New Alternatives in Seafood Restructured Products

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A general overview, focusing on new trends in the different techniques used in restructured seafood product processing has been described in this work. Heat induced gelation has been more widely studied in scientific literature than cold gelation technology. This latter technology includes the use of hydrocolloids (alginates and glucomannan) or enzymes (microbial transglutaminase) for making both raw and cooked restructured products. In restructuration processes, fortification processing with some functional ingredients is studied, giving as a result extra value to the products as well as increasing the variety of new seafood products. The process of alleviating heavy metals and organic pollutants from the raw material used has also been studied in the present paper.

Keywords: restructured seafood, cold gelation, microbial transglutaminase, sodium alginate, glucomannan, dietary fibre, fatty acids, pollutant alleviation.

INTRODUCTION

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Restructured seafood products are foods made from minced and/or chopped muscle, which are used with or without additional ingredients or additives, to make other products with either a new appearance or texture or both. For some time now, there have been products on the market made from under-valued fish species or by-products in the form of fish fingers or other shapes, designed basically to attract children. Seafood products are also being elaborated by different restructuration techniques to mimic other more highly-valued products. The main reason for restructuring fish muscle has been due to the limitations on high value seafood product supplies and, therefore, the necessity to make the best use of existing resources. Many of these resources are under-utilized species that would otherwise have commercial problems because of their size, composition, bony structure, unattractive appearance or texture. Additionally, fish by-products (muscle trimmings, sawdust from frozen fish sawing, etc.), from regular processing of more expensive species, can be used as raw material for the elaboration of restructured seafood products (Borderías and Pérez-Mateos, 1996; Ramirez et al., 2011).

Although there are many restructured products on the market made with chopped muscle pieces or mince, most of them, especially the seafood analogues, are elaborated from *surimi*. This is a stabilized myofibrillar protein paste, obtained after washing and refining the mince, which has good long-term frozen storage stability and excellent gel-forming ability. When fish mince or *surimi* is used, it is frequently necessary to create different types of structures in order to endow the final product with better textural properties. For that purpose, gelation is induced to develop myotome-like fibres (CSIC, 2006) or other structures. Restructured products can also be very good carriers for functional ingredients, since there are fewer technical restrictions compared with inclusion via, for example, dietary modulation in fish muscle.

The aim of this paper is to describe the current technology used in the elaboration of restructured seafood products, which includes heat-induced gelation, cold gelation adding different ingredients, new possibilities for adding functional ingredients and contaminant alleviation of raw material. As there is a lot of published information about heat-induced gelation in the elaboration of restructured products, this review will only touch on this subject lightly.

HEAT-INDUCED GELATION

This is the most common procedure used in the industry to process restructured seafood products, from both *surimi* and minced muscle. Heat gelation involves the gelation of myofibrillar proteins, mostly myosin. It happens after the addition of enough salt permits the myofibrillar proteins to unfold, thereby inducing the exposure of their reactive surfaces, and thus facilitating interactions, so intermolecular bonds are formed. When these kinds of bonds are sufficient in number a three-dimensional network is formed resulting in a gel. Different types of bonds take place to form this network, such as hydrogen bonds, ionic interactions, hydrophobic interactions and covalent bonds (Lanier et al., 2005). It is important to note that hydrophobic interactions and covalent bonds are the interactions responsible for gel thermostability, an important characteristic to be considered for food products that will be cooked.

If after unfolding by salting, the dough is kept at a low temperature (0 to 50°C), myofibrillar proteins can form a softer and more deformable gel, named *suwari* gel; this process is termed *setting*. This type of gel network is formed as a result of the enzymatically catalysed formation of non-disulphide covalent bonds between the amino acids glutamine and lysine on unfolded proteins under the effect of a calcium dependant transglutaminase, naturally present in fish muscle (Lanier et al., 2005). The temperature

at which proteins are unfolded and bonded in this process is different depending on the species. So in cold-water fish better results are obtained at temperatures of around 25°C, whereas *suwari* gels made with warm-water species show better mechanical properties when incubated at higher temperatures nearer to 40°C (Ramírez et al., 2011). An alternative way of developing *suwari* in fish gels is by protein denaturation induced by high pressure, where gels with a more deformable texture and a shinier colour are obtained. In this case, the hydrophobic bonds play a predominant role (Perez-Mateos et al., 1997). Subsequent heating of *suwari* gels at 80-90°C, results in the formation of stronger gels than without low-temperature pre-incubation (Lanier et al., 2005). To get different textures from heat gelation a number of ingredients and additives are used. These additives can act, both by interacting with the proteins or as fillers in the protein network (Park, 2005; Ramírez et al., 2011).

COLD GELATION

In recent decades, there has been an increasing demand for minimally processed products. For this reason, the development of cold gelation technology is becoming an interesting option for elaborating restructured fish products with a raw aspect. Products elaborated by this method are very versatile and can be commercialised in many different ways, such as fresh ready-to-cook fish fillets, small fillet pieces to put into brochettes, carpaccio-like *sushi* or ready-to eat-dishes, and also in marinated or smoked products (CSIC, 2006; Moreno et al., 2008, 2010a, b). There are many different ways of elaborating restructured products by using cold technology because of the different binding agents available. In cold gelation, protein muscle aggregation is mostly due to the action of different bindings that are able to act at sufficiently low temperatures without inducing changes in the raw aspect of the product (Moreno et al., 2008).

Moreover, by using these binding agents, it is also possible to manufacture added-value products in which final texture will also depend on muscle size and quality, composition, the specific binding agent employed and the treatment applied (Moreno et al., 2009a, 2010a, c).

Two of the most widely used binding additives in fish restructuring are alginates (mainly sodium alginate) and microbial transglutaminase (MTGase)., Recently however, new scientific studies have suggested the incorporation of konjac glucomannan in seafood mince in order to make it possible to elaborate structures from non-functional raw material.

Alginates

Alginates are hydrocolloids that play an important role in the structure, stability and functional properties of several processed foods. Alginates are alginic acid salts that are extracted from brown seaweeds (*Laminaria sp.*, *Macrocystis sp.*, etc.) known by the name of Kelp. They are linear polysaccharides composed of β -D-manuronic acid and α -L-guluronic acid.

The use of alginates in food technology (Sodium alginate was approved as E-401 by the European Food Safety Authority in 2007) started to become of interest in the 1960's, when they were used to elaborate protective coatings, such as casing substitutes, and for structure/texture modification in fresh and further processed products, initially in meat. It is also considered a GRAS substance by the United States Department of Commerce. Alginate coatings were also reported to increase shelf-life by delaying odour development, dehydration, and off-colour in fresh products such as apples (Nussinovitch, 1998). Furthermore, alginates can be added to minced fish muscles as it

is the alginate molecule network that communicates texture to minced muscle (Pérez-Mateos and Montero, 2000; Lu et al., 2010).

In order to form a thermostable network, alginates require the presence of different polyvalent ions. Calcium is the divalent ion most commonly used to elaborate alginate thermostable gels, because its interaction with alginates is superior to that of other polyvalent cations (Rourke et al., 1997; Boles and Shand, 1998). In this process, the solubilized sodium alginate forms thermostable gels capable of binding comminuted or ground fish muscle, producing as a result structures of different textures (Moreno et al., 2008, 2010a). The alginate network interacts with myofibrillar proteins, but these interactions seem to be mainly electrostatic between the anionic groups on the alginate and the positively charged groups on the proteins (Rourke et al., 1997; Montero et al., 2000). The interaction between alginates and proteins is determined by the hydrocolloid concentration and calcium ion sources. In a comparative study of cold gelation of minced fish batters supplemented with different calcium sources and sodium alginate, Moreno et al. (2011) indicated that restructured products could be made firmer with calcium chloride than with the other calcium sources (calcium lactate and caseinate). On the other hand, the proportion of sodium alginate, calcium source, setting time and conditions were the main variables used to achieve different kinds of texture, and needed to be properly analysed in order to optimize the process. According to this study, depending on the texture desired in hake muscle (*Merluccius capensis*) restructuration, the concentration of calcium chloride was shown to be more effective at 0.1% than at higher concentrations, while sodium alginate was commonly ranged from 0.05% to 0.5% (Moreno et al., 2008, 2011). Moreover, the presence of salt and fat up to levels of 3% and 5% respectively did not significantly affect the texture of alginate/muscle systems (Beriaín et al., 2011; Moreno et al., 2011).

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150 **Transglutaminase**

151 Transglutaminases (TGases, EC 2.3.2.13, glutaminyl-peptide, amine- γ -glutamyl-
152 transferase) belong to a class of enzymes which catalyse the acyl transfer reaction
153 between the γ -carboxamine group of a peptide-bound glutaminyl residue and a primary
154 amino group of various substrates (Motoki and Seguro, 1998).

155 It is important to distinguish between endogenous transglutaminase (TGase)
156 which is widely distributed in plants and animal tissues, and microbial transglutaminase
157 (MTGase). In relation with fish muscle, endogenous transglutaminase (TGase)
158 promotes protein cross-linking in myofibrillar proteins in *surimi* and seafood mince at
159 temperatures lower than 50°C.

160 Microbial transglutaminase is secreted by *Streptoverticillium mobaraense* or
161 *Streptomyces mobaraense* (Motoki and Seguro, 1998). It is the principal commercially
162 available purified transglutaminase for food (Ajinomoto Handbook, Activa®).
163 Although, MTGase is considered a GRAS substance by the United State Department of
164 Commerce, the European Food Safety Authority (FSA, 2007) is still studying its use in
165 the food industry.

166 The great advantage of MTGase over endogenous TGase is that MTGase
167 activity is totally independent of Ca^{2+} . MTGase optimum temperature is around 20-50°C
168 depending on the specific enzyme preparation (Ajinomoto Handbook, Activa®) and the
169 substrate, although there is activity at 5°C in fish muscle (Moreno et al., 2010a,b). This
170 fact makes MTGase a good option in the preparation of gelled products at low
171 temperature without inducing changes in the raw aspect of the final product (Moreno et
172 al., 2008) and also with raw materials of different qualities and salt content. In this
173 sense, Moreno et al. (2009a) have reported that variations in muscle protein quality (due

to different frozen storage histories) is extremely important when elaborating restructured products by joining pieces of muscle, but not when the purpose is to gel a finely homogenate muscle. Moreover, the pH of the mince used to elaborate the restructured products should be controlled, especially if the added salt content is particularly low (Moreno et al., 2010c). It is also important to note that MTGase activity is clearly potentiated by adding sodium caseinate at around 1.0%, especially in cold gelation processes. Additionally, the incorporation of soy protein isolate and whey protein improves MTGase activity, though clearly in a less significant way when compared with sodium caseinate (Kuraishi et al., 1997).

The combination of transglutaminase and isostatic high pressure, whether or not coupled with the heating process, has been studied by various authors (Ashie and Lanier, 1999; Moreno et al., 2009b) and has led to the conclusion that a setting ranged between 25-40 °C prior to high pressure treatment is very suitable for the preparation of gels with improved mechanical properties. In this sense, it has been demonstrated that transglutaminase activity, both endogenous and microbial, is not hindered by 100-600 MPa; and even remains active during a subsequent setting process (Lee and Park, 2002). However, pressures higher than 200 MPa could induce changes in the raw colour, leading to a cooked appearance; an important point that should be taken into account when considering the final properties of the product (Moreno et al., 2009b).

Glucomannan

When seafood muscles have been previously processed by heating, as is the case of canned tuna, cooked octopus, “sawdust” from fish block sawing, etc., the protein functionality of the by-products is lost to a great extent. This is a problem, if these by-products are going to be used in seafood restructuration, as the gelation will be weak,

even though MTGase is used. The same happens if a certain level of fat content is high in the fish muscle that is going to gel (Moreno et al., 2009a; 2010b). To overcome these problems, konjac glucomannan has recently been studied by Herranz et al. (2012a, b) for use as a gelling agent where minced muscle would act as filler (CSIC, 2011).

Konjac glucomannan (KGM) is a neutral hydrocolloid extracted from the tuber of *Amorphophallus konjac* C. Koch. It has been used as a food and food additive in China and Japan for more than 1000 years. It has been recognized as safe (GRAS) since 1994 (Takigami, 2000; Zhang et al., 2005), was authorized as a binder in meat and poultry products by the U.S. Department of Agriculture (USDA) in 1996, and has been approved as E-425 by the European Food Safety Authority (FSA, 2007). Apart from this, KGM provides several health benefits such as lowering blood, cholesterol and sugar levels, as well as promoting intestinal activity, immune function, etc. (Vuksan et al., 1999). In Western countries its consumption has increased recently among obese people owing to its potential as a dietary fibre because of its effective water-absorbing ability (Chua et al., 2010).

KGM consists of a linear backbone of β -1,4-linked D-mannose and D-glucose and 5-10% of acetyl substituted residues which are assumed to be responsible for the solubility of KGM in water. The loss of these acetyl groups in an alkaline media, resulting in the formation of a thermostable gel, is the basis of many traditional oriental foods such as noodles, tofu and snacks (Chua et al., 2010; Douglas et al., 2005).

Even so, the use of KGM is not very extended in the seafood industry and few references can be found in scientific literature. Park (1996) observed that the addition of 5% konjac flour to whiting (*Merluccius productus*) and pollock (*Theragra chalcogramma*) surimi reinforced gel hardness by a factor of 8-10 in both of them. This author also concluded that surimi gels containing konjac flour exhibited an important

ability to maintain consistent shear strain values against repeated freeze/thaw abuse, that konjac flour (up to 2%) increased gel lightness, and that yellowness intensified as konjac flour was added (up to 5%). Iglesias-Otero et al. (2010) used an aqueous solution of KGM (10%) in a proportion of 1% in giant squid (*Dosidicus gigas*) surimi at different pH values to improve quality. These authors concluded that at pH 10.4 the surimi gelation showed an increase in strain amplitude and a decrease in the frequency-dependence of storage (G') and loss (G'') moduli; resulting in an improvement in gel properties. On the other hand, Xiong et al. (2009) showed that 1% of KGM could work as well as a conventional cryoprotectant on myofibrillar proteins from grass carp (*Ctenopharyngodon idella*) muscle during frozen storage.

Based on the ability of glucomannan to make thermostable gels, Herranz et al. (2012a, c) studied the possibility of KGM acting as a gelling agent in minced fish, forming a network in which the fish muscle particles could act as a filler. This would provide the possibility of using a wide variety of seafood muscles whose completely denaturalized protein, due to previous processes, had made it either incapable of gelling or that its gel-forming ability was poor. Hence, a wide range of restructured products after gelating in the form of fibre or myotomes could be elaborated and commercialized as fresh, frozen or cooked products (Borderías et al., 2012). Furthermore, due to the fat binding properties of this gel (unpublished data), the nutritional characteristics of these products could be improved so that functional ingredients such as omega-3 polyunsaturated fatty acids (n-3 PUFAs) from fish oils and conjugated linolenic acid (CLA) from vegetable oils could be included in the network formed by KGM. Additionally, based on the large amount of water (around 86%) that restructured muscle elaborated with a mixture of KGM and muscle contains, these products could be used in hypocaloric diets.

Before the application of glucomannan as a gelling agent in the elaboration of restructured seafood products, an exhaustive study on glucomannan gelation was carried out to choose the type and concentration of glucomannan and alkali, as well as the setting time and neutralization conditions of the process (Herranz et al., 2012a; Herranz et al., 2012c). From these studies, it was concluded that the best conditions to solubilize the glucomannan and obtain aqueous glucomannan solutions (AGD) were 30 minutes at 60°C in a homogenizer and 5% of KOH at 0.6 N to raise pH values to 11.8-12.0, resulting in complete deacetylation. After the alkali was added, a two-step setting was applied to obtain a thermostable gel (first 1 hour at 30°C followed by another of 4 hours at 5°C). After that, the gel was placed in a buffer until neutralization. When this gel was heated at 50-70°C, the gel became even more stable (Herranz et al., 2012b).

Next, a study of fish muscle analogue prototypes elaborated with a solution of glucomannan and non functional muscle was carried out (Herranz et al. 2012c). The solution was made up of aqueous glucomannan dispersions (AGD) at different concentrations (3 and 6%, v/v) and non-functional mince ("sawdust") obtained from sawing frozen hake (*M. capensis*) blocks. The different glucomannan:sawdust ratios were 50:50 and 25:75 w/w and the processing was the same as that for glucomannan gelation alone, reported above. The different determinations carried out (lightness, water binding capacity, puncture test and sensorial analysis) showed that the gel formed by 6% AGD (w/v) added in a proportion of 25:75 to the non-functional muscle, produced a prototype with textural and flavour characteristics similar to those of fish (hake) muscle (Herranz et al., 2012c).

FUNCTIONAL RESTRUCTURED SEAFOOD PRODUCTS

In recent years, an increasing interest in the design of functional products has been noted. However, the potential for the development of functional seafood products has not yet been fully exploited (Careche et al., 2011). The fact that during restructured seafood processing, small pieces or mince are reshaped provides an excellent opportunity for including some bioactive ingredients in this fish muscle matrix. Some functional ingredients such as dietary fibres (DFs), phytosterols, carotenoids, natural antioxidants, n-3/n-6 fatty acids and vitamins are widely used in the design of functional foods and new ones are emerging e.g. antioxidant DFs. The present production of functional seafood products is limited compared to other foods, as seafood is mainly used as a source of bioactive components (e.g. fish oils or protein powders) to develop alternative functional foods.

Dietary fibres in fishery products

DFs are regarded as the most widely used functional ingredients in foods, but they have rarely been incorporated in seafood. The addition of DFs to fishery products is of great interest, not only as a means to further complement their healthy characteristics, but also as a means of improving the technological properties of the products such as water binding, gelling, etc. (Borderías et al., 2005). Thus, most of the DFs used in seafood products are soluble (e.g. carrageenan, guar, inulin) and usually selected because of their high water binding, emulsifying, thickening or gel-forming ability (Park, 2005).

In the case of insoluble DFs, experience of their use in seafood products is more limited (Borderías et al., 2005). However, nowadays interest in the use of insoluble DFs and other DF sources, mainly from fruits, with a well-balanced soluble:insoluble ratio has increased significantly. Other types of DFs of particular interest in this area are

antioxidant DFs that are mainly obtained from fruits and seaweeds, which contain both DF (>50%) and natural constituents with specific antioxidant capacity (Saura-Calixto, 1998). The addition of antioxidant DFs to restructured fish products could serve not only to include DF in seafood diets but also delay lipid oxidation of polyunsaturated fatty acids (PUFA), both of which appear in the list of positive compounds for nutritional claims in the EU regulation (EC, 2006; EU, 2010). A Consumer Product Test has also shown that there is an opportunity to develop new seafood products enriched in DFs well-adapted to consumer preferences (Borderías et al., 2008; Careche et al., 2008a). This fact can be considered as a new business opportunity for some industries. Therefore, this section reviews the inclusion of these insoluble DFs, both high cellulosic and antioxidant DFs, in restructured seafood products.

Commercial insoluble dietary fibres

Wheat DF (Vitacel®) is one of the insoluble DFs (*iDFs*) most widely-used as a food ingredient. It is composed of 74% cellulose, 26% hemicellulose and <0.5 % lignin, which endows it with highly insoluble properties and fat binding capacity. It is white in colour and neutral in taste and smell. It was previously stated that in Alaska pollock (*T. chalcogramma*) and giant squid (*D. gigas*) *surimi* gels, the inclusion of wheat DF at concentrations of 3 and 6%, fibre grain sizes 80 and 250 µm long, and at constant final moisture conferred an extra whiteness on the products (Sánchez-Alonso et al., 2006, 2007d). Moreover, from the point of view of gelation, these gels can be regarded as good quality gels, even though water retention properties and hence mechanical properties and texture were altered. This would be as a consequence of the proportional reduction of protein content in the gel, which may also be associated with the irregular network formed due to the addition of the non-gelling wheat DF compound. The

particle size of the added wheat DF is also relevant, thus it is not necessarily the case that the higher the water holding capacity of the DF the better the formulation. In fact, this can have the effect of dehydrating the protein. This was found to be the case with wheat DF, where two separate moisture compartments can occur in the matrix, indirectly impairing the gel-forming capacity of the muscle protein (Sánchez-González et al., 2009). Furthermore, wheat DF makes softer and more deformable gels and this attenuates the gummy texture which is not well-accepted in Western countries (Sánchez-Alonso et al., 2007d). In minced hake (*Merluccius merluccius*) and horse mackerel (*Trachurus trachurus*) fish muscle, Sánchez-Alonso et al. (2007a) studied the incorporation of wheat DF levels of 3 to 6%. In this case, the main technological advantages of including wheat DF in minced fish was its effectiveness in binding water of battered products, thus preventing the coating from breaking and the cooked portions from deforming. Besides this, the restructured products are softer and less cohesive, which may be an advantage for some end products. Sensory analysis suggests that the addition of a 3% wheat DF concentration is better for minced fish muscle (Sánchez-Alonso et al., 2007a).

The above-cited authors in association with a company producing restructured fish products, successfully included wheat DF in two commercial products, and a Consumer Product Test was performed with 500 consumers in order to check the sensory acceptability as well as the concept of DF inclusion in seafood products. The results showed that products with information about DF content and its health benefits generated positive expectations in the consumers. Consumers only detected differences in attributes related to texture, thus the optimization step and scaling up was performed successfully, developing a product which could be labelled “high in dietary fibre” (Careche et al., 2007; Dopico et al., 2007; Borderías et al., 2008).

Another commercial cellulose DF used as an ingredient in restructured seafood products is a cellulose powder (Solka-Floc®) that effectively reduces the usage level of cryoprotectants, sweetness and the amount of modified starch needed in *surimi* products, thereby achieving a non-starch texture. This kind of cellulose also prevents gelled products from becoming rubbery and dry during frozen storage by reducing freeze syneresis and improving water binding (Yoon and Lee, 1990). There is also inner pea DF (Swelite®) on the market that is beneficial in safeguarding the textural properties of hake (*M. capensis*) and sea bass (*Dicentrarchus labrax*) heat-induced gel products (Cardoso et al., 2007, 2011). However, effects can differ between fish species, for instance, this DF did not improve the gel strength of mackerel (*Scomber scombrus* and *S. japonicus*) *surimi* gels but instead increased hardness and other related parameters (Cardoso et al., 2009).

Non-commercial dietary fibres: antioxidant dietary fibres

Although antioxidant DFs (ADFs) have been non-commercial until now, it is important to consider them in this review because of their beneficial properties. Some of the most interesting ADFs are grape concentrates produced from various white or red grape by-products (skin, seeds or pomace). Grape DF has a high content of total DF (>70%) and a relatively high content of soluble DF in comparison with total DF. Moreover, the presence of associated polyphenolic compounds (>5%) endows the DFs with antioxidant properties (Sánchez-Alonso et al., 2007b, 2008b). As a source of antioxidant DF, these ADFs were added to restructured products made from horse mackerel, a semi-fatty fish species very prone to oxidation (Sánchez-Alonso et al., 2007b, c, 2008b; Sánchez-Alonso and Borderías, 2008). Both white and red grape DF added to minced horse mackerel muscle significantly increased its water holding

capacity. It also reduced thaw drip after frozen storage, and significantly increased the cooking yield due to the reduction in drip loss during heating; cohesiveness and hardness were also reduced. Furthermore, adding 2% grape DF produced an acceptable product in sensory terms and delayed lipid oxidation in frozen minced muscle stored at -20°C for up to 180 days (Sánchez-Alonso et al., 2007b, 2008b; Sánchez-Alonso and Borderías, 2008).

On the other hand, Careche et al. (2008b) reported that the inclusion of DF, alone or together with natural antioxidants associated with the DF, led to sensorially acceptable restructured products made of fish muscle that fulfilled the requirement of “source of dietary fibre” and/or “high content of fibre” according to EU regulations (EC, 2006). Antioxidant DFs could also act by delaying PUFA oxidation with the subsequent nutritional and sensory advantages associated with these products.

Some consumer studies have been conducted on fish mince (*Salmon sp.*) with added grape DF (Careche et al, 2008a, b), and one of the consequences arising from the results of these consumer studies was the interest in using alternative sources of ADF, as consumers preferred their seafood products to be supplemented with DFs of marine origin e.g. seaweed concentrates (Careche et al., 2008b). Thus, Díaz-Rubio et al. (2011) studied the nutritional profile of *Fucus spp.* seaweed ADF and its technological effect on minced horse mackerel (*T. trachurus*) during frozen storage. This ADF is composed mainly of DF (>60%) with a relatively high soluble content (16% of total DF) and more than 5% of associated phenolic compounds. The study demonstrated that minced fish samples supplemented with *fucus* ADF (1% and 2%) had lower lipid oxidation than those without it, and the total drip after thawing and cooking was reduced during frozen storage. Fish samples with 1% *fucus* DF did not differ in flavour from the samples without DF (Díaz-Rubio et al., 2011).

Milled seaweeds could be used as a good source of DF in restructured fish products. Senthil et al. (2005) reported that up to 10% of red seaweed, *Eucheuma* (*Kappaphycus alvarezii*) powder could be added without adversely affecting the appearance, texture and acceptability of fish (*Pampus argentius*) cutlets. Recently, a range of innovative seafood products enriched with coloured ADFs has been developed from seaweed or grapes, based on a new product idea, similar to the “Swiss roll, that the authors have named “brazo de gitano” (CSIC, 2010). One of these prototypes is made up of a covering of *surimi*-based gel enriched with wheat DF, stuffed with a mixture of minced fish muscle and freeze-dried seaweed (*Ulva rigida*) (Sánchez-Alonso et al., 2008a). The seafood prototype contains sufficient DF to be labelled as a “source of DF” (EC, 2006). The foregoing shows that it is possible to make restructured colourful products with an attractive image and sensory acceptability, thereby overcoming the main barriers discouraging the application of some very interesting antioxidant DFs (grapes, seaweeds) (Sánchez-Alonso et al., 2008a).

Fatty acids in seafood restructured products

Lipids are one of the ingredients which can be incorporated into *surimi* products as a texture modifier, colour enhancer, or processing aid (Park, 2005). In fact, vegetable and animal lipids are often added to *surimi* products since fish muscle can produce a stable emulsion with them. In this context, food products with added PUFAs (ω -3) are in increasing demand due to their demonstrated health benefits, confirmed by the appearance of PUFAs in the positive list of compounds for nutritional claims in EU regulations (EU, 2010). Pérez-Mateos et al. (2004) reported little changes in sensory properties and lipid oxidation of fortified *surimi* seafood (crab analogue) during 2 months of chilled storage in gels containing 1.5-2.5 % ω -3 fatty acids by adding PUFAs

from different sources (fish oil concentrate, menhaden oil, or purified marine oil). Recently, Pietrowski et al. (2011, 2012) demonstrated that the nutritional value, colour and gelation of Alaska pollock *surimi* seafood can be enhanced, without altering texture properties, by the addition of 9 % omega-3 PUFA-rich oils (flaxseed, algae, menhaden, krill). Also, cod (*Gadus morhua*) *surimi* was fortified with ω -3 fatty acids (500 mg of ω -3 fatty acids per serving of *surimi* ~85 g) from algal oil with good oxidative stability and gel strength (Park et al., 2004). Cardoso et al. (2010) successfully developed a ready-to-eat minced fish product from hake containing dietary fibre (inner pea DF) and fortified with ω -3 fatty acids (2.9 % deodorized cod liver oil).

Another type of functional fatty acid is conjugated linoleic acid (CLA) which refers to a mixture of different isomers of linoleic acid (18:2n-6) commonly found in beef, lamb and dairy products. A commercial CLA oil was added to Alaska pollock *surimi* gels instead of sunflower oil, at a dosage (3.7 grams of CLA isomers per 100 grams of sample) that has been reported to have beneficial health effects (Whigham, Watras and Schoeller, 2007) without modifying appearance, mechanical properties or water binding capacity (Sánchez-Alonso and Ayo, 2007).

CONTAMINANT ALLEVIATION DURING MINCE PROCESSING

Although it is well known that fish and shellfish contain high levels of pollutants due to the accumulative process of biomagnification, scarcely any literature has been found about its elimination. The uptake and distribution of pollutants in seafood varies depending on factors such as physiological and environmental conditions, age, size, sex, body weight, etc. (Pourang et al., 2004). Heavy metals are distributed in diverse tissues (muscle, internal organs, fatty tissues, etc.), but in most cases, the highest concentration of heavy metals is found in the hepatopancreas, probably because of its high

metallothionein content (Pourang et al., 2004; Ren et al., 2008). In this regard, the higher levels of heavy metals in molluscan (Mediterranean mussel, *Mytilus galloprovincialis*) than in fish (rainbow trout, *Oncorhynchus mykiss*) could be due to the fact that mussel metallothioneins are more reactive than fish proteins to metal binding (Vergani et al., 2005). Persistent organic pollutants accumulate mainly in the fatty tissue due to its lipophilic nature.

Contaminant alleviation in seafood restructured products could be done during the washing stage of the pieces of muscle or, even better, once the muscle has been minced owing to the small size of the minced particles.

Chelating washing technique to reduce heavy metal contamination

Among the main procedures for alleviating heavy metals is the chelating washing technique, where ligands bind to toxic metals from the muscle by forming strong water-soluble complexes that can be eliminated by washing. The efficiency of the process can be affected, on the one hand, by the chelating agent type and concentration along with the pH-effect and ionic strength, and on the other hand, by the heavy metal type and concentration and its speciation state. In some cases, sequential extraction steps might be necessary and even the use of mixtures of different chelating agents.

Based on the knowledge that metals have a high affinity for thiol groups of enzymes and proteins, Schwartz (2008) patented a chelant extraction process for removing heavy metals (such as mercury, lead, uranium, cadmium, etc.) from food by a vacuum soaking process using different types of ligands such as concentrated proteins (legume protein, pulse legume protein, vegetable protein, grain protein, bacterial metalloregulatory protein) and organic acids (phytic acid) and other chelants (EDTA,

BAL, DMSA, DMPS, etc.). The author suggested that another way of incorporating ligands could be by adding them to a coating, sauce, or marinade to be eaten with the fish, thereby avoiding its absorption by the digestive tract.

Mercury is the most studied metal in fish muscle. Cohen and Schrier (1975) carried out a study in carp (*Ciprinus carpio*) and sheepshead (*Archosargus probatocephalus*) by using a dilution of 1.2 g of sodium borohydride (for 1 ppm of mercury in 100 g of fish protein concentrate) for 20-30 min at room temperature. Spinelli et al. (1973) studied the effectiveness of cysteine in reducing the mercury content of comminuted fish and fish protein concentrate from Pacific halibut (*Hippoglossus stenolepis*) and hake (*M. productus*) where it was reported that the amount of mercury extracted was related to the tissue pH and the concentration of cysteine (0-0.5%). A similar study was carried out in precooked “yellow tuna” where it was reported that about 66-75% of mercury was removed from shredded muscle by a two-hour extraction at 5°C by washing with dilute hydrochloric acid containing 0.33% cysteine hydrochloride. The same procedure was used for thick slices but with a 24-hour extraction period (Yannai and Saltzman, 1973).

It is important to bear in mind that the amount of mercury extracted was related to the ligand concentration (in order to facilitate effective competition) and the medium pH. In general, it seems that the lower the solution pH, the better the alleviation of mercury from the fish muscle. Mercury removal efficiency by 0.5 % cysteine hydrochloride solution could be about 80 % in tuna slices under the most favourable conditions (pH, stirring, extraction time, muscle:water ratio, etc.), but no higher, due to the difficulty in breaking some strong bonds (Schab et al., 1978). This is in agreement with the findings of Hajeb and Jinap (2009) carried out in minced mackerel (*Rastrelliger brachysoma*) muscle. These authors reported that mercury reduction

significantly decreased (up to 81%) depending on the pH of the mixed solution used (citric acid, sodium hydroxide and salt) during the washing treatment. However, Gong et al. (2011) reported no decrease in mercury levels in protein isolates from whitefish (*Coregonus clupeaformis*) and walleye (*Sander vitreus*) muscles prepared by pH shifting in the presence of 5 mM sodium citrate; it only decreased with the addition of 0.5% cysteine.

Regarding the other heavy metals (selenium, cadmium, lead, arsenic), Cohen and Schrier (1975) suggested that they can be reduced in the same manner as mercury. In this respect, Ren et al. (2008) studied the removal of cadmium from scallop hepatopancreas by washing four times with a weak acid solution (2% acetic acid or citric acid) which produced a reduction in the cadmium level from approx. 39 ppm to <0.6 ppm. Besides using quelants, in a recent study carried out by Topcu and Bulat (2010), it was reported that two strains of *Enterococcus faecium* showed detoxification properties and could be used in the production of fermented functional foods because of their ability to bind cadmium and lead from the aqueous medium of food (pH 5).

Decontamination of persistent organic pollutants

Persistent organic pollutants (POPs) are bioaccumulated in the fatty fraction of fish and seafood. Therefore, a reduction in the fat content would reduce the content of these undesirable compounds in the raw material. This can be done by simply washing the mince and then decanting the separate fat fraction. These liposoluble compounds can also be removed by oil extraction, using dissolvent oils such as soybean oil, olive oil or fish oil (Baron et al., 2007; Oterhals and Nygård, 2008). As fish oil decontamination does not imply the alteration of nutritionally valuable compounds, it

can be added as raw material in order to increase the levels of omega -3 fatty acids in foods.

Most of the studies related to removing pollutants have been performed on fishmeal and fish oil using adsorbent treatment with activated carbon (Kawashima et al., 2009; Maes et al., 2010; Oterhals et al., 2007) combined with short-path distillation (Berntssen et al., 2010; Oterhals and Berntssen, 2010), by supercritical CO₂ extraction (Kawashima et al., 2009), and deodorization using heat (Maes et al., 2010).

A recent way of reducing the fat content in fish muscle is by using the pH-shift process in which the protein content is solubilized in acid or alkaline medium and then precipitated and isolated from the rest of the components. Following this technique, Marmon et al. (2009) obtained more than a 70% reduction of organic contaminants (dioxins, and polychlorinated biphenyls) in the protein isolate from Baltic herring (*Clupea harengus*). In this case, most of the pollutants were found in the floating fat emulsion layer after the first centrifugation of the acid and alkaline homogenates. The authors tried to maximize the extraction of fat-soluble contaminants by the addition of solvent (ethanol), oil (olive, fish and soybean), calcium chloride and citric acid. However, the modified acid and alkaline pH-shift method will need further investigation in order to improve its efficiency.

CONCLUSIONS

The present review has attempted to summarize the great potential that restructured seafood technologies offer in order to elaborate added-value products. In this sense, it is important to remark that, on the one hand, the methodology will clearly depend on raw material properties (mainly protein content and functional qualities) especially when using thermal induced and cold gelation technology. On the other hand,

seafood restructuring offers the opportunity of adding functional components not only to reinforce nutritional and health characteristics, but also to remove unpleasant components, as is the case of fats, colours, contaminants, etc. Moreover, restructured seafood products provide an opening for the creation of new presentations that will widen the range of marketing possibilities, aimed at the new type of consumer who is looking not only for healthy food but for food with a new feel-good factor.

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