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Effect of transglutaminase concentration in curing solution on the physicochemical properties of salted large yellow croaker (*Pseudosciaena crocea*)

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ABSTRACT

This study investigated the effect of transglutaminase (TGase) added to curing solution on the physicochemical properties of salted fish. Large yellow croaker was salted in the curing solution containing 0–2.0% TGase at 10 °C for 48 h. The hardness, moisture content and immobilized water ratio of fish salted with 1.0% TGase were 629.94 g, 59.14%, and 95.34% respectively, which decreased with increasing or decreasing TGase concentration. The scanning electron microscopy image showed that a compact structure on the meat surface of fish salted containing 1.0% TGase. A similar microstructure was found in the internal meat of fish salted with 0.5% TGase. The hardness of fish salted with 0.5% TGase after roasting was 1135.97 g, which was higher than that of fish salted without TGase. In conclusion, high-quality salted large yellow croaker can be obtained by adding TGase in curing solution.

Introduction

Large yellow croaker (*Pseudosciaena crocea*) is an important economic marine-cultured fish in China (Li et al., 2020). The fish is rich in protein, EPA and DHA and is favored by consumers because of its tender texture and delicious taste (Zhou & Wang, 2019). The fish is mainly cultured in Ningde, Fujian, China, where the total annual output reached 204,576 tons in 2020 (Bureau, 2021). Large yellow croaker can be processed into various products, including salted fish, dried fish, and canned fish. In China, large yellow croaker is mainly processed into frozen products after being salted (Zhang, Yang, Guo, Li, & Chi, 2012). In Korea, yellow croaker (*Larimichthys polyactis*) is mainly processed as salted and dried products (Kim, Oh, Lee, Yoon, & Lee, 2020).

As a traditional method for seafood processing, salting not only provides the characteristic flavor of salted fish but also extends the shelf life of the product (Hong, Luo, Zhou, & Shen, 2012). Sensory quality is an important quality index of salted fish; salted products with high commercial value usually require high sensory scores on hardness and chewiness (Chen, Okazaki, Suzuki, Nguyen, & Osako, 2016). Texture attributes, such as hardness, of salted fish is related to protein function (Liang, Xie, Li, Luo, & Hong, 2021). During salting, the fish meat swells because salt ions in the curing solution weaken the interactions between the opposite charged side chains of proteins (Thorarinsdottir, Arason, Geirsdottir, Bogason, & Kristbergsson, 2002). Proteins solubilized in extracellular connective tissues induce swelling, which reduces the hardness of salted fish (Jiang, Nakazawa, Hu, Osako, & Okazaki, 2019). Therefore, researchers attempted to improve the texture properties of salted fish products by increasing the salt concentration in the curing solution (Chen, Okazaki, & Osako, 2016) and by drying the fish after salting (Kim et al., 2020).

Transglutaminase (TGase) is a safe and effective cross-linking agent that can catalyze the acyl-transfer reaction between glutamine and lysine residues to form intra and intermolecular protein aggregates (Li, Gui, Huang, Feng, & Luo, 2018). The hardness and breaking strength of salted Alaska pollock roe were increased by adding 0.5% TGase in the curing solution (Chen, Takahashi, Geonzon, Okazaki, & Osako, 2019). Studies have investigated improvements in the physical properties of restructured meats and products (Lee, Jang, Kang, & Chin, 2017) and surimi gels (Li et al., 2018) by adjusting TGase; however, limited information is available on the effect of TGase on the physicochemical properties of salted large yellow croaker.

The addition of sorbitol and polyphosphate in curing solution can

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improve the salting effect on meat products (Antoine, Marshall, Sims, O'keefe, & Wei, 2000; Casales & Yeannes, 2016). In the present study, TGase was added to curing solution containing 15% sorbitol, 10% salt, 0.5% sodium tripolyphosphate, and 0.5% sodium pyrophosphate. Texture profile analysis (TPA) and determination of moisture content, water holding capacity (WHC), cooking loss, and water distribution in salted large yellow croaker were conducted. The secondary structure, thermal stability, and microstructure of proteins in salted fish were evaluated to reveal the effect of TGase on the characteristics of the surface and inside meat. Finally, the effect of TGase in the curing solution on the texture properties of roasted fish meat was investigated.

Materials and methods

Materials

Freshly dead large yellow croakers were purchased from Ningde Sanfang Paddy Field Aquatic Products Co., Ltd (Fujian, China). The average length and weight of the croakers were 32.39 ± 1.25 cm and 377.96 ± 12.63 g, respectively. The croakers were placed in a polyethylene foam box with crushed ice and transported to the laboratory within 24 h. TGase (100 U/g) was purchased from Nanning Pangbo Biological Engineering Co., Ltd. (Nanning, China). Salt, sorbitol, sodium tripolyphosphate, and sodium pyrophosphate were of food grade. All other chemical agents used were of analytical grade.

Preparation of salted large yellow croaker

The croakers were descaled, beheaded, eviscerated, filleted, and washed thoroughly in running water. The fillets were salted in curing solution containing 0.5% sodium tripolyphosphate, 0.5% sodium pyrophosphate, 10% salt, and 15% sorbitol added with various concentration of TGase (0–2.0%, w/v) at 10 °C for 48 h. The ratio of fillet to the curing solution was 1:3 (w/v). The fillet samples were salted and drained for 5 min for subsequent analysis.

TPA

TPA was performed according to the method reported by Sáez et al. (2020) with some modifications. Small pieces (20 mm × 20 mm × 15 mm) were cut from the salted fillets and measured by a texture analyzer (TA-XT Plus, Surrey, UK) equipped with a 36 mm cylindrical probe (P/36R). The samples were compressed for two cycles with an interval of 5 s and a trigger force of 5 g to 30% strain at a constant speed of 1.0 mm/s. TPA parameters including hardness, chewiness, springiness, and cohesiveness were calculated from the force–deformation curves. The TPA of the roasted fish was also measured after the salted croakers were roasted at 180 °C for 20 min and cooled to room temperature.

Determination of moisture content, WHC, and cooking loss

The fillet sample was dried at 105 °C for 24 h to a constant weight, and moisture content was determined according to AOAC International (2005).

WHC was determined according to the method descried by Chen et al. (2020) with slight modifications. The fillet samples (about 2 g) were wrapped with filter paper, placed at the bottom of the centrifuge tube, and centrifuged at 3000 × g for 20 min at 4 °C. WHC was expressed as percentage of water loss from the fillet samples.

Cooking loss was measured according to the method of Feng, Bansal, and Yang (2016) with slight modifications. The fillet samples (about 2 g) were placed in a beaker filled with 50 mL of water, incubated at 95 °C for 10 min, and then cooled on ice. Water on the surface was wiped by filter paper. Cooking loss was expressed as percentage of water loss from the fillet samples.

Low-field nuclear magnetic resonance (LF-NMR)

LF-NMR measurement was carried out with an LF-NMR analyzer (MesoMR20-060H-I, Suzhou Niumag Analytical Instrument Co., Suzhou, China) according to the method described by Tan, Ye, Chu, and Xie (2021). The fish meat samples (2.0 cm × 2.0 cm × 1.5 cm) were examined in a 40 mm-diameter cylindrical glass tube. Transverse relaxation time (T_2) was measured using the Carr–Purcell–Meiboom–Gill (CPMG) sequence at 32 °C under four scans and 6000 echo repetitions. The acquisition parameters were set as follows: spectral width of 200 kHz, preamplifier gain of 0, sampling repetition time of 3000 ms, and echo time of 0.30 ms. T_2 and peak ratio (P_2) were obtained through transformation of the CPMG exponential decay curve by using simultaneous iterative reconstruction technique.

Sodium dodecyl sulfate–polyacrylamide gel electrophoresis (SDS-PAGE)

SDS-PAGE was performed using 4% stacking gel and 8% separating gel according to the method of Fang et al. (2021). The fish meat (0.5 g) was homogenized with 50 mL mixture of 2% β-mercaptoethanol, 8 mol/L urea, 2% SDS, and 20 mmol/L Tris-HCl (pH 8.8). The homogenate was agitated at room temperature for 24 h and then centrifuged at 10,000×*g* for 15 min. The supernatant was mixed with SDS-PAGE sample buffer containing 4% SDS, 20% glycerol, with or without 10% β-mercaptoethanol, and 0.125 mol/L Tris-HCL (pH6.8) to a final protein concentration of 1 mg/mL. The samples (8 μg) were loaded on the polyacrylamide gel and subjected to electrophoresis at a constant current of 8 mA per gel. After electrophoresis, the gel was stained with 0.025% (w/v) Coomassie Bright Blue R-250 and destained with 30% methanol and 10% acetic acid.

Fourier-transformed infrared spectroscopy (FTIR)

The FTIR spectra of the freeze-dried samples were recorded using an ATR-FTIR spectrometer (Nicolet iS50, Thermo Nicolet Ltd., USA) following the method of Tan et al. (2021). The FTIR spectra were collected within the wavenumber of 4000–400 cm^{-1} with a resolution of 4 cm^{-1} and a scanning frequency of 32. Single-beam spectra of the samples were obtained against air as background. The OMNIC 6.0 data collection software program (Thermo Fisher Scientific Inc., USA) was used to convert the ATR spectra to absorbance and analyze the position, peak height, and area of the spectral band.

Differential scanning calorimetry (DSC)

The thermodynamic properties of the salted large yellow croakers were measured using a differential scanning calorimeter (Q2000, TA Instruments, New Castle, DE, USA) following the method in a previous report (Tan and Xie, 2021). The samples (approximately 5 mg) were accurately weighed and tightly sealed in aluminum pans. An empty sealed aluminum pan was used as reference. The thermal transition temperature (T_m) of the samples was measured from 20 °C to 95 °C at a heating rate of 5 °C/min.

Scanning electron microscopy (SEM)

The microstructures of the salted large yellow croakers were determined using SEM as previously described by Fang et al. (2021). Slices (4.0 mm × 4.0 mm × 2.5 mm) were cut from the surface and inside of the salted fish meat and immersed in fixative solution (2.5% glutaraldehyde, 0.1 mol/L phosphate buffers, pH 7.2) at 4 °C for 24 h. The fixed samples were thoroughly rinsed with 0.1 mol/L phosphate buffer (pH 7.2) and gradually dehydrated in gradient concentrations of ethanol solution. The dehydrated samples were dried in a vacuum freeze dryer (FD-1–50, Biocool Corporation, Beijing, China). The samples were fixed on the copper specimen holder with conductive adhesive and sprayed

with a layer of gold by using a sputter coater (Sputter Coater SC7620, Quorum, England). The microstructures of the samples were observed using a scanning electron microscope (Phenom-World Pro, Netherlands) at an acceleration voltage of 10 kV.

Statistical analysis

All experimental data were analyzed by one-way ANOVA and Duncan's multiple-range tests on SPSS 17.0 software (SPSS Inc., Chicago, IL, USA). The significance level of the difference among treatments was set at $p < 0.05$.

Results and discussion

TPA

The effect of TGase concentration on the texture of salted large yellow croaker is shown in Table 1. After the croakers were salted in the curing solution containing salt, sorbitol, sodium tripolyphosphate, and sodium pyrophosphate, the hardness of the sample was 423.84 g, which increased by up to 629.94 g when the curing solution was added with 1.0% TGase. Similarly, Chen et al. (2019) reported that the mechanical properties of salted Alaska pollock roe were improved by adding TGase in the curing solution due to the formed covalent cross-linking among proteins. However, the hardness slightly decreased with further increase in the TGase concentration. The cross-linking on the fish meat surface possibly inhibited the penetration of TGase into the internal meat, resulting in decreased hardness. A similar change in chewiness was observed in the salted croaker. A high chewiness level is related to the compact protein network structure of gels (Kou et al., 2018). Thus, the addition of an appropriate amount of TGase to curing solution could improve the compact network structure of the fish meat (Table 1). However, no significant differences in springiness and cohesiveness were found among croakers cured with various TGase concentrations.

Moisture content, WHC, and cooking loss

The effect of TGase concentration on the moisture content of salted large yellow croaker is shown in Fig. 1(A). The moisture content in the croaker salted in the curing solution containing 0.5% TGase was 46.93%, which was similar to that in the salted fish without TGase. The moisture content in the fish cured with 1.0% TGase reached 59.14% but slightly decrease when the TGase concentration was increased. In general, the texture of salted fish products is related to moisture content; that is, low moisture content leads to high hardness (Yang et al., 2017). In the present study, the salted fish meat had high moisture content of 46.93%–59.14% and hardness of more than 550 g (Table 1 and Fig. 1A). Hence, the addition of TGase to curing solution could improve the texture of salted fish probably due to the formed covalent cross-linking among proteins. The WHC of the fish meat refers to the ability of fish meat to maintain water under the action of external forces and reflects the ability of proteins to bind to water (Jiang, Jia, Nakazawa, Hu, Osako, & Okazaki, 2019). The TGase concentration in the curing solution had no significant effect on the WHC of the salted fish (Fig. 1B), indicating that TGase did not affect the interaction between protein and water. These findings are consistent with previous reports of Zhao, Wang et al. (2021), who investigated the effect of TGase on microwave 3D-printed surimi products. Furthermore, the water loss of the salted fish meat during cooking gradually decreased with increasing TGase concentration (Fig. 1C). Hence, the formation of protein cross-linking on the fish meat surface induced by TGase could hinder water loss from the internal fish meat. Consumers expect the prepared product to have high moisture content but low cooking loss rate.

Table 1
Effects of TGase concentration on TPA parameters of salted large yellow croaker.

TGase concentration (%)	Hardness (g)	Chewiness	Springiness	Cohesiveness
0	423.84 ± 35.55^{c}	166.00 ± 6.09^{b}	0.66 ± 0.03^{a}	0.59 ± 0.02^{a}
0.5	582.18 ± 25.38^{b}	218.36 ± 31.16^{a}	0.64 ± 0.05^{a}	0.58 ± 0.02^{a}
1.0	629.94 ± 27.26^{a}	213.00 ± 23.48^{a}	0.59 ± 0.05^{a}	0.57 ± 0.02^{a}
1.5	552.64 ± 22.38^{b}	223.23 ± 25.42^{a}	0.67 ± 0.05^{a}	0.61 ± 0.03^{a}
2.0	560.23 ± 15.00^{b}	195.50 ± 10.63^{ab}	0.61 ± 0.03^{a}	0.57 ± 0.02^{a}

Data are expressed as mean ± standard deviation.
Values with different letters in the same column indicate significant differences at $p < 0.05$.

LF-NMR

The water mobility and distribution in the salted large yellow croaker meat were analyzed using LF-NMR technology (Table 2 and Fig. S1). In general, the relaxation time of T_{21} corresponds to bound water that combines tightly with protein, T_{22} refers to immobilized water trapped in a protein network, and T_{23} corresponds to free water outside the protein network (Zhao, Chen et al., 2021). No significant difference was found in the T_{21} of the fish meat salted with<1.5% TGase, while the mobility of bound water in the fish meat salted with 2.0% TGase was higher than that in the other samples. This finding could be due to the incomplete removal of easily flowing bound water by the low salt concentration, and the penetration of salts was inhibited by the formed protein crosslinking on the fish meat surface at high TGase concentration. The T_{22} and T_{23} of the fish meat salted with 0.5% TGase were higher than those of the other samples and gradually decreased with decreasing or increasing TGase concentration in the curing solution. Hence, the mobility of the immobilized and free water was inhibited by the formation of protein network on the meat surface.

The peak ratios of bound water (P_{21}), immobilized water (P_{22}), and free water (P_{23}) in the salted large yellow croaker without TGase were 7.72%, 91.34%, and 0.95%, respectively (Table 2), which were similar to those in stinky mandarin fish fermented with 6% salt at 15 °C for 5 d (Yang et al., 2017). The free water mobility was decreased by salting; the salt induced the swelling of myofibrillar proteins, leading to the diffusion and immobilization of free water into meat fibers (Qin et al., 2017). Therefore, the peak ratio of P_{22} in the salted croaker without TGase was close to that of fresh seabass (Zhao, Chen et al., 2021). In the present study, the peak ratio of P_{21} in the salted croaker was decreased, while those of P_{22} and P_{23} were increased significantly by the addition of TGase up to 1.0% (Table 2). Hence, the ratios of water with different mobility levels were mainly affected by the formation of protein networks in the internal fish meat. Dong et al. (2020) investigated the effect of TGase on the 3D printing quality of surimi, they suggested that TGase-induced protein–protein interaction was more superior than protein–-water interaction.

SDS-PAGE

The protein patterns of the salted large yellow croaker are shown in Fig. S2. The band intensity of high-molecular-weight fractions (HMWF) on the meat surface of the salted fish without TGase was less than that of the sample with TGase. No significant difference was found in the band intensity of MHC on the meat surface of fish salted with 0–1.5% TGase, which was higher than that in fish meat salted with 2.0% TGase (Fig. S2). Hence, HMWFs were formed through cross-linking by TGase during curing. However, different results were found in the internal meat samples of the salted fish. The MHC band intensity in fish salted with 0.5% TGase was the least, but it increased with increasing TGase concentration in the curing solution. Only the HMWF band intensity in fish salted with 2.0% TGase was less than that in fish salted without

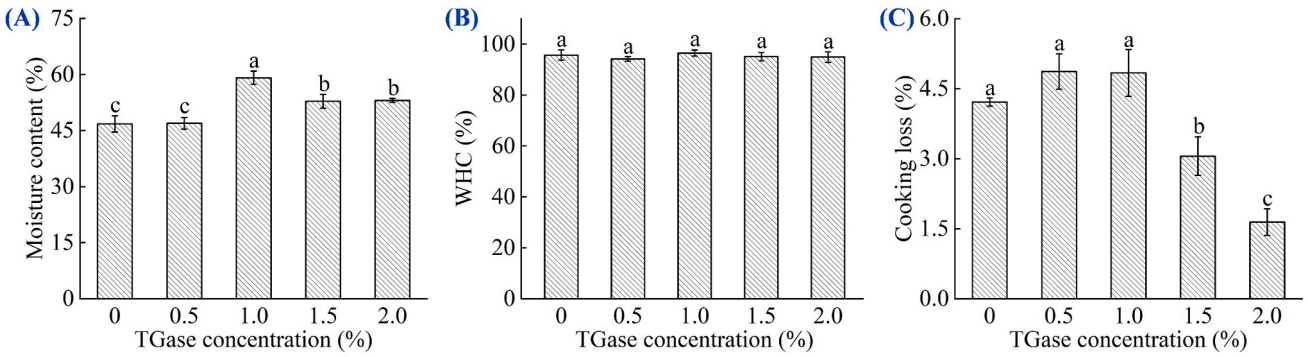

Fig. 1. Effect of TGase concentration on moisture content (A), WHC (B) and cooking loss (B) of salted yellow large croaker. Error bars show standard deviation. Different letters indicate significant differences at $p < 0.05$.

Table 2
Effects of TGase concentration on the LF-NMR parameters of salted large yellow croaker.

TGase concentration (%)		0	0.5	1.0	1.5	2.0
Transverse relaxation time (ms)	T_{21}	0.31 ± 0.02^{b}	0.26 ± 0.02^{b}	0.23 ± 0.02^{b}	0.28 ± 0.09^{b}	0.66 ± 0.08^{a}
	T_{22}	67.56 ± 5.18^{b}	79.42 ± 6.54^{a}	75.65 ± 3.12^{a}	53.50 ± 5.27^{c}	37.89 ± 5.28^{d}
	T_{23}	758.31 ± 41.00^{b}	902.24 ± 60.74^{a}	750.56 ± 64.21^{b}	730.99 ± 49.41^{b}	741.77 ± 48.73^{b}
Peak ratio (%)	P_{21}	7.72 ± 1.23^{a}	4.68 ± 0.52^{b}	3.36 ± 0.35^{c}	3.58 ± 0.29^{c}	3.20 ± 0.17^{c}
	P_{22}	91.34 ± 1.94^{c}	93.37 ± 3.65^{b}	95.34 ± 3.21^{a}	95.73 ± 4.98^{a}	95.68 ± 1.84^{a}
	P_{23}	0.95 ± 0.05^{c}	1.96 ± 0.08^{a}	1.31 ± 0.13^{b}	0.99 ± 0.18^{c}	1.02 ± 0.22^{c}

Data are expressed as mean ± standard deviation.
Values with different letters in the same column indicate significant differences at $p < 0.05$.

TGase. Fig. S2 suggests that TGase promoted the cross-linking of proteins on the meat surface of the fish. The high concentration of TGase inhibited the penetration of TGase and reduced the cross-linking of proteins inside the fish meat during curing.

FTIR spectroscopy

The amide A band represents N—H, C—H, and O—H groups involved in hydrogen bond formation, while the amide B band originates from methylene (—CH_2) asymmetric stretching (Sun et al., 2021). The peak at 1744 cm^{-1} belongs to the stretching vibration of C═O, which is associated with lipids in fish meat (Liang et al., 2020). The amide I band is mainly attributed to the C═O stretching vibration, C—N stretching, and N—H bending of proteins. The amide II band arises from the N—H bending and C—N stretching vibrations of peptide bonds (Candoğan, Altuntas, & İğci, 2021). Furthermore, changes in the intensity at peak height indicate changes in the contents of corresponding functional groups (Cebi, Durak, Toker, Sagdic, & Arici, 2016).

As shown in Fig. 2(A), typical absorption peaks of amide A, amide B, amide I and amide II were found on the meat surface of the croaker salted without TGase. No obvious changes in typical absorption peaks were observed among samples salted in curing solutions containing 0.5%–1.5% TGase. When the curing solution was added with 2.0% TGase, only the amide I absorption peak shifted from 1647 cm^{-1} to 1654 cm^{-1} (Fig. 2A) probably due to the increased number of N—H groups through the indirect effect of TGase (Liu et al., 2017). Compared with the surface meat of fish salted without TGase, the intensity of amide A in internal meat sample decreased significantly, while the intensities of peaks at 1744 cm^{-1} and amide II increased. Hence, the internal meat had lower water content but higher fat and protein contents than the surface meat. In the internal fish meat sample, the absorption peak at 2851 cm^{-1} is caused by the methylene (—CH_2) symmetric stretching, which is also associated with high lipid contents (Volpe, Coccia, Siano, Di Stasio, & Paolucci, 2019). Furthermore, the characteristic absorption peaks at 1078, 1042, and 889 cm^{-1}, which represent phosphate and sorbitol (Liang et al., 2020), only appeared in the surface meat sample. In addition, the addition of TGase to the curing solution had no significant effect on the FTIR spectra of the internal meat sample of salted fish.

DSC

The thermal transition temperature (T_m) of salted large yellow croaker was measured by DSC (Fig. 2C and 2D). Two characteristic peaks were observed at 41.77 °C–49.06 °C and 59.88 °C–74.30 °C, corresponding to the denaturation temperatures of myosin and actin, respectively (Sun, Sun, Xia, Xu, & Kong, 2019).

In the surface meat sample, the T_m values of myosin and actin in the salted fish without TGase were 41.94 °C and 59.77 °C, respectively, which were increased to a certain extent by the addition of TGase to the curing solution (Fig. 2C). The T_m values of myosin and actin in the internal meat of salted fish without TGase were 48.32 °C and 69.46 °C, respectively (Fig. 2D). A similar phenomenon was reported by Thorarinsdottir et al. (2002), who found that the T_m values of myosin and actin in the salted cod (*Gadus morhua*) fillets were approximately 50 °C and 70 °C, respectively. TGase concentration had no significant effect on the T_m of myosin in the internal meat sample of the salted fish. The T_m of actin gradually increased with increasing TGase concentration in the curing solution. The T_m of protein aggregates increased after being catalyzed by TGase (Wang, Liu, Ye, Wang, & Li, 2015). This phenomenon could be due to the fact that myofibrillar proteins on the meat surface can easily absorb water and swell during curing, thereby decreasing the protein content per unit volume and the effect of TGase. Therefore, the thermal stability of the surface meat was significantly lower than that of internal meat under the same curing condition (Fig. 2C and D).

SEM

The microstructures of salted fish meat were evaluated (Fig. 3). In the surface meat of fish salted without TGase, loose and bunched microstructures were observed. Jiang et al. (2019) found a similar phenomenon in salted bigeye tuna (*Thunnus obesus*) meat and reported that it could be due to the swelling of the filament lattice. When croakers were salted in curing solution containing 0.5% TGase, fiber bundles were formed on the meat surface because TGase induced the aggregation of myofibrillar proteins (Liang et al., 2020). When the TGase concentration in the curing solution was increased to 1.0%, a compact and

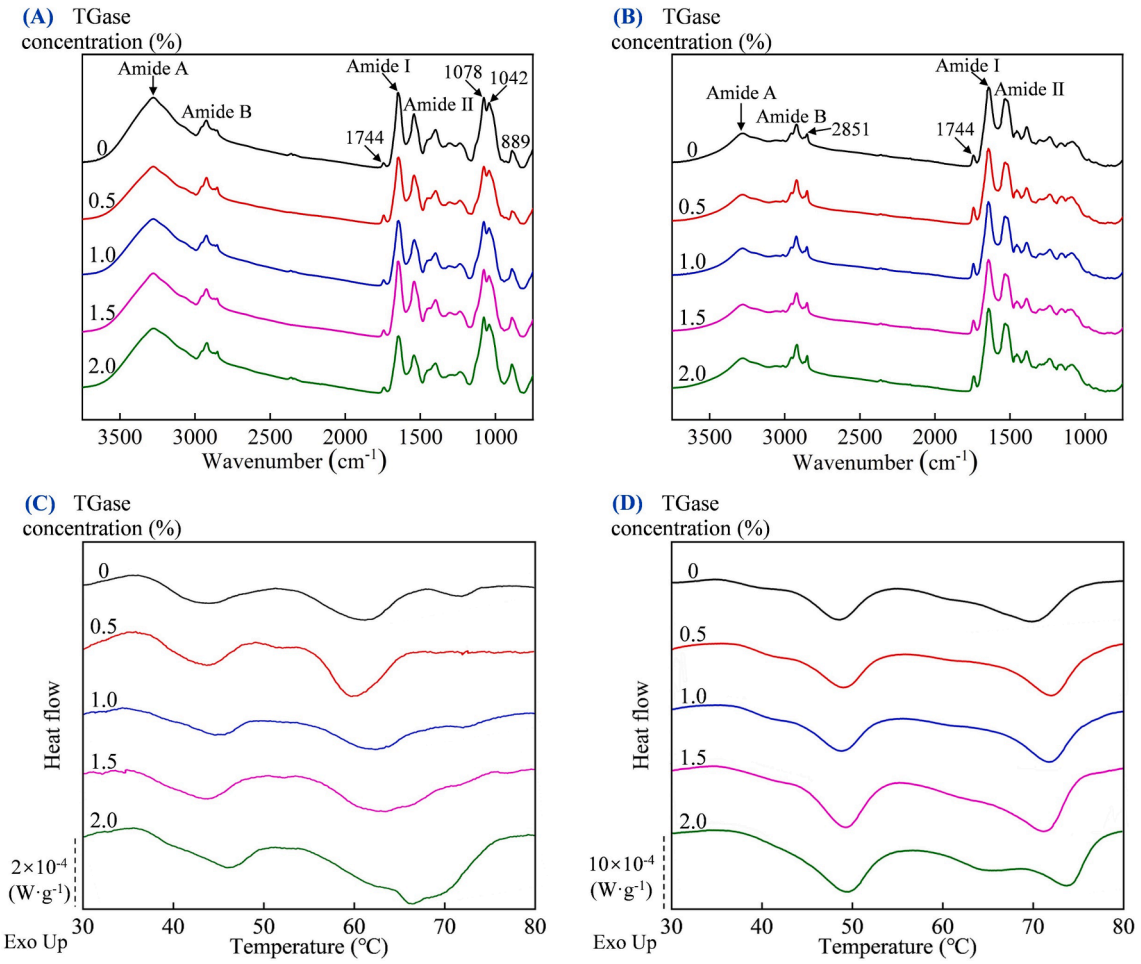

Fig. 2. FTIR spectra and DSC thermograms of salted yellow large croaker meat (A: FTIR spectra of salted fish meat surface; B: FTIR spectra of salted fish meat inside; C: DSC thermograms of salted fish meat surface; D: DSC thermograms of salted fish meat inside). (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

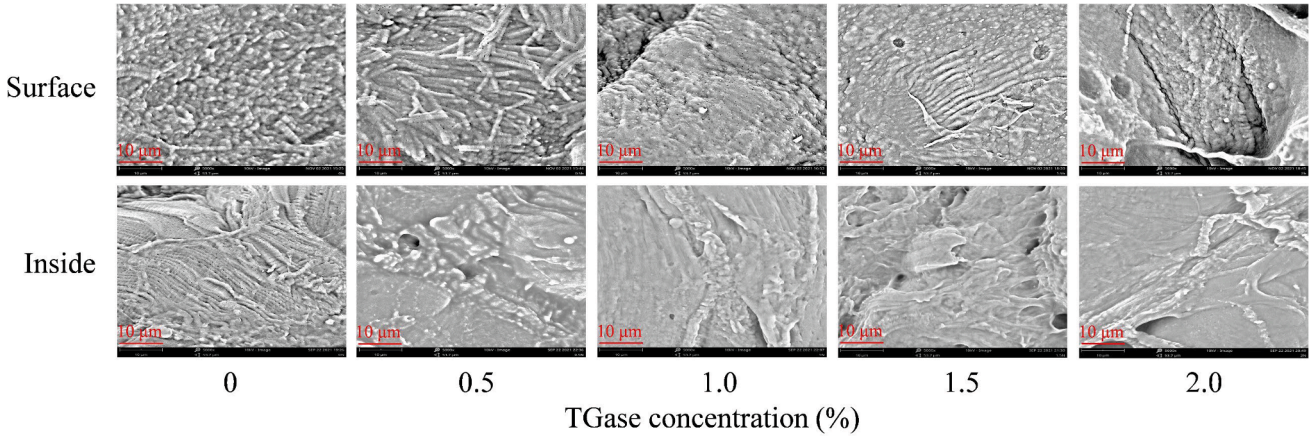

Fig. 3. SEM images of salted yellow large croaker (5000× magnifications). (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

creased perimysium structure appeared on the fish meat surface and fiber bundles disappeared. However, no obvious changes were observed when the TGase concentration was further increased. Hence, the protein aggregates formed in the curing solution with 1.0% TGase were sufficient to inhibit the fracture of the fish myofibrillar structure during salting. Compared with the surface meat sample, a certain regular fiber structure was found in the internal sample of fish salted without TGase, and the gap between fibers was smaller. When the curing solution was added with 0.5% TGase, the formed fiber structures disappeared and compact structures were formed by fiber aggregates. When the concentration of TGase in the curing solution was further increased, no obvious changes were found in the microstructure of the salted fish

meat. The network structure in the internal meat sample was significantly denser than that on the surface meat at the same TGase concentration added to the curing solution. This finding could be due to the fact that the water-soluble components were wrapped by the internal myofibrillar protein network (Xia et al., 2019). The water absorption and swelling degree of internal myofibrillar proteins were lower than those of surface proteins. In general, the more compact the structure is, the higher the hardness of the salted fish meat will be (Yalçın & Şeker, 2016). The highest hardness was found in the fish sample salted with 1.0% TGase (Table 1). The microstructure in the surface and internal fish samples salted with more than 1.0% TGase was similar to that of fish salted with 1.0% TGase, but the hardness decreased to a certain extent. This finding is mainly due to the fact that the cross-linking of internal meat proteins was inhibited by the increased TGase concentration in the curing solution (Fig. S2). The dense myofibrillar protein network was reported to be conducive to bind to immobilized water in red seabream (*Pagrus Major*) fillets (Cao et al., 2018). A similar phenomenon was observed in the present study; that is, the immobilized water content in the fish meat salted without TGase was lower than that in the samples with TGase (Table 2) due to the formation of dense networks.

TPA of roasted fish

The texture properties of the salted croaker after roasting were analyzed (Table 3). The hardness of the roasted fish meat salted without TGase was 871.15 g, which was increased to 1135.97 g when the curing solution was added with 0.5% TGase. Hence, the addition of TGase affected the texture properties of the roasted croaker. However, no significant differences in hardness was found among roasted fish salted with curing solution containing 0.5%–2.0% TGase. The cross-linking and network structure formed by the protein in the internal meat were relatively close when the curing solution contained more than 0.5% TGase (Figs. S2 and 3). Moreover, the myofibrils of the salted fish shrank after roasting (Warner, Ha, Sikes, & Vaskoska, 2017), thereby decreasing the effect of TGase on hardness. A similar trend in the chewiness of the roasted fish meat was found, but no significant differences in springiness and cohesiveness were observed. Meanwhile, the hardness and chewiness of the roasted fish meat were significantly higher than those of the samples before roasting (Tables 1 and 3) probably due to water loss and protein denaturation during roasting (Sun et al., 2020).

Conclusion

The quality of salted large yellow croaker can be improved by salting in curing solution with TGase. The hardness, chewiness, and moisture content of croaker salted with curing solution containing 1.0% TGase were higher than those in the other samples and gradually decreased with decreasing or increasing TGase concentration. TGase could promote the covalent cross-linking of MHC, form dense protein network structures, and reduce the mobility of immobilized water and free water during curing. When the curing solution was added with more than 1.0% TGase, a compact perimysium structure was formed on surface fish meat. The structure inhibited the penetration of TGase, resulting in reduced protein cross-linking in the internal fish meat. Hence, the addition of 1.0% TGase to the curing solution not only produces salted croaker with good texture but also ensures better texture of roasted fish. In future studies, large yellow croaker will be pretreated with phosphate solution containing TGase to penetrate into fish meat first and obtain high meat quality.

CRediT authorship contribution statement

Li Huang: Methodology, Data curation. **Linfan Shi:** Writing – review & editing. **Zhongyan Ren:** Supervision. **Gengxin Hao:** Supervision. **Wuyin Weng:** Conceptualization, Writing – review & editing.

Table 3
Effects of TGase concentration on TPA parameters of salted large yellow croaker after roasting.

TGase concentration (%)	Hardness (g)	Chewiness	Springiness	Cohesiveness
0	871.15 ± 52.91^{b}	204.12 ± 22.77^{c}	0.57 ± 0.06^{a}	0.41 ± 0.07^{a}
0.5	1135.97 ± 64.25^{a}	240.44 ± 22.42^{b}	0.56 ± 0.04^{a}	0.39 ± 0.05^{a}
1.0	1229.24 ± 93.22^{a}	291.24 ± 21.52^{a}	0.60 ± 0.02^{a}	0.40 ± 0.04^{a}
1.5	1260.69 ± 97.16^{a}	297.29 ± 28.40^{a}	0.61 ± 0.02^{a}	0.38 ± 0.05^{a}
2.0	1266.77 ± 94.71^{a}	303.96 ± 29.50^{a}	0.59 ± 0.03^{a}	0.41 ± 0.05^{a}

Data are expressed as mean ± standard deviation.
Values with different letters in the same column indicate significant differences at $p < 0.05$.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.fochx.2022.100277.

References

Antoine, F. R., Marshall, M. R., Sims, C. A., O'keefe, S. F., & Wei, C. I. (2000). Phosphate pretreatment on smoke adsorption of cold smoked mullet (*Mugil cephalus*). *Journal of Aquatic Food Product Technology, 9*(2), 69–81. https://doi.org/10.1300/J030v09n02_07

Candoğan, K., Altuntas, E. G., & İğci, N. (2021). Authentication and quality assessment of meat products by fourier-transform infrared (FTIR) spectroscopy. *Food Engineering Reviews, 13*(1), 66–91. https://doi.org/10.1007/s12393-020-09251-y

Cao, M., Cao, A., Wang, J., Cai, L., Regenstein, J., Ruan, Y., & Li, X. (2018). Effect of magnetic nanoparticles plus microwave or far-infrared thawing on protein conformation changes and moisture migration of red seabream (*Pagrus major*) fillets. *Food Chemistry, 266*, 498–507. https://doi.org/10.1016/j.foodchem.2018.06.057

Casales, M. R., & Yeannes, M. I. (2016). Mass transfer modeling during marination of anchovy fillets in ternary and multicomponent solutions. *Journal of Aquatic Food Product Technology, 25*(5), 760–769. https://doi.org/10.1080/10498850.2014.924168

Cebi, N., Durak, M. Z., Toker, O. S., Sagdic, O., & Arici, M. (2016). An evaluation of Fourier transforms infrared spectroscopy method for the classification and discrimination of bovine, porcine and fish gelatins. *Food Chemistry, 190*, 1109–1115. https://doi.org/10.1016/j.foodchem.2015.06.065

Chen, J., Deng, T., Wang, C., Mi, H., Yi, S., Li, X., & Li, J. (2020). Effect of hydrocolloids on gel properties and protein secondary structure of silver carp surimi. *Journal of the Science of Food and Agriculture, 100*(5), 2252–2260. https://doi.org/10.1002/jsfa.10254

Chen, C., Okazaki, E., & Osako, K. (2016). Textural improvement of salt-reduced Alaska pollack (*Theragra chalcogramma*) roe product by $CaCl_2$. *Food Chemistry, 213*, 268–273. https://doi.org/10.1016/j.foodchem.2016.06.084

Chen, C., Okazaki, E., Suzuki, T., Nguyen, H. T. N., & Osako, K. (2016). Objective quality evaluation of commercial spicy pollack roe products in terms of mechanical and biochemical properties. *Food Science and Technology Research, 22*(3), 337–347. https://doi.org/10.3136/fstr.22.337

Chen, C., Takahashi, K., Geonzon, L., Okazaki, E., & Osako, K. (2019). Texture enhancement of salted Alaska pollock (*Theragra chalcogramma*) roe using microbial transglutaminase. *Food Chemistry, 290*, 196–200. https://doi.org/10.1016/j.foodchem.2019.03.114

China Fishery Bureau. (2021). China fishery statistical yearbook. Beijing, China: Chinese Agriculture Express. pp 22+26. (in Chinese).

Dong, X., Pan, Y., Zhao, W., Huang, Y., Qu, W., Pan, J., … Prakash, S. (2020). Impact of microbial transglutaminase on 3D printing quality of *Scomberomorus niphonius* surimi. *LWT - Food Science and Technology, 124*, Article 109123. https://doi.org/10.1016/j.lwt.2020.109123

Fang, Q., Shi, L., Ren, Z., Hao, G., Chen, J., & Weng, W. (2021). Effects of emulsified lard and TGase on gel properties of threadfin bream (*Nemipterus virgatus*) surimi. *LWT, 146*, Article 111513. https://doi.org/10.1016/j.lwt.2021.111513

Feng, X., Bansal, N., & Yang, H. (2016). Fish gelatin combined with chitosan coating inhibits myofibril degradation of golden pomfret (*Trachinotus blochii*) fillet during cold storage. *Food Chemistry, 200*, 283–292. https://doi.org/10.1016/j.foodchem.2016.01.030

Hong, H., Luo, Y., Zhou, Z., & Shen, H. (2012). Effects of low concentration of salt and sucrose on the quality of bighead carp (*Aristichthys nobilis*) fillets stored at 4 ℃. *Food Chemistry, 133*(1), 102–107. https://doi.org/10.1016/j.foodchem.2012.01.002

Jiang, Q., Jia, R., Nakazawa, N., Hu, Y., Osako, K., & Okazaki, E. (2019). Changes in protein properties and tissue histology of tuna meat as affected by salting and subsequent freezing. *Food Chemistry, 271*, 550–560. https://doi.org/10.1016/j.foodchem.2018.07.219

Jiang, Q., Nakazawa, N., Hu, Y., Osako, K., & Okazaki, E. (2019). Microstructural modification and its effect on the quality attributes of frozen-thawed bigeye tuna (*Thunnus obesus*) meat during salting. *LWT - Food Science and Technology, 100*, 213–219. https://doi.org/10.1016/j.lwt.2018.10.070

Kim, B. S., Oh, B. J., Lee, J. H., Yoon, Y. S., & Lee, H. I. (2020). Effects of various drying methods on physicochemical characteristics and textural features of yellow croaker (*Larimichthys Polyactis*). *Foods, 9*(2), 196. https://doi.org/10.3390/foods9020196

Kou, X., Luo, D., Li, Y., Xu, B., Zhang, K., Li, P., … Liu, J. (2018). Effect of inulin with different degree of polymerisation on textural and rheological properties of wheat starch–Effect of inulin on gel properties of starch. *International Journal of Food Science & Technology, 53*(11), 2576–2585. https://doi.org/10.1111/ijfs.13852

Lee, H. C., Jang, H. S., Kang, I., & Chin, K. B. (2017). Effect of red bean protein isolate and salt levels on pork myofibrillar protein gels mediated by microbial transglutaminase. *LWT - Food Science and Technology, 76*, 95–100. https://doi.org/10.1016/j.lwt.2016.10.039

Li, Q., Gui, P., Huang, Z., Feng, L., & Luo, Y. (2018). Effect of transglutaminase on quality and gel properties of pork and fish mince mixtures. *Journal of Texture Studies, 49*(1), 56–64. https://doi.org/10.1111/jtxs.12281

Li, X., Liu, C., Wang, J., Zhou, K., Yi, S., Zhu, W., … Li, J. (2020). Effect of hydroxyl radicals on biochemical and functional characteristics of myofibrillar protein from large yellow croaker (*Pseudosciaena crocea*). *Journal of Food Biochemistry, 44*(1), Article e13084. https://doi.org/10.1111/jfbc.13084

Liang, F., Lin, L., He, T., Zhou, X., Jiang, S., & Lu, J. (2020). Effect of transglutaminase on gel properties of surimi and precocious Chinese mitten crab (*Eriocheir sinensis*) meat. *Food Hydrocolloids, 98*, Article 105261. https://doi.org/10.1016/j.foodhyd.2019.105261

Liang, Y., Xie, Y., Li, D., Luo, Y., & Hong, H. (2021). Dynamics of water mobility, salt diffusion and hardness changes in bighead carp fillets during low-salting. *LWT, 135*, Article 110033. https://doi.org/10.1016/j.lwt.2020.110033

Liu, F., Chiou, B. S., Avena-Bustillos, R. J., Zhang, Y., Li, Y., McHugh, T. H., & Zhong, F. (2017). Study of combined effects of glycerol and transglutaminase on properties of gelatin films. *Food Hydrocolloids, 65*, 1–9. https://doi.org/10.1016/j.foodhyd.2016.10.004

Qin, N., Zhang, L., Zhang, J., Song, S., Wang, Z., Regenstein, J. M., & Luo, Y. (2017). Influence of lightly salting and sugaring on the quality and water distribution of grass carp (*Ctenopharyngodon idellus*) during super-chilled storage. *Journal of Food Engineering, 215*, 104–112. https://doi.org/10.1016/j.jfoodeng.2017.07.011

Sáez, M. I., Vizcaíno, A., Galafat, A., Anguís, V., Fernández-Díaz, C., Balebona, M. C., … Martínez, T. F. (2020). Assessment of long-term effects of the macroalgae Ulva ohnoi included in diets on Senegalese sole (*Solea senegalensis*) fillet quality. *Algal Research, 47*, Article 101885. https://doi.org/10.1016/j.algal.2020.101885

Sun, L., Du, H., Wen, J., Zhong, C., Liu, G., Miao, S., & Cao, M. (2021). Physicochemical properties of acid-soluble collagens from different tissues of large yellow croaker (*Larimichthys crocea*). *International Journal of Food Science & Technology, 56*(10), 5371–5381. https://doi.org/10.1111/ijfs.15172

Sun, Q., Sun, F., Xia, X., Xu, H., & Kong, B. (2019). The comparison of ultrasound-assisted immersion freezing, air freezing and immersion freezing on the muscle quality and physicochemical properties of common carp (*Cyprinus carpio*) during freezing storage. *Ultrasonics Sonochemistry, 51*, 281–291. https://doi.org/10.1016/j.ultsonch.2018.10.006

Sun, S., Wang, S., Lin, R., Cheng, S., Yuan, B., Wang, Z., & Tan, M. (2020). Effect of different cooking methods on proton dynamics and physicochemical attributes in Spanish mackerel assessed by low-field NMR. *Foods, 9*(3), 364. https://doi.org/10.3390/foods9030364

Tan, M., & Xie, J. (2021). Exploring the effect of dehydration on water migrating property and protein changes of large yellow croaker (*Pseudosciaena crocea*) during frozen storage. *Foods, 10*(4), 784. https://doi.org/10.3390/foods10040784

Tan, M., Ye, J., Chu, Y., & Xie, J. (2021). The effects of ice crystal on water properties and protein stability of large yellow croaker (*Pseudosciaena crocea*). *International Journal of Refrigeration, 130*, 242–252. https://doi.org/10.1016/j.ijrefrig.2021.05.040

Thorarinsdottir, K. A., Arason, S., Geirsdottir, M., Bogason, S. G., & Kristbergsson, K. (2002). Changes in myofibrillar proteins during processing of salted cod (*Gadus morhua*) as determined by electrophoresis and differential scanning calorimetry. *Food Chemistry, 77*(3), 377–385. https://doi.org/10.1016/S0308-8146(01)00349-1

Volpe, M. G., Coccia, E., Siano, F., Di Stasio, M., & Paolucci, M. (2019). Rapid evaluation methods for quality of trout (*Oncorhynchus mykiss*) fresh fillet preserved in an active edible coating. *Foods, 8*(4), 113. https://doi.org/10.3390/foods8040113

Wang, Y., Liu, A., Ye, R., Wang, W., & Li, X. (2015). Transglutaminase-induced crosslinking of gelatin-calcium carbonate composite films. *Food Chemistry, 166*, 414–422. https://doi.org/10.1016/j.foodchem.2014.06.062

Warner, R., Ha, M., Sikes, A., & Vaskoska, R. (2017). Cooking and novel postmortem treatments to improve meat texture. In *New Aspects of Meat Quality* (pp. 387–423). Woodhead Publishing. https://doi.org/10.1016/B978-0-08-100593-4.00016-3.

Xia, M., Chen, Y., Guo, J., Huang, H., Wang, L., Wu, W., … Sun, W. (2019). Water distribution and textual properties of heat-induced pork myofibrillar protein gel as affected by sarcoplasmic protein. *LWT - Food Science and Technology, 103*, 308–315. https://doi.org/10.1016/j.lwt.2019.01.009

Yalçın, M. Y., & Şeker, M. (2016). Effect of salt and moisture content reduction on physical and microbiological properties of salted, pressed and freeze dried turkey meat. *LWT - Food Science and Technology, 68*, 153–159. https://doi.org/10.1016/j.lwt.2015.12.032

Yang, S., He, Y., Yan, Y., Xie, N., Song, Y., Yan, X., & Ding, Z. (2017). Textural properties of stinky mandarin fish (*Siniperca chuatsi*) during fermentation: Effects of the state of moisture. *International Journal of Food Properties, 20*, 1530–1538. https://doi.org/10.1080/10942912.2016.1233433

Zhang, X. Y., Yang, X. S., Guo, Q. Y., Li, X. Y., & Chi, H. (2012). Effect of moisture content on storage properties in light salted *Pseudosciaena crocea*. *Science and Technology of Food Industry, 33*(09). https://doi.org/10.13386/j.issn1002-0306.2012.09.008

Zhao, X., Chen, L., Wongmaneepratip, W., He, Y., Zhao, L., & Yang, H. (2021). Effect of vacuum impregnated fish gelatin and grape seed extract on moisture state, microbiota composition, and quality of chilled seabass fillets. *Food Chemistry, 354*, Article 129581. https://doi.org/10.1016/j.foodchem.2021.129581

Zhao, Z., Wang, Q., Yan, B., Gao, W., Jiao, X., Huang, J., … Fan, D. (2021). Synergistic effect of microwave 3D print and transglutaminase on the self-gelation of surimi during printing. *Innovative Food Science & Emerging Technologies, 67*, Article 102546. https://doi.org/10.1016/j.ifset.2020.102546

Zhou, F., & Wang, X. C. (2019). Effect of heat extraction on water-soluble taste substances in processing products of chilled large yellow croaker (*Pseudosciaena crocea*). *Food Science & Nutrition, 7*(12), 3863–3872. https://doi.org/10.1002/fsn3.1213