Contents lists available at ScienceDirect





Ultrasonics - Sonochemistry

journal homepage: www.elsevier.com/locate/ultson

# Ultrasound or microwave vacuum thawing of red seabream (*Pagrus major*) fillets



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# ARTICLE INFO

Keywords: Ultrasound thawing Vacuum thawing Microwave thawing Pagrus major Seabream Red seabream

# ABSTRACT

Ultrasound assisted vacuum thawing (UVT) or microwave vacuum thawing (MVT) with red seabream fillets were compared to fresh, chill storage thawing, vacuum thawing, microwave thawing and ultrasound thawing. The thermal stability and gelation properties were studied with DSC and dynamic rheology, respectively. Raman spectra before and after H/D isotope exchange and intrinsic fluorescence were used to measure protein secondary and tertiary structure. Low-field NMR was done to measure water migration. The two thawing techniques both retained actin thermal stability and generally retained more stable tertiary structures than the other thawing methods. MVT showed a desirable viscoelasticity of muscle proteins and UVT had a relatively stable secondary structure. There were no significant changes in free water. Thus, UVT and MVT could be used to improve the physicochemical properties of proteins during thawing of fillets.

# 1. Introduction

Freezing includes three main operations: actual freezing, frozen storage and thawing [1]. In particular, physicochemical and microbiological changes occur during thawing, which is slower than freezing. This is further complicated with sub-optimally thawing operations [2]. Development of more efficient, convenient, safe and low-cost thawing methods would benefit the food industry.

Red seabream (*Pagrus major*) is a high protein, low fat and very tender tasting fish. It is popular with customers in China, Japan, and South Korea [3]. Because the shelf-life is short due to protein degradation, lipid oxidation, and decomposition [4], so fillets are being frozen and would benefit from better thawing methods being developed for the end-users.

Traditional thawing includes room temperature thawing, chill storage thawing, vacuum thawing, warm saltwater thawing, and static water thawing [5]. Some novel thawing techniques include microwave thawing [6,7], ultrasound assisted thawing, pressure ohmic thawing [8], ohmic thawing [9,10], radio-frequency thawing [11,7], highpressure thawing [12], high voltage electric field thawing [13,14], and acoustic thawing [15]. Microwave thawing has a number of advantages such as high efficiency, energy saving, and simple controls. It is being used both commercially and by consumers. Problems include localized

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https://doi.org/10.1016/j.ultsonch.2018.05.001

Received 18 March 2018; Received in revised form 30 April 2018; Accepted 3 May 2018 Available online 04 May 2018

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heating and low thermal conversion. Ultrasound has been shown to work better with frozen foods than with unfrozen foods [16]. Ultrasound thawing is more uniform than microwave thawing. The front of the phase change from ice to water moves faster than most conventional thawing methods which will decrease the possibility of water loss, protein denaturation, and microbial contamination [17]. Gambuteanu and Alexe [18] reported that an ultrasound frequency of 25 kHz and an ultrasound power of 0.6 W/cm<sup>2</sup> can shorten the time frozen food is between  $\sim -5$  and  $\sim -1$  °C, the most critical temperatures. Besides, Gambuteanu and Alexe [19] came to a conclusion that the physicochemical and microbiological changes showed a non-significant difference between ultrasound thawing and conventional thawing for pH, total drip loss, moisture content, thiobarbituric acid reactive substances (TBARS) and microbial growth, i.e., it was faster without negative changes. However, ultrasound thawing has some disadvantages such as high power consumption, localized heating and poor penetration [20].

There are many possible methods that might improve microwave or ultrasound thawing, such as increasing the ultrasound frequency or moving the frozen food one or many times when thawing in a microwave oven. However, there are some disadvantages in them. Increasing frequency is energy consuming and moving frozen food once or many times is inconvenient. Vacuum thawing is a method that requires a relative low temperature compared with other thawing methods. It has

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less affect on oxidation reactions and microbial reproduction. The low temperature can reduce energy consumption. In this work, microwave and ultrasound thawing were combined with vacuum thawing. The focus was on comparing the effects of microwave vacuum thawing (MVT) and ultrasound vacuum storage (UVT) with those of chill storage, vacuum, microwave, and ultrasound thawing of red seabream fillets. The main emphasis was on the properties of the muscle proteins.

# 2. Materials and methods

# 2.1. Reagents

Bovine serum albumin was purchased from Solarbio Science & Technology Co., Ltd. (Beijing, China). All other chemicals unless specifically mentioned were bought from Fengchuan Chemical Reagent Technologies Co., Ltd. (Tianjin, China) as analytical grade or better. Deionized water (200 L, Sanda Shui Beijing Technology Co., Ltd., Beijing, China) was used during all experimental work.

# 2.2. Sample preparation

Live red seabream was purchased from a local fish market in Jinzhou, China (red scales, farmed in an offshore area of the Bohai sea for 2 yr, weight ranged from  $\sim 0.9$  to 1.0 kg and length was  $\sim 25$  to 30 cm). Seven fish were transported to the laboratory within one h and were immediately stunned with one or two blows on the head with a wooden club once they arrived avoiding stress response. Fish were then wiped off with tissue paper. One skin-on and one skin-off fillet were obtained from each fish. Every fillet was cut with a knife into 4 parts which were about  $8 \text{ cm} \log \times 4 \text{ cm}$  wide and weighed about 100 g. Except for the fillets of one fish, which was designated as the fresh sample (FS), the other fillets were packaged in polyethylene bags ensuring all pieces of one fish were all in one bag. These fillets were frozen and stored in  $-20 \pm 1$  °C freezer (BCD-649WE, Qingdao Haier Co., Ltd., Qingdao, Shandong, China) for at least 24 h. After being frozen, the pieces of the 6 fish were thawed using 6 different treatments: chill storage thawing (CST), microwave thawing (MT), ultrasound thawing (UT), vacuum thawing (VT), UVT and MVT. The same part of each of the 7 fish was used for each different measurement.

#### 2.3. Fillets thawing

#### 2.3.1. Chill storage thawing (CST)

One fillet was put in a 4  $\pm$  0.5 °C refrigerator. A temperature recorder with a temperature sensor (Elitech RC-4, Jingchuang Electric Co., Ltd., Xuzhou, Jiangsu, China) was used to detect the fillet's core temperature changes. The pieces were thawed for a while to assure the surface of the pieces were thawed so the sensor could be put into the core of one piece. In order to fix the temperature sensor, the temperature sensor's wire was taped to the container. The end of thawing was reached when the sample reached 0 °C. The system for endpoint determination was used with all thawing treatments except the microwave vacuum thawing. Pieces were then used for 9 different measurements. The thawed pieces were put in the ~4 °C refrigerator to await the other treatments so measurements could be done at one time with all 7 samples. All samples were thawed and the experiments started on the day after freezing.

#### 2.3.2. Microwave thawing (MT)

A microwave oven (NN-DF392B, Panasonic,  $350 \times 299 \times 199$  mm, Osaka, Japan) was used. A fish piece was placed in a 250 mL drinking glass and put into the microwave oven and thawed to 0 °C at 2450 MHz and a micro power of 300 W according to the manufacturer. Each piece was done separately as was also done for the other thawing methods.

#### 2.3.3. Ultrasound thawing (UT)

The UT was done using an ultrasonic cleaning machine (KQ-400KDE,  $300 \times 240 \times 380$  mm, Kunshan Ultrasonic Instrument Co., Ltd., Kunshan, Jiangsu, China). The fish was placed in a 500 mL beaker with 250 mL deionized water. Because of the density difference of ice and water, the fillet floated on the water. This beaker was put into the machine filled with about 5 L deionized water. The power was set at 200 W, the frequency was 40 kHz and the temperature of the water in the sonicator was controlled at 10 °C using added ice to maintain temperature according to the temperature monitor of ultrasonic cleaner.

# 2.3.4. Vacuum thawing (VT)

VT was done using a 1 L vacuum flask which could be brought to 0.06 MPa gauge with a water-circulation multifunction vacuum pump (SHB-III, Great Wall Scientific Industry and Trade Co., Ltd., Zhengzhou, Henan, China), with a rubber tube connecting it with the flask. The floating fillet was in 500 mL deionized water. The thawing to 0  $^{\circ}$ C was done with the vacuum flask in the ultrasonic cleaning machine (not turned on) to control temperature as described in 2.3.3.

## 2.3.5. Ultrasound vacuum thawing (UVT)

UVT was a combination of ultrasound and vacuum thawing. The vacuum flask was placed in the ultrasonic cleaning machine with about half of the vacuum flask immersed in the external water.

#### 2.3.6. Microwave vacuum thawing (MVT)

MVT was a combination of microwave and vacuum thawing although identical equipment was not used. A microwave vacuum drying oven (ORW08S-3Z, Nanjing Aorun Microwave Technology Co., Ltd., Nanjing, Jiangsu, China) was used. To prevent moisture loss, the power was set at 300 W and the frequency was 2450 MH<sub>Z</sub> according to the manufacturer. The temperature was set at 10 °C. The temperature detection was done once a min by opening the microwave vacuum drying oven and inserting the temperature sensor.

#### 2.4. Physicochemical changes in fish protein

#### 2.4.1. Dsc

Skin off fish pieces were chopped with a knife and about 5–10 mg of finely chopped fish was placed in an aluminum pan keeping a good contact with the pan bottom. The measurements were done using a differential scanning calorimeter (Q2000, TA Instruments Co., Ltd., Shanghai, China) with an empty aluminum pan as a reference. The sample aluminum pan and empty aluminum pan both were heated from 20 to 85 °C at 3 °C/min using the instrument's temperature settings. The denaturation temperatures were the temperatures of the enthalpy peaks' maximum point and the enthalpies of the transitions were the area under the peaks both estimated using the TA Universal Analysis software (Q2000, TA Instruments Co., Ltd., Shanghai, China) provided with the instrument.

# 2.4.2. Dynamic rheology

A rheometer (Discovery HR-1, TA Instruments Co., Ltd. Shanghai, China) equipped with a 40 mm diameter parallel plate was used to analyze the viscosity and elasticity of the samples. The truncation gap was set at 1 mm; a strain of 5%, and a frequency of 1 Hz were used. Finely chopped skin off white muscle ( $\sim$ 3 g) was placed on the plate and the rheometer cover put on. Paroline (Fengchuan) was put around the opening to prevent moisture loss. Samples were heated from 20 to 90 °C at 2 °C/min. Storage modulus (G') and loss modulus (G'') were obtained as calculated by the software provided with the instrument.

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