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Discarded fish on the Spanish Mediterranean coast: influence of season on fatty acids profiles

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Abstract

This work focused on determining the fatty acid (FA) composition of commonly discarded fish on the Spanish Mediterranean coast in winter and summer to assess their potential use as raw sources of very long-chain polyunsaturated fatty acids (VLCPUFAs). Total *n*-3 VLCPUFA content significantly varies depending on capture season, and values were higher in winter for *Pomadasys incisus* (1.36 g/100 g), *Chelidonichthys lucerna* (1.67 g/100 g) and *Trachinus draco* (2.04 g/100 g), while *Pagellus acarne* had larger *n*-3 PUFA amounts in summer (3.89 g/100 g). Generally for most species, monounsaturated FA, eicosapentaenoic acid (20:5*n*-3) and total FA had higher values in summer, while both the *n*-3 VLCPUFA fraction and DHA content were higher in winter. Knowledge of these changes allows the processes for their proper use as valuable PUFA sources to be adjusted. The discarded fish herein studied could be generally considered to be raw *n*-3 VLCPUFAs sources.

Keywords: Fish composition; discarded fish; fatty acid profiles; Mediterranean coast; *n*-3 very long-chain PUFAs; seasonal changes.

Introduction

Global extractive fishing catches have remained at around 90 million tons per year for the last two decades (FAO, 2018). It has been estimated that about 10.8% of this production is discarded as by-products (Gilman *et al.*, 2020). Fish discard is the portion of total organic matter of animal origin that is wasted or dumped at sea. The main reasons for discarding fish are certain species' low commercial value (no target species) and legal restrictions for species of commercial interest because fish is regulated by quotas (Total Allowable Catch, TAC) and size-regulated fish (Minimum Conservation Reference Size (MCRS)).

The European Commission has proposed plans to promote the use of more selective fishing practices and to, thus, reduce the amount of discarded fish. The discarded fish biomass is a rich source of high-quality compounds of pharmaceutical, nutritional or biotechnological interest (Rincón Cervera *et al.*, 2014; Nascimento *et al.*, 2015; Blanco *et al.*, 2018). Furthermore, using fish discards not only mitigates the drawback of their disposal, but also prevents negative effects on biological resources and ecosystems.

The marine biomass is generally rich in *n*-3 very long-chain polyunsaturated fatty acids (*n*-3 VLCPUFA), such as eicosapentaenoic acid (EPA, 20:5*n*-3) and docosahexaenoic acid (DHA, 22:6*n*-3) (Nascimento *et al.*, 2015). These nutrients are conditionally essential for humans because of their well-proven health benefits (Simopoulos, 2004), particularly those related to the prevention of cardiovascular and neurological diseases. In addition, both EPA and DHA are precursors of lipid mediators (eicosanoids and docosanoids, respectively), which have anti-inflammatory properties (Calder *et al.*, 2017).

Nowadays, searching for alternative sources of *n*-3 VLCPUFA is mandatory, and growing interest is being shown in fish discard and seafood industrial waste given their high content of these important nutrients (Orban *et al.*, 2011; Rincón-Cervera *et al.*, 2014; Nascimento *et al.*, 2015; Blanco *et al.*, 2018; Kandyliari *et al.*, 2020), and also because using discarded fish can prevent overfishing commonly consumed *n*-3 VLCPUFA-rich fish species.

The lipid content and fatty acid (FA) profile of fish vary according to different variables like seasonality, geographical area, size, sex and sexual maturation (Shirai *et al.*, 2002). The most significant environmental factors are water temperature, fluctuation in food availability and

reproductive cycle (Kołakowska *et al.*, 2003).

The main types of fishing gear operating in the Alboran Sea are artisanal (small scale) fisheries. Nevertheless, the most significant one in terms of both the volume of catches and economic value is bottom trawling. Trawling fishing gear can have a stronger impact on the marine ecosystem and may lead to higher discarding rates than other gear operating at open sea (Sánchez *et al.*, 2004; Tsagarakis *et al.*, 2014; Urra *et al.*, 2017). The main discarded species of Alboran fisheries include some of the species herein analyzed, such as seabream (*Pagellus acarne*, *P. erythrinus*), mackerel (*Trachurus mediterraneus*, *T. trachurus*) and horse mackerel (*T. trachurus*) as the main discarded species (Carbonel *et al.*, 2018). In addition to these species, red porgy (*Pagrus pagrus*), bogue (*Boops boops*) and picarels (*Spicara* spp.) are also some species discarded by bottom trawl fisheries in the Western Mediterranean subregion (Sánchez *et al.*, 2004; FAO, 2018).

Both the lipid fraction and FA content of discarded marine fish have been reported in some species, such as the gadiform family (Falch *et al.*, 2006 a, b), sea sardine (*Sardina pilchardus*), mackerel (*S. colias*), horse mackerel (*T. trachurus*) (García-Moreno *et al.*, 2013), bogue (*Boops boops*) (Orban *et al.* 2011, Morales-Medina *et al.*, 2015) and nakedband gaper (*Champsodon nudivittis*) (Ozyilmaz, 2016). The FA profiles from fish processing industry by-products have also been studied (Rincón-Cervera *et al.*, 2014; Nascimento *et al.*, 2015; Kandyliari *et al.*, 2020). However, more research is needed to improve knowledge on FA composition and its seasonality for a wide range of species discarded on the Mediterranean coast. Furthermore, profound knowledge of the total amount of these species' discarded portion is necessary to assess their potential as EPA and DHA sources, and to ensure their large-scale production (Rustad *et al.*, 2011).

This study elucidates the biometric parameters and FA composition of discarded fish species from pelagic and benthic habitats on the East Mediterranean coast of Spain caught in winter (January) and summer (June) to hypothesize that these species could be potential marine sources of *n*-3 VLCPUFA.

Material and Methods

Fish collection and sampling

Fish were taken from a bottom trawl vessel operating on the East Mediterranean coast of the Iberian Peninsula (N Alboran Sea) (Fig. 1) in winter (January) and summer (June) of 2018. The surface water temperature was 15.3°C and 20.0°C, respectively. The discarded fish species were identified according to the available literature. Samples of each species (8 fish each) were stored separately in polyethylene bags with identifying labels and preserved in ice until they reached the laboratory. Then fish were frozen at -20°C until further processed.

Thirteen discarded species were selected, most from the demersal habitat: blackspot sea bream (*Pagellus bogaraveo*), common pandora (*Pagellus erythrinus*), axillary seabream (*Pagellus acarne*), red porgy (*Pagrus pagrus*), bogue (*Boops boops*), red gurnard (*Chelidonichthys cuculus*), tub gurnard (*Chelidonichthys lucerna*), greater weever (*Trachinus draco*) and bastard grunt (*Pomadasy incisus*). Other species were pelagic (Mediterranean horse mackerel, *Trachurus mediterraneus*; horse mackerel, *Trachurus trachurus*) or benthopelagic (blotched picarel, *Spicara maena* and picarel, *Spicara smaris*). Table 1 lists the analysed species, including their habitats, depth range, feeding type, reproductive characteristics, data of sizes upon first maturity and legal fishing limits and Minimum Conservation Reference Size (MCRS, Annex III to Regulation EC No. 1967/2006).

Biometric parameters

Eight fish of each species were selected to determine the biometric parameters.

Total length (TL, cm) from the tip of the snout to the tip of the longer lobe of the caudal fin was recorded by a measuring board. Total body weight (TBW, g), liver weight (LW, g), digestive weight (DW, g) and fillet weight (FW, g) were recorded using a balance to the



Fig. 1: Map of the Northern Alboran Sea (Geographical Subarea 1 according to General Fisheries Commission for the Mediterranean, GFCM division. Obtained from D-maps.com website: https://d-maps.com/carte.php?num_car=33776&lang=en).

Table 1. List of fish species analysed in this work including habitats and reproduction characteristics.

Scientific name	Habitats	Code	Usual depth range	Types of bottom	feeding	Reproduction	Spawning season	Range of size at first maturity (cm) ¹	Size at first maturity (cm) ²	MCRS (cm) ³
<i>Pagellus bogaraveo</i>	benthopelagic	SBR	150 - 300 m	rocks, sand, mud	Omnivorous, feeding mainly on crustaceans, mollusks, worms and small fish	protandrous hermaphroditism	January to June	31.4	20-30	33
<i>Pagellus erythrinus</i>	benthopelagic	PAC	50 - 100 m	rock, gravel, sand, mud	Small crustaceans, mollusks, small fishes	protogynous hermaphrodite	From April to July	13-16	12.8-25.2	15
<i>Pagellus acarne</i>	benthopelagic	SBA	40 - 100 m	seagrass beds and sand	Small crustaceans, mollusks, small fishes	protandrous hermaphroditism	From April to September	16	18-21.7	17
<i>Pagrus pagrus</i>	demersal or semipelagic	RPG	10 - 80 m	rock, rubble, sand	Crustaceans, fishes, and mollusks	protogynous hermaphroditism	spring	24	24	18
<i>Boops boops</i>	demersal	BOG	0 - 100 m	sand, mud, rocks, seaweeds	Omnivorous (crustaceans) also planktophagous	protogynous hermaphroditism	from January to May	14.3	11.9 - 17	11 ⁴
<i>Trachurus mediterraneus</i>	pelagic	HMM	5 - 250 m		Small crustaceans, shrimps, small fishes	dioic, external fertilization, multiple spawner	from March to April	20	19.1	15
<i>Trachurus trachurus</i>	pelagic	HOM	100 - 200 m		Fish, crustaceans, and cephalopods	dioic, external fertilization, multiple spawner	From May to September	21-30	19- 30	15
<i>Spicara maena</i>	pelagic-neritic	BPI	30 - 130 m	<i>Posidonia</i> beds, sand, muddy	Zooplanktivorous preference to animal food	protogynous hermaphroditism	from March to June	10.3	11.5-13	11,5
<i>Spicara smaris</i>	pelagic-neritic	SPC	15 - 170 m	<i>Posidonia</i> beds, muddy bottoms	Zooplanktivorous preference to plant food	protogynous hermaphroditism	from March to June	9.1	11-13	9 - 11 ⁵
<i>Chelidonichthys cuculus</i>	demersal	GUS	30 - 250 m	sand and gravel, crag, and rocks	Benthic crustaceans, other invertebrates and bottom-dwelling fishes	dioic, external fertilization	from January to April ⁸	26.6 (16.8 ⁷)	25	15 ⁶
<i>Chelidonichthys lucerna</i>	demersal	GUU	20 - 318 m	sand, muddy sand, gravel bottoms	Fish, crustaceans and mollusks	dioic, external fertilization	from May to September ⁹	24.7 (14.4 ⁷)	18-21.6	>18 ⁹

Continued

Table 1 continued

Scientific name	Habitats	Code	Usual depth range	Types of bottom	feeding	Reproduction	Spawning season	Range of size at first maturity (cm) ¹	Size at first maturity (cm) ²	MCRS (cm) ³
<i>Trachinus draco</i>	demersal	WEG	1 - 30 m	sandy, muddy or gravelly bottoms	Small invertebrates and fishes	Oviparous	June and August	12		14
<i>Pomadasys incisus</i>	coastal demersal	BGR	10 - 100 m	hard bottom and sand	Bottom and near-bottom invertebrates	Oviparous	from August to October	?	14.9-16.1	15-33

¹ Data from FishBase² Tsikliras, Stergiou, 2014.³ Minimum Conservation Reference Size (MCRS) (Annex III to Regulation (EC) No 1967/2006)⁴ Real Decreto 560/1995, de 7 de abril, por el que se establece las tallas mínimas de determinadas especies pesqueras.⁵ Ordre del Conseller d'Agricultura i Pesca, de dia 3 de desembre de 1985, per la qual es determina la talla mínima del gerret⁶ Vallisneri *et al.*, 2012⁷ Follesa, Carbonara, 2019.⁸ Ordines *et al.*, 2014.⁹ Özdemir *et al.*, 2019.

nearest ± 0.01 g. The data from the linear and weight determinations were used to calculate the following indices: Condition factor (%) = $100 * TBW/TL^3$; HIS: Hepatosomatic index (%) = $100 * LW/TBW$; DSI: Digestivesomatic index (%) = $100*(DW/TBW)$ and Fillet yield % = $100*(2*FW/TBW)$.

Fatty acid analyses

Simultaneous oil extraction and transesterification were carried out according to a previous work (Guil-Guerrero *et al.*, 2013). Samples of the thawed whole fish ($n=3$) were homogenized using a laboratory blender. Then a representative aliquot (50 mg wet weight) was weighed in a 10-ml test tube. After transesterification, fatty acid methyl esters (FAME) were analyzed by a Focus GC (Thermo Electron, Cambridge, UK), equipped with a flame ionization detector and an Omegawax 250 capillary column (30 m x 0.25 mm ID x 0.25 μ m film thickness) (Supelco, Bellefonte, PA, USA). The oven temperature was 90°C (1 min), 10°C/min to 100°C (3 min), 6°C/min to 260°C (5 min). The injector temperature was set at 250°C with a split ratio of 50:1 and an injection volume of 4 μ L. The detector temperature was set at 260°C and the carrier gas (nitrogen) flow was 1 mL/min. The peak area of the internal standard was used as a reference to quantify the mass of each FA in the resulting chromatograms. Fatty acid profiles were reported as FA percentages of the total FA available in samples. Peaks were identified by their retention times, obtained for known FAME standards (PUFA No. 1, 47033 from Sigma, St. Louis, USA).

Quality index

FA were grouped into families (SFAs, MUFAs, $n-6$ and $n-3$ PUFAs), which were calculated and expressed as percentages and the total amount of each FA group as 100 g of fish tissue.

Both the Atherogenic Index (AI) and Thrombogenic Index (TI) were calculated according to Ulbricht & Southgate (1991), and the PUFA/SFA Index was also obtained.

IA indicates the relation between the main pro-atherogenic FA and the main anti-atherogenic FA.

$$IA = (12:0 + 4*14:0 + 16:0) / [(n-6 + n-3) PUFA + 18:1 + \text{the sum of the other MUFA}],$$

IT shows the tendency of clots forming in blood vessels. This is defined as the relation between the pro-thrombogenic (saturated) and the anti-thrombogenic FA (MUFA, $n-6$ PUFA, $n-3$ PUFA).

$$IT = (14:0+16:0+18:0) / (0.5*18:1 + 0.5*\Sigma MUFAs + 0.5*PUFAs n-6 + 3* PUFAs n-3 + (n-3/n-6)).$$

Statistical Analysis

The results are reported as mean value \pm S.D.

The effect of the categorical variables “fish species” and “sampling time”, and their possible interactions, were determined for each parameter by fitting a general linear statistical model (GLM analysis), which relates

the measured parameters to predictive factors. Data were processed by a one-way ANOVA and least-square means were tested for differences by Fisher's least significant difference (LSD) procedure. When the results were expressed as a percentage (e.g. FAs), data were normalized using the arcsine transformation of their square root prior to the statistical analysis. Unless otherwise specified, a 95% significance level was considered to indicate statistical differences ($P < 0.05$). All the statistical analyses were conducted with specific software (Statgraphics Plus 4.0, Statistical Graphics Corp., Rockville, Maryland, USA).

Results

Biometric Parameters

The results of the biometric parameters determined for the different fish species in several sampling months are shown in Table 2. The smallest size (length and weight) of the fish sampled in January was found for *P. bogaraveo*, *P. erythrinus* and *C. cuculus*, whereas the biggest size went to *B. boops*, *T. draco* and *P. incisus*. In June, low values were still observed for *C. cuculus* and high ones for *B. boops*. *P. bogaraveo* and *P. erythrinus* obtained intermediate values and *P. acarne* was the biggest species. The *P. pagrus* specimens also had high weight values in all the measurements.

The condition factor (CF) relates total length and total weight, and indicates how elongated a fish is. The CF values were above 1.0 in most of the studied species, except *B. boops*, *T. mediterraneus*, *T. trachurus*, *S. smarís* and *T. draco* whose values were below 0.8. The highest CF values in January were for *P. pagrus*, *C. cuculus* and *P. incisus*, while *P. acarne* and *C. cuculus* had the highest CF values in June. The last species also showed the highest HSI in January and June.

Fatty acid content

FA content (g FA/100 g tissue) and proportion (% of total FAs) were included for the data collected in January and June (Table 3 for Sparidae pelagic members, Table 4 for *Trachurus* and *Spicara* spp. and Table 5 for demersal species).

The species with the highest FA contents in January were *P. incisus* (7.3 g FA/100 g fish) and *T. draco* (5.7 g FA/100 g fish), whereas the lowest FA contents were found for *P. bogaraveo* (1.3 g FA/100 g fish) and *P. acarne* (1.9 g FA/100 g fish).

The June sampling gave higher FA contents than in January. *P. acarne* (10.9 g FA/100 g fish) and *B. boops* (3.0 g FA/100 g fish) had the highest and lowest FA content, respectively. In general, this parameter significantly increased in June for all the species. However, this fact was not verified in the species caught only in June, and not in January.

Fatty acid profiles

When considering the FAs grouped according to their degree of unsaturation, saturated FAs (SFAs) were between 30% and 38% of total FAs, MUFAs were approximately between 10% and 33%, and PUFAs went from 17.4 to 46.8% for the *n*-3 series and from 5.2 to 11.3% for the *n*-6 series.

In January, the lowest proportion of SFAs (29.7%) was found in *S. smarís* and the highest (38.0%) in *P. acarne*. MUFAs had low values in *P. erythrinus* and *S. smarís* (over 9.7%), with high values in *S. maena* (33%) and *P. incisus* (40.2%). The *n*-3 PUFA proportion ranged from 17.4% in *P. incisus* to 46.9% in *S. smarís*. *S. smarís*, *P. acarne* and *P. pagrus* had the highest *n*-6 PUFA proportions (10.6, 10.8 and 11.3%, respectively), while the lowest proportions went to *T. mediterraneus* and *T. draco* (over 5%). The *n*-3/*n*-6 ratio ranged from 3.3 in *P. incisus* to 7.4 in *T. mediterraneus*.

Significantly high MUFA, and low SFA and PUFA fractions, were observed in *Sparidae* spp in June compared to January (Table 3). The drop in *n*-3 PUFA was less marked than that of *n*-6 PUFA and, consequently, the *n*-3/*n*-6 ratio was higher in summer in most of the species of this family, except *P. erythrinus*. The other evaluated pelagic or demersal species displayed different trends, but the *n*-3/*n*-6 ratio significantly rose in all cases.

The lowest proportion of SFAs in June was for *C. cuculus* (29.1%), *S. smarís* and *P. erythrinus* (around 29.8%), and the highest (34.0%) for *T. trachurus*. MUFAs had low values in *B. boops* (8.4%) and high values in *P. acarne* (22.4%) and *P. pagrus* (19.2%). The *n*-3 PUFA proportion ranged from 34.9% in *P. acarne* to 50.4% in *B. boops*. The highest *n*-6 PUFA proportion was for *P. erythrinus* and *P. pagrus* (11.6 and 11.3%), while the lowest went to *T. trachurus* (4.5 %). The *n*-3/*n*-6 ratio ranged from 3.6 in *P. erythrinus* to 9.6 in *T. trachurus*.

Quality parameters

The lowest *n*-3/*n*-6 ratios in January were for *P. incisus* (3.3), *P. pagrus* (3.4), *S. maena* (3.7) and *P. acarne* (3.9), whereas the highest values were found in *T. draco* (6.9) and *T. mediterraneus* (7.4). In June, the highest values went to *B. boops* (8.5) and *T. trachurus* (9.6), and *P. erythrinus* had the lowest value (4.4).

The AI values in January ranged from 0.33 (*S. smarís*) to 0.56 (*T. mediterraneus* and *S. maena*), and values increased in June for *P. Bogaraveo*, *T. trachurus* and *S. smarís*. TI ranged from 0.19 (*S. smarís*) to 0.42 (*P. incisus*) in the January sampling, and values lowered for several species in June (*B. boops*, *S. maena* and *C. cuculus*).

The highest PUFA/SFA ratio in the January sampling was observed in *S. smarís* (1.59), followed by *T. trachurus* (1.35), *P. bogaraveo* (1.30) and *P. erythrinus* (1.31), while the lowest value was for *S. maena* (0.80). The ratio significantly lowered in all the species sampled in June.

The FA groups (g/100 g of tissue) are depicted in Figure 2. In the January sampling, *P. incisus* had the highest

Table 2. Biometric parameters of different species in the January and June sampling (mean \pm SD; n=8).

	<i>Pagellus bogaraveo</i> Blackspot sea bream	<i>Pagellus erythrinus</i> Common pandora	<i>Pagellus acarne</i> Axillary seabream	<i>Pagrus pagrus</i> Red porgy	<i>Boops boops</i> Bogue	<i>Trachurus mediterraneus</i> horse mackerel	<i>Trachurus trachurus</i> Horse mackerel	<i>Spicara maena</i> Blotched picarel	<i>Spicara smaris</i> Picarel	<i>Chelidonichthys cuculus</i> Red gumard	<i>Chelidonichthys lucerna</i> Tub gumard	<i>Trachinus draco</i> Greater weever	<i>Pomadourus incisus</i> Bastard grunt
Total length [#] (cm)													
JAN	11.8 ^{ab} \pm 0.4	10.6 ^a \pm 1.4	14.8 ^{bc} \pm 2.0	13.3 ^b \pm 1.2	20.8 ^f \pm 0.6	11.8 ^{de} \pm 0.4	10.6 ^a \pm 0.7	14.8 ^{bc} \pm 1.7	13.3 ^b \pm 0.4	11.4 ^{ab} \pm 1.0	17.5 ^d \pm 0.5	22.5 [±] 1.5	20.5 ^c \pm 0.7
JUN	15.9 ^{bc*} \pm 0.2	16.4 ^{bc*} \pm 0.8	16.7 ^{bc} \pm 1.2	14.5 ^b \pm 1.0	21.2 ^d \pm 1.5	-	11.6 ^a \pm 0.9	16.5 ^{bc} \pm 0.8	18.0 ^{**} \pm 1.4	11.4 ^a \pm 1.1	-	-	-
Total weight (g)													
JAN	17.19 ^{ab} \pm 0.4	15.0 ^a \pm 5.4	27.1 ^d \pm 2.7	22.6 ^e \pm 1.3	74.4 ^h \pm 3.5	17.2 ^{ab} \pm 3.8	15.0 ^a \pm 3.8	37.1 ^f \pm 2.7	33.8 ^e \pm 2.5	17.7 ^{ab} \pm 5.3	45.5 ^g \pm 0.7	74.5 ^h \pm 37.5	121.5 ⁱ \pm 7.8
JUN	49.3 ^{cd*} \pm 4.6	53.9 ^{g*} \pm 3.9	62.0 ^{g*} \pm 3.5	54.7 ^g \pm 2.5	83.3 ^f \pm 1.5	-	13.1 ^{ab*} \pm 3.2	44.4 ^{g*} \pm 4.9	49.6 ^{cd*} \pm 3.0	18.4 ^b \pm 1.5	-	-	-
Condition factor ¹													
JAN	1.0 ^{bc} \pm 0.2	1.2 ^{de} \pm 0.1	0.9 ^b \pm 0.1	1.3 ^{ef} \pm 0.2	0.8 ^{ab} \pm 0.1	1.0 ^{bc} \pm 0.1	1.2 ^a \pm 0.1	1.1 ^{cd} \pm 0.1	1.4 ^f \pm 0.0	1.2 ^{de} \pm 0.1	0.9 ^b \pm 0.0	0.6 ^a \pm 0.0	1.4 ^f \pm 0.1
JUN	1.2 ^{bc} \pm 0.2	1.2 ^{bc} \pm 0.3	1.4 ^{g*} \pm 0.2	1.3 ^{cd} \pm 0.0	0.9 ^a \pm 0.3	-	1.0 ^{ab*} \pm 0.2	1.0 ^{ab} \pm 0.2	0.8 ^{ab*} \pm 0.1	1.2 ^{bc} \pm 0.1	-	-	-
HSI ²													
JAN	0.7 ^{ab} \pm 0.3	0.8 ^{abc} \pm 0.2	1.1 ^{abc} \pm 0.2	1.2 ^c \pm 0.1	0.7 ^{ab} \pm 0.2	0.7 ^{ab} \pm 0.4	0.8 ^{abc} \pm 0.1	0.8 ^{abc} \pm 0.2	1.1 ^c \pm 0.1	2.5 ^c \pm 1.1	0.9 ^{bc} \pm 0.0	1.2 ^c \pm 0.0	1.9 ^d \pm 0.1
JUN	0.8 ^a \pm 0.1	1.0 ^b \pm 0.3	1.0 ^b \pm 0.6	1.5 ^c \pm 0.6	0.8 ^a \pm 0.6	-	0.9 ^{ab} \pm 0.1	1.0 ^b \pm 0.6	1.0 ^b \pm 0.6	2.7 ^d \pm 1.0	-	-	-
DSI ³													
JAN	3.9 ^e \pm 0.3	3.2 ^{cd} \pm 0.1	3.0 ^{cd} \pm 0.3	4.5 ^f \pm 1.2	4.6 ^f \pm 1.2	3.9 ^{bcd} \pm 0.3	3.2 ^{cd} \pm 0.1	2.8 ^{bc} \pm 0.3	3.5 ^{de} \pm 0.6	3.5 ^{de} \pm 1.5	2.3 ^{ab} \pm 0.0	1.5 ^a \pm 1.3	3.3 ^{cd} \pm 0.4
JUN	4.8 ^{cd*} \pm 2.4	3.6 ^{ab*} \pm 1.5	4.5 ^{de} \pm 1.4	5.7 ^{e*} \pm 1.8	5.5 ^{e*} \pm 0.6	-	4.5 ^{e*} \pm 0.5	3.4 ^{ab*} \pm 1.2	3.1 ^{ab*} \pm 0.2	2.0 ^{a*} \pm 0.4	-	-	-
Fillet yield ⁴													
JAN	43.4 ^b \pm 0.6	49.1 ^d \pm 1.4	46.7 ^{cd} \pm 3.3	58.4 ^e \pm 2.9	58.0 ^e \pm 2.4	43.4 ^b \pm 1.6	49.1 ^d \pm 1.4	48.7 ^{cd} \pm 2.3	48.4 ^{cd} \pm 2.9	44.7 ^b \pm 1.6	67.8 ^e \pm 0.1	61.6 ^d \pm 4.6	20.1 ^a \pm 1.6
JUN	61.7 ^{cd*} \pm 5.9	57.4 ^{bc*} \pm 3.9	56.5 ^{bc*} \pm 2.4	56.4 ^{bc*} \pm 0.4	63.6 ^{g*} \pm 1.9	-	52.9 ^{ab*} \pm 3.2	65.9 ^{gh*} \pm 1.9	64.8 ^{gh*} \pm 1.87	49.9 ^{ab*} \pm 2.8	-	-	-

Values with different superscripts indicate significant differences (p<0.05) attributable to different species in a sampling time * indicate significant differences attributable to sampling time for each specie. (p<0.05).

¹Condition factor % = 100 * (Total weight (g)/(Total length (cm))) where weight in g and length in cm, ²Hepatosomatic index % = 100*(Liver weight (g)/Total weight (g)),

³Digestive-somatic index % = 100*(Digestive weight (g)/Total weight (g)), ⁴Fillet yield % = 100*(2* Lateral fillet weight (g)/Total weight (g)).

Table 3. Muscle fatty acid content (g FA/100 g wet weight) and profiles of main FA (FA% of total FAs) of Sparidae species in January and June sampling (Values are reported as mean ± SD, n=3)

	Pagellus bogaraveo		Pagellus erythrinus		Pagellus acarne		Pagrus pagrus		Boops boops	
	January	June	January	June	January	June	January	June	January	June
FA content (g FA/100 g fish)	1.8 ^a ± 0.2	5.4 ^{c*} ± 0.4	2.4 ^b ± 0.1	3.5 ^B ± 0.4	1.9 ^a ± 0.3	10.9 ^{D*} ± 0.4	2.0 ^{ab} ± 0.3	5.5 ^{AB**} ± 0.2	2.2 ^b ± 0.1	3.0 ^{A*} ± 0.1
FA profile (% of total FA)										
14:0 (MA)	2.3 ^{bc} ± 0.2	4.7 ^{D*} ± 0.4	1.4 ^a ± 0.1	1.5 ^A ± 0.3	1.8 ^{ab} ± 0.1	3.9 ^{C*} ± 0.1	2.2 ^B ± 0.1	3.7 ^C ± 0.2	2.8 ^c ± 0.1	2.2 ^B ± 0.2
15:0 (PDA)	1.6 ^b ± 0.2	0.9 ^{AB*} ± 0.4	0.8 ^a ± 0.0	1.3 ^{B*} ± 0.4	2.4 ^c ± 0.2	0.9 ^{AB*} ± 0.4	0.8 ^b ± 0.0	0.7 ^A ± 0.2	2.5 ^c ± 0.1	0.7 ^{A*} ± 0.1
16:0 (PA)	20.6 ^{bc} ± 0.7	17.1 ^{A*} ± 0.3	21.6 ^{bc} ± 0.3	19.1 ^{B*} ± 0.3	25.8 ^d ± 0.1	18.2 ^{AB*} ± 0.2	19.9 ^a ± 0.1	18.1 ^{AB*} ± 0.8	21.9 ^{bc} ± 0.3	21.3 ^c ± 0.8
16:1n-7(POA)	1.7 ^a ± 0.1	5.6 ^{B*} ± 0.4	3.0 ^{ab} ± 0.1	5.4 ^{B*} ± 0.1	2.7 ^{ab} ± 0.3	6.5 ^{C*} ± 0.1	5.4 ^c ± 0.1	7.0 ^{CD*} ± 0.1	3.8 ^{bc} ± 0.0	2.7 ^{A*} ± 0.1
18:0 (SA)	9.2 ^b ± 0.1	8.4 ^{B*} ± 0.2	9.3 ^c ± 0.6	7.7 ^{A*} ± 0.2	8.0 ^b ± 0.8	7.3 ^A ± 0.1	8.5 ^b ± 0.6	7.6 ^A ± 0.1	6.7 ^a ± 0.1	7.5 ^{A*} ± 0.1
18:1n-9 (OA)	7.4 ^b ± 0.1	8.3 ^C ± 0.1	5.6 ^a ± 0.2	6.8 ^{B*} ± 0.1	5.4 ^a ± 0.3	11.9 ^{E*} ± 0.1	7.6 ^b ± 0.2	8.9 ^{CD*} ± 0.2	7.7 ^b ± 0.1	5.3 ^{A*} ± 0.2
18:1n-7 (VA)	2.2 ^a ± 0.2	5.7 ^D ± 0.3	3.9 ^b ± 0.1	3.0 ^{A*} ± 0.1	-	4.8 ^C ± 0.1	3.8 ^b ± 0.1	4.3 ^B ± 0.1	3.6 ^b ± 0.1	2.4 ^A ± 0.1
18:2n-6 (LA)	1.6 ^{ab} ± 0.1	1.4 ^B ± 0.2	1.4 ^{ab} ± 0.1	1.5 ^B ± 0.6	2.9 ^c ± 0.1	1.4 ^B ± 0.1	2.4 ^{bc} ± 0.1	1.1 ^{A*} ± 0.1	2.9 ^{cd} ± 0.2	1.4 ^B ± 0.1
18:3n-3 (ALA)	1.0 ^a ± 0.1	0.8 ^A ± 0.1	0.8 ^b ± 0.0	0.9 ^{AB} ± 0.1	0.7 ^{ab} ± 0.2	1.1 ^B ± 0.0	0.5 ^a ± 0.1	1.5 ^{C*} ± 0.1	0.6 ^a ± 0.1	0.9 ^{AB} ± 0.1
18:4n-3 (SDA)	0.2 ^{ab} ± 0.1	-	0.1 ^a ± 0.3	-	-	0.9 ^C ± 0.1	-	0.4 ^A ± 0.2	0.3 ^b ± 0.1	0.7 ^B ± 0.2
20:1n-9 (EEA)	0.2 ^a ± 0.1	3.5 ^{B*} ± 0.1	1.1 ^b ± 0.3	-	0.2 ^a ± 0.1	3.4 ^{B*} ± 0.1	1.3 ^b ± 0.1	3.3 ^{B*} ± 0.1	-	0.4 ^a ± 0.1
20:4n-6 (AA)	5.0 ^{bc} ± 0.3	2.6 ^{B*} ± 0.1	6.3 ^c ± 0.1	4.4 ^{C*} ± 0.2	4.8 ^b ± 0.5	2.5 ^{AB} ± 0.1	4.9 ^b ± 0.2	2.7 ^{B*} ± 0.1	2.9 ^a ± 0.1	2.3 ^{A*} ± 0.1
20:4n-3 (DGLA)	0.4 ^a ± 0.1	0.7 ^{A*} ± 0.0	-	-	0.2 ^a ± 0.1	0.7 ^A ± 0.1	1.6 ^b ± 0.1	0.8 ^{B*} ± 0.1	-	0.6 ^A ± 0.1
20:5n-3 (EPA)	7.6 ^b ± 0.1	12.7 ^{C*} ± 0.4	11.3 ^d ± 0.2	8.1 ^{B*} ± 0.1	4.6 ^a ± 0.3	11.4 ^{C*} ± 0.2	7.7 ^b ± 0.1	13.2 ^{D*} ± 0.1	9.5 ^c ± 0.5	7.4 ^{A*} ± 0.1
22:5n-3 (DPA)	2.3 ^{bc} ± 0.1	4.9 ^B ± 0.2	3.5 ^c ± 0.1	4.8 ^B ± 0.1	3.5 ^c ± 0.2	4.1 ^B ± 0.1	3.2 ^c ± 0.1	4.1 ^{B±} ± 0.1	1.8 ^a ± 0.2	2.6 ^A ± 0.2
22:6n-3 (DHA)	32.7 ^c ± 0.1	18.5 ^{B*} ± 0.2	27.6 ^b ± 0.5	28.2 ^D ± 0.3	31.7 ^c ± 0.3	17.7 ^{A*} ± 0.2	25.7 ^a ± 0.2	19.8 ^{C*} ± 0.2	27.6 ^b ± 0.8	39.1 ^{E*} ± 0.8
Unidentified	0.3 ± 0.3	0.6 ± 0.2	0.5 ± 0.1	0.9 ± 0.1	0.4 ± 0.2	0.6 ± 0.1	0.7 ± 0.1	0.6 ± 0.3	0.5 ± 0.6	0.7 ± 0.3
SFA ¹	33.7 ^b ± 0.6	31.0 ^{B*} ± 0.0	33.1 ^b ± 0.1	29.6 ^{A*} ± 0.6	38.0 ^c ± 0.4	30.3 ^{AB*} ± 0.5	31.4 ^a ± 0.9	30.1 ^{AB} ± 0.8	33.9 ^b ± 0.5	31.6 ^{B*} ± 0.3
MUFA ²	10.4 ^a ± 0.3	18.1 ^{C*} ± 0.1	9.7 ^a ± 0.1	13.3 ^{B*} ± 0.5	10.4 ^a ± 0.0	22.4 ^{D*} ± 0.8	14.4 ^b ± 0.2	19.2 ^{CD*} ± 0.6	13.0 ^b ± 0.4	8.4 ^{A*} ± 0.2
n-3 PUFA ³	43.9 ^c ± 0.9	37.5 ^{B*} ± 0.3	43.1 ^d ± 0.6	41.9 ^{de} ± 0.5	40.5 ^c ± 0.5	34.9 ^a ± 1.1	38.5 ^a ± 0.2	39.3 ^{BC} ± 0.1	39.3 ^a ± 0.7	50.4 ^{E*} ± 0.4
n-6 PUFA ⁴	9.5 ^b ± 0.3	7.1 ^{B*} ± 0.5	9.8 ^{ab} ± 0.6	11.6 ^C ± 0.6	10.8 ^{bc} ± 0.7	6.4 ^{A*} ± 0.2	11.3 ^c ± 0.4	6.3 ^{A*} ± 0.1	9.6 ^a ± 0.1	6.0 ^{A*} ± 0.1
n-3/n-6	4.6 ^c ± 0.2	5.3 ^{B*} ± 0.4	4.4 ^c ± 0.2	3.6 ^{A*} ± 0.2	3.9 ^{ab} ± 0.2	5.5 ^{B*} ± 0.08	3.4 ^a ± 0.3	6.2 ^{C*} ± 0.0	4.1 ^{bc} ± 0.3	8.5 ^{D*} ± 0.1
AI ⁵	0.46 ^b ± 0.04	0.57 ^{B*} ± 0.02	0.43 ^a ± 0.02	0.38 ^{A*} ± 0.02	0.52 ^b ± 0.01	0.53 ^B ± 0.02	0.45 ^a ± 0.01	0.50 ^B ± 0.04	0.53 ^b ± 0.01	0.46 ^{B*} ± 0.03
TI ⁶	0.22 ^a ± 0.01	0.23 ^C ± 0.00	0.22 ^a ± 0.01	0.20 ^{AB} ± 0.00	0.26 ^c ± 0.01	0.24 ^D ± 0.01	0.23 ^b ± 0.01	0.21 ^{B*} ± 0.01	0.23 ^b ± 0.01	0.19 ^{A*} ± 0.01
PUFA/SFA	1.30 ^c ± 0.02	0.57 ^C ± 0.01	1.31 ^c ± 0.01	0.58 ^C ± 0.01	1.07 ^a ± 0.03	0.33 ^A ± 0.00	1.23 ^b ± 0.04	0.50 ^B ± 0.01	1.06 ^a ± 0.03	0.46 ^B ± 0.042

Values with different superscripts indicate significant differences attributable to different species; lower case in January sampling and capital letter in June sampling; * indicate significant differences attributable to sampling time for each specie. (p<0.05).
¹Saturated fatty acids, ²monounsaturated fatty acids, ³polyunsaturated fatty acids n-3, ⁴polyunsaturated fatty acids n-6, ⁵AI: Atherogenic Index=(12:0 + 4*14:0 + 16:0) / [(n-6 + n-3) PUFA + 18:1 + the sum of other MUFA], ⁶TI: Thrombogenic Index=(14:0+16:0+18:0) / (0.5*18:1 + 0.5*ΣMUFA + 0.5*PUFA + 3*PUFA n-3 + (n-3/n-6)).

Table 4. Muscle fatty acid content (g FA/100 g wet weight) and profiles of main FA (FA% of total FAs) of *Trachurus* and *Spicara* spp in January and June sampling (Values are reported as mean \pm SD, n=3)

	<i>Trachurus mediterraneus</i>		<i>Trachurus trachurus</i>		<i>Spicara maena</i>		<i>Spicara smaris</i>	
	January	June	January	June	January	June	January	June
FA content (g FA/100 g fish)	3.3 ^b \pm 0.4	-	2.4 ^a \pm 0.1	6.0 ^{c*} \pm 0.7	3.4 ^b \pm 0.1	4.5 ^B \pm 0.2	2.2 ^a \pm 0.2	4.1 ^{A*} \pm 0.2
FA profile (% of total FA)								
14:0 (MA)	2.5 ^a \pm 0.2	-	2.7 ^{bc} \pm 0.1	3.3 ^B \pm 0.4	3.3 ^c \pm 0.1	3.1 ^B \pm 0.1	-	2.1 ^A \pm 0.2
15:0 (PDA)	2.2 ^c \pm 0.2	-	0.7 ^b \pm 0.1	0.6 \pm 0.4	0.4 ^{ab} \pm 0.1	0.7 \pm 0.1	0.4 ^a \pm 0.1	0.8 \pm 0.2
16:0 (PA)	23.4 ^b \pm 0.1	-	22.7 ^{ab} \pm 0.5	21.7 ^B \pm 0.3	24.4 ^c \pm 0.4	23.0 ^C \pm 0.2	22.1 ^a \pm 0.8	20.6 ^A \pm 0.1
16:1n-7(POA)	3.8 ^b \pm 0.1	-	2.3 ^a \pm 0.1	3.1 ^{A*} \pm 0.1	7.8 ^c \pm 0.2	4.8 ^B \pm 0.4	1.5 ^b \pm 0.0	3.1 ^{A*} \pm 0.5
18:0 (SA)	8.4 ^b \pm 0.4	-	8.0 ^b \pm 0.1	8.5 ^B \pm 0.4	4.7 ^a \pm 0.4	6.2 ^{A*} \pm 0.1	7.2 ^b \pm 0.1	6.4 ^{A*} \pm 0.1
18:1n-9 (OA)	6.8 ^b \pm 0.3	-	5.8 ^a \pm 0.1	10.7 ^B \pm 0.3	22.9 ^c \pm 0.3	10.8 ^{B*} \pm 0.1	5.7 ^a \pm 0.1	8.5 ^{A*} \pm 0.6
18:1n-7 (VA)	2.8 ^b \pm 0.2	-	2.4 ^{ab} \pm 0.1	3.0 ^{B*} \pm 0.1	-	3.1 ^B \pm 0.1	1.9 ^a \pm 0.1	2.6 ^A \pm 0.1
18:2n-6 (LA)	1.5 ^a \pm 0.3	-	2.0 ^b \pm 0.1	1.6 ^A \pm 0.2	1.7 ^{ab} \pm 0.3	2.1 ^B \pm 0.1	5.3 ^c \pm 0.2	2.2 ^{B*} \pm 0.1
18:3n-3 (ALA)	0.8 ^b \pm 0.1	-	0.4 ^a \pm 0.1	1.2 ^{B*} \pm 0.2	0.8 ^b \pm 0.1	1.2 ^B \pm 0.2	0.8 ^a \pm 0.1	0.9 ^A \pm 0.1
18:4n-3 (SDA)	0.5 ^{ab} \pm 0.2	-	0.1 ^a \pm 0.0	0.8 ^B \pm 0.1	0.6 ^b \pm 0.0	0.5 ^A \pm 0.1	0.4 ^{ab} \pm 0.2	0.5 ^A \pm 0.1
20:1n-9 (EEA)	2.0 ^b \pm 0.1	-	1.1 ^a \pm 0.1	0.4 ^{a*} \pm 0.1	1.8 ^b \pm 0.3	0.9 ^B \pm 0.1	1.2 ^a \pm 0.1	1.8 ^{c*} \pm 0.1
20:4n-6 (AA)	1.6 ^a \pm 0.2	-	2.0 ^{ab} \pm 0.2	1.2 ^A \pm 0.1	2.9 ^b \pm 0.1	1.9 ^{B*} \pm 0.0	2.0 ^{ab} \pm 0.1	2.0 ^B \pm 0.1
20:4n-3 (DGLA)	0.6 ^a \pm 0.1	-	3.4 ^c \pm 0.1	0.7 ^{a*} \pm 0.0	1.9 ^b \pm 0.1	0.7 ^A \pm 0.1	0.3 ^a \pm 0.0	1.2 ^B \pm 0.2
20:5n-3 (EPA)	6.7 ^{ab} \pm 0.2	-	7.1 ^b \pm 0.1	9.0 ^{b*} \pm 0.3	7.1 ^b \pm 0.1	8.9 ^{AB*} \pm 0.1	6.0 ^b \pm 0.2	8.7 ^{A*} \pm 0.1
22:5n-3 (DPA)	2.8 ^c \pm 0.1	-	3.3 ^d \pm 0.2	2.3 ^{B*} \pm 0.0	1.8 ^{ab} \pm 0.1	1.4 ^A \pm 0.1	1.2 ^b \pm 0.0	1.8 ^{AB*} \pm 0.1
22:6n-3 (DHA)	30.1 ^b \pm 0.5	-	31.6 ^c \pm 0.2	30.1 ^B \pm 0.4	14.5 ^c \pm 0.5	28.2 ^{A*} \pm 0.1	38.9 ^d \pm 0.3	32.9 ^C \pm 0.5
Unidentified	0.3 ^a \pm 0.1	-	0.6 ^b \pm 0.4	0.4 ^A \pm 0.3	0.2 ^a \pm 0.1	0.8 ^B \pm 0.0	0.7 ^b \pm 0.1	0.6 ^{AB} \pm 0.3
SFA ¹	36.5 ^c \pm 0.9	-	32.4 ^b \pm 2.0	34.0 ^{C*} \pm 0.4	32.7 ^b \pm 0.8	32.9 ^B \pm 0.3	29.7 ^a \pm 1.0	29.8 ^A \pm 0.0
MUFA ²	13.6 ^b \pm 0.3	-	10.5 ^{ab} \pm 0.2	14.1 ^A \pm 0.3	33.6 ^c \pm 0.7	16.4 ^{B*} \pm 0.6	9.8 ^a \pm 0.4	14.9 ^{AB*} \pm 0.5
n-3 PUFA ³	40.9 ^{bf} \pm 0.1	-	45.9 ^c \pm 0.1	43.3 ^B \pm 0.5	26.0 ^b \pm 0.6	40.3 ^{A*} \pm 0.1	46.9 ^c \pm 0.3	45.3 ^c \pm 0.8
n-6 PUFA ⁴	5.6 ^a \pm 0.1	-	7.7 ^c \pm 0.2	4.5 ^{a*} \pm 0.3	6.9 ^b \pm 0.2	6.1 ^B \pm 0.4	10.6 ^d \pm 0.5	6.5 ^{b*} \pm 0.4
n-3/n-6	7.4 ^d \pm 0.5	-	5.9 ^c \pm 0.4	9.6 ^{b*} \pm 0.3	3.7 ^a \pm 0.2	6.7 ^{A*} \pm 0.4	4.5 ^b \pm 0.6	7.0 ^{a*} \pm 0.5
AI ⁵	0.55 ^b \pm 0.01	-	0.53 ^b \pm 0.01	0.66 ^{B*} \pm 0.05	0.56 ^c \pm 0.06	0.56 ^B \pm 0.01	0.33 ^a \pm 0.02	0.43 ^{a*} \pm 0.01
TI ⁶	0.25 ^c \pm 0.01	-	0.22 ^b \pm 0.01	0.22 ^B \pm 0.01	0.32 ^d \pm 0.06	0.23 ^{B*} \pm 0.05	0.19 ^a \pm 0.01	0.19 ^A \pm 0.02
PUFA/SFA	1.12 ^b \pm 0.02	-	1.35 ^c \pm 0.06	0.56 ^B \pm 0.01	0.80 ^a \pm 0.03	0.56 ^B \pm 0.02	1.59 ^d \pm 0.05	0.43 ^A \pm 0.03

Values with different superscripts indicate significant differences attributable to different species; lower case in January sampling and capital letter in June sampling; * indicate significant differences attributable to sampling time for each specie. (p<0.05).

¹Saturated fatty acids, ²monounsaturated fatty acids, ³polyunsaturated fatty acids n-3, ⁴polyunsaturated fatty acids n-6, ⁵AI: Atherogenic Index = (12:0 + 4*14:0 + 16:0) / [(n-6 + n-3) PUFA + 18:1 + the sum of other MUFA], ⁶ TI: Thrombogenic Index = (14:0+16:0+18:0) / (0.5*18:1 + 0.5*ΣMUFA + 0.5*PUFA n-6 + 3*PUFA n-3 + (n-3/n-6)).

Table 5. Muscle fatty acid content (g FA/100 g wet weight) and FA profiles (FA% of total FAs) of demersal species in January and June sampling (Values are reported as mean \pm SD, n=3)

	<i>Chelidonichthys cuculus</i>		<i>Chelidonichthys lucerna</i>		<i>Trachinus draco</i>		<i>Pomadasys incisus</i>	
	January	June	January	June	January	June	January	June
FA content (g FA/100 g fish)	2.9 ^a \pm 0.2	4.8 ^{cd*} \pm 0.1	4.7 ^b \pm 0.2	-	5.8 ^c \pm 0.1	-	7.3 ^{ab} \pm 0.2	-
FA profiles (FA% of total FA)								
14:0 (MA)	2.7 ^b \pm 0.2	2.2 \pm 0.2	1.8 ^a \pm 0.1	-	3.1 ^c \pm 0.3	-	4.1 ^d \pm 0.2	-
15:0 (PDA)	0.9 \pm 0.2	0.7 \pm 0.2	0.6 \pm 0.1	-	0.4 \pm 0.3	-	0.6 \pm 0.2	-
16:0 (PA)	22.3 ^b \pm 0.1	19.8* \pm 0.3	21.2 ^a \pm 0.6	-	21.4 ^a \pm 0.4	-	22.3 ^b \pm 0.8	-
16:1n-7(POA)	2.9 ^a \pm 0.2	4.6 \pm 0.1	8.9 ^a \pm 0.2	-	6.0 ^b \pm 0.1	-	12.0 ^a \pm 0.3	-
18:0 (SA)	7.1 ^c \pm 0.1	6.5* \pm 0.1	7.3 ^c \pm 0.6	-	5.8 ^a \pm 0.8	-	6.9 ^b \pm 0.8	-
18:1n-9 (OA)	10.9 ^a \pm 0.1	11.9* \pm 0.3	11.7 ^b \pm 0.2	-	16.9 ^c \pm 0.3	-	28.2 ^d \pm 0.2	-
18:1n-7 (VA)	3.0 ^b \pm 0.1	3.5 \pm 0.1	4.0 ^a \pm 0.1	-	2.4 ^a \pm 0.1	-	-	-
18:2n-6 (LA)	3.2 ^b \pm 0.1	1.2* \pm 0.0	3.4 ^b \pm 0.1	-	2.3 ^{ab} \pm 0.1	-	1.9 ^a \pm 0.1	-
18:3n-3 (ALA)	0.8 ^b \pm 0.1	0.4 \pm 0.0	0.5 ^a \pm 0.1	-	0.9 ^b \pm 0.3	-	0.6 ^a \pm 0.2	-
18:4n-3 (SDA)	0.6 ^a \pm 0.2	0.5 \pm 0.1	0.3 ^a \pm 0.3	-	1.1 ^b \pm 0.4	-	1.7 ^c \pm 0.2	-
20:1n-9 (EEA)	1.1 ^c \pm 0.0	0.5 \pm 0.0	1.0 ^a \pm 0.2	-	0.7 ^{ab} \pm 0.1	-	0.9 ^b \pm 0.2	-
20:4n-6 (AA)	2.0 ^a \pm 0.1	2.8* \pm 0.0	2.8 ^b \pm 0.4	-	2.0 ^a \pm 0.1	-	2.2 ^a \pm 0.2	-
20:4n-3 (DGLA)	1.2 ^b \pm 0.3	0.8 \pm 0.1	1.2 ^b \pm 0.1	-	1.0 ^b \pm 0.2	-	0.4 ^a \pm 0.1	-
20:5n-3 (EPA)	8.3 ^c \pm 0.2	9.6* \pm 0.3	10.2 ^d \pm 0.4	-	5.9 ^a \pm 0.4	-	7.0 ^b \pm 0.3	-
22:5n-3 (DPA)	1.9 \pm 0.1	3.7* \pm 0.2	2.2 \pm 0.2	-	2.1 \pm 0.1	-	1.9 \pm 0.1	-
22:6n-3 (DHA)	27.7 ^d \pm 0.6	27.9 \pm 0.4	21.2 ^b \pm 0.0	-	26.1 ^c \pm 0.8	-	7.6 ^a \pm 0.3	-
Unidentified	0.5 \pm 0.4	0.7 \pm 0.1	0.2 \pm 0.1	-	0.5 \pm 0.2	-	0.5 \pm 0.2	-
SFA ¹	30.1 ^a \pm 0.4	29.1* \pm 0.4	30.3 ^a \pm 0.6	-	30.4 ^a \pm 0.1	-	33.3 ^b \pm 0.6	-
MUFA ²	16.8 ^a \pm 0.3	17.6* \pm 0.4	24.6 ^b \pm 0.1	-	24.3 ^b \pm 0.2	-	40.2 ^c \pm 0.8	-
n-3 PUFA ³	40.5 ^c \pm 0.4	42.4 \pm 1.1	35.1 ^b \pm 0.2	-	35.8 ^b \pm 0.5	-	17.4 ^a \pm 0.5	-
n-6 PUFA ⁴	7.6 ^b \pm 0.9	6.5 \pm 0.2	7.4 ^b \pm 0.4	-	5.2 ^a \pm 0.6	-	5.3 ^a \pm 0.6	-
n-3/n-6	5.2 ^b \pm 0.3	6.6* \pm 0.4	4.7 ^b \pm 0.4	-	6.9 ^c \pm 0.2	-	3.3 ^a \pm 0.4	-
AI ⁵	0.52 ^c \pm 0.01	0.40* \pm 0.02	0.44 ^a \pm 0.01	-	0.51 ^b \pm 0.04	-	0.60 ^b \pm 0.03	-
TI ⁶	0.23 ^a \pm 0.00	0.20* \pm 0.01	0.24 ^b \pm 0.01	-	0.23 ^a \pm 0.00	-	0.42 ^c \pm 0.0	-
PUFA/SFA	1.21 ^c \pm 0.05	0.43* \pm 0.03	1.14 ^b \pm 0.06	-	1.17 ^b \pm 0.05	-	0.52 ^a \pm 0.01	-

Values with different superscripts indicate significant differences attributable to different species; lower case in January sampling and capital letter in June sampling; * indicate significant differences attributable to sampling time for each specie (p<0.05).
¹Saturated fatty acids, ²monounsaturated fatty acids, ³polyunsaturated fatty acids n-3, ⁴polyunsaturated fatty acids n-6, ⁵AI: Atherogenic Index = (12:0 + 4*14:0 + 16:0) / [(n-6 + n-3) PUFA + 18:1 + the sum of other MUFA], ⁶TI: Thrombogenic Index = (14:0+16:0+18:0) / (0.5*18:1 + 0.5*ΣMUFA + 0.5*PUFA n-6 + 3*PUFA n-3 + (n-3/n-6)).

values for SFA (1.20 g/100g), MUFA (2.87 g/100g) and *n*-6 PUFA (0.39 g/100 g) contents. The *n*-3 PUFA values were also high for this species (1.36 g/100 g), but *C. lucerna* and *T. draco* had larger amounts (1.67 g/100 g and 2.04 g/100 g, respectively). In June, SFA and MUFA displayed significant higher values. It is necessary to emphasize the marked increase observed in *P. acarne* for all the FA groups, and *n*-3 PUFAs are highlighted.

Discussion

Discarded species

Five of the species included in the present study (*P. bogaraveo*, *P. erythrinus*, *P. acarne*, *P. pagrus*, *B. boops*) belong to the family Sparidae. They are demersal or semipelagic, and live above the continental shelf, usually down to a depth of about 100 m, although young individuals can be found nearer the shore. They feed on crustaceans, fish and mollusks (See Table 1). The first four species are among the main target species in the Mediterranean for their high commercial value (García *et al.*, 2012). The commercial importance of bogue (*B. boops*) varies vastly depending on the area. In Spain, its commercial value is limited and it is, therefore, frequently discarded (Carbonell *et al.*, 2018).

Trachurus spp. are pelagic fish of less commercial interest. They sometimes represent a very high percentage of the total capture for purse seine fishing, but are absent in trawl fishing in the Alboran Sea (García *et al.*, 2012). *T. mediterraneus* is frequently located in shallow areas, while *T. trachurus* frequent bigger habitats. Species' food habits are influenced by life cycle. Juveniles feed principally on plankton, while larger individuals prefer fish as prey.

Blotched picarel (*S. maena*) and picarel (*S. smaris*) belong to the family Centranchidae and Greater weever (*T. draco*) is one of four species of the family *Trachinidae*. Although this species is of very little commercial interest, the scarcity of fishing resources, the high demand for fish products and new manufacturing techniques have led to a growing interest being shown in this species, and it is landed in practically all the ports along the Alboran Sea (Portillo Stempel *et al.*, 2008). The commercial value of *C. cuculus* and *T. draco* is low on the Spanish coast, and they are sold together in a mixed fish category known as "morralla" (Ordines *et al.*, 2014).

Bastard grunt (*P. incisus*) is one of the five species of the family *Haemulidae*, a species that is frequently discarded for its low commercial value (Pajuelo *et al.*, 2003). Its presence in the Mediterranean Sea is relatively recent owing to the progressive warming of Mediterranean waters, which is why it has been considered an indicator of changing marine conditions toward "tropicalisation".

Biometric parameters

Most sampled species did not exceed the Minimum Conservation Reference Size (MCRS, Annex III to Regulation EC No. 1967/2006). In this study, their size was smaller than the minimum legal size, under which fish should not be caught (Table 1), so they are discarded, and they include *P. bogaraveo*, *P. erythrinus*, *P. acarne*, *P. pagrus*, *T. mediterraneus*, *T. trachurus* and *C. cuculus*. The summer samples of *P. acarne* (16.7 cm) and *P. erythrinus* (16.4 cm) were below the limits set by Tsikliras & Stergiou (2014) (21.7 cm and 25.2 cm, respectively). Therefore, the caught individuals were below the maturity size.

The larger *B. boops* size indicates that samples were

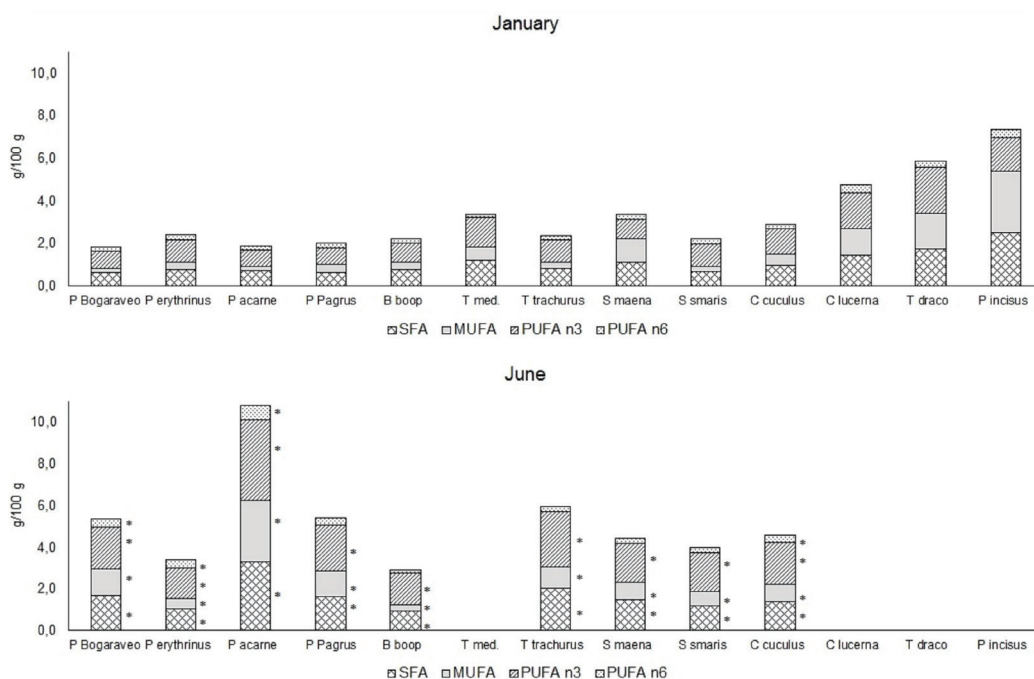


Fig. 2: Fatty acid content of whole body of discarded fish species (g/100 g wet weight).

longer than upon their first maturity, but were discarded because their commercial value on the Spanish coast is low (Carbonell *et al.*, 2018).

The *C. lucerna* specimens also had a longer total length (17.5 cm), which came close to the proposed minimum landing size (>18 cm, Özdemir *et al.*, 2019). The total length of *Spicara* spp., *T. draco* and *P. incisus* samples was also longer than their MCRS (11.5 cm for *S. maena*, 11 cm for *S. smaris*, 14 for *T. draco*, 15.3 cm for *P. incisus*) for both sampling times, and the reason why these species are discarded is clearly their low commercial value. The condition factor (CF) relates length and weight, and most values were above 1.0, except for *B. boops*, *T. mediterraneus*, *T. trachurus*, *S. smaris* and *T. draco*, with values below 0.8. The length-weight ratio is normally affected by several factors; e.g., sampling season, food abundance, size, age, sex, reproductive period, muscle development and fat storage. The CF values did not significantly change between the winter and summer samplings for most studied species, which indicates isometric growth patterns. However, positive allometric growth was seen for *P. bogaraveo*, *P. acarne*, and *P. pagrus*, whose weight increased at a higher rate than their body length. Their CF values were significantly higher in June than in January, possibly because of the more marked increase in Digestive-somatic index (DSI) and Fillet yields (FY) rather than in these species' length.

The length-weight ratio within one species can fluctuate between seasons due to the influence of temporal and/or spatial factors, such as temperature, salinity, food availability and composition. Such variations may also be due to the influence of reproductive processes on individual growth, although the species herein studied had not yet reached the reproductive stage.

Conversely, *S. maena* and, more importantly, *S. smaris* showed negative allometric growth, caused by a considerable increase in length and a significant decrease in digestive weight (perivisceral fat loss). This scenario may be due to changes in fish body shape that becomes more elongated throughout their life span. Reproductive status can also be influenced because the samples of both species were longer than they were upon first maturity. However, no mature gonads were found in any sample, possibly because it was the end of the reproductive season (from May to June).

FA contents

The species with higher FA contents (*C. lucerna*, *T. draco* and *P. incisus*) are essentially demersal. However, their habitat changes throughout the year. In spring, they are found in shallower water (El-Serafy *et al.*, 2015 for *C. lucerna*; Portillo Stempel *et al.*, 2008 for *T. draco* and Fehri-Bedoui & Gharbi, 2008 for *P. incisus*). The larger sizes shown by the samples of these species (bigger upon first maturity) indicated that they were adults, which could be related to their higher FA content. As these individuals were caught in January outside their spawning season, the presence of mature gonads was not observed

in any sample. Several authors have reported that the larger fish size is, the higher lipid contents are (García Mesa *et al.*, 2014).

A significant increase in the FA content was observed in June, which highlights *P. acarne* (10.9 g/100 g fish) and *T. trachurus* (6.0 g/100 g fish). These increases coincided with heavier body weight, especially in *Sparidae* species, whose weight practically doubled during the summer sampling. In some exception cases, the increases noted in the FA content and weight of *B. boops* were not as remarkable as they were in the other analyzed species, possibly because these individuals were adults in both samples, while the others were juveniles. Conversely, Morales-Medina *et al.* (2015) indicated significant changes in the lipid content throughout the year in *B. boops* (from 1.0% in spring to 6.0% in autumn), which of similar size to those herein analyzed.

The general increased FA content in summer agrees with those reported by several authors and could be because of increased feed intake owing to a higher metabolic rate at higher water temperatures (Yildiz *et al.*, 2006). In addition, environmental changes and annual fluctuations influence food availability and composition which, in turn, affect the fish chemical composition (Orban *et al.*, 2011), while the fish spawning season can also influence seasonal variations.

Fatty acid profiles

The proportion of SFAs was similar in most of the species studied in the January sampling (from 29.7% to 33.0% of total FA), but higher values were observed in *P. acarne* (38.0%). The most abundant FA of this family was palmitic acid (PA, 16:0), which agrees with previous works. Its accumulation has been explained by it constituting the major energy source for metabolism and growth (El Oudiani *et al.*, 2019). PA reached its highest values in *S. maena* (24.4%) and *P. acarne* (25.8%). The *S. maena* case could be related to its above-mentioned bigger size and higher FA content. The differences found for *P. acarne* compared to other Sparidae family members may be due to varying feeding habits. Young fish from this family are pelagic, and this habitat is partitioned among different species to avoid diet overlapping.

For MUFA, all the species contained mainly oleic acid (OA, 18:1n-9), which agrees with previous reports (Aydn *et al.*, 2013). High levels of palmitoleic acid (POA, 16:1n-7) were also found for several species, such as *S. maena* (7.8%), *T. draco* (6.0%), *P. incisus* (12.0%) and *C. lucerna* (8.9%). This spells a higher MUFA proportion in these species, especially in *P. incises*, whereas the total MUFA proportion was higher than SFA and PUFA, unlike the trend observed in most of the remaining species (PUFA>SFA>MUFA). High MUFA levels in fish can indicate that they are carnivorous, and their diet is based on zooplankton, crustaceans (e.g. copepods) and mollusks (El Oudiani *et al.*, 2019) because MUFA generally derive from marine copepods and other macrozooplankton (Arai *et al.*, 2015).

n-6 PUFA was represented mainly by linoleic acid (LA, 18:2n-6) and arachidonic acid (ARA, 20:4n-6), which accounted for 9-11% of the total FAs in most studied species. In general, less *n*-6 PUFAs was found than other FA groups, which may reflect their lower level in marine lipids. This contrasts with that observed in the terrestrial environment. Conversely, the *n*-3 PUFA proportion was high (between 35% and 46% of total FAs) in most species, which coincides with previous studies on marine fish. DHA (22:6n-3) and EPA (20:5n-3) were the most representative *n*-3 PUFAs, and DHA was more abundant than EPA in most analyzed discarded species. These FA accumulate from marine microalgae and reach higher values in omnivorous fish (Loukas *et al.*, 2010; Zhang *et al.*, 2020) Significantly lower values of *n*-3 PUFAs were observed in *S. maena* and *P. incisus*, and they had lower DHA proportions.

MUFA and EPA significantly increased in June and, conversely, the remaining PUFA (especially DHA) lowered in most species, except in *B. boops* and *S. maena*, which displayed an inverse trend. Interestingly, the SFA fraction decreased in June only in Sparidae species.

Several researchers have reported an increase in PUFAs and a decrease in MUFAs and SFAs in fish sampled in winter *versus* summer (Celik, 2008; Batičić *et al.*, 2009; Batičić *et al.*, 2011; Suárez *et al.*, 2015) owing to the higher degree of FA unsaturation needed to preserve fluidity of membranes at low temperature (Henderson & Tocher, 1987). The changes observed in the present study can be explained by lower feed intake in winter, which causes lipid reserve depletion. Thus both SFAs and MUFAs are catabolized to a greater extent, which could be why an increase in the PUFA fraction was seen (Shirai *et al.*, 2002; Orban *et al.*, 2011).

El Oudiani *et al* (2019) have reported high MUFA levels in autumn for *S. scombrus*, although Soriguer *et al.* (1997) found an inverse trend for the same species (high MUFA levels in winter). Such differences may be due to different fish physiological states or changes in diet composition during distinct seasons.

DHA is an essential FA located in the phospholipids of cell membranes and is preferentially preserved rather than metabolized for energy production (Bell *et al.*, 1986). Therefore, when DHA amounts do not suffice for structural needs in winter, DHA synthesis from EPA takes place and leads to the observed low EPA levels in January. The high MUFAs and EPA and lower DHA and *n*-6 PUFAs percentages observed in the June sampling could confirm this hypothesis.

Quality index

The *n*-3/*n*-6 ratio is a good marker for assessing the nutritional value of any seafood, and high values indicate good quality lipids: *n*-3 PUFA perform anti-inflammatory and anti-aggregatory activities (Calder *et al.*, 2017). Our results fell within the established range from 3.3 to 14.4 (Henderson & Tocher, 1987), and *T. mediterraneus* and

T. draco stood out (7.4 and 6.9). The lowest values were found in the species with high MUFA and low *n*-3 PUFA levels: 3.7 in *S. maena* and 3.3 in *P. incisus*. *P. pagrus* and *P. acarne* obtained high AA percentages and their *n*-3/*n*-6 ratios were also relatively low (3.4 and 3.9). Values above 3.0 were found in most marine fish species (Celik, 2008). Values higher than 3.5 have been reported to help to lower cholesterol levels in blood and to improve the plasma lipid profile (Morales-Medina *et al.*, 2015).

The PUFA/SFA ratio is another index normally used to evaluate lipid quality, and values above 0.45 are considered positive in human diet given their potential to lower serum cholesterol (Zhang *et al.*, 2020). In the January sampling, the PUFA/SFA ratio ranged between 0.80 in *S. maena* and 1.59 in *S. smarvis*, and values lowered in the June sampling for all species. All the discarded species included in the present study obtained PUFA/SFA ratios above the recommended value, except *P. acarne* (0.33) in the June sampling. This finding agrees with results of Özogul *et al.*, (2009) for several fish species from the Mediterranean Sea.

Atherogenic (AI) and thrombogenic (TI) indices are determined by the relative contents of several FAs and indicate the capacity of the lipid fraction to protect against aggression in the endothelium of blood vessels (atheroma formation), and to produce thrombosis or embolism, respectively. Their determinations are based on the functional effects of FAs on cholesterol metabolism. The most important hypercholesterolemic SFAs are C14:0 and C16:0, while hypocholesterolemic ones are C18:1n-9, C18:1n-7 and PUFAs. High values for these indices indicate an increased risk of platelet aggregation and of thrombus and atheroma formation. All values were favorable (<1) and, therefore, the lipids of the studied species can be considered healthy (Morales-Medina *et al.*, 2015). The highest AI values were 0.56 (*T. mediterraneus* and *S. maena*) in the January sampling and 0.66 (*T. trachurus*) in June, whereas the higher value for TI was 0.42 (*P. incisus*) in January, and all values were lower in June. These values fell within the range indicated by other authors for Mediterranean fish (Özogul *et al.*, 2009; Orban *et al.*, 2011; Morales-Medina *et al.*, 2015) as promising marine food to prevent cardiovascular disorders.

The amount of different FAs (g of FA/ 100 g tissue) was one of the most relevant pieces of information that this study provides because it indicates the FA concentration in the fish body. The largest *n*-3 PUFA amounts in January were for *P. incisus*, *C. lucerna* and *T. draco* (1.36, 1.67 and 2.04 g/100 g, respectively). In June, significantly larger quantities of *n*-3 PUFA were observed in *P. acarne* (3.89 g/100 g). The lowest values were obtained in the winter sampling for Sparidae fish (0.8-1.0 g/100 g). Similar values have been reported for fish in the Mediterranean Sea (Loukas *et al.*, 2010; Morales-Medina *et al.*, 2015) and in the South Pacific (Rincón-Cervera *et al.*, 2020). These findings indicate that, despite seasonal fluctuations, discarded fish is a raw source of bioactive FA, such as EPA and DHA.

Conclusion

High FA contents were found in *C. lucerna*, *T. draco* and *P. incisus*, which also had high body weight values. This fact could not be related to changes in the reproductive cycle because these individuals were caught in January, outside their spawning season. The FA content increased in the summer sampling compared to winter in most studied species, especially *P. acarne* and *T. trachurus* which, apart from a significantly heavier body weight, could be due to increased food availability.

The *n*-3 PUFA percentages were high (between 35% and 46% of total FAs) in most studied species, and EPA, and specially DHA, were the most valuable PUFAs found. However, significantly low *n*-3 PUFA values were observed in *S. maena* and *P. incisus*. In summer, a significant increase in MUFA and EPA, and a decrease in PUFA and DHA percentages, took place in most species. The capture season influenced the total *n*-3 PUFA fraction, with the highest values in winter for *P. incisus* (1.36 g/100 g), *C. lucerna* (1.67 g/100 g) and *T. draco* (2.04 g/100 g), and in summer for *P. acarne* (3.89 g/100 g).

Despite seasonal fluctuations, the discarded fish here-in analyzed can be considered not only healthy for humans according to their FA composition, but also a raw source for the production of edible oils rich in EPA and DHA.

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Supplementary data

The following supplementary information is available online for the article:

Table S1. Fresh fish commercially available in Almeria from January 01 to December 31, 2018.