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# Seasonal effects in the nutritional quality of the body structural tissue of cephalopods

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## Abstract

The most commonly-consumed cephalopods around the world (the common cuttlefish, Sepia officinalis, European squid, Loligo vulgaris, common octopus, Octopus vulgaris and musky octopus, Eledone moschata) were evaluated in terms of seasonal variations in proximate and fatty acid compositions. The arms of the octopuses were used for this study, whereas the mantle of the other species (squids and cuttlefish) were used for the analyses. The lipid contents of species were found to be very low and considered as lean. The lowest lipid content was obtained from E. moschata (0.60–0.68%), whereas L. vulgaris gave the highest level of lipid  $(1.34-1.92)$  throughout all seasons. Unlike lipid content, protein contents of cephalopods did not change across the seasons. The fatty acid compositions of each species ranged from 28.18% to 35.28% saturated (SFA), 4.36–9.47% monounsaturated (MUFAs) and 43.58–56.55% polyunsaturated acids (PUFAs). The highest proportions of fatty acids in cephalopods were myristic acid (C14:0, 0.96–2.96%), palmitic acid (C16:0, 15.53– 25.20%), heptadecanoic acid (C17:0, 1.05–2.56%), stearic acid (C18:0, 4.32–9.96%), oleic acid (*cis18:1 n*–9, 1.80–4.29%), *cis*-11-eicosenoic acid (C20:1, 2.07–4.69%), linoleic acid (C18:2 n–6, 0.17–1.95%), arachidonic acid (C20:4 n–6, 1.48–11.65%), cis-5,8,11,14,17-eicosapentaenoic acid (EPA, C20:5 n–3, 7.86–16.97%) and cis-4,7,10,13,16,19-docosahexaenoic acid (DHA, C22:6 n–3, 20.99–39.00%). The results indicated that these cephalopod species are excellent protein sources and very rich in  $n-3$  fatty acids.  $© 2007 Elsevier Ltd. All rights reserved.$ 

Keywords: Nutritional quality; Cephalopods; GC

## 1. Introduction

The cephalopods represent an important economic seafood for human consumption and contribute to 14% of the world fisheries, according to [FAO \(2004\)](#page-5-0). Due to their nutritional and market value, cephalopod aquaculture has also shown an increase during the past few years ([Almansa et al., 2006\)](#page-4-0).

Seafood is generally the main contributor of  $n-3$  PUFA in the human diet. Lipids of marine fish species are generally characterised by high levels of long-chain  $n-3$ polyunsaturated fatty acids ([Steffens, 1997](#page-5-0)). Among the

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polyunsaturated fatty acids, EPA (eicosapentaenoic acid, C20:5  $n-3$ ) and DHA (docosahexaenoic acid, C22:6  $n-3$ ) are the dominant  $n-3$  fatty acids in marine fish ([Ackman,](#page-4-0) [1989](#page-4-0)). These fatty acids are of great importance to humans for the prevention of coronary heart disease ([Conner, 2000;](#page-5-0) [Kinsella, 1987; Mozaffarian, Bryson, Lemaitre, Burke, &](#page-5-0) [Siscovick, 2005\)](#page-5-0). General recommendations for daily dietary intakes of DHA/EPA are 0.5 g for infants, and 1 g/day for adults [\(Kris-Etherton, Harris, & Appel,](#page-5-0) [2002](#page-5-0)). Although cephalopods contain low levels of fat, it is rich in n-3 fatty acids ([Ozyurt, Duysak, Akamca, &](#page-5-0) [Tureli, 2006; Passi, Cataudella, Di Marco, De Simone, &](#page-5-0) [Rastrelli, 2002; Zlatanos, Laskaridis, Feist, & Sagredos,](#page-5-0) [2006](#page-5-0)).

The fatty acid composition of fish reflects the fatty acid composition of their natural foods [\(Grigorakis, Alexis,](#page-5-0)

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[Taylor, & Hole, 2002; Henderson & Tocher, 1987; Van Vli](#page-5-0)[et & Katan, 1990](#page-5-0)). Diet, location and season are the major factors affecting the fatty acid composition [\(Gruger, 1967,](#page-5-0) [chap. 1\)](#page-5-0), while seasonal changes in water temperature and nutrients are the major factors affecting composition of fish muscle. From the nutritional point of view, less attention has been paid to the fatty acid profile of cephalopods. Therefore, it is important to determine the level of lipid and fatty acid profile of these species. The current sudy is a comprehensive seasonal comparison of the biochemical composition (protein, lipid, moisture, ash and fatty acids) of four cephalopod species. The most popular and commonly-consumed cephalopods in the Mediterranean sea are the common cuttlefish, Sepia officinalis (Cephalopoda, Sepiidae), European squid, Loligo vulgaris (Cephalopoda, Loliginidae), common octopus, Octopus vulgaris (Cephalopoda, Octopodidae) and musky octopus, Eledone moschata (Cephalopoda, Octopodidae).

#### 2. Materials and methods

# 2.1. Sample preparation

After catching the cephalopods by trawl in spring, autumn and winter (fishing is forbidden in summer), the cephalopod species were immediately iced and transported to the laboratory. These cephalopod species were caught in the eastern Mediterranean sea. The species of cephalopods identified and their mantle length and total weight were measured (Fig. 1). Mean values of mantle lengths and body weights of the captured species were  $10.7 \pm 2.71$  cm and  $126 \pm 82.3$  g for S. *officinalis*,  $15.3 \pm 4.38$  cm and  $87.7 \pm 1.3$ 66.1 g for *L. vulgaris*,  $8.08 \pm 1.60$  cm and  $99.2 \pm 49.2$  g for *E. moschata*, and  $10.3 \pm 3.82$  cm and  $370 \pm 448$  g for O. vulgaris. A minimum of three individuals from each species were gutted, filleted and minced for analyses. Arms of the octopuses were used for this study whereas mantles of the other species (squids and cuttlefish) were used for the analyses.

#### 2.2. FAME analyses

Lipid extraction was made according to the [Bligh and](#page-5-0) [Dyer method \(1959\)](#page-5-0). Methyl esters were prepared by transmethylation using  $2 M KOH$  in methanol and *n*-heptane, according to the method described by [Ichihara, Shibahara,](#page-5-0) [Yamamoto, and Nakayama \(1996\),](#page-5-0) with minor modification. Extracted oil (10 mg) was dissolved in 2 ml *n*-heptane, followed by 4 ml of 2 M methanolic KOH. The tube was then vortexed for 2 min at room temperature. After centrifugation at 4000 rpm for 10 min, the *n*-heptane layer was taken for GC analysis.

#### 2.3. Gas chromatographic condition

The fatty acid composition was analysed by GC with autosampler (Clarus 500, Perkin Elmer, Norwalk, CT) equipped with a flame ionisation detector and a fused silica capillary column (30 m  $\times$  0.32 mm  $\times$  0.25 µm BP20; SGE, Ringwood, Australia). The oven temperature was initially 140 °C, for 5 min, rising to 200 °C at a rate of  $4 \text{ }^{\circ}C/\text{min}$ and then to 220 °C at a rate of 1 °C/min, while the injector and the detector temperature were set at  $220^{\circ}$ C and 280 °C, respectively. The sample size was 1  $\mu$ l and the carrier gas was controlled at 16 psi. The split ratio used was 1:100. Fatty acids were identified by comparing the retention times of FAME with the standard 37 component FAME mixture. Two replicate GC analyses were performed and the results were expressed in GC area%, as a mean value  $\pm$  standard deviation.



Fig. 1. Total length (TL) and mantle length (ML) of the cephalopods.

# 2.4. Statistical analysis

For data analysis, each sampling season was subjected to one-way analysis of variance, at the 5% confidence level, using the Duncan multiple range test.

# 3. Results and discussion

## 3.1. Proximate composition

Table 1 shows English and taxonomic names of the cephalopods species studied. Table 2 shows the seasonal variations in the proximate compositions of cephalopods. Although all species showed variations in the total lipid content of their body structural tissue throughout all of the seasons, no significant seasonal variation in lipid content was observed in any species apart from O. vulgaris  $(p < 0.05)$ . The lowest lipid content was observed for E. moschata  $(0.60-0.68\%)$  whereas L. vulgaris gave the highest level of lipid (1.34–1.92%), throughout all seasons. The lipid contents of species were found to be very low and the species were all considered as lean. Similar results were found in cephalopod molluscs (cuttlefish, octopus and squid) by [Zlatanos et al. \(2006\)](#page-5-0). In general, lipid content of cephalopods was observed to increase in autumn. This may be due to the fact that nutrients in summer and

Table 1

English and taxonomic names of cephalopod species

Name in English	Scientific name
Common cuttlefish	Sepia officinalis
European squid	Loligo vulgaris
Common octopus	Octopus vulgaris
Musky octopus	Eledone moschata

Table 2

The proximate compositions (%) of edible parts of cephalopods

autumn are more abundant than in other seasons. A considerable amount of research has been done on seasonal changes in the lipid level of various species and it was concluded that the major factor affecting the lipid level was the abundance of food [\(Ackman, 1995; Robards, Anthony,](#page-4-0) [Rose, & Piatt, 1999; Rosa, Nunes, & Sousa Reis, 2002\)](#page-4-0). It has also been indicated that the lipid content of fish changes due to species, gender, geographical origin and season ([Rasoarahona, Barnathan, Bianchini, & Gaydon,](#page-5-0) [2005](#page-5-0)).

Unlike lipid contents, protein contents of cephalopods did not change throughout all of seasons. Generally, E. moschata had the lowest level of protein while L. vulgaris gave the highest protein content throughout the year (Table 2). In spring and winter, L. vulgaris gave the highest protein level while S. officinalis gave the highest level in autumn. S. officinalis gave a significantly higher protein content ( $p < 0.01$ ) in autumn than in spring and winter, while the lowest protein content was obtained from L. vulgaris  $(p < 0.01)$  in autumn, compared to spring and winter (Table 2).

The results indicate that these cephalopod species are excellent protein sources and low in fat content. Moisture content of cephalopods significantly fluctuated ( $p \le 0.01$ ) across all seasons. Ash contents of cephalopods were also found to be significantly different ( $p \le 0.05$ ) throughout all of the seasons, except for E. moschata ( $p > 0.05$ ).

# 3.2. Fatty acid profiles

[Table 3](#page-3-0) shows seasonal variations in fatty acid compositions of cephalopods. The results of the fatty acid composition analysis show that cephalopods are very rich in  $n-3$ fatty acids. The fatty acid compositions of each species ranged from 28.18–35.28% saturated (SFA), 4.36–9.47%



Means followed by different letters within the same row are significantly different:  $*p < 0.01$ ,  $p < 0.05$ , ns: not significant.

<span id="page-3-0"></span>



For each species, values in the same row with differing letters are significantly different ( $p < 0.05$ ).

<span id="page-4-0"></span>monounsaturated (MUFAs) and 43.6–56.6% polyunsaturated fatty acids (PUFAs). Among the species, the percentages of SFA in L. vulgaris, MUFA in O. vulgaris and PUFA in S. *officinalis* were found to be higher than others throughout all seasons. The highest proportions of fatty acids in cephalopods were myristic acid (C14:0, 0.96– 2.96%), palmitic acid (C16:0, 15.5–25.2%), heptadecanoic acid (C17:0, 1.05–2.56%), stearic acid (C18:0, 4.32– 9.96%), oleic acid (*cis*-C18:1 n–9, 1.8–4.29%), *cis*-11-eicosenoic acid (C20:1, 2.07–4.69%), linoleic acid (C18:2 n-6, 0.17–1.95%), arachidonic acid (C20:4 n-6, 1.48–11.65),  $cis$ -5,8,11,14,17-eicosapentaenoic acid (EPA, C20:5  $n-3$ , 7.86–16.97%) and cis-4, 7,10,13,16,19-docosahexaenoic acid (DHA, C22:6  $n-3$ , 20.99–39.0%). These results are in agreement with previous studies on fatty acids of cephalopods ([Ozyurt et al., 2006; Reale et al., 2006; Rosa, Pere](#page-5-0)[ira, & Nunes, 2005; Zlatanos et al., 2006\)](#page-5-0). It was also observed that the proportion of these fatty acids changed significantly throughout the seasons (Table 4).

The proportions of  $n-3$  PUFAs (ranging from 29.27% for E. moschata to 50.44% for L. vulgaris in autumn) were higher than those of  $n-6$  PUFAs (ranging from 2.30 in winter to 4.01 for *L. vulgaris* in autumn). The UK department of Health recommends a maximum dietary ratio of

Table 4

The results of the analysis of variance on the seasonal variation in the fatty acid composition of cephalopods

Fatty acids	S. officinalis	L. vulgaris	O. vulgaris	E. moschata
$(\%)$	(p)	(p)	(p)	(p)
C14:0	ns	**	ns	ns
C15:0	ns	$**$	ns	ns
C16:0	ns	**	ns	ns
C17:0	ns	**	ns	ns
C18:0	ns	**	ns	ns
C20:0	ns	ns	ns	ns
C22:0	$**$	$**$	*	ns
<b>SFA</b>				
C14:1	$\ast$	**	** **	ns
C15:1	ns *	ns **		ns
C16:1			** *	ns
C17:1	ns	ns *	$\ast$	ns
C18:1 $n-9$	ns	**	**	ns
C20:1	ns			ns *
C22:1 $n-9$	**	**	**	
C24:1	ns	ns	ns	ns
<b>MUFA</b>	**			
$C18:2 n-6$		ns	ns	ns
C18:3 $n-6$	ns *	ns	ns	ns
$C18:3 n-3$		ns	ns	ns
C20:2	ns *	ns	ns	ns *
C20:3 $n-6$		ns **	ns	
C20:3 $n-3$	ns $**$	**	ns $**$	ns
C <sub>20</sub> :4 $n-6$		$**$		ns *
C20:5 $n-3$	ns **		ns **	$**$
C22:2		ns		
C22:6 $n-3$	ns	*	*	*
PIIFA				

PUFA

\*\*p < 0.01,  $p$  < 0.05, ns: not significant.

 $n-6/n-3$  of 4.0 [\(HMSO, 1994\)](#page-5-0). Values higher than the maximum value are harmful to health and may cause cardiovascular diseases [\(Moreira, Visentainer, de Souza, &](#page-5-0) [Matsushita, 2001](#page-5-0)). In this study, the ratio of  $n-6/n-3$ was found to be much lower than 4. A recommended minimum value of PUFA/SFA ratio is 0.45 [\(HMSO, 1994](#page-5-0)), which is lower than those obtained from all species studied. The highest PUFA/SFA ratio was obtained from all species in spring, except E. moschata, which had the highest PUFA/SFA ratio in autumn, whereas the lowest values were found in winter for all species, except E. moschata (in spring).

MUFA have been paid attention during the last decades due to their beneficial effects on cardiovascular heart disease ([Kalogeropoulus, Andrikopoulos, & Hassapidou,](#page-5-0) [2004](#page-5-0)). [Keys et al. \(1986\)](#page-5-0) indicated that populations which consumed olive oil had a low rate of CHD, especially Mediterranean countries. Therefore, inclusion of higher dietary MUFA has been suggested by NCEP (2001). The levels of MUFA in this study were found to be low compared to SFA and PUFA, ranging from 4.36% to 9.47%. Consumption of cephalopods would also contribute the intake of MUFA.

Palmitic acid (C16:0) was the primary saturated fatty acid, whereas oleic acid (C18:1  $n-9$ ) and cis-11-eicosenoic acid (C20:1) were the most represented MUFAs. The major fatty acids identified as polyunsaturated fatty acids were eicosapentaenoic acid (EPA, C20:5  $n-3$ ) and docosahexaenoic acid (DHA, C22:6  $n-3$ ). These results are supported by the findings of other researchers ([Ozyurt et al.,](#page-5-0) [2006; Reale et al., 2006; Zlatanos et al., 2006\)](#page-5-0). The highest proportions of EPA were obtained from S. officinalis  $(16.13)$  in winter, *O. vulgaris*  $(16.97)$  in spring, and *L. vulgaris* (14.31) and *E. moschata* (12.23) in autumn.

A high proportion of DHA was found in all species of cephalopods in this study, compared to fish species ([Ozogul](#page-5-0) & Ozogul, 2007; Ozogul, Ozogul, & Alagöz, 2007). Three species of cephalopods had the highest DHA in spring  $(33.0\%$  for *S. officinalis*,  $39.0\%$  for *L. vulgaris* and  $29.6\%$ for *O. vulgaris*), the exception being *E. moschata*  $(28.2\%)$ in autumn). Although cephalopods contained small amounts of fat, EPA and DHA were found to be high in all species ([Kalogeropoulus et al., 2004; Ozyurt et al.,](#page-5-0) [2006; Reale et al., 2006; Zlatanos et al., 2006\)](#page-5-0), increasing the value of cephalopods compared to cultured or wild fish species.

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