

# Texture and Structure Measurements and Analyses for Evaluation of Fish and Fillet Freshness Quality: A Review

Jun-Hu Cheng, Da-Wen Sun, Zhong Han, and Xin-An Zeng

**Abstract:** Recently, food safety and quality have become critical issues of great concern throughout the world. Fish is one of the most vulnerable and perishable aquatic products. The evaluation of fish and fillet freshness is therefore very significant in research and development for providing premium and supreme quality for human health and acceptance by consumers, as well as for international trade. The texture and structure of fish muscle are important freshness quality attributes that depend on several parameters such as hardness, cohesiveness, springiness, chewiness, resilience, and adhesiveness, as well as the internal cross-linking of connective tissue and the detachment of fibers. This review aims to present recent advances of texture and structure measurements and analyses, including sensory evaluation and instrumental methods, for indicating and evaluating fish freshness quality. Factors affecting these measurements are detailed and correlations between texture and structure are discussed. Moreover, the limitations and challenges of fish texture and structure measurements are described and some viewpoints about current work and future trends are also presented.

## Introduction

Freshness is one of the most significant aspects for evaluation of fish quality as freshness is directly linked to appearance, texture, and taste of the perception of consumers. Generally speaking, fish are processed and frozen to guarantee palatability and safety, to improve their shelf life and convenience, and to maintain and prolong their freshness for various culinary supplies and for the fish processing industry due to vital economic importance (Campus and others 2010). However, now-a-days, one great challenge existing in the fish product processing industry is to acquire trustworthy and effective information on fish freshness throughout all the production process chains, which should eventually afford a guaranteed and premium quality of fish and fish products for human consumption and international trades (Damez and Clerjon 2008). In addition, freshness is a multifaceted quality attribute of fish that notably affects the eating quality and further influences the acceptability and partiality of consumers (Sharifian and others 2011). There are many external and internal influencing factors that affect the freshness quality of fish, including processing parameters and different stages of postmortem transformation such as the early stage of rigor, rigor mortis, and end of the rigor, the

autolysis process, and microbiological spoilage after death. Such transformation stages consist of physical, chemical, physicochemical, and biochemical processes, followed by bacterial spoilage, protein degradation, and ATP decomposition, which accelerate the loss of freshness, destroy the structure of muscle, and degrade the quality of fish (Ayala and others 2010). Among these factors influencing the freshness quality, textural and structural measurements play a critical role in the evaluation of fish quality.

Generally speaking, during the postmortem condition, the muscle of fish is very prone to become soft, which further affects the textural quality of fish muscle. Therefore, the texture of fish is a main feature used to appreciate the freshness quality (Chéret and others 2006). Textural parameters are also frequently employed to examine and evaluate fish quality along the fish value chain, which mainly displays in assessment of influences of handling and processing methods on the shelf life of fish products and partiality and satisfactoriness of consumers. It has been confirmed that storage temperature during handling and operating process generally has a distinctive effect on fish texture measurement (Allen and others 2003; Pearce and others 2011). The most common texture defects are muscle softening and gaping forming caused by pre- and postmortem treatment. The existing problems are mostly associated with the changes of chemical compositions and the degradation of muscle proteins (Aussanasuwannakul and others 2010). Moreover, there are other numerous interacting factors along with physical factors (species, age and size, feeding ingredients, sample heterogeneity, and gaping), chemical factors (water content and distribution, fat content and distribution, and collagen content), and diverse treatments (storage time and temperature, freezing, chilling, high-pressure processing (HPP), and salting and

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MS 20130791 Submitted 6/12/2013, Accepted 8/15/2013. Authors Cheng, Sun, Han, and Zeng are with Academy of Contemporary Food Engineering, College of Light Industry and Food Sciences, South China Univ. of Technology, Guangzhou 510006, P.R. China. Author Sun is also with Food Refrigeration and Computerized Food Technology, Agriculture and Food Science Centre, Univ. College Dublin, Natl. Univ. of Ireland, Belfield, Dublin 4, Ireland. Direct inquiries to author Sun (E-mail: [dawen.sun@ucd.ie](mailto:dawen.sun@ucd.ie)).

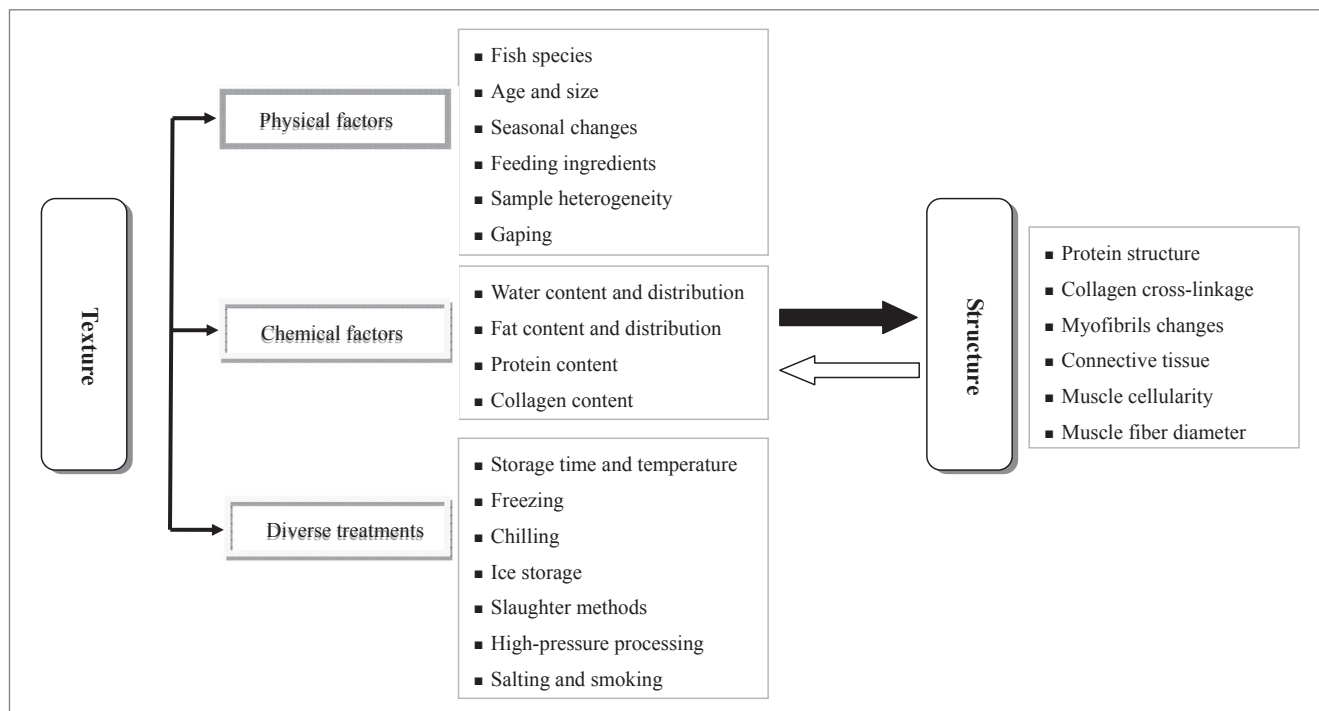


Figure 1—Factors affecting fish texture and structure measurements.

smoking) (Hultmann and Rustad 2004; Pearce and others 2011). The detailed factors influencing fish texture measurements are shown in Figure 1.

With respect to the structure of fish muscle, myotome as one of most important constituents of muscle is made up of huge numbers of single muscle fibers and this especial structure is linked by intramuscular connective tissues that are also named membranous myocommata (Sharifian and others 2011). Figure 2 illustrates the basic microstructure of fish muscle. On the other hand, in the connective tissue of fresh fish muscle, collagen is the most significant constituent, amounting from 3% to 10% of the protein and plays a vital role in maintaining fillet integrity and muscle cohesiveness (Aussanasuwannakul and others 2012). During the postmortem situation, autolysis of fish muscle caused by collagenases and other proteases usually result in the changes of the collagen (Suárez and others 2005).

Obviously, there is a close and subtle relationship between texture and structure or microstructure. The variation of textural properties of fish muscle derived from intrinsic differences mainly depends upon the structure of fish muscle tissue, which is attributed to the internal factors related to the structures of contractile protein, the framework of connective tissue, lipid oxidation, and some external factors, such as methods of sample handling and conditions of cold storage (Aussanasuwannakul and others 2012). Among texture attributes, firmness also termed as hardness, an essential evaluating parameter of fish freshness is closely associated with the human visible acceptability of fish products. This vital index depends largely on the structure of connective tissue (Casas and others 2006). Some studies (Johnston and others 2000; Thompson 2002; Purslow 2005) have indicated that textural attributes pose significant positive correlations with the density of muscle fiber.

Some general reviews have previously been conducted on food texture and structure, showing relationships between particles and food texture perception (Engelen and others 2005), between food

structure and rheology (Fischer and Windhab 2011), between food tenderness and collagen content (Lepetit 2007), and between food rheology and tribology (Chen and Stokes 2011). With regard to fish texture and structure measurements, in an early review, Coppes and others (2002) presented texture measurements in fish and fish products and the correlations between sensory and instrumental measurements. However, no review is available to specifically address the measurements of fish and fish fillet texture and structure for analyzing and evaluating fish freshness quality. Therefore, the aim of this paper was largely to review the measurement methods, influencing factors, and correlations between fish texture and structure.

## Texture Measurements

As discussed previously, texture is about sensory interpretation and expression of the structure or interior construction of products linked to their response to stress and haptic attributes (Coppes and others 2002; Lepetit 2007). Therefore, texture is commonly measured and presented as some mechanical properties, manifesting as performance of hardness/firmness, gumminess, resilience, cohesiveness, springiness, adhesiveness, and viscosity by the vision, hearing, somesthesia, and kinesthesia of human sense in the muscle based on the hand, finger, tongue, jaw, or lips (Hagen and others 2007). Figure 3 shows an understanding of fish texture related to fish processing from farm to fork for final consumption.

## Measurement methods

Based upon the interpretation of fish texture, the measurement methods of texture are of obvious significance for indicating product freshness, and these methods are related to sensory and instrumental measurements.

**Sensory methods.** Texture of raw fish fillet is generally measured in the industry by the “finger method,” which is an indication of the suitability for further processing and is mainly dependent on

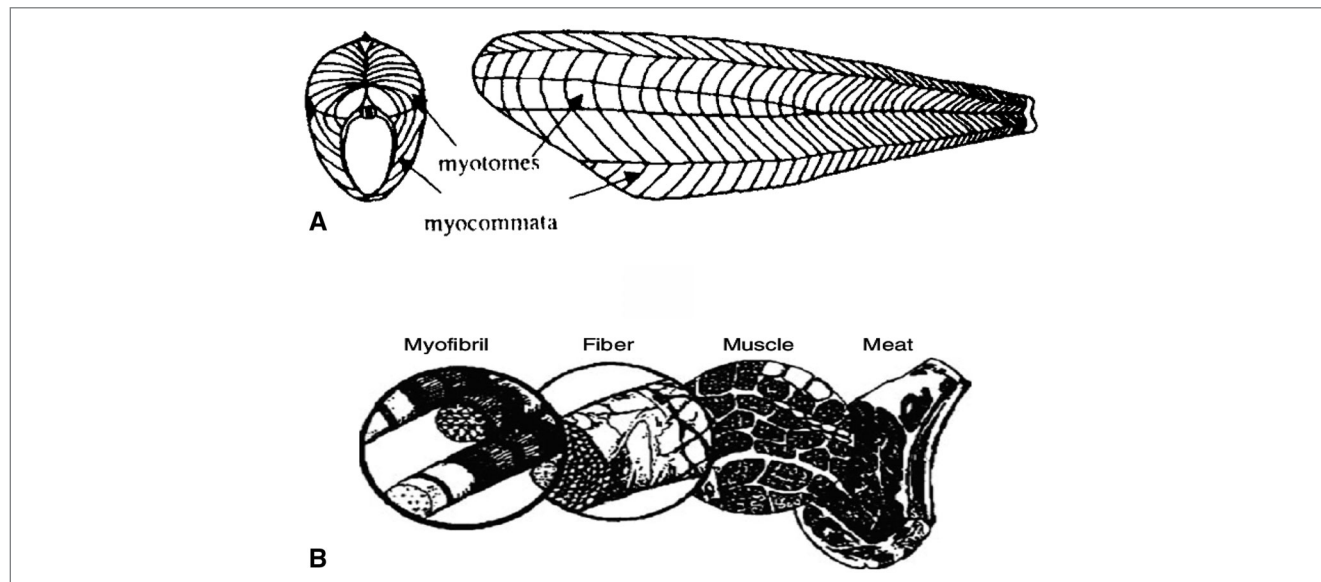


Figure 2—Basic microstructure of fish muscle. (A) Construction of fish muscle: myotomes and myocommata (Hyldig and Nielsen 2001); (B) an example of hierarchical scales in muscle tissue (Aguilera 2011).

the firmness (Sigurgisladdottir and others 1999). The evaluation of firmness is usually performed through pressing on the skin or the fillet of fish by finger, and this method depends to a large extent upon subjective assessments of the expert panel. At present, the popular improvement of a quality index method of sensory evaluation with different scores showing the degree of firmness of fish texture indicating the freshness quality has been implemented in several European countries (Barbosa and Vaz-Pires 2004; Cardenas and others 2007; Sant'Ana and others 2011; Massa and others 2012; Cyprian and others 2013).

**Instrumental methods.** Compared with sensory evaluation, it has been proved that the textural measurements by instrumental analysis methods are better and more precise by reasons of reducing the variations during measurements arising from human factors. The puncture, compression, shear, and tension are 4 main instrumental techniques and methods that are used to measure and evaluate the texture of fish from force–deformation curve, displaying the value of force, deformation, slope, and area (Casas and others 2006). On the other hand, in order to decrease the variations of the choice of sampling points and the influence of chemical composition, providing accurate, reliable, and objective methods are necessary and of interest for the fish processing industry. The Warner–Bratzler and Kramer shear compression cell based on shearing and cutting devices are 2 typical methods that have been widely developed (Jonsson and others 2001; Cavitt and others 2004, 2005; Alizadeh and others 2007). Most importantly, double-compression as another effective method is capable of performing a texture profile analysis (TPA) model obtained from a force–time curve, which can offer a meaningful interpretation to a series of textural parameters illustrated in Figure 3 (Rahman and Al-Farsi 2005; Herrero and others 2007). Currently, TPA for fish texture measurement as a standard method is still generally referred to in the literature for textural description and analysis (Sigurgisladdottir and others 1999; Martinez and others 2004; de Huidobro and others 2005; Chen 2009; Chen and Stokes 2011). Table 1 summarizes the measurements of fish texture by instrumental methods.

### Factors affecting fish texture measurements

It is a fact that factors affecting fish texture measurements mostly involve physical and chemical factors, and diverse treatments. Among these factors, the effects of fish species, physicochemical properties, different handling methods, and HPP are focused on here and are detailed below.

**Effects of fish species.** Atlantic salmon (*Salmo salar*) is one of the most extensively studied fish species in the world owing to its significance in aquaculture, fisheries, and ongoing conservation efforts to protect declining populations (Bourret and others 2013). Hence, the texture measurements of Atlantic salmon have been widely investigated using instrumental methods. For example, in an early investigation, instrumental texture analyses of raw Atlantic salmon fillet muscle at slaughter and ice storage after 4 d were carried out and it was shown that the force against compression at about 5 mm was lower in sample groups starved for a shorter time than in those groups starved for a longer time lasting for 58 d (Einen and Thomassen 1998). In another work, Casas and others (2006) measured the textural characterizations of Atlantic salmon preserved on ice at 2 °C using diverse instrumental methods performed at 3 different positions along the fillet. The force, energy, and the slope of the force–deformation curve as variables were obtained, and the results showed that the tail region was relatively firmer than the other locations of the fillet, and the compression test with a cylindrical probe was proved to be most appropriate method to obviously discriminate the location changes. On the other hand, in order to evaluate the textural properties and gaping in salmon fillets kept at 0 to 4 °C, a new tensile test was developed and data obtained from this method were shown to have a strong correlation with gaping severity (Ashton and others 2010). Hence, it could be concluded that this novel method is capable of making an important complement to mechanical analysis of fish texture, specifically for prediction and detection of gaping during fish processing.

Studies on other fish species have also been conducted. Ocaño-Higuera and others (2011) measured the textural properties of rayfish muscle using the Warner–Bratzler shear force method in

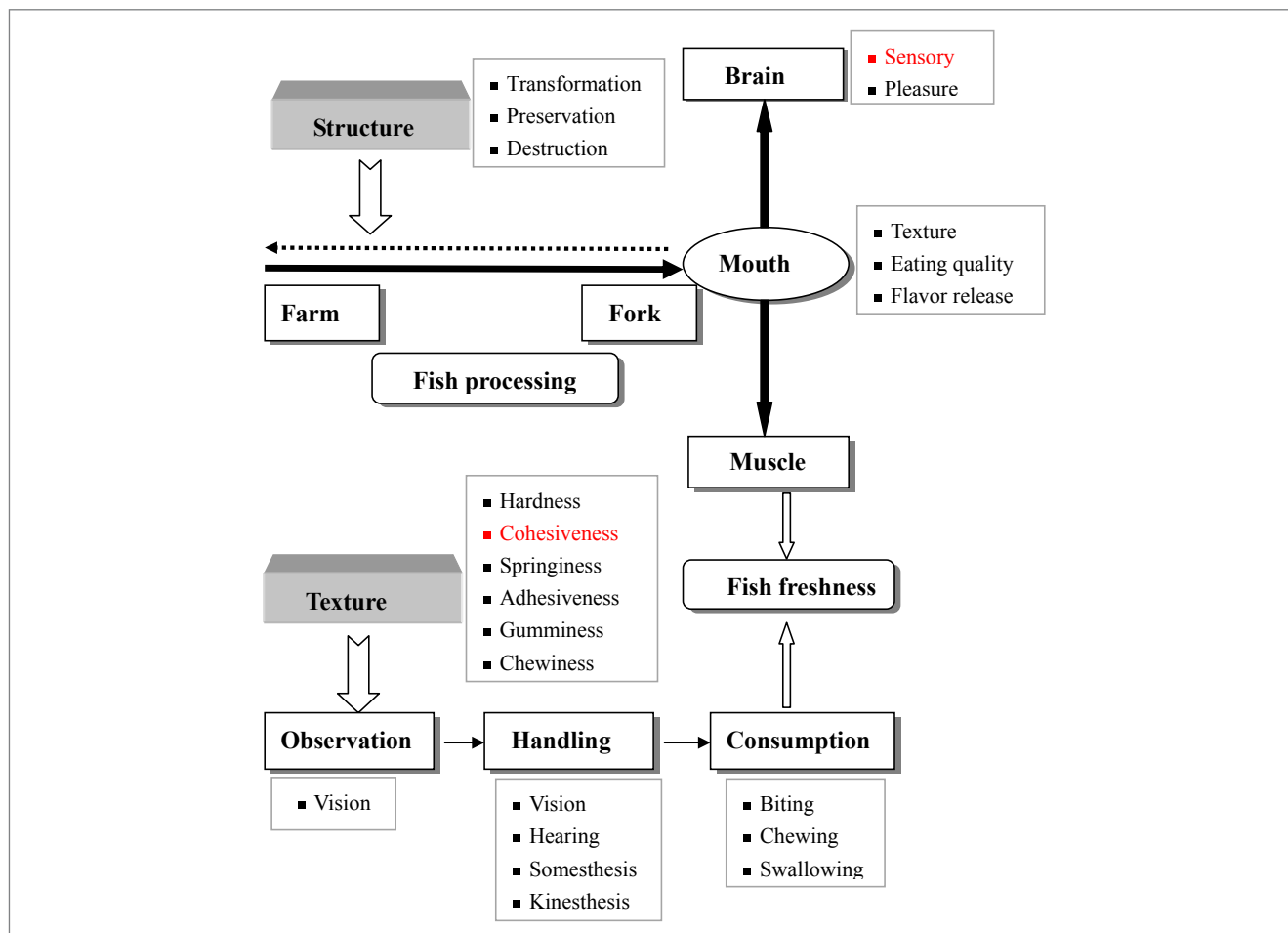


Figure 3—An understanding of fish texture and structure.

a universal testing machine with a speed of 3 mm/s, and it was recorded and observed that a significant decrease ( $P < 0.05$ ) in texture with an increase in storage time was obtained, showing the value of the necessary force ranging from 16.18 to 5.60 kgf. Jain and others (2007) also investigated the textural properties of Indian Rohu fish during ice storage and it was shown that the hardness of fish skin varied from 95.778 to 48.714 N for the period of 8 d of ice storage time. During 5 d of ice storage, there was a small difference in skin hardness and then the hardness was dropped obviously beyond 5 d storage. With respect to the value of stiffness, there was also a decrease from 4.634 to 2.003 N/mm during ice storage. Moreover, Ayala and others (2010) measured the textural parameters of the dorsal muscle of the left location of sea bream fillet by compression using a texturometer and the results indicated that, except springiness, all parameters changed with postmortem storage, and most of them decreased significantly ( $P < 0.05$ ) within 5 d of storage. In particular, the values of hardness, gumminess, and chewiness all decreased sharply by about one-half, in comparison with the values observed at prerigor. It was strange that there were no significant changes for the values of above 3 parameters in the subsequent stages, while adhesiveness increased significantly ( $P < 0.05$ ) with the increasing storage time. In a recent investigation, reported by Aussanasuwannakul and others (2012), the assessment of the firmness of rainbow trout fillet using the variable-blade attachment and the Allo-Kramer shear method was studied, and the data obtained from the experiments

showed that fillet refrigerated at 4 °C for 0 d was firmer than that preserved for 14 d.

**Effects of physicochemical properties.** The alterations of physicochemical properties of fish can straightforwardly affect textural quality. Mørkøre and Einen (2003) investigated the relationship between sensory data of Atlantic salmon and those of instrumental texture analyses using 4 different instrumental probes with different features of 12.5- and 23-mm-dia cylinders, a 25.4-mm-dia sphere, and a Warner-Bratzler blade. Results demonstrated that sensory hardness had a significant correlation with instrumental results, depending on the precise prediction of raw salmon by the 12.5-mm-dia cylinder, and of smoked salmon by the 23-mm-dia cylinder. In an earlier study, Andersen and others (1999) made a preliminary exploration on the relationship between fish textural features and the fat content. The Instron compression test was conducted to measure the textural properties of farmed rainbow trout based on fat content and ice storage time, and it was found that the high-fat group displayed less resistance against compression during the period of ice storage. Suárez and others (2005) studied the relation between fish firmness and the collagen content in sea bream muscle under different storage conditions. It was observed that the collagen content in fish muscle decreased somewhat through the whole storage time, which was closely linked to the firmness of fish muscle. With regard to collagen solubility, in the 1st few hours of postmortem, it was noted that the content of pepsin-soluble collagen increased a bit and acid-soluble collagen

Table 1—Measurements of fish texture and structure based on instrumental methods.

Fish name	Species	Test/properties	Instruments	Temperature	References
Atlantic salmon	<i>Salmo salar</i>	Compression	Texture analyzer, TA-XT2	Cold storage at 4 °C	Einen and Thomassen (1998)
Ray fish	<i>Dasyatis brevis</i>	Shear force	WB	Frozen at -86 °C	Ocaño-Higuera and others (2011)
Sea bream	<i>Sparus aurata</i>	Compression	Texturometer, QTS-25	Refrigerated at 4 °C	Ayala and others (2010)
Atlantic salmon	<i>Salmo salar</i>	Firmness	Texture analyzer, TA-XT2	Chilling regime at 0.4 °C	Sigholt and others (2006)
Rainbow trout	<i>Oncorhynchus mykiss</i>	Compression	Instron	Storage on the ice	Andersen and others (1999)
Gilthead sea bream	<i>Sparus aurata</i>	Compression	Texture analyzer, TA-XT2	Refrigerated at 2 °C	Ayala and others (2011)
Gilthead sea bream	<i>Sparus aurata</i>	Stress relaxation	Texture analyzer, TA-XT2	Ice bath at 0 to 2 °C	Campus and others (2010)
Sea bream	<i>Sparus aurata</i>	Compression	Texture analyzer, TA-XT2	Refrigerated at 4 °C for 14 d	Caballero and others (2009)
Sea bass fillets	<i>Dicentrarchus labrax</i>	Shear force	Texture analyzer, TA-XT2	Cold storage at 4 °C for 42 d	Fuentes and others (2012)
Atlantic salmon	<i>Salmo salar</i>	Compression	Texture analyzer, TA-XT2	Ice storage at 2 °C	Casas and others (2006)
Sea bream	<i>Sparus aurata</i>	Firmness	Texture analyzer, MA RK-10	Stored at -80 °C	Suárez and others (2005)
Rainbow trout	<i>Oncorhynchus mykiss</i>	Shear force	VB, AK	Refrigerated at 2 °C and frozen at -25 °C	Aussanasuwannakul and others (2010)
Rainbow trout	<i>Oncorhynchus mykiss</i>	Firmness	VB, AK	Refrigerated at 4 °C	Aussanasuwannakul and others (2012)
Abalone sections	<i>Haliotis</i>	Texture attributes	Texture analyzer, TA-XT2	Refrigerated at 4 °C	Briones-Labarca and others (2012)
Catfish	<i>Ictalurus punctatus</i>	Shear force	Texture analyzer, TA-XT2	Stored at 4 °C for 14 d	Rawdkuen and others (2010)
Grouper fillets	<i>Epinephelus coioides</i>	CSA	Microscope, CX21FS1	Refrigerated at 4 °C	Sharifian and others (2011)
Sea bream	<i>Sparus aurata</i>	CSA	LM, TEM	Refrigerated at 2 °C	Ayala and others (2011)
Pacific bluefin tuna	<i>Thunnus orientalis</i>	CSA	LM, TEM	Refrigerated at 4 °C for 5 d	Roy and others (2012)
Atlantic salmon	<i>Salmo salar</i>	CSA, ED	LM, TEM	Superchilled at -30 °C	Kaale and Eikevik (2012)
Abalone sections	<i>Haliotis</i>	CSA, ED	LM, SEM	Refrigerated at 4 °C	Briones-Labarca and others (2012)
Catfish	<i>Ictalurus punctatus</i>	CSA, ED	LM, SEM	Refrigerated at 4 °C for 14 d	Rawdkuen and others (2010)

CSA, cross-sectional area; ED, equivalent diameter; WB, Warner-Bratzler blade; VB, variable blade; AK, Allo-Kramer shear attachments; LM, light microscopy; TEM, transmission electron microscopy; SEM, scanning electronic microscopy.

content declined slightly, while insoluble collagen decreased starting from 96 h, corresponding to the loss of fish firmness. In addition, Jonsson and others (2001) measured textural properties and expressible moisture of fresh Atlantic salmon fillets on 7 locations along the fillet using 4 different instrumental methods and the results showed that, compared with the method using the flat-ended cylinder and Kramer shear compression cell, the puncture method with the spherical probe and the shearing device by Warner-Bratzler were more suitable for differentiating the textural properties of raw salmon fillets among different points. The expressible moisture exhibited a significant ( $P < 0.05$ ) linear correlation with the spherical probe and the Kramer shear compression cell with the correlation coefficient of 0.83 and 0.77, respectively. In another investigation, Hultmann and Rustad (2002) measured the textural properties and protein solubility of salmon and cod fillets during ice storage. It was reported that, in this storage condition, salmon muscle was easy to become softer and less elastic than that of cod, and the breaking strength and hardness of fillets both decreased, while cohesiveness of salmon increased, contributing to the solubility of protein mainly related to the increase of the fraction of salt-soluble proteins.

**Effects of handling methods.** Different handling methods play a significant role in fish texture measurements and dramatically influence the freshness of fish. Sigholt and others (2006) studied the ef-

fect of handling stress on the texture of farm-raised Atlantic salmon and showed that the handling stress had an effective influence ( $P < 0.001$ ) on the firmness of salmon fillet. Within the 0.4 °C chilling period, the breaking strength was lower in the stressed group than in the control group, while no difference emerged within the 3.3 °C chilling. In the meantime, Hagen and Solberg (2010) described the effect of fasting on the textural attributes of farmed Atlantic cod fillet, showing that fasting for 11 wk did not result in the loss of biomass, but an increase of 370% in the fillet texture. Compared to the fed control group, the texture of the fasting group was firmer by 15%. In order to investigate suitable methods for handling the fish prior to slaughter, Roth and others (2006) made a preliminary exploration on the texture changes of Atlantic salmon by 3 diverse handling methods related to stunned by electricity, stunned with CO<sub>2</sub>, and percussion prior to slaughter. Compared with the other 2 stunning methods, stunning with CO<sub>2</sub> method played an obvious role in the fish texture, resulting in an earlier onset and resolution of rigor mortis, thus accelerating the softening of the muscle tissue. In addition, Michalczyk and Surówka (2009) studied textural characteristics of trout muscle tissue under gravading treatment and reported that compared with untreated fish, treated samples showed larger cohesiveness and chewiness value as well as lower stress decay during relaxation. It could be concluded that from



the dynamic oscillatory measurements, the gravading process to a certain extent decreased the elastic properties of minced fish samples.

Packaging methods also affect fish product texture. Fuentes and others (2012) compared air, vacuum- and modified-atmosphere packaging methods on the textural properties of smoked sea bass under the condition of cold storage at 4 °C for 42 d, and it was revealed that different packaging methods posed a greater effect on the hardness, gumminess, and chewiness of fish samples, especially for vacuum- and modified-atmosphere packaging leading to visible decrease on textural parameters. In a recent work, Aubourg and others (2013) also studied the changes in textural properties of Atlantic mackerel during freezing and frozen storage pretreated by HPP. It was observed that hardness value of fresh muscle sample increased by 53.8 N after freezing, and after 3 mo of frozen storage, it also amounted to 65.1 N. Adhesiveness of the fresh sample was higher than that of frozen sample with the amplification of 38.8 gs, while the values of springiness and cohesiveness had no significant change. Concerning to the value of chewiness, it was gradually increased from 1.33 to 6.12 N during 1 mo frozen storage and then declined to 2.84 N after 3 mo.

**Effects of HPP.** HPP is a valuable and effective preservation technique that provides a low or moderate temperature for prolonging shelf life of foods and minimizing the loss of freshness quality by inactivation of microbial activities (Campus and others 2010). Hence, this technique has been extensively studied in fish industry. Reported by Chéret and others (2006), the effects of HPP (up to 500 MPa, 5 min) on the textural properties of sea bass fillets treated by different refrigerated storage time (0, 7, and 14 d) were studied. It was observed that when the treated pressure was above 300 MPa, higher values of fish hardness were obtained after storage than in the untreated sample, and it has been confirmed that this technique had the ability to improve the textural quality of refrigerated fish fillet. In another work, the effect of HPP on the texture of abalone muscle during cold storage was carried out by TPA test. The results of textural measurements obtained revealed that there was no significant difference ( $P > 0.05$ ) on hardness and springiness between control samples and treated samples with the pressure of 500 and 550 MPa at day 0, while a significant ( $P \leq 0.05$ ) difference on cohesiveness and chewiness occurred at day 0. At day 30, the firmness of control samples was radically affected by storage time, showing a sharp decrease by 94% of the hardness value in control samples and indicating that rubbery texture was developed in the control abalone tissue during storage (Briones-Labarca and others 2012).

Besides mechanical measurements of fish texture, there are some useful attempts on fish texture measurements using non-destructive methods and techniques such as image analysis (Hu and others 2012), low-field nuclear magnetic resonance spectroscopy for measurements of textural changes in fish muscle during frozen storage (Steen and Lambelet 1997), and digital imaging and stereoscopy technique for estimation of fish fillet firmness (Quevedo and Aguilera 2010; Grigorakis and Dimogianopoulos 2012). However, great challenges exist with respect to improving the accuracy and reliability of these methods for fish texture determination.

## Structure Measurements

As discussed earlier, unlike common meats such as chicken, beef, and pork, the muscle of fish has its own special structure

due to the alternating muscular sheets termed as myotomes that are separated and anchored by connective tissue. The following 2 typical combinations of reactions occur postmortem that lead to the softening of fish muscle: biochemically induced reactions related to the deterioration of myofibrils and collagen by enzymolysis, and physical reactions usually generating gaping owing to the isolation of myotomes, which result in the destruction of muscular structure (Chéret and others 2006). It is common knowledge that the period of rigor mortis starts after the fish dies and that the final fish freshness quality rests with this critical period. Originally, due to the separation of the myotomes, the mechanical phenomenon of gaping occurs, which is associated with the contraction of muscle fibers supported and maintained by the skeleton and the connective tissue. The collapse of the connective tissue over a certain pressure range can easily generate gaping. Hence, gaping is a consequence of the breakdown of muscle fibers and connective tissue to attach the muscle architectures along the fillet, which destroys the structure and microstructure of fish and results in the loss of freshness quality of fish and fish fillets (Hagen and others 2007).

## Measurement methods

As described and discussed earlier about fish structure, measurements of fish structure and microstructure are of great concern for further interpretation of fish freshness. Microscopy as an effective measurement tool has been widely used to control and probe the alterations of fish muscle structure and microstructure. There are 2 kinds of microscopes for food determination: optical microscopy and electron microscopy (James 2009). Optical microscopy can provide the simplest and most effective way to obtain enlarged images of muscle tissues and describe meat and meat product structures (Damez and Clerjon 2008). The use of electron microscopy can shed light on a specimen and produce a magnified image and provide a much greater resolution capability than that of optical microscopes. Common methods for investigating and detecting the exterior and interior structures are primarily related to transmission electron microscopy, scanning electron microscopy (Tunick 2010), confocal laser scanning microscopy (Wilkinson and others 2000), and environmental scanning electron microscopy (Dürrenberger and others 2001; Stokes 2003; Straadt and others 2007). Table 1 shows the applications of optical microscopy and electron microscopy for fish structure and microstructure measurements.

## Factors affecting structure measurements

The structure and microstructure measurements of fish are subject to the effects of internal and external factors (Figure 3) such as collagen cross-links, protein structure, and cold storage related to different freezing and chilling processes in combination of other techniques, as well as salting and smoking.

**Effects of cold storage.** It is a fact that fresh fish when well handled and kept at relatively low temperature scan slow down and restrain bacterial growth, enzymatic breakdown, lipid oxidation, and other forms of spoilage (Roy and others 2012). With the aim of improving the freshness quality of fish, diverse storage methods, such as flake ice and slurry ice, are necessary to prolong the shelf life of fish products and avoid the corruption of fish (Jain and others 2007). Moreover, it has been widely proved that refrigerated storage as one of the simplest preservative methods for short-time management and storage is broadly employed to maintain fish freshness before consuming it or using it for various technological processes and is also frequently used in supermarkets

or grocery stores for retail needs of fish muscle slices (Tan and Fok 2009; Sharifian and others 2011).

Effects of refrigerated storage on fish structure have been studied far and wide. Løje and others (2007) investigated the effect of chilled storage on the structure of smoked salmon fish, and the results showed that the cells of a smoked salmon sample were more firmly constrained than those of a control sample. As the growth of chilled storage of the smoked sample, the extracellular space between the cells gradually became wider. In the meanwhile, Sharifian and others (2011) studied the effects of cold storage on the microstructure of grouper fillets and results showed that, at day 0, the muscle fibers of control samples displayed fairly homogeneous and relatively normal shapes of the cross-section. After 7 d of cold storage, in comparison to the control, slight shrinkage of extracellular space and fibers were observed, and after 14 d of storage, these changes were enhanced, which were characterized as the degradation of per-cellular connective tissue. In another work, Roy and others (2012) measured the changes of structure and ultrastructure of cultured Pacific bluefin tuna muscle slices during chilled storage and indicated that the changes of the fish muscle slices were due to loss of myofibers to myofiber adhesion, detachment of the sarcolemma, increase of intermyofibrillar spaces, and adjustment of hexagonal arrangement of thick compared with thin contractile myofilaments in myofibrils. In addition, Briones-Labarca and others (2012) combined HPP treatment and cold storage to study the microstructure changes of abalone muscle and their results showed that unpressurized muscle of abalone exhibited a honeycomb structure that was encircled by thin and thick perimysium. However, pressure treatment played an important role in attacking the fish muscle fibers, causing rearrangement of perimysium and changes of the honeycomb structure due to the size decrease in endomysium. In another work by Alizadeh and others (2009), the impact of freezing methods on the microstructure of Atlantic salmon tissue was studied. Muscle fibers of unfrozen salmon sample showed relatively homogeneous and expected shapes. Slow freezing led to formation of relatively large ice crystals, thus seriously damaging the tissue. On the other hand, air-blast freezing ( $-30\text{ }^{\circ}\text{C}$ , 4 m/s) increased the freezing rate, resulting in a better preservation of the microstructure.

**Effects of salting and smoking.** Salting and smoking as effective means in the fish processing are usually used for control and improve the quality of fish and fish fillet, and further result in some influences on the muscle structure. Reported by Sigurgisladdottir and others (2000a), some variations were observed about the muscle microstructure treated by smoking fresh and freezing–thawing Atlantic salmon, showing that the frozen–thawed fish muscle fibers became shrunken and the extracellular space became larger compared to the fresh sample. The small muscle fiber diameters shrank to a lesser extent than those with larger fiber diameters. Consideration of the effect of smoking, the space between fiber and fiber and the fiber shrinkage was increased to a higher degree in salmon muscle that was frozen before smoking than muscle smoked from fresh salmon sample. In addition, Sigurgisladdottir and others (2000b) also studied the effects of diverse salting and smoking methods on the microstructure changes of salmon fillets and during the salting and smoking processes, the decrease of the cross-sectional area of the muscle fibers was noticed, and there was obvious difference in the cross-sectional area of fibers treated by dry-salted and brine-salted fish fillets. The cross-sectional area of fibers obtained from the former treatment was smaller than the latter treatment due to the more shrinkage during dry-salting process. In addition, in order to study the changes of transversal

and longitudinal sections of the fish muscle, a further study of examining the changes in the microstructure of fresh and smoked Atlantic salmon fillets with light microscopy and image process technique was carried out by Sigurgisladdottir and others (2008). It was noticed that, during the salting and smoking processes, salmon muscle fibers gradually shrank, while it seemed to have no changes about the length of sarcomere, and the dispersions of a large number of fat globules were observed among the muscle fibers after smoking. In a further study, Thorarinsdottir and others (2011) aimed to study the effects of different procedures of pre-salting and salting on the microstructure of cod product. It was observed that salting process resulted in the shrinkage of fiber diameter and enlargement of intercellular space, which were in accordance with the study of Løje and others (2007), despite not exactly the same processing methods.

**Effects of HPP.** As discussed previously, studies on the applications of HPP technique to evaluate fish freshness quality have been focused on the effects of this treatment on the changes of muscle structure. Campus and others (2010) studied the effects of this technique on structural changes of gilthead sea bream muscle tissues. Immunoblotting studies performed on the main structural proteins revealed that an obvious degradation of desmin could be prohibited when treated at 400 MPa, suggesting that high-pressure treatments caused negative enzyme activity on proteins with the functions of maintaining the tissue integrity and texture quality.

### Correlating Texture and Structure

Textural properties generally place emphasis on physical information and sensory perceptions of fish freshness, whereas the structure and microstructure focuses on subtle changes in internal characteristics and provides more information and further interpretation for textural alterations induced by external conditions such as high pressure, salting, smoking, freezing, and so on. Therefore, a close correlation exists between fish texture and structure. Damez and Clerjon (2008) related fish structure to textural sensory properties such as pastiness, crustiness, palatability, chewiness, juiciness, and tenderness. In fact, fish muscle texture closely associates with 2 factors: the myofibrillar structure strongly affected by farming and rearing situations and connective tissue. However, these 2 main factors play their respective roles in evaluation of the overall textural properties of fish muscle (Fuentes and others 2012). It has been confirmed that collagen fibers in connective tissue have the function of maintaining the textural toughness and the integrity of fish muscle. Meanwhile, gaping as a most damaging problem of texture commonly occurs when the muscle fibrils and collagen fibers collapse (Andersen and others 1999). Therefore, a large number of studies on the relationships between fish texture and structure have been reported for further interpretation of the loss of freshness.

Sea bream (*Sparus aurata*) is one kind of teleost fish widely distributed in the Mediterranean Sea and Atlantic Ocean and has attracted great interest in aquaculture due to its high commercial value. Suarez and others (2011) examined the effects of diverse storage temperatures on postmortem textural changes of sea bream muscle and showed that storage at  $1\text{ }^{\circ}\text{C}$  induced a prolongation of the firmness compared with  $4\text{ }^{\circ}\text{C}$ . Inversely, the rate of degradation of muscle collagen was faster at  $4\text{ }^{\circ}\text{C}$  compared with  $1\text{ }^{\circ}\text{C}$ , thus affecting the cross-linkage of connective tissue and becoming a major contributing factor to the firmness loss of fish texture. Furthermore, postmortem changes in the muscle of sea bream during ice storage have also been reported by Caballero and others

(2009), who showed that, within 7 d of storage, the parameters of hardness, gumminess, and chewiness showed greater changes than brittleness. Most importantly, the variability of muscle filament proteins including desmin, actin, and dystrophin, as well as endoproteases and the corresponding endogenous inhibitor detected by immunohistochemical technique indicated that the changes of muscle structure were interrelated with the textural properties during 14 d cold storage. Regarding the reasons, it could be explained that, after the fish died, the dystrophin disappeared quickly and this loss of dystrophin resulted in the detachment of myofibers and myocommata, and the reduction of textural hardness, while actin and desmin collapsed until the muscle tissue showed a deteriorated sensory appearance and texture. In addition, another study reported by Ayala and others (2011) was aimed to study the effects of 2 different treatment methods (1 group refrigerated at 2 °C, and the other group 1st vacuum-packed and then refrigerated) on the muscle structure and texture of gilthead sea bream fillets. It was observed that the group refrigerated had the lowest values, and most of the textural parameters were negatively correlated with the separations of fibers in both groups. Further study showed that the sarcolemma–endomysium of the 2nd group vacuum-packed was progressively disrupted and then vanished after 22 d.

Other fish species have also been investigated. The changes in texture and structure of Atlantic salmon muscle stored on ice were reported by Taylor and others (2006), who revealed that the loss of attachment of muscle fibers seriously caused the decrease in textural and this action was associated with the detachment of myofibers from the myocommata. Further interpretations about the changes were due to the fact that the destruction of fish fillet texture depended on 2 devastating breaks from several distinct structures, with the 1st break being from the looseness of cell cytoskeleton and the loss of fiber–fiber attachment, and later the other break being from the collapse of connective tissue and the detachment of fibers. In another work, Badii and Howell (2002) studied the changes in texture and structure of cod and haddock fillets stored at –10 and –30 °C. It was observed that fish muscle hardness increased with the growth of storage temperature and time, and the solubility, hydrophobicity, and aggregation of protein badly affected the protein denaturation and textural and structural changes treated during frozen storage. In addition, textural properties of farmed giant catfish muscle in different locations were investigated, and it was noticed that with an increase in storage time, there was continuous decrease in shear force for both dorsal and ventral parts (Rawdkuen and others 2010). The study also showed that, compared with the ventral part, the dorsal part had lower resistance to compression by the blade. The same trend of toughness with hardness values was observed in both dorsal and ventral parts. Decreasing toughness was obtained with increasing refrigeration time. Microstructures of both transverse and longitudinal sections of the fish muscles were also illustrated. The starting muscle (day 0) had a well-organized structure of the muscle bundle. After 1 wk of refrigerated storage, the muscle bundles were less attached and there was a loss of muscle interaction. After a period of time (day 7 to 14), muscle bundles became larger and showed gaps between them.

### Limitations and Challenges

Better understanding of fish structure and its changes during diverse treatments is essential to make significant advances in delivering more information on texture with excellent freshness quality. However, this can only be achieved by measuring techniques that

allow monitoring and quantifying these changes. According to the definition of texture, it is a typical sensory parameter and is extremely difficult to evaluate and judge accurately due to its complexity, multifactor features, and uncertainty. Sensory response to fish mechanical stimulus related to texture is different and can be easily influenced by adaptation, adjustment, and exhaustion, and also the level of the trained sensory panel. In contrast with instrumental measurements and analyses, sensory evaluation is time consuming, laborious, tedious, subjective, and expensive. Thus, it has been confirmed that sensory perception has its own limitation in assessing large samples and in/online evaluation and detection (Chen and Opara 2013). Moreover, sample pretreatment and preparation methods easily have an effect on the repeatability, reliability, and accuracy of texture measurements due to the fact that the fish fillet usually seems to be heterogeneous. Therefore, looking for a representative, desirable, and uniform sample for experimental research is rather difficult, and currently, a better approach for measurements of textural properties is cut the raw fish into fillets, which to a certain extent may depend upon the most appropriate location within the fillet. In addition, chemical constituents and nonhomogeneous distribution such as fat, moisture, and collagen of fish can differently affect the perception of texture features. Measurement of quality parameters often induces variations along the fish fillet from head to tail. Until recently, the measurement of food microstructure has been essentially based on light or electron microscopy, which often requires considerable sample preparation time, and due to its cost and complexity of the equipment, such a technique is mainly used for academic purposes but seldom in a food industrial environment. Hence, there is still much work to be done in understanding the role of texture and structure on the sensory properties of fish freshness.

### Conclusion

Texture and structure measurements and evaluations for fish and fish fillet are of significance in freshness quality control and assurance, and product development in the seafood industry. This review is aimed to indicate and evaluate the freshness quality of fish and fillets with the measurement of texture and structure. Both sensory and mechanical methods can be used for fish texture measurements. Compared with sensory methods, mechanical methods are more objective and reliable. Mechanical methods include puncture, compression, shear, and tensile techniques with Warner–Bratzler devices, Kramer shear compression cell, and the TPA test. Meanwhile, the main factors in terms of fish species, physicochemical properties, handling methods, and HPP on fish texture measurement are discussed and analyzed. The structure of fish plays a vital role in reflecting the internal subtle changes of connective tissue of fish muscle, usually measured by optical microscopy and electron microscopy. Most importantly, measurement of fish structure could provide more information and further interpretation for texture alterations, which are induced by external conditions such as freezing, chilling, salting, smoking, and others. Therefore, a close correlation has been observed and is here discussed between fish texture and structure. Although fish texture can be measured and evaluated through sensory and instrumental methods, it is difficult to come to an agreement on which is the best method and there is no single method universally accepted and applied in fish and fish product industry. Thus, computer vision and spectroscopy could be potential innovative methods for nondestructive and online measurements of fish texture and structure.



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