Valorization of recurrently discarded fish species in trawler fisheries in North-West Spain

Blanco, M. ¹, Domínguez-Timón, F. ¹, Pérez-Martín, R.I. ², Fraguas, J. ², Ramos-Ariza, P. ², Vázquez, J.A. ² and Borderías, A.J. ¹, Moreno, H.M. ¹*

²Marine Research Institute (IIM). Spanish National Research Council (CSIC). C/ Eduardo Cabello 6, 36208 Vigo, Pontevedra, Spain

*Corresponding author: hmoreno@ictan.csic.es

Abstract

The progressive elimination of fish discards established by the European Union Council in 2013 has stimulated the valorization of flesh from discarded high-quality species with good protein functional properties but which frequently have excessive fish-bones, fat, strange flavours, soft texture, etc. The present study therefore focuses on valorization of the extracted muscle (minced muscle), from several fish species frequently discarded in north-western Spanish fisheries (Atlantic Ocean): Blue whiting (Micromesistius poutassou), Mackerel (Scomber scombrus), Red scorpionfish (Scorpaena scrofa), Pouting (Trisoreptus luscus) and Gurnard (Trigla spp.). Valorization of these discarded fish resources is a key objective for the survival of the fishery sector in this area. In this regard two clear objectives have been defined for this study: on the one hand, to examine the behaviour of the mince during 6 months of frozen storage by means of physicochemical and sensory analyses, and on the other, to test consumer acceptance of
three technologically different products (burgers, nuggets and structured fingers) prepared with fish mince from different species. Results indicate that protein aggregation started at the outset of frozen storage but progressed very slowly, with the exception of non-washed blue whiting and red scorpionfish minces. Moreover, during frozen storage lipid oxidation increased slightly in all samples; the increase was highest in minced mackerel, a fatty fish, but no rancid flavour was detected. All mince samples presented acceptable physicochemical properties and good sensory acceptability after six months of frozen storage. Acceptability of final products made with these minces was high in all cases. Burgers were more acceptable for consumers aged over 40 and fingers and nuggets more for younger people.

**Keywords:** Landing obligation, fish discards, fish mince, fish muscle valorization, frozen storage.
Introduction

In 2013 EU Regulation No 1380/2013 established as an objective the progressive elimination of fish discards by introducing the obligation to land (LO: landing obligation) all specimens of all species under TAC regulations (TAC: total allowance captures). When necessary, the new Common Fishery Policy also aims to make the best use of unwanted catches. There are three main reasons for discarding: (a) fish species of low commercial value, (b) fish under minimum conservation reference sizes and (c) non-quota fish species; however, most of those discarded specimens possess significant amounts of bioactive compounds besides good nutritional value, containing high quality proteins, fats or minerals among others (García-Moreno et al. 2013; Blanco et al. 2015; Vazquez et al. 2017). In this context, one of the most challenging aspects of the LO is therefore the need to develop different alternatives for valorization of those discarded species, which must now be kept on board, with the ultimate aim of maximizing the return on fishing captures, alleviating the costs imposed by the new regulation, and contributing to long-term environmental, economic and social sustainability.

Together with the most commercially valuable species captured by Galician fishing fleets (North-West of Spain) (hake, sole, monkfish, megrim, etc.), other non-target species are unintentionally caught and are mostly discarded (Ordóñez-Del Pazo et al. 2014; Blanco 2015). In view of the level of interest shown by fishermen and fish producer organizations, together with their known high nutritional value, the present study focuses on some habitually discarded species such as Blue whiting (Micromesistius poutassou), Mackerel (Scomber scombrus), Red scorpionfish (Scorpaena scrofa), Pouting (Trisoreptus luscus) and Gurnard (Trigla spp.). Another key factor in selecting these species was their capture stability throughout the year.
The valorization of fish muscle proteins from high-quality discarded species has been studied, and these proteins are considered suitable for the preparation of different seafood products with enhanced added value in comparison to the traditional preparation of fish meals or fish oils (www.valdescar.com; Moreno et al. 2010a,b; Moreno et al. 2009). However, this is the first study analysing the preparation of different ready-to-cook products from muscle of the above mentioned species. For this purpose, fish muscle was first mechanically extracted, followed or not by fish mince washing depending on the types of products to be prepared; finally, natural additives (cryoprotectant and antioxidant) were added, and samples were frozen to produce a new raw material suitable for use in preparing different seafood products with different textures and flavours.

The aim of the present study was then to investigate frozen stability, up to 6 months at -20°C, of minces from those 5 fish species discarded in the Galician coastal trawler fishery, and also to obtain a consumer evaluation of three types of products, chosen as examples of different manufacturing procedures.

Materials and Methods

Raw material

The selected fish species, Blue whiting (*Micromesistius poutassou*), Mackerel (*Scomber scombrus*), Red scorpionfish (*Scorpaena scrofa*), Pouting (*Trisoreptus luscus*) and Gurnard (*Trigla spp.*), were captured by a commercial trawler during a one-week trip, operating off the Northwest coast of the Iberian Peninsula (areas VIIIc and IXa ICES) in September 2016. Immediately all the catch was on board, 100 kg of each selected species was separated from commercial ones and distributed in 12 kg iced boxes. Once
in port, all the fish was landed and transferred to the pilot plant where it was kept on ice until further processing.

**Fish processing**

Fish were gutted and headed by hand within 24 hours of landing. Afterwards, fish meat was mechanically separated from bones and skin (Baader 694, Germany). Then the minced muscle from each species was divided into two batches. One was mixed with cryoprotectant (4% sorbitol, Panreac Química SLU) and antioxidant (400ppm, Antiox 1 Tocopherol, Altaquímica, S.A.), placed in a freezer-tray and frozen in a plate freezer at -20 ºC. The other batch was washed with iced water (1:5 p/v) and slowly shaken for 10 minutes. Then, the fat was removed from the surface, and the mince was separated from the water using a rotary sieve. At this point the moisture content of the meat had increased from about 76-79 % to 90-92 %; it was therefore essential to remove the excess of water with a screw press (Baader 523, Germany) up to around 79-82 % depending on the species (Table 1). Finally, a kneader was used to add the sorbitol and antioxidant to the mince. As in the unwashed batch, the mince was placed in a tray and frozen in a plate-freezer (-20 ºC). Frozen mince blocks (two for each species) were placed in plastic bags and stored in cold rooms at -20 ºC.

**Physicochemical and sensory analyses of fish mince**

Mince from the ten frozen blocks was subjected to physicochemical and sensory analyses and also used as raw material to prepare the final restructured products.

*Proximate analyses and muscle yield.*

Ash, fat, crude protein and moisture contents of washed and unwashed mince from all species were determined in quadruplicate (AOAC 2000). Crude protein content was
measured with a LECO FP-2000 Nitrogen Determinator (Leco Corporation, St Joseph, MI, USA). Muscle yield was determined as the percentage of muscle obtained from the headed and gutted fish.

SDS-PAGE in polyacrylamide/agarose gels: Analysis of myosin cross-linking

Myosin heavy chain cross-linking was studied by observing the decrease in the MHC monomer and the increase in MHC polymer contents, separated on agarose/polyacrylamide gel (Konno and Imamura, 2000). Fish mince was analysed at the beginning of the study and after six months of frozen storage (-20°C).

Differential scanning calorimetry (DSC).

Thermal behaviour of the myosin from different fish minces was monitored using a differential scanning calorimeter (DSC Q1000, TA Instruments, New Castle, USA). The temperature (°C) and transition (ΔH-J/gdm-) enthalpies were determined for each fish mince at the beginning of the study and after 6 months of frozen storage.

Apparent viscosity ($\mu_{app}$)

The apparent viscosity, as indicative of protein functional quality, of all fish minces was determined according to Borderías et al. (1985) using a solution of muscle in a 5% NaCl solution (1:5 p/v). Measurements were carried out in triplicate at the beginning of the study and after 3 and 6 months of frozen storage. The results were expressed in centipoises (cP).

Cooking Loss (CL)
Cooking loss was performed as described by Moreno et al. (2008). CL was expressed as percent of water released per 100 g of sample.

Lipids oxidation

Thiobarbituric acid reactive substances (TBARS) were determined at the beginning of the study and after 3 and 6 months of frozen storage, in order to evaluate lipid oxidation (Vyncke, 1975). The results were expressed as mg of malonaldehyde equivalents per kg of sample (mg MDA/kg muscle).

Sensory analysis

Fish Mince

The appearance, taste and texture of the fish minces were analysed by 10 trained panellists in a standard sensory panel room using a non-structured 10-point scale. Samples were previously thawed, cut into portions 0.8 cm thick and grilled for 2 minutes before sensory analysis.

Consumer test: final restructured products

The test consisted of 40 adult frequent seafood consumers. Consumers were asked to judge the general acceptability of the products by assigning scores on a 5-point scale to each product. The analysis was segmented according to sex and age. Three different types of products were made with the fish mince. Depending on the final product, different minces were used to achieve the best physicochemical and sensory properties. Various formulations were tested (results not shown), but only the most appropriate are included in the results section. Test products were prepared as follows:
Burgers: These were prepared using unwashed fish mince and washed Mackerel mince. The suggested formula was: mince (84.9%), Alaska Pollock Surimi grade KA (10%), modified starch (4%), salt (0.8%) and garlic powder (0.3%). The mince was gently blended in a blending machine with the rest of the components to preserve mince particle structure. The burgers were shaped in a manual burger machine. The burgers (~50 g) were frozen and stored at -40ºC until use for the consumer evaluation test, when they were cooked on a grill.

Nuggets: These were prepared using washed minces of Red scorpion, Blue whiting and Pouting (all lean species whose muscle has better gel-forming ability). The formula was: 80% mince, 15.2% surimi (97% surimi+3% salt), 4% modified starch and 0.8% total dough. The surimi was homogenized in a vacuum cutter (Stephan UMC 5, Stephan Machinery, Germany) at chilled temperature for 10 minutes with 3% salt. Then the rest of the ingredients were added and the whole homogenized for 5 minutes more. After gelation, cubiform portions of around 3 cm were battered and pre-fried at 180ºC for 20 seconds to fix the batter. The nuggets were then frozen at -40ºC.

Structured Fingers: These were prepared to imitate the natural structure of fish muscle, which is organized in flakes (imitation of myotomes and myosepta) (CSIC, 2007). Two formulae were required to make this product: for myotome (A): 85.2% mince, 10% surimi (97% surimi +3% NaCl), 4% modified starch, 0.8% salt; and for myosepta (B): 11.62% sodium caseinate, 1.5% CaCO3, 0.2% fish gelatin (Juncá S.A., Girona, Spain), 3% Microbial Transglutaminase Activa WM (Ajinomoto Co.; Tokyo, Japan) and 83.68% water. Slabs of around 1 cm were formed after homogenizing. A thin layer of formula (B) was spread on the formula (A) slabs. “Sandwiches” were formed from four
slabs of formula (A) with the corresponding layers of formula (B) in between. Lastly, pieces 1 cm thick were transversally cut from the slabs. These portions (like broad fingers) were battered and pre-fried at 180 °C for 20 seconds, then frozen at -40°C.

**Statistical Analysis**

One-way analysis of variance was carried out with the SPSS® computer programme (SPSS Inc., Chicago, IL, USA), and differences between pairs of means were evaluated by the Tukey test using a 95% confidence interval.

**Results and Discussion**

**Proximate analyses and muscle yield**

Table 1 shows the proximate composition of the fish mince and the muscle yield after mechanical extraction. As observed, proximate composition profiles were very similar in all samples, the sole exception being mackerel, with a larger amount of lipids (p<0.05) and not considered a lean species. The moisture content of washed mince was higher than unwashed. It was decided to maintain the water content between 74 and 82 %, which is similar to the natural content of fish muscle.

In general, lipid content was lower in washed than in unwashed minces (p<0.05). This result is consistent with the process considering that the loss of fat is a consequence of the washing process, as fat floats on the water and is removed with the washing water (Park et al., 2013). The protein fraction was proportionally lower in washed than in unwashed mince (p<0.05). This result could be explained firstly by leaching-out of most of the soluble proteins (lower molecular weight) during the washing step, and secondly
due to the fact that the amount of water was greater in washed minces and so the percentage of protein was relatively smaller (Park et al., 2013).

Modification of protein structure during the process

*SDS-PAGE in polyacrylamide/agarose gels: Analysis of myosin cross-linking.*

As shown in the SDS-PAGE profile (figure 1), the MHC (Myosin high chain) band is denser in washed mince due to the fact that this fraction is proportionally greater with respect to the total protein. This could be due to the loss of other protein fractions of lower molecular weight (more soluble) in the washing step (Park et al., 2013). Washed mince showed more bands in the higher molecular weight area of electrophoretic gels (>250 kDa), which could be molecules of polymerized or aggregated myosin (Núñez-Flores et al. 2018). This may also be related to the washing process, which could affect the protein muscle structure.

Comparison of the electrophoretic profiles of fish minces just after freezing (day 0) (a) and after 6 months of frozen storage (b) showed more bands with molecular weight over 250 kDa in the latter, indicating some polymerization or aggregation (Konno and Imamura 2000, Núñez-Flores et al. 2018). Also, after 6 months of frozen storage the MHC band appeared slightly denser than at day 0, indicating that native myosin had probably been slightly aggregated as a consequence of frozen storage.

*Differential scanning calorimetry (DSC)*

DSC analysis was used to determine preservation of the protein native structure. In general, the denaturation enthalpy was higher in washed samples than in unwashed ones.
(Table 2), indicating some aggregation due to the washing process, as also observed in the SDS-PAGE profile.

After 3 months of frozen storage, denaturation enthalpy tended to increase, most probably as a result of protein aggregation and/or polymerization during frozen storage (Schubring 2009). After 6 months of frozen storage the enthalpy had increased in most of the minces, though the increment between month 3 and 6 was smaller than the increment between months 1 and 3. This means that the aggregation started just at the beginning of the frozen storage period (Huidobro and Mohamed 1998). The enthalpy maxima recorded in these fish minces was similar to that found in good-quality Alaska Pollock surimi (Grade KA) (Núñez-Flores et al. 2018), meaning that the protein was not extensively denatured in any sample.

**Functional properties of minces**

**Cooking loss**

Table 3 shows data from the cooking loss analysis. Cooking loss was significantly lower at the beginning (month 0) than after frozen storage (months 3 and 6). The increase was particularly evident after 3 months of frozen storage, after which it remained stable. During frozen storage myofibrillar proteins, mainly myosin, become aggregated, leading to changes in their structure. As a consequence, some water bound to the proteins is exuded, an effect which could be maximized by extreme temperatures (Damodaran and Paraf 1997). Also, cooking loss was higher in washed than in unwashed minces. The washing process removes undesirable water-soluble materials, such as fats, inorganic salts and some proteins, and at the same time proteins bonded to
water molecules (Park et al., 2013). These water molecules are weakly linked to
proteins, which are easily lost during cooking.

Apparent viscosity ($\mu_{\text{app}}$)

Protein functional quality was measured by apparent viscosity (Barroso et al. 1998). As
shown in Table 4, washed minces exhibited higher apparent viscosity than unwashed
minces, indicating very good functional properties of the protein present in the muscle
of those species. Most of the minces registered higher apparent viscosity at the
beginning of frozen storage, which could be due to the lack of aggregation as reported
in the cooking loss section. However, after six months of frozen storage viscosity was
still high, with the sole exceptions of unwashed Blue Whiting and Red Scorpionfish
mince. Blue Whiting belongs to the Gadidae family, whose muscles produce
formaldehyde and DMA from trimethylamine oxide (TMAO) during frozen storage,
inducing protein aggregation and hence a reduction in protein functionality and
viscosity (Rey-Mansilla et al. 1999). Although Red Scorpionfish does not belong to the
Gadidae family, it is probable that its muscle also contains some protein-denaturant
substance. This effect was not observed in washed Blue Whiting or Red Scorpionfish
mince, since TMAO is leached out during the washing process.

Lipid oxidation during frozen storage

Table 4 shows lipid oxidation as measured by TBA index (mg of MDA/kg of muscle).
As observed, the TBA index was lower in washed than in unwashed mince. This could
be due to the removal of part of the lipid content floating in the water during the
washing process, as reported by Park et al. (2013).
After six months of frozen storage, lipid oxidation increased in all minces, particularly in mackerel, which is a fatty fish. The concentration of TBARS in good quality frozen fish is typically between 5 and 8 mg MDA/kg, whereas levels of 8 mg MDA/kg are generally regarded as the limit of acceptability for most species (Schormüller 1968). The values of the present samples (Table 4) are below those limits, indicating good quality of the frozen fish. Moreover, Baron et al. (2007) reported that lipid hydroperoxides and secondary oxidation products (volatiles) increased dramatically after 13 months of storage at −20 °C. Nazemroaya et al. (2011) reported similar results, suggesting that lipid oxidation was the main reason for the short frozen storage life of fish.

**Sensory analysis**

*Fish mince.*

The scores in the sensory analysis for the different parameters studied are shown in Table 5. Although all muscles scored well for acceptability at the beginning of frozen storage, washed muscles scored lower than unwashed. Moreover, the highest scores were awarded to Blue whiting, Mackerel and Gurnard, both washed and unwashed. After six months of frozen storage scores were slightly lower, but all samples still scored around the middle of the scale for the three sensory characteristics analysed (general appearance, texture and taste).

**Consumers test: final restructured products**

Based on the variation in protein structure and functionality, and also lipid oxidation during frozen storage, several products were prepared following the methodology
described in sections 4.1 to 4.3. For each final product, different fish minces were used so as to obtain products with desired sensory properties.

“Burgers” were made using unwashed mince from all species to preserve a pronounced fishy flavour. Minces from washed Blue whiting, Pouting and Red scorpionfish were used to make the “nuggets” and “structured fingers”. These two products are very well accepted by young people, who tend to like the white colour of the meat. The methodology for manufacture of these products entailed protein gelation, so that washed minces were more suitable given their better protein functionality and whiter colour.

The highest scores (between 4.5 and 5) were awarded to “fingers” of Pouting and Red scorpionfish, and above all Mackerel “burgers”. Consumers aged between 20 and 40 preferred the structured “fingers” (4 and 4.5 points), and the over-40s liked the Mackerel “burgers” best. Consumers aged from 45 to 70 selected Mackerel “burger” and the structured “fingers” as the best, followed by Red scorpionfish “nuggets”. This last fact is striking given that nuggets are a kind of food specially designed for children.

Conclusions

The various target species are abundant and are seasonally regular enough to be a reliable source of by-catch in coastal Galician trawler fisheries for the industry to make marketable fish products. Some of these species, which are too bony or have a strong taste due to the abundance of fat, are not highly appreciated by consumers. However, it has been concluded that they are suitable for processing in the form of mince to make derivative products. The proposed technology is simple, entailing washing of the mince (optional), further mechanical draining followed by kneading or homogenizing to
incorporate the cryoprotectant and the natural antioxidant, and finally freezing. All the
minces performed well over 6 months of frozen storage. This was the limit for protein
functionality in the case of unwashed Blue Whiting and Red Scorpionfish mince, and
also in the case of Mackerel because of lipid oxidation. The three products, made with
different textures and appearances, were well accepted by consumers. It is worth noting
that products with strong flavours, like mackerel, were more acceptable to older people.
Moreover, older people preferred well-known products like burgers and younger people
preferred products such as fingers and nuggets.

Acknowledgments Financial support from project VALDESCAR (Fondo Europeo
Marítimo y Pesca; Secretaría General de Pesca (MAPAMA)) is acknowledged. The
assistance of PhD. Ruth Núñez-Flores is also appreciated.

References
AOAC (2000.) Official methods of analysis of the AOAC. Association of Official
Analytical Chemists Inc, Arlington
Baron CP, Kjaersgard IVH, Jessen F, Jacobsen C (2007). Protein and lipid oxidation
during frozen storage of rainbow trout (Oncorhynchus mykiss). J Agric Food
Blanco, M. 2015. Valorización de descartes y subproductos de pintarroja (Scyliorhinus


Moreno HM, Carballo J, Borderías AJ (2009) Study of two different cold restructuring processes using two different qualities of hake (Merluccius capensis) muscle,
https://doi.org/10.1002/jsfa.3592


