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Title: Seventeen years analysing mislabelling from DNA barcodes: towards hake sustainability

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10 ABSTRACT

Mislabelling is a common threat to fisheries sustainability. Over the last decades, molecular 11 tools have been established as the main resource to detect mislabelling. This study focuses 12 on these efforts in the genus Merluccius. A meta-analysis approach is taken in order to 13 detect trends on mislabelling directions for the last 17 years. A total of 1291 DNA-identified 14 hake products from 45 different studies were compiled. An increase in number of 15 publications using DNA forensics to detect mislabelling in hake can be seen along the 16 studied time period. However, representation of different hake species varies, i.e. Pacific 17 hakes are underrepresented. Different risk of mislabelling has been identified depending on 18 19 the regions: Highest risk of mislabelling was found in African hake species (only 20.53% of African hake were correctly labelled). Furthermore, a high amount of hake products with 20 incomplete labels (e.g. not reporting the species) were unevenly distributed. Directionality 21 in mislabelling was detected for all cases between sympatric species. Differences in 22 mislabelling rates were found for different regions (Africa, Europe, Pacific America and 23 South America). While a decrease in mislabelling was reported between 2011 and 2014, this 24 has not being sustained over time, as more recent data show an increase in mislabelling 25 rates. Altogether, rigorous monitoring of product authenticity is called for, with special 26 27 attention to the more vulnerable species.

28 *Keywords*: Mislabelling, Fraud, Hake, *Merluccius*, DNA-authentication, meta-analysis.

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30 **1. Introduction**

Mislabelling and food fraud are high-profile issues that have been reported and covered both in the media and by the academic community. It is a common concern affecting different types of food products (Di Pinto et al., 2015; Stamatis et al., 2015). The seafood sector is one of the most affected: instances of fraud have been widely reported (e.g.

35 (Bénard-Capelle et al., 2015; Galal-Khallaf, Ardura, et al., 2016; Guardone et al., 2017; Helyar et al., 2014; Khaksar et al., 2015). Regulations of different countries demands products to be 36 37 correctly labelled. For instance, EU regulation (EU 1379/2013) requires fish labels to individually identify each commercial seafood product to the species level. Labels under this 38 39 regulation must include a scientific name and a commercial designation according to a standardized list with approved names in the official languages of the territories. 40 41 Information regarding production method and region and date of capture are also required. 42 However, these regulations are not always followed.

Illegal, unreported and unregulated fishing (IUU) constitutes about one-fifth of the 43 global catch (Flothmann et al., 2010). IUU products reach the market despite considerable 44 efforts made against them, for example, using eco-labels (Yokessa & Marette, 2019). 45 Evidences of commercial mislabelling hiding covered exploitation of substitute species 46 suggest that this practice happens worldwide (e.g. Ardura et al., 2010; Helyar et al., 2014; 47 Machado-Schiaffino, Martinez, & Garcia-Vazquez, 2008; Muñoz-Colmenero et al., 2017; Von 48 Der Heyden et al., 2010). If the product of IUU fishing is sold fraudulently mislabelled as 49 50 another species, it is impossible to determine the real catch, thus, hindering sustainable management of the resource (Galal-Khallaf et al., 2016; Marko et al., 2011). Substitutions 51 52 from one species to another are frequent all along the supply chain (Helyar et al., 2014; 53 Muñoz-Colmenero et al., 2016). Hence, mislabelling is accumulative, and it is carried along 54 from the point where it takes place onwards, up to the final consumer's plate (Gordoa et al., 55 2017). This is a serious threat for the sustainability of fishing resources, which has been 56 reported for different case studies; e.g. the high level of mislabelling in red snapper (Lutjanus campechanus) inflates stock estimates, further damaging the already depleted 57 58 stocks (Marko et al., 2004; Spencer & Bruno, 2019).

59 Mislabelling is also a problem for the consumer when they buy a substitute species 60 instead of the desired one. Furthermore, inadequate labelling raises health concerns, as potential allergens or unnoticed toxic species may pass unreported (Giusti et al., 2018; 61 Guardone et al., 2017; Muñoz-Colmenero et al., 2013; Sheth et al., 2010; Triantafyllidis et 62 al., 2010), as well as potential contaminants, which may be more frequent in certain areas 63 (Filonzi et al., 2010; Garcia-Vazquez et al., 2011). The economy and consumer's preferences 64 are often behind fish fraud, since substitutes are generally less appreciated species (Muñoz-65 Colmenero et al., 2017). Moreover, mislabelling covering exploitation of vulnerable species 66 undermines consumer's awareness and conservation efforts (Cawthorn et al., 2011; 67 Christiansen et al., 2018; Garcia-Vazquez et al., 2012; Marko et al., 2011). Thus, it has not 68 69 only ecological but also social consequences (Crona et al., 2016; Lam & Pauly, 2010; Levin et al., 2014; López de la Lama et al., 2018; Mariani et al., 2014). 70

DNA authentication of seafood products is key to detect mislabelling. It allows for species identification even in processed products where species recognition by other methods, such as morphological identification, would be impossible. Furthermore, DNA-

74 based identification methods require less taxonomic expertise and are easy and quick to apply. While other authentication methods exist, i.e. protein based, or immunological 75 76 methods, the use of molecular tools has therefore been established as the main tool to 77 detect the mislabelling, allowing for a growing identification of fraud and the raise of 78 awareness against it. This may have an effect on mislabelling rates, that seem to decrease 79 over time, as suggested by Mariani et al. (2014) for cod. However, no standardized 80 procedure or routine genetic controls have been implemented. This has been pointed out by experts (Barcaccia et al., 2016; Clark, 2015; Griffiths et al., 2014; Mariani et al., 2015; 81 Pérez et al., 2018), as well as advised by FAO (2018). Although not routinely applied, there is 82 83 abundant literature for the development of genetic markers and application of molecular 84 techniques for detecting mislabelling, with different methods and markers being developed 85 constantly (Böhme et al., 2019; Haynes et al., 2019).

In this review, we will focus on the genus Merluccius (Rafinesque, 1810), as it is of great 86 87 interest due to its high economic value. Hake is one of the most consumed fish in Europe, especially in Spain, where most of the landings have been reported (Eurostat, 2020). The 88 89 family Merlucciidae -to which hakes (Merluccius) belong- is among the five taxa in which 90 mislabelling risk has been studied the most (Luque & Donlan, 2019). This genus comprises 91 12 species distributed along the Atlantic and the West and Southern part of the Pacific 92 Ocean. Total amount of hake landings pass the million tonnes per year. Currently, North 93 Pacific Hake (M. productus), Argentine Hake (M. hubbsi) and Cape hakes (M. capensis and 94 M. paradoxus) are the most caught species (FAO, 2020a), and many of the Merluccius 95 species have stocks under high fishing pressure (FAO, 2020b). Declines in their stocks have occurred since the 1990s (Pitcher & Alheit, 1995). As they are sensitive to overfishing 96 97 pressure (H Arancibia & Neira, 2008), determining IUUs affecting them can play a significant role on undermining stocks assessment and management decisions. Many of the hake 98 99 species overlap their range of distribution with at least another congeneric species (see the geographic pattern of distribution in Fig. 2, as described in Pitcher & Alheit, (1995)), being 100 101 generally caught in mixed-stock fisheries. Due to this and their similar morphology, accidental mislabelling occurs between sympatric species, e.g. M. albidus and M. bilinearis, 102 or M. capensis and M. paradoxus (Garcia-Vazquez et al., 2009, 2011; Machado-Schiaffino et 103 al., 2008; Muñoz-Colmenero et al., 2015)). However, fraud has been reported in several 104 105 species from different regions (Barendse et al., 2019; Cawthorn et al., 2015; Delpiani et al., 2020; Muñoz-Colmenero et al., 2015; Pérez et al., 2018), as substitutions occur as well 106 107 between allopatric Merluccius species. First studies developing molecular markers in hake oriented to detecting mislabelling are from the beginning of the 2000s (e.g. Castillo et al., 108 109 2003; Perez & Garcia-Vazquez, 2004), and allowed for studies focusing in fraud at market level, which have continued since. 110

Using a meta-analyses approach, the aim of this review is to depict a global picture of labelling issues discovered from DNA-based authentication in hakes, to assess the directionality of the mislabelling, and to identify which species are the most underreported thus at risk of undetected overexploitation. Research gaps in the knowledge of commercial
 fraud in the different species of hake and the evolution of mislabelling rates in the last
 seventeen years have also been assessed.

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118 2. Material and methods

119 *2.1 Data search and compilation*

120 In order to carry out a meta-analysis through relevant literature, key words were employed to standardize the search and optimize the results. Google Scholar and Web of 121 122 Science were the engines employed for the literature search. We aimed at research articles reporting molecular identification of mislabelling in *Merluccius* hake species. The key words 123 used were "Mislabelling", "Hake", "Merluccius", "IUU", "Molecular Markers", "Gadoids" and 124 "Fish fraud identification". Publications not including samples of the Merluccius genus were 125 126 not considered. In addition, articles which had been referenced in the reviewed literature were also added. 127

The following data from each of the research articles analysed were compiled: species reported on the label (both common name and scientific name); species assigned by molecular tools; genetic marker employed; country of purchase of the product; sampling year; reference article. Products were classed in one of these three categories: correctly labelled, mislabelled, or incompletely labelled (for example not stating the scientific species name or not reaching species level). All compiled data of labels and substitute species can be found in an online repository (Blanco Fernandez et al., 2020).

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136 *2.2 Data analysis*

Data from the different papers were combined in order to focus on different aspects 137 of problematic mislabelling cases. First, we analysed the commercial products by genetically 138 ascertained species. Samples were classified attending to the type of substitute species 139 assigned by DNA-based techniques. Substitute species were classified into the following 140 categories: "Different genus" i.e. not Merluccius, "Allopatric" i.e. Merluccius species with no 141 overlapping geographical distribution, and "Sympatric" i.e. Merluccius species exhibiting the 142 same or at least overlapping distribution. Since hakes of the genus Merluccius can be 143 morphologically similar, sympatric substitutions could be due to an accidental mistake in 144 sorting onboard, if the species are caught in mixed fisheries, or at landings. In contrast, 145 substitutions by an allopatric or a non-Merluccius species are more probably deliberate. 146 147 Results were analysed by region of origin of the species (Europe, Africa, South Atlantic 148 America, North Atlantic America and Pacific America).

To test the risk of mislabelling in each species, only the subset of data corresponding to complete labels – to a species level- was employed. Rates on mislabelling along time were analysed according to the year of sample collection. Samples were grouped in four time intervals comprehending similar time spans (2002-2006, 2007-2010, 2011-2014 and 2015-2019), and analysed by regions. In the case of those articles that did not state the year, we assumed samples to have been collected the year before publication. Time intervals were optimized to include as many studies as possible. For this analysis, three studies carried out along a span of several years without individual sample dating were excluded: Borrell et al., (2016); Shehata et al., (2018); Stamatis et al., (2015).

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159 *2.3 Statistics*

The substitution status identified from DNA (number of samples containing: the 160 same genus and distribution as sympatric; same genus and different distribution as 161 allopatric; a different genus; mixed or ambiguous species; no substitutes) was compared 162 163 between species using principal component analysis (PCA) with correlation option. The relative contribution of each type of substitutions (no substitution or correct, sympatric, 164 165 allopatric, mixture, other genus - no Merluccius) to the variance of the metadata set was assessed from loadings. Components 1 and 2 were plotted to visualize the relationships 166 167 among variables and how different hake species were distributed according to their mislabelling. The different substitutions were plotted as diagonals with length proportional 168 169 to the respective weight in the analysis.

Contingency chi square tests were performed for every regional group of species to test for differences among periods in the proportion of mislabelled (versus correctly labelled) samples i.e. if there was a temporal change in mislabelling in any region. Statistics was performed with PAST free software (Hammer et al., 2001).

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175 **3. Results**

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177 *3.1 Data overview*

Using the key words indicated above we retrieved 45 articles spanning 2003-2020. In 178 179 these articles, 1291 commercial hake samples were analysed and provided 1452 individuals or pieces genetically ascertained. Samples were originated from a total of 22 countries, 180 although not evenly distributed, as most of the samples came from Spain (n=660), followed 181 by Italy (n=156), South Africa (n=91) and Portugal (n=73). The interest in hake mislabelling is 182 reflected in a significant increase of publications over time (linear trend y=0.0058x with 183 r=0.52, 18 d.f., p=0.026), and of commercial products analysed (y=0.0065x + 0.0066 with 184 r=0.67, 18 d.f., p=0.002), with a valley in 2013-2014 and further sustained growth (Fig. 1). 185 186 The search was done in July 2020, thus the real figure of 2020 could be higher.

187 Samples were identified at species level using either individual or a combination of different molecular markers: the mitochondrial 16SrDNA (used in 1.8% of products), 188 189 Cytochrome b (49.1%), cytochrome oxidase subunit 1 or COI (63.5%), and Control Region (5.3%); and/or the nuclear ITS1 (3.6%) and 5SrDNA (25.1%). Of all samples identified from 190 191 DNA, 1097 were labelled down to a *Merluccius* species level, while the rest were incomplete (i.e. "Hake" or "Merluccius spp") or incorrectly labelled as a different genus. Most samples 192 193 with species labels corresponded to M. merluccius (European hake) (n=207), M. bilinearis (Silver hake) (n= 172; 171 from a single study (Garcia-Vazquez et al. 2009)), and M. hubbsi 194 (Argentine hake) (n=172). South African *M. paradoxus* (Deep water Cape hake) and *M*. 195 capensis (Shallow water Cape hake) samples were abundant as well, although many were 196 ambiguously labelled with both scientific names together. The Pacific hakes M. productus 197 (North Pacific hake) and *M. gayi* (South Pacific hake) were underrepresented with only 55 198 199 and 39 samples, respectively.

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201 *3.2 Incomplete labels*

Considering all the compiled data, only 761 out of 1291 hake products were 202 203 completely labelled down to a single species level (either correct or incorrect from DNA analyses). Products with no clear specification of the fish group (n=28) or simply labelled as 204 205 "fish" (n=4) were identified as several Merluccius species (M. productus, M. bilinearis, M. paradoxus, M. merluccius, M. hubbsi, M. qayi). A total of 307 products exhibited labels at a 206 207 genus level, stated either as Merluccius spp. or as generic, common or vernacular names (i.e. "Merluza", "Bakalairos" (Triantafyllidis et al., 2010), or "European Hake"). Species 208 209 contributing to incomplete labels were principally M. paradoxus (31.57%), M. hubbsi 210 (17.74%), M. productus (7.37%) and M. capensis (6.45%). Other samples were ambiguously 211 labelled as mixes of different species: particularly, Cape hakes M. paradoxus and M. capensis were labelled as M. capensis/M. paradoxus in 182 products. From those, 88 212 213 products were genetically identified as one of the two species. This group was highly skewed towards *M. paradoxus*, which accounted for 87.5% of the samples identified from 214 215 DNA.

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217 *3.3 Mislabelling risk by species*

The different DNA-ascertained hakes found in this study had different risks of being used as a substitute species (Table 1). For this analysis, we considered only those products identified as a single *Merluccius* species by DNA analysis (n=1079 products), excluding those containing mixes of species (see Table 2).

In general, mislabelling risk varied among species (Fig. 2). Higher risk of being employed as a substitute was found for African species (only 22.52% of the products containing African species were correctly labelled) and American Pacific ones (45.07%

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correctly labelled) than for species native to other regions (Table 1). For example, more than
83% of the European *M. merluccius* were not substitutes of any other species. In contrast,
the highest risk of a species to be a substitute was found for *M. paradoxus*, for which only
9.2% of the products were correctly labelled (226 out of 249 products were incorrectly
labelled, the majority as mixtures of species or ambiguously labelled as *M. capensis/M. paradoxus*).

Merluccius angustimanus (Panama hake), M. polli (Benguela hake) and M. albidus 231 232 (offshore silver hake) were not reported on any label. However, both M. albidus and M. polli were found from DNA assignations as substitutes of sympatric species (20 products in total, 233 234 10 of each species; note the small sample sizes). M. albidus was a substitute of M. bilinearis 235 and M. polli of M. senegalensis. M. angustimanus appeared in two products labelled as 236 Gadus sp. (Cod). Since M. angustimanus and M. albidus were not found as substitutes of allopatric species (Table 1), we could infer that the particular mislabelling found in Panama 237 238 and offshore silver hakes is probably accidental.

In the case of sympatric species (see Fig. 2), the risk of substitution was 239 asymmetrical; in other words, directional mislabelling was found. In the pair of North 240 American hakes M. albidus/M. bilinearis, M. albidus was the substitute. In the triplet M. 241 merluccius/M. senegalensis/M. polli, M. polli was the substitute species. In the M. 242 capensis/M. paradoxus pair, the latter species was the main substitute. In sympatric South 243 American hakes, *M. hubbsi* was the main substitute of *M. australis* but not the other way 244 around (Fig. 2). On the other hand, substitution of allopatric species occurred for most 245 246 species, up to 23.1% in the case of *Merluccius australis* (Table 1).

Merluccius species also appeared as substitutes of other fish in a few samples (N=16; 247 Table 1). M. merluccius, M. paradoxus, M. hubbsi, M. gayi, M. productus and M. 248 angustimanus were sold as substitutes of cod (as seen above), G. chalcogrammus (Alaska 249 250 Pollock), Pleuronectes platessa (European plaice), Sarda (Tuna) and Solea solea (Common 251 sole). Reciprocally, 40 products labelled as hakes contained substitutes belonging to other genera of Gadiformes (Gadus chalcogrammus, Gadus morhua, Macruronus magellanicus, M. 252 253 novaezelandiae, Coryphaenoides acrolepis, Phycis phycis, Pollachius virens, and Urophycis tenuis), also Scombriformes (Thunnus sp., Katsuwonus pelamis), Perciformes (Dissostichus 254 255 eleginoides), Pleuronectiformes (Limanda aspera) and Siluriformes (Pangasianodon 256 hypophthalmus).

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The situation of each species regarding its use as a substitute of other species –which can endanger its conservation- can be seen in the PCA plot (Fig. 3). PC1 and PC2 explained more than 80% of the total variance (Table 3). Allopatric substitutions and mix/ambiguous labels had higher weights in PC1 and correct labels and non-*Merluccius* substitutions in PC2, indicating greater differences among species for these types of mislabelling than for sympatric substitutions. In Fig. 3, it is possible to observe that the European *M. merluccius* is 264 in quadrant I close to the diagonal representing correct labelling, while the Argentinean M. hubbsi is in the same quadrant but in the middle of non-Merluccius and allopatric 265 266 substitutions. *M. paradoxus*, together with *M. capensis* in quadrant IV, is clearly weighting for sympatric substitutions and mix/ambiguous labelling. Quadrant II contains M. bilinearis 267 268 and *M. gayi* that are substitutes of other *Merluccius* in none or very few products, being instead used in species mixes or ambiguously labelled products. The species under-269 270 represented in the collection of articles analysed are located together in quadrant III of the plot: M. angustimanus, M. albidus and M. polli. They share the plot with M. senegalensis, M. 271 productus and M. australis, all of them with intermediate levels of correct labelling and not 272 used in sympatric substitutions (Fig. 3). 273

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275 *3.4 Evolution of mislabelling rate*

276 A total of 1079 samples were included in this analysis after excluding samples that could not be assigned to a time period. As it can be seen in Fig. 4, commercial hake 277 278 mislabelling seems to be increasing at a global level over the last decade, being that increase 279 statistically significant (Table 3). Mislabelling decreased in 2011-2014 in comparison with 280 the 2007-2010 period (even more in South American hakes), but increased again in 2015-2019 (Fig. 4, Table 3). However, the evolution was not the same for all the species. 281 Significant temporal changes across the studied periods (2002-2006, 2007-2010, 2011-2014 282 and 2015-2019) were found for species native to all the regions except for Atlantic North 283 American hakes. It should be noted that Atlantic North American and Pacific samples were 284 unequally represented among time periods in this meta-analysis, with some periods in blank 285 (Table 3). 286

In the specific case of African species, the recent increase (2015-2019) of incorrect labelling (Table 3) included an increase of products not labelled down to species level, principally of the most exploited African hakes, *M. capensis* and *M. paradoxus*. However, even when the 98 products labelled as mixed cape hakes are taken out of the analysis, the increase in mislabelling rates is still significant (Chi-square = 25.909, *p* << 0.001).

In the case of South American and European species, the changes of mislabelling along the timeline were parallel with the mentioned valley for the period 2011-2014. Mislabelling was higher in South American hakes, with more than 40% found in 2007-2010, while in European *M. merluccius* it was near 20% (Table 3).

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297 **4. Discussion**

In this study we have detected several facts of importance regarding the accuracy of labelling in commercial hake. The first evidence is that, after a period of apparent and significant reduction of mislabelling in the period 2011-2014, it is increasing again in the last 301 years. This is a call of attention that indicates the need for a more strict control of marketed hake. The decrease of mislabelling by 2014 could perhaps be explained from a significant 302 303 number of articles followed by press releases between 2008 and 2012 that revealed high level of fraud in hake markets in the previous years (e.g. Triantafyllidis et al. 2010, Garcia-304 305 Vazquez et al. 2011, Cawthorn et al. 2012 and others). Mariani et al. (2014) found that media attention on the results of forensic analysis of marketed cod was followed by 306 307 significant improvement in cod labelling, and our results in hake would concur with theirs. In 308 addition, the appearance of strict EU legislation about seafood labelling in 2013 could also 309 contribute to explain the decrease of mislabelling around that date.

Mislabelling being a discrepancy between a commercial product and its label, indeed, 310 depends on the information that must be displayed on the label. However, labelling related 311 laws are heterogeneous, and vary from one country to another. The European Union has 312 common legal frames for seafood labelling with strict label requirements. European 313 Regulation (EU) 1379/2013 requires labels to provide the scientific name of the species as 314 315 well as a commercial denomination (common name). However, despite common regulation, there is a lack of harmonization and standardization across the EU and each country may 316 implement control measures differently (Griffiths et al., 2014). In the U.S., Food and Drug 317 Administration (FDA) establishes the obligation of identity labelling in food commodities 318 (Code of Federal Regulations CFR Title 21 Subchapter B Part 101 – Food Labelling, and Part 319 320 123 Fish and Fishery Products), with a common name that must appear in the list of recognized species with their scientific names. On the other hand, regulation is scarce in 321 322 some regions. For instance, many regulations do not require adding the species name but only a list of accepted species that fall under a wider term (i.e. "Hake" for all Merluccius 323 324 species; (Hofherr et al., 2016)). An important source of uncertainty found in this study comes from incompletely labelled products. While regulations vary widely between 325 326 countries, allowing for common names in the labels that may include several species, incomplete labels are present in countries (e.g. Italy and Spain) where the scientific name of 327 328 the species is required by current regulations.

329 Within the Merluccius genus, we found that some North Atlantic species (European and Silver hakes) are relatively rare as substitute species, while African species and North Pacific 330 hake have much higher risk of appearing as substitutes of other species or be ambiguously 331 labelled. Reasons for high risk of mislabelling are varied: accidental misclassification of 332 species that are fished together; deliberate fraud for purposes of catching over quota; 333 obtaining higher economic benefit when the label species is more expensive than the 334 335 substitute; exploiting protected species, and more (e.g. Donlan & Luque, 2019; Muñoz-Colmenero et al., 2015). In the market, European hake is generally more expensive than the 336 337 African species (Muñoz-Colmenero et al. 2015), thus its lower use as a substitute can be easily explained by economic reasons. M. bilinearis has been sold occasionally as a 338 339 substitute of the more expensive M. merluccius (Sánchez et al., 2009), but given its smaller 340 size (commonly 37 cm in average versus 45 cm of *M. merluccius*, 50 of *M. capensis* or 80 of 341 *M. australis*; Cohen et al. 1990) it is not the best substitute of larger and more appreciated 342 species.

Since most samples were purchased in Europe, the conclusions of this meta-analysis 343 should not be taken as universal. Mislabelling may differ greatly in other regions that have 344 been less studied, thus mislabelling in the species marketed there may be overlooked. This 345 would be the case for Pacific hake species that are underrepresented in this meta-analysis in 346 347 comparison to catch reports. For example, M. productus catch was 299270 tonnes between 2014 and 2017 (FAO, 2020a). Only 47 products of this species are represented in this review; 348 in contrast, for the same period we found 172 samples of M. merluccius analysed 349 forensically while the catch of this species was 104180 tonnes, less than one half of M. 350 productus catch. Increased efforts in DNA analysis of Pacific species would be 351 352 recommended.

The prevalent use of incomplete labels hinders sustainability of affected fisheries 353 (Cawthorn et al., 2012). This is well documented for Cape hakes, which are usually managed 354 as a single stock (FAO, 2011; Wilhelm et al., 2015). Typically, M. paradoxus has been 355 described as the predominant landings in the eastern and south coast, and M. capensis in 356 the west coast of South Africa (FAO, 2011). However, this is not supported by the data, as 357 there is an unbalance in the occurrence of both species in products identified from DNA that 358 could not be explained from the reported catches. Managing two species together without 359 accounting for their differences in biology and ecology may lead to an overestimation of the 360 available stock (Kathena et al., 2016). This is likely to occur for all overlapping species and 361 362 has been reported for other taxa where species that are morphologically similar are caught together (Crego-prieto et al., 2010; Iglésias et al., 2010). In particular, special focus should 363 be put on hakes whose distributions largely overlap, like silver hakes in the west North 364 Atlantic and all the African hakes. Moreover, there was no record of products labelled as M. 365 polli, M. angustimanus or M. albidus although we found those species from DNA analyses. 366 Special efforts should be made to cover these species. 367

In our meta-analysis we focused on hake products marketed for human consumption. 368 However, the presence of hakes in products destined to other uses may go unnoticed. For 369 example, forensic studies on fish meals are scarce, although the content of the pellets is 370 371 frequently not disclosed and may hide the use of overexploited or endangered species (A 372 Ardura et al., 2012; Galal-Khallaf, Osman, et al., 2016; Martín et al., 2010; Pegels et al., 373 2013; Prado et al., 2012; Vlachavas et al., 2019). Thus, we would encourage the analysis of 374 fishmeal pellets DNA using specific primers to detect hake species. On the other hand, other 375 types of products may contain hake, like those based on gelatin (e.g. candies and other foodstuffs); Muñoz-Colmenero et al. (2016) found traces of different hakes in marshmallows 376 377 and jelly gummies. Expanding forensic analysis of these and other commodities would also be recommended for detection of commercial niches where hake is actually employed, 378 declared or not. 379

380 Correct fisheries management is essential for the sustainability of the stocks. This includes detecting IUUs, as can be done from forensic analysis of commercial products 381 382 (Ogden, 2008). Signs of improvement reported for some hake fisheries by 2014 (e.g. M. merluccius and M. hubbsi; (Antelo et al., 2012; Lorenzo & Defeo, 2015), coinciding with 383 384 lower mislabelling detected in our analysis, do not seem to be maintained for the later years (2015-2019). Precisely in these years mislabelling increased again in Africa, Europe and 385 386 South Atlantic America (Fig. 4). Indeed, this is just an observation and cause-effect cannot be inferred from it, but a continuous monitoring of the label accuracy is called for in order 387 to avoid irreparable declines in stock resources. The conservation status of Merluccius 388 species is diverse, as their fisheries are. M. senegalensis is considered as endangered species 389 (Iwamoto, 2015c), while M. merluccius is vulnerable (Di Natale et al., 2011), and M. 390 bilinearis is near threatened (Carpenter, 2015). In the case of M. gayi, the data is deficient 391 (Iwamoto, T., Eschmeyer, W., Alvarado, J., Bussing, 2010a) to evaluate its status. The status 392 of M. australis, M. hubbsi and M. paradoxus are not evaluated yet, and the rest are of 393 394 species are considered of least concern (M. angustimanus (Iwamoto et al., 2010), M. productus (Iwamoto, T., Eschmeyer, W., Alvarado, J., Bussing, 2010b), M. capensis 395 (Iwamoto, 2015a), M. polli (Iwamoto, 2015b) and M. albidus (McEachran & Polanco 396 Fernandez, A Russell, 2015). At least the species catalogued as endangered, vulnerable and 397 near threatened, i.e. M. senegalensis, M. merluccius and M. bilinearis, should be targets of 398 399 specific campaigns for forensic control of mislabelling.

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401 **5. Conclusions**

After a decline in mislabelling rates from 2011 to 2014, our data shows a new rise in 402 403 recent years. All Merluccius species are not equally affected by mislabelling; species from Africa and from the Pacific seem to be used as substitutes more frequently. Furthermore, 404 405 special attention must be given to incomplete labelling, which is a great source of uncertainty, masking mislabelling between sympatric species and hindering correct 406 407 management of stocks. Notoriously, this is reflected for Cape hakes, where DNA identifications found deep Cape hake to be the predominant species in commercial 408 409 products, despite this not being reflected in catches reports. More research is needed for 410 other species, particularly; Pacific hakes appear underrepresented, as well as species with 411 lower commercial interest, i.e. M. polli. Ensuring the correct labelling of products helps the detection of IUUs, hence stricter labels and monitoring should be implemented with special 412 413 attention to threatened species. There is an urgent need for an international harmonization 414 in seafood labelling, in order to have a better control of mislabelling to ensure consumer 415 rights and fisheries sustainability worldwide.

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417 **CRediT authorship contribution statement**

Carmen Blanco-Fernandez: Conceptualization, Methodology, Data curation, Formal
 analysis, Writing - original draft. Eva Garcia-Vazquez: Conceptualization, Formal analysis,
 Writing - review & editing, Supervision, Funding acquisition. Gonzalo Machado-Schiaffino:
 Conceptualization, Methodology, Writing - review & editing, Supervision, Funding
 acquisition.

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424 **Declaration of competing interest**

- 425 The authors declare that there is no conflict of interest.
- 426

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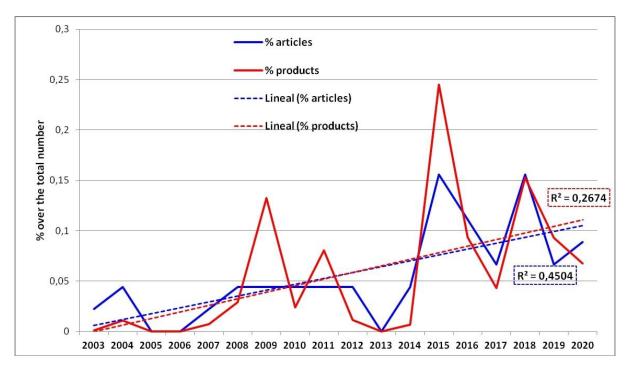


Fig. 1. Evolution of publications about hake mislabelling in the two last decades, presented as the proportion of the total number of articles published and products analysed between 2003 and 2020 by year.

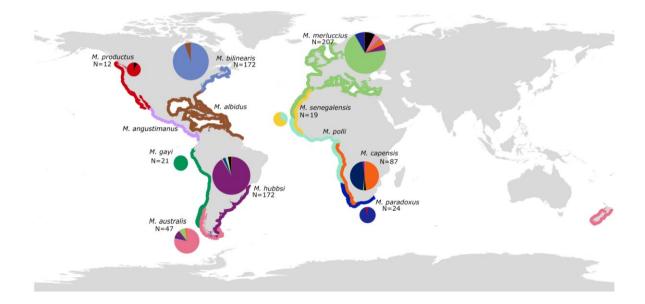


Fig. 2. Meta-analysis of mislabelling in individual commercial hake samples (total N = 761) identified from DNA. Pie charts represent the proportion of each DNA-authenticated species marketed under the same species name. Colour codes of DNA-identified species: *Merluccius*

albidus (brown), M. angustimanus (light purple), M. australis (pink), M. bilinearis (light blue), M. capensis (orange), M. gayi dark (green), M. hubbsi (dark purple), M. merluccius (light green), M. paradoxus (dark blue), M. polli (blue green), M. productus (red). The distribution of each species is shaded with the same colour codes.

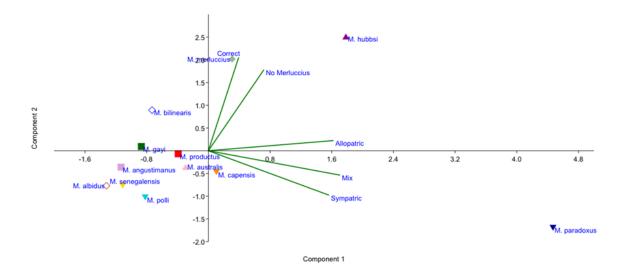


Fig. 3. PCA scatter plot showing the different *Merluccius* species in relation to the five types of substitution considered (no substitution or correct labelling, substitutes of sympatric, or allopatric species, substitutes of species of another genus or no *Merluccius*, ambiguously labelled as a species mixture or "mix"). Diagonals are proportional to the relative weight of each type of substitution in the analysis.

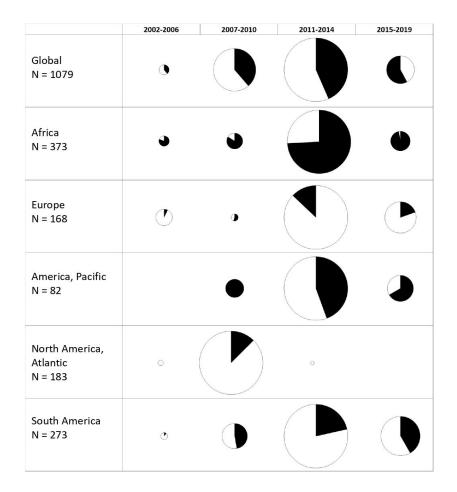


Fig. 4. Evolution of mislabelling by geographic region over the last 17 years. White represents correct labels and black is mislabelling. Pie sizes are proportional to the sample sizes. Mislabelling includes incomplete labels and labels indicating different species.

–Table 1. Hake mislabelling by species, presented as % of mislabelling in samples ascertained from DNA. Categories for each species (as determined by DNA analyses) include different types of labelling: hake samples which were mislabelled outside of *Merluccius* genus, samples mislabelled as an allopatric *Merluccius* species, samples mislabelled as a sympatric species, samples with a label which does not indicate species, and correctly labelled samples. N corresponds to the total amount of sample products which have been identified by DNA as each *Merluccius* species. Species' regional distribution is provided: Africa, Europe, Atlantic North America, Pacific North America and South America; as well as the total mislabelling per region counting all species of the region (in bold). Allopatric *Merluccius*: no overlapping distribution, versus a sympatric *Merluccius* species. Raw data can be found in the repository table (Blanco Fernandez et al., 2020).

		Incorrec				
DNA identified species	A different genus (%)	Other <i>Merluccius</i> (no overlapping distribution)	Other overlapping <i>Merluccius</i> species	Label does not contain a single species name	Correctly labelled	N (sample size)
Africa	0.53	9.60	12.27	57.07	20.53	375
M. senegalensis	0.00	4.17	0.00	41.67	54.17	24
M. polli	0.00	30.00	60.00	10.00	0.00	10
M. capensis	0.00	12.64	0.00	45.98	47.13	92
M. paradoxus	0.80	8.43	16.06	65.46	9.24	249
Europe	2.91	4.65	0.00	9.30	83.14	172
M. merluccius	2.91	4.65	0.00	9.30	83.14	172
America, Pacific	9.76	3.66	0.00	47.56	39.02	82
M. angustimanus	100.00	0.00	0.00	0.00	0.00	2
M. productos	6.38	6.38	0.00	63.83	23.40	47
M. gayi	9.09	0.00	0.00	27.27	63.64	33
North America, Atlantic	0.00	1.09	5.46	4.92	88.52	183
M. albidus	0.00	0.00	100.00	0.00	0.00	10
M. bilinearis	0.00	1.16	0.00	5.20	93.64	173
South America, Atlantic	2.46	9.82	1.40	18.60	67.72	285
M. australis	0.00	23.08	0.00	5.77	71.15	52
M. hubbsi	3.00	6.87	1.72	21.46	66.95	233

Table 2. Principal component (PC1, PC2 and PC3) Eigenvalue, % of the total variance and loadings.
The substitution types most contributing to each component are marked in bold.

	PC 1	PC 2	PC 3
Eigenvalue	2.7	1.52	0.49
% variance	54.1	30.5	9.9
No Merluccius	0.244	0.604	-0.754
Allopatric	0.551	0.076	0.252
Sympatric	0.532	-0.332	-0.022
Mix	0.58	-0.183	-0.041
Correct	0.133	0.697	0.605

Table 3. Evolution of mislabelling by geographic region over the last 17 years. Mislabelling is given in percentage (%) over the total number of samples analysed each period (N), and includes incomplete labels and labels indicating different species. Contingency Chi-square value and its associated p is given. Significant p-values are in italics.

		2002-2006	2007-2010	2011-2014	2015-2019	Chi square	<i>p</i> -value
Africa	Ν	47	62	192	72	23.38	3.37E-05
	%	80.85	83.87	70.83	97.22		
Europe	Ν	28	9	85	46	13.09	0.004
	%	7.14	55.56	12.94	19.57		
America (Pacific)	Ν	0	16	45	21	15.69	0.0004
	%	-	100	44.44	66.67		
North America (Atlantic)	Ν	14	168	1	0	2.12	0.35
	%	0	12.5	0	-		
South America	Ν	18	55	121	79	19.16	0.0002
	%	11.11	47.27	21.49	41.77		
GLOBAL	Ν	107	310	444	218	21.19	9.60E-05
	%	39.25	38.71	43.47	57.80		