p value of 0.01, 0.03, 0.04 and 0.01 respectively). However, there were no statistically significant differences for K and Mg contents ($p > 0.05$).

4. Discussion

According to the first part of results devoted to *D. annularis* collected from the Gulf of Gabes area (Tunisia), it can be concluded that the global chemical composition (moisture, ash, fat and protein contents) undergoes large fluctuation in response to a variety of factors. The protein content remained at high levels over the year (superior to 16 g/100 g of fresh fish). This tendency supports the reports of many studies [17,18] according to which the protein content of fish changes very little with season. Recently, [19] reported the same observation for the Mediterranean *Sardinella aurita* species. However, fat contents showed high variation. According to [20], these changes are likely associated with the preparation for reproductive activities. It was reported that sexual maturity stage of the fish affects the fat content due to increased consumption of fat reserves during the spawning period. In addition, the fat high variation can be explained by the gonadic maturation. [21] reported that breeding and

during the gametogenic cycle, fishes consume protein and fat reserves located in the muscle. The variation of fat and moisture contents of *D. annularis* showed that fat content varies widely with seasons, and in inverse proportion to moisture content. It is a fact widely recognized that moisture content in pelagic fish was inversely related to fat content [22]. Moreover, *D. annularis* showed minimal fat contents at the spring-beginning of summer and a maximal value at the end of September- beginning of autumn. These results allow us to conclude that the reproductive strategy of the *D. annularis* can be described as conservative, because storage of protein occurs before gametogenesis, and protein seasonal changes are negatively correlated with gonad development. The composition of a particular species often appears to vary depending on fishing location and seasons, but the basic causes of composition variations are usually related to the variation in the amount and quality of food available for fish eats and the amount of movement it makes. For example, fish usually stop feeding before they spawn, and draw on their reserves of fat and protein. Again, when fish species are overcrowded, there may not be enough food to go round; intake will be low and composition will change accordingly. Reduction in a basic food resource, plankton for example, can affect the whole food chain. The nutrition of *D. annularis* is multivariate (worms, crustaceans, molluscs, echinoderms, hydrozoans and algae). It is present at variable amounts all the year, Upwelling and mixing also occurs within the storm systems of the west-wind belt. The mixing by winter storms brings nutrients to the surface; this results in plankton blooms in early summer when the sun is high over the horizon and delivers the light necessary for photosynthesis. Also, the sun warms the surface waters and provides a measure of stability, so phytoplankton is kept in sunlit surface waters, rather than being mixed downward into the dark regions. Generally, regions characterized by a succession of strong mixing alternating with warming and much sunlight yields the highest productivity. So, the fat content is higher in autumn and winter and lower in spring and summer which coincides with the period of the gonadic maturation. Therefore, the fat content varies significantly with the seasons. Furthermore, the sex has no effect on the variation of the moisture, fat and protein contents. This result is in agreement with other research [5].

In the second part of this work, we analyzed the fatty acids composition of *D. annularis* lipids. Oleic acid (C18:1) was the most abundant fatty acids over year. This result is in accordance with other studies [23,24]. This observation was typical because oleic acid has exogenous origin and usually indicates the fish diet type. Palmitic acid (16:0) is the second major fatty acid in all seasons. Other studies on Mediterranean fish reported that this fatty acid is the most abundant [8,19]. This fatty acid was found to be the key metabolite in fish [25]. The fatty acids C22:6 (varies between 6.68% and 10.67%) and C18:0 (Varied between 6.20% and 9.88%) were reported to have the highest percentages after C18:1 and C16:0. These results were in accordance with those reported for other fish species [26,27,28], but they were different from those showed in other studies [19]. For example, according to [8], the second most abundant fatty acid in the anchovy and in picarel was C22:6 with value of 12.23 and 14.23%,

respectively (obtained in February). Moreover, it is very important to note that fatty acid profile differs from males to females. Saturated and monounsaturated fatty acids are the most abundant fatty acids in *D. annularis*. This result is in agreement with data reported by other studies [29]. Generally, SFAs and MUFAs are abundant in fish from warm or temperate regions, compared to PUFAs which show high levels in fish from cold regions [30]. It is also important to mention that the PUFAs have a high level over year (values varied between 15% and 22%). Interestingly, total omega-3 content of the total fatty acids (11%< ω -3<18%) was superior to *Sparus aurata* fish species belonging to the same family (0.9-8.6%) [31]. However, *D. annularis* oil is comparable to other commercial marine fish oils such as Atlantic mackerel, *Scomber scombrus* (about 18%). Therefore, Tunisian *D. annularis* seems to be a good source of omega-3 fatty acids recognized for their health benefits.

The analysis of the mineral content showed that Ca, K, Na and Mg were the most abundant elements, however Zn and Fe were at very low levels in *D. annularis* muscles. As reported for protein and lipid, mineral content varied with seasons. Generally, the most abundant elements are essential to cellular metabolism and present at high concentrations in biological tissues [32]. However, Zn and Fe at low rate are involved in enzyme activities. Thus, this fish can serve as a good source of essential elements [33,34].

5. Conclusion

Diplodus annularis species from the Gabes Gulf (Tunisia) was found to be a good source of protein and fat. The chemical composition showed a large fluctuation over the year in response to various factors: season, gonadic maturation and sex. Fat content was lower in June for males and in April for females, but higher in September for both sexes. It varies with season and gonadic maturation in inverse proportion to moisture content. The major fatty acids in *D. annularis* lipids were palmitic acid, stearic acid, oleic acid, eicosapentaenoic acid, docosahexaenoic acid and myristic. The high amounts of saturated and monounsaturated fatty acids in the screened species are almost in agreement with other studies. The high nutritional quality of this fish species indicates its importance as a source of energetic and nutritional values over the year. The significant seasonal differences observed in the mineral contents are mainly related to the fish diet but also to various environmental factors.

References

- [1] Aitken, A., Mackie, I., Merrit, J. and Windsor, M.L., "Fish Handling and Processing". Ministry of Agriculture, Fisheries and Food, Torry Research Station Edinburgh, Scotland, UK. 1982.
- [2] Simopoulos, A., "Nutritional aspects of fish," In: Luten J, Orrensen B, Oehlenschla T, Ger J (eds), *Seafood from Producer to Consumer, Integrated Approach to Quality*. London, UK: Elsevier Science. pp 589-607. 1997.
- [3] Gonzalez, S., Flick, G.J., O'keefe, S.F., Duncan, S.E, Mclean, E. and Craig, S.R., "Composition of farmed and wild yellow perch (*Perca flavescens*)", *Journal of Food Composition and analysis*, 19. 720-726. 2006.
- [4] Haliloglu, H.I., Bayir, A., Sirkeciogil, A.N., Aras, N.M. and Atamanalp, M., "Comparison of fatty acid composition in some

- tissue of rainbow trout (*Oncorhynchus mykiss*) living in seawater and freshwater". *Food Chemistry*, 86: 55-59, 2004.
- [5] Ketata Khitouni, I., Abdelmouleh, A., Bouain, A. and Boudhrioua Mihoubi., N., "Variations of the chemical compositions of five coastal catch fish species of the Gulf of Gabes (Tunisia)". *Cybiurn* 34,175-183. 2010.
- [6] Kozlova, T.A., "Seasonal cycles in total chemical composition of two Lake Baikal benthic-pelagic sculpins (*Cottocomephorus cottoides*)", *Journal of Fish Biology*, 50, 734-743. 1997.
- [7] Gockse, M.A., Tasbozan, O., Celik, M, and Tabakoglu, S.S., "Seasonal variation in proximate and fatty acid compositions of female common sole (*Solea solea*)". *Food Chemistry*, 88: 419-423. 2004.
- [8] Zlatanov, S, and Laskaridis, K., "Seasonal variation in the fatty acid composition of three Mediterranean fish-sardine (*Sardina pilchardus*), anchovy (*Engraulis encrasicolus*) and picarel (*Spicara smaris*)". *Food Chemistry*, 103. 725-728, 2007.
- [9] Noël, L., Chafey, C., Testu, C., Pinte, J., Velge, P, and Guerin, T., "Contamination levels of lead, cadmium and mercury in imported and domestic obsters and large crab species consumed in France: differences between white and brown meat". *Journal of Food Composition and analysis*, 24, 368-375. 2011.
- [10] Roy, P.K, and Lall, S.P., "Mineral nutrition of haddock *Melanogrammus aeglefinus* (L.): a comparison of wild and cultured stock". *Journal of Fish Biology*, 68: 1460-1472. 2006.
- [11] Direction Générale de la Pêche et de l'Aquaculture DGPA, "Annuaire des statistiques des produits de la pêche. Ministère de l'Agriculture, Tunisie", 2009.
- [12] Fischer, W., Bauchot, M. L, and Schneider, M., "Fiches F.A.O. d'identification des espèces pour les besoins de la pêche "Révision" Méditerranée et Mer noire" Zone de pêche 37. Volume I et II. Vertébrés. Rome, F.A.O. 1530 p, 1987.
- [13] AOAC, "Official Methods of analysis Washington". DC. Association of official Analytical Chemists. 2000.
- [14] Bligh, E.G, and Dyer, W.J., "A rapid method for total lipid extraction and purification", *Canadian Journal of Biochemistry and Physiology*, 37, 911-917, 1959.
- [15] Aubourg, S., Gallardo, J.M, and Sotelo, C., "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, 1991.
- [16] Jabeen, F, and Chaudhry, A.S., "Chemical compositions and fatty acid profiles of three freshwater fish species". *Food Chemistry*, 125, 991-996, 2011.
- [17] Emokpae, A.O., "Preliminary studies on the chemical and weight composition of some commercially important species of fish and shrimp caught in the Nigerian inshore waters". *International Journal of Food Science and Technology*, 18, 271-283. 1983.
- [18] Njinkoue, J.M., Barnathan, G., Miralles, J., Gaydoud, E.M, and Sambe, A., "Lipids and fatty acids in muscle, liver and skin of three edible fish from the Senegalese coast: *Sardinella maderensis*, *Sardinella aurita* and *Cephalopholis taeniops*. *Comparative Biochemistry and Physiology*, 131, 395-402. 2002.
- [19] Ben Rebah, F., Abdelmouleh, A., Kammoun, W, and Yezza, A., "Seasonal variation of lipid content and fatty acid composition of *Sardinella aurita* from the Tunisian coast", *Journal of the Marine Biological Association of the United Kingdom*, 90: 569-573. 2010.
- [20] Rajasilta, M., "Relationship between food, fat, sexual maturation, and spawning time of Baltic herring (*Clupea harengus membras*) in the Archipelago Sea. *Canadian Journal of Fisheries and Aquatic Sciences* 9, 644-654, 1992.
- [21] Barber, B.J, and Blake, N.J., "Intra-organ biochemical transformations associated with oogenesis in the bay scallop, *Argopecten irradians concentricus* (Say), as indicated by ¹⁴C incorporation". *Biological Bulletin* 168, 39-49. 1985.
- [22] Grigorakis, K., Alexis, M.N, Taylor, A, and Hole, M., "Comparison of wild and cultured gilthead sea bream (*Sparus aurata*): composition, appearance and seasonal variations". *International Journal of Food Science Technology*, 37: 1-8. 2002.
- [23] Ackman R.G. Fish lipids. Part1. In: Connell JJ (ed.), *Advances in fish science and technology*, Farnham, Surrey: Fishing News Books Ltd. Pp, 86-103, 1980.
- [24] Ackman, R.G. "Fatty acids". In: Ackman RG (ed), *Marine Biogenic Lipids, Fats and Oils*. Boca Raton: CRC Press. pp 145-178, 1989.
- [25] Andrade, A.D., Rubira, A.F, Matsushita, M, and Souza, N.E., "ω3 Fatty acids in freshwater fish from south Brazil. *Journal of American Oil Chemistry*, 72, 1207-1210, 1995.
- [26] Karakoltsidis, P. A., Zotos, A. and Constantinides, S. M., "Composition of the commercially important Mediterranean finfish, crustaceans and mollusks. *Journal of Food Composition and Analysis*," 8, 258-273, 1995.
- [27] Guner, S., Dincer, B., Alemdag, N., Colak, A, and Tufekci, M., "Proximate and selected mineral content of commercially important fish species from the Black sea". *Journal of the Science of Food and Agriculture* 78, 337-342, 1998.
- [28] Luczynska, J., Borejszo, Z, and Luczynski, M.J., "The composition of fatty acids in muscles of six freshwater fish species from the Mazurian great lakes (Northeastern Poland)". *Archives of Polish Fisheries* 16, 167-178, 2008.
- [29] Gabriel, F., Lozano, B., Benedito-Palos, L., Navarro, J.C., Kaushik, S, and Perez-Sanchez, J., "Prediction of fillet fatty acid composition of market-size gilthead sea bream (*Sparus aurata*) using a regression modelling approach". *Aquaculture* 319, 81-88, 2011.
- [30] Dey, I., Buda, C., Wiik, H., Halver, J.E, and Farkas, T., "Molecular and structural composition of phospholipid membranes in livers of marine and freshwater fish in relation to temperature". *Proceedings of the National Academy of Sciences of the United States of America*, 90, 7498-7502, 1993.
- [31] Cardinal, M., Cornet, J., Donnay-Moreno, C., Gouygou, J.P, Bergé, J.P, Rocha, E., Soares, S., Escórcio, C., Borges, P, and Valente, L.M.P., "Seasonal variation of physical, chemical and sensory characteristics of sea bream (*Sparus aurata*) reared under intensive conditions in Southern Europe". *Food Control*, 22, 574-585, 2011.
- [32] Wagner, A, and Boman, J., "Biomonitoring of trace elements in muscle and liver tissue of freshwater fish". *Spectrochimica Acta part B-Atomic Spectroscopy*, 58, 2215-2226, 2003.
- [33] Johnson, M, and Fischer, J., "Role of minerals in protection against free radicals". *Food Technology*, 48, 112-120, 1994.
- [34] Lal, S, Macro and trace elements in fish and shellfish. In: Ruiter A (ed), *Fish and Fishery Products: Composition, Nutritive Properties and Stability*. Wallingford, CN, USA: CAB International. pp 187-214, 1995.