1	Mercury contents in relation to biometrics and proximal composition and
2	nutritional levels of consumed fishes from Western Mediterranean Sea (Almeria bay).
3	
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16	Keywords: Food safety; Mercury; Mediterranean seafood; Fish biometrics; Trophic
17	relationship.
18	
19	Abstract
20	The total liver and muscle mercury, and muscular composition, biometrics and trophic levels
21	were determined in four species (Mullus surmuletus, Merluccius merluccius, Auxis rochei and
22	Scomber japonicus) of Mediterranean Sea (Almeria Bay, Spain). The mercury levels did not
23	exceed the Maximum Residue Limit, being the <i>M</i> . merluccius the one which showed the highest
24	level in muscle. Great variations of Hg content among individuals were observed in not
25	gregarious species. A positive correlation between Hg and trophic level or length was found in
26	muscle but not in liver. The organ (liver or muscle) with major Hg accumulation depends on
27	species; in M. merluccius was the muscle and in S japonicus was the liver. The results indicate
28	that the Hg levels in fish depend on intra and inter species factors that should be taken into
29	account in the monitory systems of Hg levels.
30	
31	1. Introduction
32	Pollution is one of the current problems facing the environment. In this context mercury (Hg) is
33	one of the six worst pollutants on our planet, according to the UN's International Chemical
34	Safety Programme (Keeler et al., 2006) as it has adverse effects on most living organisms (Dietz
35	et al., 2000), and also neurological damage, especially in children (Counter et al., 2002; Zhang
36	and Wong, 2007). It may enter the environment from human activities, like mining and smelting
37	processes (Gutiérrez et al., 2016; Kaitantzian et al., 2013; Odumo et al., 2014) or from fossil

38 combustion (Kelepertzis and Argyraki, 2015; Rodriguez Martin et al., 2014; Rodríguez Martín 39 et al., 2013). In any case, human activities have substantially raised the heavy metals levels in 40 the last decades (Rodríguez Martín et al., 2015). The global inventory of Hg sources for the year 41 2000 was 2,200 ton year<sup>-1</sup> (Pacyna et al., 2006). Nowadays, about 2,200–4,000 ton year<sup>-1</sup> of 42 anthropogenic Hg is emitted to the atmosphere (Kim and Kim, 1999). Coastal ecosystems are 43 conditioned by growing anthropogenic impact. Consideration to pollution of the Mediterranean 44 Sea is asociated to semi-enclosed sea intensification of contamination effects (Aston and 45 Fowler, 1985). Same fish species from Atlantic Ocean contain half Hg than specimens from 46 Mediterranean Sea (Aston and Fowler, 1985). Della Croce et al. (1997) have established that the 47 amount of Hg discharged into the Mediterranean is 130t per year. The fish and shellfish being 48 the food that most concern the risk associated with exposure to Hg (Zamani-Ahmadmahmoodi 49 et al., 2014).

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51 On the other hand, the potential health benefits related to fish consumption are due to the 52 presence of proteins, minerals, vitamins and the presence of unsaturated essential fatty acids related (UFAs), especially omega-3 UFAs including eicosapentaenoic acid (EPA) and 53 54 docosahexaenoic acid (DHA). Nowadays the intake of fish is recommended as a healthy 55 nutritional habit. Although people may be exposed to any form of mercury in various circumstances, diet is the mainly way of Hg accumulation (Hall et al., 1997) and seafood is one 56 57 of main way exposition (OMS, 2017). The Hg content varies among fish species (Groth, 2010), 58 feeding (Minganti et al., 2010) and length (Branco et al., 2004) or age (Monteiro and Lopes 59 1990; Storelli, 2005a, 2005b). The Hg is incorporated to the food chain provoking a 60 biomagnification, increasing Hg content with trophic position, being in adult top-predator more 61 than a million times higher than water concentration (Grigal, 2003; Ullrich et al., 2001;).

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63 The Mediterranean Sea supplies of fish to more than 480 million people (EEA 2017), where 64 Mullus surmuletus, Merluccius merluccius, Auxis rochei and Scomber japonicus are four of the 65 most consumed species. Red mullet (Mullus surmuletus) is a target species of Mediterranean 66 demersal fisheries that can be catched through different gears (Reñones et al., 1995). In Almeria 67 Fish Market, *M. surmuletus* is located among the 20 species with highest profits in the last 3 68 years (328.538€ among 2015-2017, IDAPES). European hake (Merluccius merluccius) in one 69 of the most importan species of the Mediterranean Sea (Chapela et al., 2007), and it's fished by 70 longline, gillnet and bottom-trawling fishery. In Almeria Fish Market, the european hake is 71 placed the 9th position of the most earnings in the last 3 years, with landings of 97.802 kg and 72 798.652 € of economic gain (2015-2017, IDAPES). Bullet tuna (Auxis rochei) is a fish species 73 exploited commercially by artisanal fisheries (purse seiners, longliners and traps). Bullet tuna is 74 the most abundant species of the tuna family in the Mediterranean, and landings in Almeria Fish Market have been of 281.565 kg with profits of 328.955 € (2015-2017,IDAPES).The Pacific
Chub Mackerel (*Scomber japonicus*) is a very important commercial species exploited by purse
seiners for direct human consumption and also for can industry and as food for bluefin tuna of
aquaculture. In Almeria Fish Market, chub mackerel is the second most landed species in
volume (kg) of small pelagic fishes after pilchard (*Sardina pilchardus*), with landings of
722579 kg and profits of 300.072 € (2015-2017, IDAPES).

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The aim of this study was to explore the potencial toxicity risk of Hg in the four species most consummed in Andalusian coast. The main goals of this work were: 1) to determine the levels of mercury contents in hepatic and muscular tissues. 2) to study the relationship between mercury content and biometric index, age, trophic level and body composition. 3) to have a first sight to the quality of fishing areas in the Bay of Almeria.

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## 89 2. Materials and methods

#### 90 2.1 Studied Fish species

Four popular fish species of the Mediterranean Sea with commercial importance were selectedfrom the Bay of Almeria (Western Mediterranean Sea)

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94 Red mullet (M. surmuletus) is a teleost fish of the Order Perciformes of wide distribution, is 95 located in the Northeast Atlantic and Mediterranean, mainly in very shallow areas of the 96 continental shelf (between 0-30m) of as much zones of rock as sand and mud (Lombarte et al., 97 2000; Whitehead et al., 1986). In spite of this very coastal distribution, this species can reach to 98 inhabit depths of up to 400m, depths that the other red mullet (M. barbatus) does not inhabit 99 (Lombarte et al., 2000). It is a benthic carnivore that feeds mainly on small invertebrates 100 (crustaceans, mollusks, polychaetes) that live on or in the interior of the marine substrate 101 (Gharbi and Ktari, 1979, Golani and Galil, 1991).

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European hake (*M. merluccius*) is a demersal species teleost of the order Gadiformes widely distributed in the Atlantic Northeast and throughout the Mediterranean Sea. It inhabits from 50-750 m depth, although its most occurring habitat is in the deep zone of the continental shelf, between 150-300m (Recasens et al., 1998). *M. merluccius* is an opportunistic carnivore that feeds on adult stages of large decapods, euphausiidae and teleost fishes such as myctophids, performing daily vertical migrations to feed (Bozzano et al., 1997; Cartes et al., 2009).

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110 Bullet tuna (*A. rochei*) is a small tuna of the Order Perciformes that is distributed by the 111 temperate and tropical waters of the whole world, including the Mediterranean. *A. rochei* 

- mainly feeds on zooplankton organisms, with particular preference for planktonic crustaceans
  such as amphipods and eufasiaceans, as well as small cephalopods, fish larvae and even adult
  fishes for larger bullet tuna sizes (> 35 cm) (Mostarda et al., 2007)
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La The Pacific Chub Mackerel (*S. japonicus*), is a small pelagic of the Order Perciformes. It is a neritic pelagic species that is distributed mainly between 50-200m depth, in temperate and tropical waters of the northern and southern hemisphere, including the Mediterranean(Collette and Nauen, 1983) The diet of *S. japonicus* is mainly based on different stages of fishes (including cannibalism, decapods, and annelids (Castro, 1993).

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## 122 **2.2 Field collection**

123 In March 2016, the four species were caught during several commercial surveys in Almeria 124 Bay, Western Mediterranean, in the Southeast Coast of Andalusia (Figure 1). Two of the 125 species were small pelagics fished by the purse seine vessel "El Chapu"; the Pacific Chub 126 Mackerel (Scomber japonicus) in the fishing ground "La Terraila", characterised by an isobat of 127 100 m depth, and the Bullet tuna (Auxis rochei), in the fishing ground named "El Cantillo" with 128 depths ranging between 50-180 m. The other 2 species were fished by the bottom-trawler "Jose 129 y Fernanda", the most popular demersal species in the area, European hake (Merluccius 130 merluccius), in the fishing ground "Medio Canto" near the slope, at depths 130-200m, and the 131 abundant benthonic species Red Mullet (Mullus surmuletus), in the fishing ground "La 132 Terraila", in the first 100m. The fishes were transported to the laboratory into ice for biometrics. 133 The liver and muscle were dissected out, weighed and stored at -20°C until lyoifilization.

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#### 135 **2.3 Fish Biometric Indices and Body Composition**

Fishes were weighted (Wt) and measured (Total length (Lt)). Age of the fishes was calculated following the formula of von Bertalanffy 's growth of other studies for each species (Table 1). Trophic level for each species was calculated through the fractioning index TROPH presented in Stergiou and Karpouzi (2001) where original studies are presented for red mullet and european hake, and from Fish base online (www.fishbase.org) for chub mackerel and bullet tuna (Table 1).This index is based in stomach contents and expresses the position of organisms within the marine food webs. The index also takes in consideration the size of the specimens

144 (1) 
$$TROPH_i = 1 + \sum_{j=1}^{G} (DC_{ij} \times TROPH_j)$$

where TROPH<sub>i</sub> is the trophic level of species (i), TROPH<sub>j</sub> is the trophic level of prey (j), DC<sub>ij</sub> is
the contribution of prey (j) in the diet of species (i) and G is the total number of prey. The
values of the TROPH range 2.0-4.5:

148	a)	2.0-2.1: Pure herbivores
149	b)	2.1-2.9: Omnivores with a preference for vegetable materia
150	c)	2.9-3.7: Omnivores with a preference for animals
151	d)	3.7-4.5: Carnivores with a preference for large decapods, cephalopods and fish

The mucle composition was evaluated according to the Association of Official Analytical Chemists (AOAC, 2000). Dry matter was determined gravimetrically after drying at 105±0.5°C (DM; method #934.01) and ash was determinated after combustion at 500°C in a mufla oven (metod #942.05) to constant weight. Content of crude protein was determined by Kjeldahl (PB; methods #954.01)(Nx6.25), and total lipid was determined by ethyl ether extraction (EE; method#920.39)

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## 161 2.4 Measurement mercury levels

162 The total Hg in all samples was determined using a direct Hg analyzer (DMA80, atomic 163 absorption spectrophotometer, Milestone, Wesleyan University, Middletown, CT, USA). 164 DMA80 provides two working ranges for Hg detection: 0-40 and 40-600 ng. Each range is 165 calibrated independently to optimise the response over the entire dynamic range. The limits of 166 detection (LOD) and quantification (LOQ) were 0.5 and 1.25  $\mu$ g kg<sup>-1</sup>, respectively. The LOQ 167 was established by the lowest calibration point; the LOD was 2.5 times lower than the LOQ 168 when the signal-to-noise ratio was higher than 10 (Carbonell et al., 2009). The analytical 169 procedure validation of the Hg analysis was performed with a certified reference material CRM 463 (tuna fish 2850  $\mu$ g kg<sup>-1</sup> DW) and ERMI-CE278 (mussel tissue 196  $\mu$ g kg<sup>-1</sup> DW). The Hg 170 171 analysis revealed good agreement between the obtained and certified values, showing an 172 average recovery of 97% and 101%, respectively. Results are in consonance with those of the 173 certified values. Two replicates were analyzed per sample.

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#### 175 **2.5 Statistical analysis**

176 A standard statistical analysis (mean, median, standard deviation, etc.) was carried out to 177 describe fish biometric and analytical body compositionresults. Statistically significant 178 differences related to the control were estimated using the analysis of variance (ANOVA). Data 179 were not transformed and significant differences between species were assessed by non-180 parametric Kruskal-Wallis tests (at  $\alpha = 0.05$ ).

To study the relationship between Hg in different tissues (muscle and liver) and fish biometric
(size and analytical body composition), we used CCorA (canonical correlation analyses).
Discovered by Hotelling (1936), this method is used considerably in ecology (Campos-Herrera

et al., 2013; Höss et al., 2011; Rodríguez Martín et al., 2014). Let Y1 and Y2 be two tables
(Mercury contents, and the response variables (Y2) based on size and body composition), with
variablesp and q, respectively, we obtain:

 $\rho(i) = cor(Y1a(i), Y2b(i)) = \frac{cov(Y1a(i), Y2b(i))}{var(Y1a(i)).var(Y2b(i))}$ 

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(2)

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190 The CCorA provide two vectors, a(i) and b(i), that are maximised. Constraints must be 191 introduced so that the solution for a(i) and b(i) is unique. As the ultimate intention is to 192 maximise the covariance between Y1a(i) and Y2b(i) and to minimise their respective variance. 193 CCorA (Kianifard, 1993) provides the relationships between biological variability and 194 sensitivity to chemical disturbance (Campos-Herrera et al., 2016; Losi et al., 2013; Takoutsing 195 et al., 2017). The graphical results of the CCorA were presented with bi-plot scaling to evaluate 196 the relationship. the first set of variableswere Hg contents in liver an muscle tissues and the 197 second set of variables are the results of Fish Biometric indices and body composition 198 parameters (Moixture, lipids, Protein, Ash, O.M. and NFE). All the statistical analyses were 199 carried out by XLSTAT (Addinsoft Version 2012.2.02) package for Windows.

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## 201 **3. Results and discussion**

#### 202 **3.1. Biomectics and muscle composition**

203 The summary statistics (mean and standard deviation) of biometrics of fish and their age and 204 trophic level are listed in Table 1 for the four study species. The Weight and Total Length (Lt) 205 were similar among individuals of each species. The highest variability was found in M. 206 merluccius. The A. rochei showed the highest values of weight and length, while the M. 207 surmuletus showed the lowest ones. The estimated age indicates that the M. merluccius, 3.5 208 years old, are the oldest fishes of this study and the M. Surmuletus are the youngest, with one 209 year old. M. Merluccius showed the highest trophic level (4.41 TROPH). On the opposite, 210 small fishes as M. Surmuletus were the ones which showed a low trophic level and similar to S. 211 *japonicus* (Table 1). The values of our species places the bullet tuna and hake with the highest 212 TROPH levels, while the red mullet and mackerel are located in lower links corresponding to 213 omnivores of animal preference (Stergiou and Karpouzi, 2001). The size of the individuals of 214 each species were similar (Table 1) with a little SD because fishes were cathed in the same set 215 of the operation gear, indicating that they were of the same school in the case of pelagic fish, S 216 japonicus and A, rochei. Regarding demersal and benthonic species, they are not gregariouss 217 and show more random geographic distributions, the bottom trawlers go across variable 218 distances capturing fishes of different stocks. As larger is an individual, there are dietary 219 changes towards larger prey, and therefore the trophic level it occupies in the marine trophic 220 chain, this is valid both within the same species and between different species (Pauly et al. al.,

221 1998, Pauly and Sa-a, 2000a). Body size is a good descriptor of the trophic level of a specimen, 222 regardless of the species (Jennings et al., 2007). In addition, the age of each species must be 223 taken into account, because their food structures grow and ingest larger prey (Deudero et al., 224 2004; Galván et al., 2009). Therefore, our species and the trophic values that they show, are 225 combining both the size and the age of the specimens, and placing the hake in the highest place 226 of the trophic chain, despite not having the greatest weight but they are the oldest fishes. 227 Following the hake, is placed the bullet tuna, that if it has the greater weight and an intermediate 228 age between the hake and mackerel. At lower levels are mackerel and mullet, small sizes and 229 ages between 1-2 years (Table 1).

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231 The study of the muscular composition is shown in Table 2. For moisture the highest value 232 correspond to M. surmuletus (82,21%) and lowest in A. rochei (65,03%) which presents the 233 highest percentage of lipids (23,69%) while the lower lipids content were observed in M 234 merluccius. The protein is major macronutrient content in all the species analysed 66% (A. 235 rochei) to 84% (M. merluccius). The organic matter and NFE showed similar values among 236 species; the lowest value was obtained in *M. merluccius* (94.67% and 5.05% respectively), and 237 the highest in S. japonicus (95.52% and 9.68% respectively). The lipids percentaje observed in 238 A rochei (Table 2) was slightly higher to the one obtained by Saito and Ishihara (1996) for this species, 4.8% for dorsal muscle and 21.6% for ventral muscle, but lower than the results of 239 240 Karunarathna and Attygalle (2010). S. japonicus showed similar lipids values to the results 241 described by Reinitz et al. (1979), which ranged between 1.5 and 19% of the wet weight, but 242 richer than the ones described by Celik (2008); M. merluccius showed aproximately double 243 lipids levels than those published by Pérez-Villarreal and Howgate, (1987). Regarding the 244 protein, the values obtained for A rochei were similar to the protein reported by Karunarathna 245 and Attygalle, (2010), however S. japonicus and M. merlucius showed lower protein levels than 246 the reported by Celik (2008) and Pérez-Villarreal and Howgate, (1987) respectively. In general, 247 the values of muscular composition are in the range described by FAO (2017) for these species, 248 nevertheless fishes are subjected to seasonal changes in body composition due to different 249 availability and composition of their prey. Moreover, the activities of the fish such as 250 reproduction and migration (influenced by water temperature and photoperiod), have an 251 influence on the chemical composition of the fish (Bandarra et al., 1997; Olsson et al., 2003 252 Grigorakis et al., 2002) and it is difficult to compare data with only sampling.

253 A strong relation between muscle composition and fat, moisture, and weight were observed.

254 The increase of weight is related with fat content while the hydrophobic properties of fat

255 provoke a decrease of moisture with the increase of fat.

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#### 257 **3.2 Mercury Fish levels**

258 Table 3 summarises the statistics of the Hg contents in muscle and liver of the four study 259 species expressed as dry weight (DW). In order to assess the changes in the concentration of 260 muscle and liver the figure 2 shows the statistically significant differences between muscle and 261 liver for each species. Hg content in muscle for *M. merluccius* fell within 555.3 - 1065.6  $\mu$ g kg<sup>-1</sup> 262  $(854.8\pm197.54 \ \mu g \ kg^{-1})$ , pointing out this species as the one with highest mercury content, with 263 statistically significant differences with A. rochei (567.7±133.44 µg kg<sup>-1</sup>), M. surmuletus 264  $(490.7\pm183.68 \ \mu g \ kg^{-1})$ , and S. japonicus  $(306.4\pm48.67 \ \mu g \ kg^{-1})$  that showed the lowest values 265 of mercury in the muscle (Table 3). The variations of Hg content among individual of same 266 species should be considered to establish an adequated number of replies. A great variation of 267 data was observed in M. surmuletus and M. merluccius while A rochei and S. japonicus didn't 268 show this variability. This could related with the ecology of each species; *M. surmuletus* and *M.* 269 merluccius are not gregarious species, them there is high intraspecific variability in their feeding 270 (Stergiou y Karpouzi, 2001) while S. japonicus and A. rochei live in schools and presumably 271 with a more homogeneous diet among individuals of the school (Menard et al. 2000).

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On the other hand, several studies have demonstrated elevated Hg concentrations in the Mediterranean fishes compared to the same species from other parts of the world (Harmelin-Vivien et al., 2009). The Hg level in fish varies among species and individuals of same species, and the factors that influence in the bioaccumulation are not completely known. The level of Hg in one species depends on trophic levels (Agah et al. 2006; Agusa et al. 2004; Anan et al. 2011), the size and age (AESAN 2010; Agusa et al 2004; Andersen y Depledge 1997; ATSDR 1999) and the duration and level of exposure (Authority EFS 2012).

280 *Merluccius merluccius* from Almeria bay has the highest concentration of Hg regarding the 281 other species studied (Table 3). In the Italian coast (Brambilla et al., 2013), this species has 282 shown mercury contents of 159  $\mu$ g kg<sup>-1</sup> in wet weight (WW), slightly lower than those measured 283 in the coast of Almeria (163  $\mu$ g kg<sup>-1</sup> WW). *M. merluccius* caught off the Central Adriatic (Di 284 Lena et al., 2017) presented even lower content (85  $\mu$ g kg<sup>-1</sup> WW). Although, Cresson et al. 285 (2015a) found levels of Hg in *M. Merluccius* in France mediterraneam coast from 290 to 1360 286  $\mu$ g kg<sup>-1</sup> (DW), similar to the levels of Hg analysed in this study (854.83  $\mu$ g kg<sup>-1</sup> DW).

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In our study *Mullus surmuletus* presents a mean content of 490.7 µg kg<sup>-1</sup> DM of Hg, (87 µg Hg kg<sup>-1</sup> WW,) which is higher than the reported for the Gulf of Lions with 202 µg kg<sup>-1</sup> DM (Harmelin-Vivien et al., 2009), for Ligurian Sea with 210 µg kg<sup>-1</sup> DM (Capelli et al., 2004), but lower than the reported in the Italian coast with 258 µg kg<sup>-1</sup> WW (Brambilla et al., 2013), in Central Adriatic 129 µg kg<sup>-1</sup> WW (Di Lena et al., 2017), and in the Gulf of Lions with 920 µg kg<sup>-1</sup> DM (Cresson et al., 2015). Also *A rochei* has presented lower levels of Hg in Almeria (198 µg kg<sup>-1</sup> WW) than the reported in the Italian coast with 221 µg kg<sup>-1</sup> WW (Brambilla et al., 2013)

or 280 µg kg<sup>-1</sup> WW (Di Lena et al., 2017). The *Scomber japonicus* species (86 µg Hg kg<sup>-1</sup> WW)
shows lower level than those obtained in other areas of the Mediterranean Sea, as Central
Adriatic 171 µg kg<sup>-1</sup> WW (Di Lena et al., 2017) or Italian coast (Brambilla et al., 2013) with 220
µg kg<sup>-1</sup> WW.

299

300 In general the liver is the organ where more mercury can be found in any terrestrial or aquatic 301 species. However, no differences in Hg levels between muscle and liver were found in A. rochei 302 and M. surmuletus whilst the M. merluccius has significant higher Hg content in the muscle 303 (854.8 µg Hg kg<sup>-1</sup> DW) than in the liver (334.90 µg Hg kg-1 DW) (Figure 2). Only the S. 304 *japonicus* showed significant higher Hg concentration in the liver 532.35 µg Hg kg-1 DW than 305 in muscle 306.39µg Hg kg-1 DW, what means a 50% higher than in muscle (Figure 2). The 306 liver has an important role in redistribution, detoxification and transformation of pollutants 307 (Yamashita, et al., 2005; Maršálek et al., 2007). Havelková et al. (2008) found that the target 308 organ to accumulate Hg depends of pollution level; the fish are from high polluted areas, the Hg 309 accumulates in liver, in low polluted areas the target organ it is the muscle. However, in our 310 study the fish was caught off from same area (Bay of Almeria), and the Hg content variability in 311 the liver depends on the species (Table 3). It has been documented that species such as *Xiphias* 312 gladius and Thunnus thynnus present higher Hg content in liver than in muscle (Storelli, 2005), whilst in shark it is the muscle the accumulative organ (Branco et al., 2007). However in catfish 313 314 there aren't differences in Hg content in organs, liver and muscle (Arantes et al., 2016).

The different ditribution of Hg depends on elimination rate and tranportation into de organs. The elimination and transport of Hg in *Siganus canaliculatus* was slowly distributed into muscle but was efficiently eliminated by the intestine. However the muscle didn't eliminate the meHg. These elimination and acculumation rates seem to be specific to each species (Pen et al., 2016).

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# 321 3.3 Assessing relationship between mercury content in tissues and Biometrics and muscle322 composition

323 As previously mentioned, no clear effect in the Hg accumulation have been shown when we 324 look at each species separately. To evaluate this, CCA allows to provide the relationships 325 between biological variability and sensitivity to chemical disturbance (Campos-Herrera et al., 326 2016; Losi et al., 2013; Takoutsing et al., 2017); thus, this method has been considerably used 327 in ecology (Campos-Herrera et al., 2013; Höss et al., 2011; Rodríguez Martín et al., 2014). 328 Numerous factors have been described that affect the accumulation of Hg. In this study the Hg 329 content of tissues has been connected with length, weight, age, trophic level, and muscle 330 composition (protein, lipids, ash...) showing a positive association with age and trophic level.

331 The CCA (Figure 3) proved the relationships between fish Biometric indices and body 332 composition parameters versus muscle and liver Hg contents as a whole which accounted for 333 89% of total variance.

No association has been found between hepatic level of Hg and trophic levels, probabily due to
the role in detoxification of the liver (Yamashita, et al., 2005; Maršálek et al. 2007) than that the
accululation.

337 On the opposite, trophic level and total fish length were the factors which was most correlated 338 with Hg contents in muscle. This correlation between trophic level and Hg muscle has been 339 previously confirmed (Snodgrass et al., 2000) and also the size of the fish (Vieira et al., 2011; 340 Storelli and Barone, 2013). Branco et al. (2004) found a tendency for Hg concentrations to 341 increase with length in shark. These results indicate that the bioaccumulation of Hg is due to 342 exposure; at higher trophic level and age, greater exposure to Hg. In the case of M. surmuletus 343 the Hg levels found were higher than what it would correspond to their age and trophic level 344 due to the fact that the habitat and feeding habits increase the exposure (Benedicto et al., 2008; 345 Storelli et al., 2005a), for that reason red mullet is considered as species indicative of the 346 environmental quality of the marine ecosystem (Machias and Labropoulou, 2002).

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In any case, our results can be interpreted in the light of there is a great variability between species still living in the same area where have been collected, but in general, bigger species and a high trophic group tend to accumulate more mercury in the muscle tissue. By the other hand, our results indicate that hake, red mullet, bullet tuna and mackerel consumption from Almeria area does not represent a risk to health.

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## **4. Conclusions**

According the results the target organ for Hg accumulation, liver o muscle, depend on the species. The age or trophic levels are related with Hg level in muscle but not with the Hg in liver. It should be noted a high variation among individuals in non-gregarious species, *M. surmuletus* and *M. merluccius*, that indicates the area and feeding of each individual are two factors to consider in addition to age or trophic levels.

360 On the other hand, according to this result, the adequate and safe monitoring systems of Hg in 361 fisheries should be designed according of the area, trophic level and gregarious habit of the 362 species.

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- 373
- 374 Figure captions:
- Figure 1: Fishing areas for four species in Almeria Bay, Western Mediterranean Sea.

Fish Biometric indices and body composition parameters

- 376
- Figure 2. Mean concentrations ( $\pm$ SD) of Hg on muscle and liver tissues per fish species. Footnote: Significant between tissues at \* 99% (p < 0.01). ns. not significantly different.

- Figure 3: Ordination diagram based on the CCorA of Hg in muscle and liver tissues versus

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