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PROCESS DEVELOPMENT FOR PRODUCTION OF SMOKED FISH FLOSS PRODUCTS FROM ATLANTIC MACKEREL AND BLUE WHITING

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ABSTRACT

Meat floss is one of the traditional meat products among Asian communities. The popular raw materials are pork and chicken. Nowadays, fish is also a favorable material because of its nutrients. In this study, Atlantic mackerel and Blue whiting were used to produce smoked fish floss with 2% salt and 1% sugar content. The research results indicated the steaming time was 10 minutes for both species and the optimal hot-drying time was 40 minutes for Atlantic mackerel and 60-70 minutes for Blue whiting. After processing, three fish floss products stored at 2-4ºC were analyzed for sensory attributes and lipid quality, water activity, colour intensities and total viable counts determined at storage time of 0 week, 2 and 4 weeks to evaluate the quality changes. The lightness went down but the yellowness rose for all products in storage. Regarding lipid changes, the PV and TBARS of two Blue whiting products and also the FFA of almost all samples, were rather stable during storage. However, for Atlantic mackerel product, the TBARS increased significantly during prolonged chilled time while the PV grew rapidly after storage for 2 weeks. Meanwhile, microbial growth and sensory quality, water content changes of all samples were not significant in the chilled storage.

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1 INTRODUCTION

1.1 Problem Statement

The Republic of Vietnam is located in Southeast Asia. Vietnam's coastline extends for some 3,260 kilometers in length with three coastal areas bordered by the East Sea, the northern, eastern and southern seas and occupies a land area of 331,210 square kilometers. There is also a network of about 2,360 rivers, estuaries countrywide and 811,700 ha of freshwater plus 635,400 ha brackish water, 125,700 ha of coves and 300,000 to 400,000 ha of wetlands. Moreover, Vietnam is a tropical country with an average temperature ranging from 21°C to 27°C. All these facts exhibit favourable conditions for the development of the fisheries sector as well as the fish processing industry in the country (GSO 2014). In 2012, Vietnam was the fourth largest exporter of fish and fishery products in the world with 6.278 US$ billions, the ninth largest with regards to marine capture fisheries production, amounting to 2.42 million tons, and the third largest aquaculture fisheries quantity by 3.09 million tons in the world (FAO 2014).

In Vietnam, the domestic consumption is mostly fresh fish, with 40% to 50% of total fresh fish production. Meanwhile, the major export seafood products by far are the frozen products (mostly low value semi-processed products), with 72.6 % of total export value in 2013. This means that the quantity and quality of added value and ready- to- eat products in seafood processing industry is low relatively (VASEP 2014, NAFIQAD 2014). Hence, improving quantity and quality of added value products, including ready to eat, snacks, fast food products, etc. and expanding those products's exporting markets were proposed as target up to 2020 for fish processing in Vietnam (NAFIQAD 2014).

Convenience is one of the big trends in the food business. Food products that save time and effort in preparation, consumption, or cleanup are of most interest for consumers. Many people's lifestyles today have led to a great need for such convenience food products. Technological innovations such as the microwave, changing household structures with more single household. Also, women are more active on the job market, which leads to less time available for preparing meals. Families will therefore depend more on convenience food, such as fast food and ready to eat food in today's modern busy life. All those factors have created opportunities for convenience food processing (Boer *et al.* 2004, Brunner *et al*. 2010).

Smoking is one of the oldest processing methods used to preserve and produce fish. Smoking has been researched and applied in the world for a long time. However, smoked food products are not common in Vietnam. There are just few smoked products produced in the country, for example smoked pork, smoked sausages and imported smoked salmon. Therefore, plus with above mentioned statements, a smoked fish floss product would be an interesting new addition to the fish products in Vietnam.

The goals of this project are to develop processing methods for smoked fish floss made from Atlantic mackerel and Blue whiting and also to study the quality changes of the products during storage time. This may contribute and add to the diversity of seafood products in Vietnam. The result of this study may be used as a reference for the producers of fish floss processing in Vietnam. This project's overall objectives are to find out optimal steaming time and hot drying time for the processing of smoked fish floss products and evaluate quality changes during storage of Atlantic mackerel (*Scomber scombrus*) and and Blue whiting (*Micromesistius poutassou*).

The tasks of this project were to:

1. Evaluate sensory attributes and measure protein-, lipid- and water content as well as free fatty acid (FFA), peroxide value (PV) and thiobarbituric acid reactive substances (TBARS), total viable count (TVC) of the raw materials (Atlantic mackerel and Blue whiting).
2. Optimize steaming time and determine the appropriate hot drying time of the processing of smoked fish floss from Atlantic mackerel and Blue whiting.
3. Evaluate quality changes of final products, including odor and flavor attributes, lipid- and water content, FFA, PV, TBARS, TVC, H_2S-producing bacteria, water activity and colour intensity changes after a storage time of 0 week, 2 and 4 weeks.

2 LITERATURE REVIEW

2.1 Drying Technology in Food Processing

Dehydration is one of the oldest methods used in food processing and preservation in the world. In general, the drying processes may be divided into two categories: in-air or in-vacuum. A vacuum is useful for removing water vapor when the products are dried without air. While, in-air processing can involve elevated temperatures which are usually used for achieving a high rate of drying as well as creating desirable sensory attributes for the products. The heat can be supplied in different ways: convection, conduction, microwave, radiation, radio-frequency, or even heating (Chen and Mujumdar 2008).

Drying has many roles in food processing and may be viewed as a thermal processing stage. The micro-structure of the food materials is particularly relevant to drying, as both liquid water and water vapor move within the structure. The micro-structure, as far as the transport of heat and chemical species are concerned, is mainly made up of compounds such as protein, fat and carbohydrate, and minerals and air. Porousness and tortuosity are usual characteristics of food (Aquilera *et al*. 2003). When drying using hot air, the surface of the material being dried is usually firmer than that of the core. Hardening is associated with shrinkage, or is a result of shrinkage, and is also well known where the dried foods are generally perceived to be ‘harder to chew’ compared with their original states before drying (Chen and Mujumdar 2008).

Color, favor, taste and shape (appearance) are the four important sensory attributes affecting people’s choice of foods (Lawless and Heymann 2010). Flavor retention of a food product after drying can also vary depending on how the drying is conducted. Drying also can remove part of undesirable odor, for instance fishy off-odor of fish in dried fish processing (Fu *et al*. 2014). Texture plays a subsequent, but also important role, once the foods are consumed. It is not clear what the relationship is between color perception and water content. Color may be intensified (colorings concentrated) as water is removed. Additionally, as in hot air drying, the surface temperature of the product can get very high, which promotes heat sensitive chemical reactions such as brown reactions (Maillard reaction), so the products have the melanoidins color. It is important to emphasise that color as a quality of the manufactured food is a ‘surface phenomena’ so the surface temperature and surface water content control should be the most important parameters to address. Also, microbes or microorganisms are inactive in the drying process, especially when a high temperature environment is applied (Horner 1997, Chen and Mujumdar 2008).

Drying is the process of removing water, involving evaporation of water vapor from a surface together with the movement of the inner of the material being dried. Moisture content and water activity (a_w) is decreasing in drying process (Doe 1998). Most of the factors which influence the acceptability and stability of foods can be related to water activity (aw). In dried and partially dried food, a_w is a critical parameter affecting food stability by influencing chemical reactions and susceptibility to spoilage during storage (Sun *et al.* 2002). In particular, the growth of microorganisms in dried fish is strongly dependent on a_w. When the a_w is reduced below about 0.62%, no bacteria or mold can grow. The growth of pathogenic and putrefactive bacteria in fish can be suppressed by decreasing the a_w below 0.91. Reducing a_w may inhibit the growth of microorganisms, and slow chemical reactions to some extent; however, many deteriorative reactions, including lipid oxidation, Maillard browning, and enzymatic reactions, still occur at relatively low a_w values (Doe and Olley 1990, Sun *et al.* 2002).

2.2 Smoking methods

Smoking is a well-known method of food preservation and is widely applied in fish processing in the world. In Europe approximately 15% of the total amount of fish for human consumption is offered on the market in the form of either cold or hot smoked products. Traditional smoking methods include treating of pre-salted, whole, eviscerated or filleted fish with wood smoke. The smoking temperature is in the range of 12-25°C during cold-smoking and 25-45°C in warm-smoking. Hot smoking is known as the traditional smoking method. In hot-smoking, the process can be undertaken in some stages, during which the temperature of the smoke ranges from 40-100°C and that in the core of the product may reach up 85°C (Andrzej and Zdzislaw 2005).

The main purpose of smoking is to preserve the fish, partly by drying and partly by adding naturally produced anti-microbiological substances such as formaldehyde and phenols (Horner 1997). Additionally, the colour and flavor attributes of fish can be desirably changed after smoking, producing high sensory quality (Kolodziejska *et al.* 2002).

2.3 Meat floss products

Meat floss, also called shredded meat or meat wool, is a dried meat product with a light and fluffy texture similar to coarse cotton, originating from China. This is one of traditional ready-to-eat meat products popular among the Asian community. It is known by different local names such as *rousong* in China, Taiwan, *serunding* in Malaysia, *abon* in Indonesia, *moo yong* in Thailand, *mahu* in Philippines. China is not only the biggest producer but also be the main market for general meat floss products. Popular raw materials for making this type product are pork and chicken, however beef, shrimp and some fish species are also suitable for meat floss processing (Huda *et al*. 2012).

In Vietnam, this type of product often is called C*ha bong* or *Ruoc* and it is used as snack food or being filled for various dishes, such as children's food, buns, pastries, rice soup, and steamed sweet rice. They often are produced at small-scale such as household level, depending on the producer's experience, so the quality is not stable (Le and Nguyen 2012, Tran and Do 2012).

Fish floss products are produced from many type of fish such as tuna, mackerel, salmon, tilapia and snakefish being combined with fish sauce, salt, sugar and some special seasons based on preferred tastes in each country or region (Liao *et al.* 2009, Guo *et al*. 2010, Le and Nguyen 2012, Tran and Do 2012). Fish floss products can also be made from flesh shreds in fish processing plants.

This can increase the utilization of by-products as well as turnover from fish processing industry (Deng *et al*. 2005).

Preparation of Meat floss

For pork and chicken floss, the washed meat is trimmed to remove outer fat membrane and tendons and then cut in to small portions. After that, cut portions are cooked in boiling water or steamed until it is tender. The meat is then crushed or torn easily into shreds because of the collagen that holds the muscle fibers of the meat together has been converted into gelatin in the former step. Thereafter, the mixture is fried and stirred under heat in a pan or a wok. After a light drying, the meat is mashed and beaten or rubbed continuously while being dried to create the shredded and fluffy form. Spices also are added while the mixture is being dried. Usually, 5 kg of raw meat will yield about 1 kg of final product. The pork floss will normally be packaged after it has cooled down to room temperature. Due to the lower moisture content, meat floss can keep without refrigeration and will not drastically change in room temperature storage (Ockerman and Marriott 2000, Liao *et al*. 2009, Huda *et al*. 2012).

For fish floss, the process is almost similar to the above one. However, deodorization may be required to remove fishy off-odor in raw fish materials and less steaming time because of low collagen and elastin content of fish meat (Guo *et al.* 2010, Tran and Do 2012).

2.4 Atlantic mackerel (*Scomber scombrus*)

The Atlantic mackerel (Figure 1) is classified as a fat fish species that belong to the Scombridae family of fish. Atlantic mackerel is common on both sides of the North Atlantic Ocean, including the Baltic Sea. Atlantic mackerel is found in cold and temperate waters. They are typically a surface –living species and swim in schools (NOAA 2014).

Figure 1. Atlantic mackerel (*Scomber scombrus*).

Mackerel is a valuable pelagic fish and most of the catch is for human consumption (Icelandic Fisheries 2015). An adult mackerel has length of 30-35 cm and weight of 300-500 g. The mackerel is a fatty fish, and the fat and water content may vary according to season. The fat content is about 6-23%, water content is 56-74% and protein content is 18-20 % throughout the year (FAO 2015). This is considered one of the healthier fish because it is not only high in omega-3 fatty acids but also be an excellent source of selenium, niacin, and vitamins B6 and B12 (NOAA 2014).

2.5 Blue whiting (*Micromesistius poutassou*)

Blue whiting (Figure 2) is one of the two species in the genus *Micromesistius* in the Cod family, is distributed in the northeast Atlantic Ocean. This is a lean fish with the fat content less than 1%. The water content is about 80-83% and the protein content is 19-20%. The average length of blue whiting is usually 22-30 cm and the weigh range is from 135g-280g. Majority of the catch is processed into fish meal. However, a small share is frozen at sea for human consumption (FAO 2015, Icelandic Fisheries 2015).

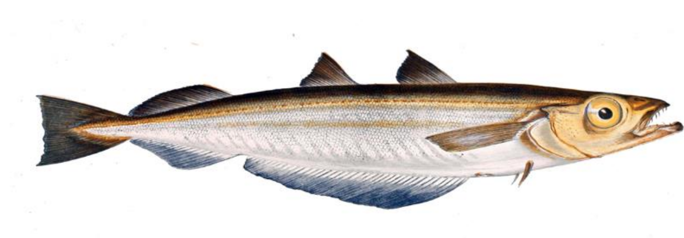

Figure 2. Blue whiting (*Micromesistius poutassou*).

3 MATERIALS AND METHODS

3.1 Material

3.1.1 Atlantic Mackerel

The Atlantic mackerel raw material used in the study was provided by the Ísfélagið Fish Processing Company, located at Vestmannaeyjar. It was caught in the southeast of Iceland on the 1st of September, 2014. The fish size was in the +400 g category.

3.1.2 Blue Whiting

There were two type of raw materials from Blue whiting used in this study. The whole frozen Blue whiting raw material was provided by Síldarvinnslan hf located in Neskaupstaður. It was caught on the 23 of September, 2014 in the southeast of Iceland. It was caught by the ship Börkur where it was kept in seawater at 0°C for 36 hours until it was processed whole and frozen. It was kept at -24°C until used. Meanwhile, the beheaded and gutted frozen Blue whiting raw material was provided by Síldarvinnslan. hf located in Neskaupstaður. It was caught on the 22nd of September, 2014 at 65°18´55“N, 08°39´14“W by the ship Polar Pelagic. It was gutted and headed on board and then frozen and kept at -24°C until used.

3.1.3 Salt

Fine sea salt (Net Wt. 10 oz.) of Badia Spices Inc., USA was used in this project.

3.1.4 Sugar

This study used white granulated sugar (1kg Net) bought from The Netherlands.

3.1.5 Smoke Agent

Smoke was produced by a Bradley Smoker. The wood flavor bisquettes were bought from Bradley Smoker Inc., Canada. Its contents included 99.98% selected hardwood and 0.2% collagen hydrolised (binder).

3.1.6 Chemicals

All of chemicals used in this project were of analytical grade and bought from Sigma-Aldrich Company.

3.1.7 Material Package

The products were packaged in round, none colour, PP seal boxes bought in Joykey Company (JETB 280 with ISO volume 0.28l).

3.2 Methodologies

3.2.1 Experimental Design

The project was undertaken in two parts (Figure 3) for two species: Atlantic mackerel (group AM) and Blue whiting (Group BW).

The first part (Part 1) included analysis on quality of initial raw materials and pre-trials were carried out to determine the appropriate steaming time and hot drying time. Both Blue whiting and Atlantic mackerel raw materials were received and thawed at 2-4°C for 20 hours in air before washing, gutting and skinning (for Blue whiting) or filleting (for Atlantic mackerel) with water at 4°C. The fish was then placed on a flat strainer for steaming at 100°C and sensory evaluation of cooked fish during steaming time 5, 10, 15 and 20 minutes. Sensory evaluation was used to determine the optimal time of this step. After steaming both fish species were cooled down and then fish meat was collected. The fish muscle was then pre-dried at 85°C for 15 minutes before being smoked at 13-16°C for 3 hours. Thereafter, the smoked fish meat was shredded and flavored with 2% of salt and 1% of sugar. The fish shreds were then spread on a sieve with thickness of 1.5cm to be hot-dried at 85°C. The fish meat was also rubbed with a fork on the surface of the sieve every 10 minutes while being dried to create the fluffy form for the final product. Pre-trials in this step were taken for measurement after 30, 40, 50 and 60 minutes hot drying time for Atlantic mackerel and 50, 60, 70, 80 minutes for Blue whiting.

In Part 2, three final products were produced from whole frozen Blue whiting (BWC group), beheaded, gutted frozen Blue whiting (BWM) and beheaded, gutted Atlantic mackerel according to optimized condition in Part 1. After drying, the fish floss was cooled down to room temperature and packed using round sealed plastic containers. They were all stored in a cooling room at 2-4°C for 0 week, 2 and 4 weeks to be evaluated for quality changes.

3.2.2 Sampling

Samples were obtained from the initial raw material for sensory evaluation and measurements of peroxide value PV, FFA, TBARS, water content, total volatile basic nitrogen (TVB-N), TVC, protein and lipid content. After the production, samples were collected of the final products after a storage time of 0 week, 2 and 4 weeks. Those samples were analysed for sensory attributes, the determination of colour, peroxide value (PV), free fatty acid (FFA), thiobarbituric acid reactive substances (TBARS), water content, lipid content, water activity (Aw), total viable count (TVC) and H_2S-producing bacteria count. Protein contents was also determined for all three products.

Total of 9kg of Atlantic mackerel, 9 kg of whole Blue whiting and 6 kg of beheaded and gutted Blue whiting raw materials were used in this project.

3.2.3 Analytic methods

Microbial analysis

Pour-plate method in measuring Total Viable Count (TVC)

Total viable counts and counts of H_2S-producing bacteria were performed on Iron Agar by the pour-plate method (according to the method used in the laboratory at Matis). 20g of fish floss product or raw fish samples were added to 180g of dilution buffer and blended in the Stomacher for one minute. A decimal solution was made from this particular mixture followed by more desired dilutions. After the decimal dilutions were made as required, a specific culture volume 1ml from each dilution was pipetted and added into culture plates. After that, the plates were added melted Iron agar medium (ca. 15ml at 45ºC) and stirred. After the agar medium solidified, plates were covered by a thin of Iron agar and the plates were then incubated at 22ºC for 48 hours. Bacteria forming black colonies (spoilage bacteria) on this medium produced H_2S from sodium thiosulphate and attained a black color. Total viable bacteria or the number of colony forming units (cfu) were counted on the plates using the Colony Counter

Chemical Analysis (Nielse and Suzanne 2010)

Total volatile basic nitrogen (TVB-N)

Samples of raw fish were determined TVB-N by steam distillation method which explained by (Malle and Poumeyrol, 1989)). 50g of raw fillets was placed in a blender then 100ml of 7.5% aqueous trichloroacetic acid solution was added and the mixture was homogenized. Filtering the mixture through a Whatman No.3 filter paper (6µm pore size), 25 mL of the filtrate was pipetted into a distillation flask with 6 mL 10% NaOH. Steam distillation was then took place using the Kjeldahl-type distillator (Struer TVN) and TVB-N was collected under a condenser in a beaker containing solution of 10 ml of 4% boric acid and indicators (0.04 ml of methyl red and bromocresol green) which turned green when alkalinized by the TVB-N. The alkalised solution was then titrated with sulphuric acid solution (0.0324N) by using 0.05ml graduated burette. Complete neutralization was obtained when the colour turned pink on addition of a further drop of sulphuric acid.

TVB-N (mg N/100g) was then calculated by using this formula: $\frac{14mg/mol \times a \times b \times 300}{25ml}$
Where a: volume of sulphuric acid (mL) b: normality of sulphuric acid (%)

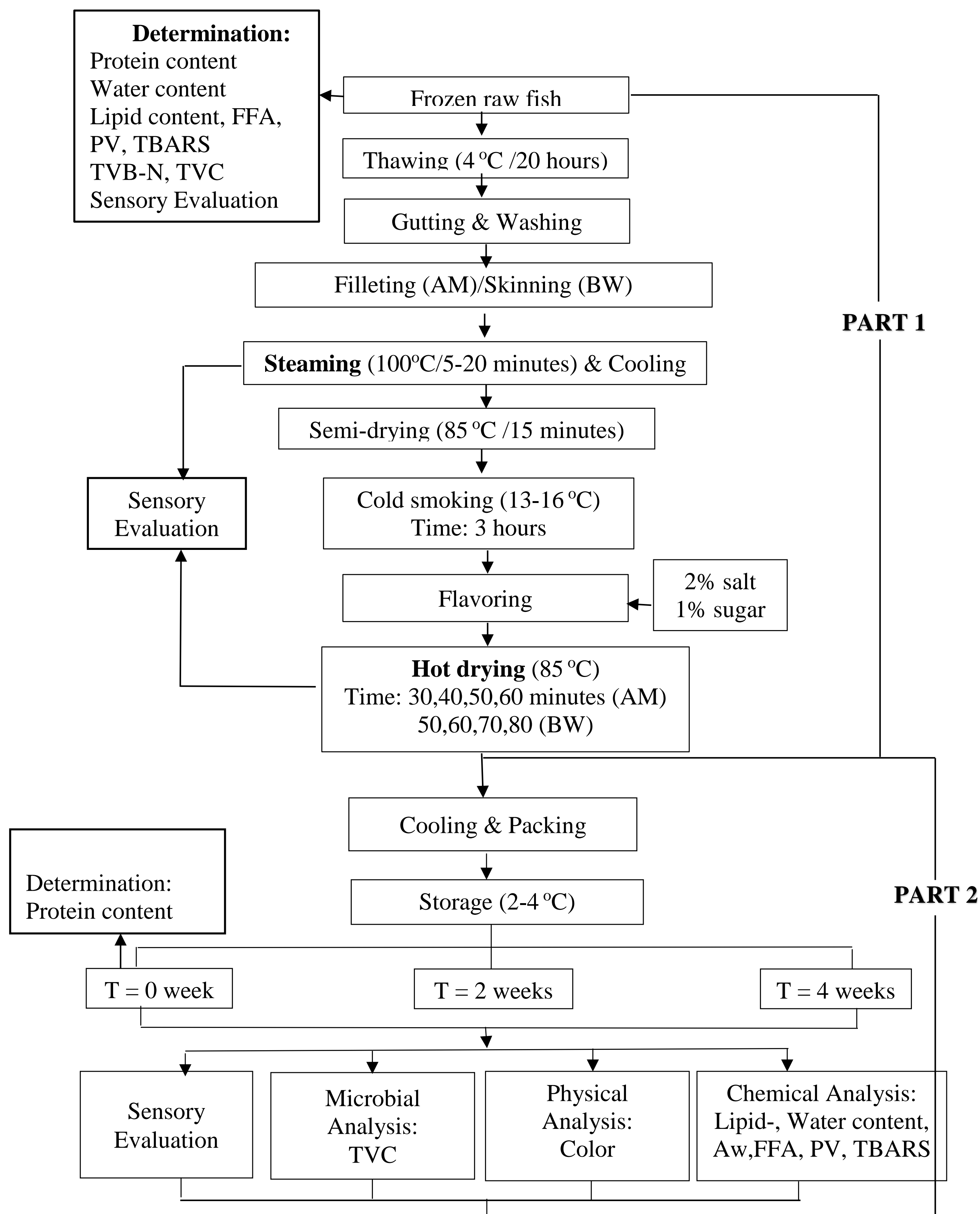

Figure 3. The flow chart of experimental design.

Water content

Water content was measured according to ISO 6496:1999 (E). Approximately 5.0 g of homogenized sample was weighed (±0.0001g) and placed in a small porcelain bowl. The porcelain bowl of sample was left to dry for 4 ± 0.1 h in the oven at 103±2 °C. The bowl was removed from the oven and allowed to cool to ambient temperature in a desiccator for about 30 minutes. The water content was calculated by the formula as follows:

$$W = \frac{m2-m3}{m2-m1} * 100\ (\%)$$

Where: m1 was the mass of the bowl (g)
m2 was the mass of the bowl, test portion (g)
m3 was the mass of the dish, dried test portion (g).

Water activity

Determination of water activity of samples were done using NOVASINA water activity meter.

Protein content

Protein content was determined by the Kjeldahl method (according to ISO 5983-2:2005). About 5 g of homogenized sample was digested in sulphuric acid in the presence of copper as a catalyst at approximately 370°C. Thereafter, the sample was placed in a distillation unit, 2400 Kjeltec Auto Sample System. The digested sample was made alkaline with NaOH and the nitrogen is distilled off as NH_3. This NH_3 was "trapped" in a boric acid solution and the amount of ammonia nitrogen in this solution was quantified by titration with standardized HCl solution. The nitrogen content was multiplied by the factor 6.25 to get the ratio of crude protein.

Lipid content

Total lipids from 25g sample were extracted according to the Bligh and Dyer (1959) Method with 50 ml of methanol, 50 ml of chloroform and 25 ml of 0.88 % KCl. 3 ml of lower phase resulting from the lipid extraction was removed solvents by using a nitrogen jet and heating at 55°C. The weight difference was the amount of lipid in these 3ml and the results was multiplied with the total volume of the chloroform used (50ml) and divided by the weight of the sample used for the lipid extraction (showing as grams lipid per 100g sample).

$$L = \frac{(m2 - m1) * 50}{3}$$

Where: m1 was the mass of the tube (g)
m2 was the mass of the tube and sample after removed solvents (g)

Free fatty acids

Free fatty acids (FFA) were measured according to method from (Lowry and Tinsley, 1976) with modification made by (Bernárdez *et al*., 2005). About 3 mL of the lower phase resulting from fat extraction (Bligh and Dyer 1959) was added in a screw cap culture tube. Any solvent present was removed at 55^0C using nitrogen jet. After cooling down, 3 mL of cyclohexane were accurately added by 1 mL of cupric acetate – pyridine reagent and vortex for ~40s. After centrifugation at 2000g for 10 min at 4°C, the upper layer was read at 710 nm in spectrophotometer. The FFA

concentration in the sample was calculated as μmol oleic acid based on a standard curve spanning a 0-20 μmol range. The results were indicated as μgram oleic acid per 100g lipid of sample.

Peroxide value (primary oxidation product)

Lipid hydroperoxides (PV) were determined with a modified version of the ferric thiocyanate method (Shantha and Decker, 1994). Total lipids were extracted from 5.0 g of samples with 10 mL ice-cold solvent (included chloroform: methanol (1:1) solution, containing 500 ppm BHT to prevent further peroxidation during the extraction process). 5.0 mL of sodium chloride (0.5 M) was added in to the mixture and homogenized for 30 seconds and then centrifuged at 5100 rpm for 5 min (TJ-25 Centrifuge, Beckmann Coulter, USA). The bottom layer (chloroform layer) was collected and transferred into 15ml test tube. 500 µL of bottom layer were collected and completed with 500 µL solvent stored at room temperature, followed by 5 µL of ammonium thiocyanate (4 M) and ferrous chloride (80 mM) mixture (1:1). The mixture was then vortex, incubated for 10 minutes at room temperature and read at 500 nm (Tecan Sunrise, Austria). A standard curve was prepared using cumene hydroperoxides. The results were indicated as μmol lipid hydroperoxides per kg of sample.

Thiobarbituric acid reactive substance (secondary oxidation product)

A modified method of Lemon (1975) was used for measuring thiobarbituric acid reactive substance (TBARS). About 5.0 g of sample was homogenized with 10.0 mL of trichloroacetic acid (TCA) extraction solution (7.5% TCA, 0.1% propyl gallate and 0.1% EDTA mixture prepared in ultrapure water) using a homogenizer for 10 seconds (Ultra-Turrax T-25 basic, IKA, Germany). The homogenized samples were then centrifuged at 5100 rpm for 20 min at 4°C (TJ-25 Centrifuge, Beckmann Coulter, USA). 0.5 mL of Supernatant was collected and mixed with the 0.5 mL thiobarbituric acid (0.02 M) and heated in a water bath at 95°C for 40 min. The samples were cooled down on ice and loaded into 96-wells microplates (NUNC A/S Thermo Fisher Scientific, Roskilde, Denmark) for reading at 530 nm (Tecan Sunrise, Austria). A standard curve was prepared using tetraethoxypropane. The results were showed as μmol of malomaldehyde diethylacetal per kg of sample.

Physical Analyses

Colour

The intensity of the colour was measured with a Minolta Chroma Meter CR-400 (Minolta, Osaka, Japan) using the CIE Lab system. The instrument recorded the L value, brightness on the scale of 0 to 100 from black to white; a value from -60 to 60, a>0 represents red component and a<0 represents green component; b value from -60 to 60, (+) stands for yellow component and (-) stand for blue component (Figure 4).

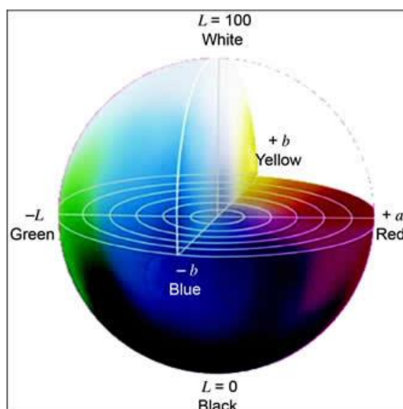

Figure 4: CIE lab colour space (Wakapon 2012)

Sensory Analysis

Generic Descriptive Analysis Method (GDA) (Lawless and Heymann 2010) was sensory evaluation method used in this study. The raw materials were evaluated sensory for odor and flavor factors. The final products were also assessed sensory for odor and flavor attributes.

Sensory panels: 10 panellists of MATIS's sensory panel having experience in sensory evaluation fish and fish products were selected. The members of the panel were previously trained specifically in GDA for detection and recognition of sensory attributes of the samples and describing the intensity of each characteristic for the given samples using an unstructured scale (0-100).

There were 16 characteristics for odor (7) and flavor (9) assessed for the cooked Atlantic mackerel fillet. 6 odor attributes and 7 flavor attributes was used to evaluate sensory quality of the cooked Blue whiting fillets. For fish floss product from Atlantic mackerel and Blue whiting, 12 sensory characteristics for odor (6) and flavor (6) were evaluated. Each attribute, as shown in Table 1 and 2 for cooked Atlantic mackerel fillets and Blue whiting fillets and in Table 3 for fish floss products, was evaluated by every panellist on a 100 point line scale anchored by the opposites 'none' to 'much'. The panellists evaluated each sample for each sampling day in duplicate while seated in separate booths under normal light in the sensory evaluation laboratory. Panellists used a computerised system for direct recording data. For raw material evaluation, each portion of fresh fish fillet was placed in a small aluminium box and then cooked at 150°C for 5 minutes in a pre-warmed oven and being covered immediately right after steamed. Meanwhile, the fish floss was collected and put in a plastic cup for sensory evaluation.

Table 1: Generic Descriptive Analysis of cooked Blue whiting fillets.

Sensory attribute	Short name	Scale anchors	Description of attribute
ODOR			
Sweet	O-sweet	none | much	Sweet odor
Shellfish, algae	O-shellfish	none | much	Shellfish, characteristic fresh odor
Dried fish	O-vanilla	none | much	Dried fish, processing odor
TMA	O-TMA	none | much	TMA odor, amine
Spoilage sour	O-sour	none | much	Sour odor, sour milk, spoilage sour, acetic acid
Rancid	O-rancid	none | much	Rancid odor
Frozen storage	O-frozen	none | much	Frozen storage odor, paper board
FLAVOR			
Salt	F-salt	none | much	Salty taste
Metallic	F-metallic	none | much	Characteristic metallic flavor of fresh fish
Sweet	F-sweet	none | much	Characteristic sweet flavor of very fresh fish
Pungent	F-pungent	none | much	Pungent flavor, bitter
Spoilage sour	F-sour	none | much	Sour taste, spoilage sour
Dried fish	F-dried fish	none | much	Dried fish, processing flavor
TMA	F-TMA	none | much	TMA flavor, amine
Frozen storage	F-frozen	none | much	Frozen storage flavor, paper board
Rancid	F-rancid	none | much	Rancid flavor

Table 2: Genetic Descriptive Analysis of cooked Atlantic mackerel fillets.

Sensory attribute	Short name	Scale	Definition
ODOR			
Fresh oil	O-oil	none || much	Fresh fishoil odor
Metallic	O-metallic	none || much	Metallic odor
Sweet	O-sweet	none || much	Sweet odor
Mouldy	O-mouldy	none || much	Mouldy odor
Butiric acid	O-butiric	none || much	Butiric acid, smelly feet
Rancid	O-rancid	none || much	Rancid odor
FLAVOR			
Fresh oil	F-oil	none || much	Fresh fishoil flavor
Metallic	F-metallic	none || much	Metallic flavor
Sweet	F-sweet	none || much	Sweet flavor
Acidic	F-acidic	none || much	Acidic, sour flavor
Mouldy	F-mouldy	none || much	Mouldy flavor
Bitter	F-bitter	none || much	Bitter flavor
Rancid	F-rancid	none || much	Rancid flavor

Table 3: Genetic Descriptive Analysis scales for smoked fish floss products.

Sensory attribute	Short name	Scale anchors	Description of attribute
ODOR			
Smoke	O-smoke	none | much	smoke odor, smoked fish
Butiric acid	O-butiric	none | much	butiric acid odor, smelly feet
TMA	O-TMA	none | much	TMA odor, amine, dried fish
Spoilage sour	O-sour	none | much	spoilage sour, sour odor, sour milk, acetic acid
Rancid	O-rancid	none | much	rancid odor
Spoilage	O-spoilage	none | much	other spoilage odor, describe in comment line
FLAVOR			
Smoke flavor	F-smoke	none | much	smoke flavor, smoked fish
Spoilage sour	F-sour	none | much	sour taste, spoilage sour
TMA	F-TMA	none | much	TMA flavor, amine, dried fish
Rancid	F-rancid	none | much	rancid flavor
Bitter	F-bitter	none | much	bitter taste
Spoilage	F-spoilage	none | much	other spoilage flavor, describe in comment line

Statistical Analysis

All data summaries and statistical analyses were conducted using STATISTICA software (Version 12.0, StatSoft, Inc. 2300 East 14th Street Tulsa, OK 74104 USA) and MS-excel 2013. The Tukey HSD test was used to compare the different means. Multiple linear Regression was used to test the correlation between quality attributes (colour, free fatty acid, peroxide value, thiobarbituric acid reactive substance and total plate counts) with storage time. Significant differences were defined at $p<0.05$.

4 RESULTS

4.1 Optimize Mackerel floss and Blue whiting floss Processing

The results of pre-trials on optimizing the steaming time are presented in Table 4 and 5. In steaming stage, high temperature was used as an essential factor for connective protein (collagen and elastin) hydrolysation. This weakened the bond within fish muscle as well as the link between muscle and bone, skin. So, after being steamed, the fish meat was collected easily. However, for mackerel, if the steaming was prolonged, collagen in the skin also hydrolyzed almost into gelatin. The skin was broken into so small parts that this made skinning step to be longer. Steaming time of 10 minutes at 100^oC was appropriate for both of the raw materials (Atlantic mackerel and Blue whiting).

Table 4: Optimized (red colour) steaming step for Blue whiting floss processing.

Time (Minutes)	Cooked	Separation of fish meat and backbone
5	Outer part	a part of the muscle linked closely with the backbone
10	fully	Easily
15	fully	Easily
20	fully	Easily

Table 5: Optimized (red colour) steaming step for Atlantic mackerel floss processing.

Time (Minutes)	Cooked	Separation of fish meat and the skin
5	Outer part	easily
10	fully	Easily
15	fully	The skin was too soft and easily broken while being removed. This led the next step taken longer time
20	fully	The skin was too soft and broken while being removed. This led the next step taken longer time

Pre-trials performed in hot drying optimization were observed fluffy texture, rancid and burnt odor and also measured the water activity. Those results are indicated in Table 6 and 7. 40 minutes was optimal hot-drying time for Atlantic mackerel floss processing. Meanwhile, the appropriate hot drying time for Blue whiting floss processing was 60-70 minutes.

Table 6: Optimized (red colour) hot-drying step for Mackerel floss processing.

Time (Minutes)	Water activity	Sensory feature
30	0.94	Wet, sticky form
40	0.92	much fluffy
50	0.91	Lightly rancid odor, rather fluffy
60	0.87	Strongly Rancid odor, the shreds was broken into fine fragments

Table 7: Optimized (red colour) hot-drying step for Blue whiting floss processing.

Time (Minutes)	Water activity	Sensory feature
50	0.95	Wet, sticky form
60	0.93	Slightly wet, much fluffy
70	0.92	much fluffy
80	0.90	Lightly burnt smell. The shreds was broken into fine fragments look like powder because they were too dry

4.2 Chemical compositions, the amount of microorganisms and sensory quality of initial raw materials

The chemical composition, total viable counts (TVC) and H_2S-producing bacteria of the raw materials were measured for skinless fillet of Blue whiting and white muscle of Atlantic mackerel. The determined results are shown in Table 8. The sensory quality of the raw materials is indicated in Appendix 1. Generally, the quality of raw material was rather good.

Table 8: Chemical compositions and the number of microorganisms of initial raw materials.

Raw material	Protein content (%)	Lipid content (%)	Water content (%)	TVB-N (g/100g)	TVC (cfu/g)	H2S-Producing bacteria (cfu/g)
BWM*	21.13±0.2	0.413±0.00	77.42±0.07	16.06±1.04	$10^{3.4}$	45
BWC*	16.7±3.0	0.69±0.18	80.4±4	18.3±0.00	10^{5}	<20
Atlantic Mackerel	19.44±0.05	15.01±0.11	63.4±0.13	14.83±0.16	< 20	< 20

*: BWC: whole frozen Blue whiting raw fish; BWM: Beheaded and gutted frozen Blue whiting raw fish.

4.3 Chemical compositions and amount of microorganisms of final products

After hot drying, the final fish floss products were analysed for chemical compositions and microbial quantities. The water activities were 0.92 both of BWM and AM. Meanwhile, BWC has water activity of 0.946. The chemical compositions and TVC of the final products are indicated in Table 9. Generally, the amount of TVC and H2S-producing bacteria of the final products were decreased after processed compared to the raw materials.

Table 9: Chemical compositions and amount of microorganisms of the final products.

Product Group	Protein content (%)	Lipid content (%)	Water content (%)	TVC (cfu/g)	H2S-Producing bacteria (cfu/g)
BWC*	39.57±0.05	1.35±0.00	52.86±0.32	< 20	< 20
BWM*	32.99±0.03	1.38±0.03	60.44±0.04	<20	<20
AM*	27.71±0.37	29.61±0.23	36.59±0.25	< 20	< 20

*: BWC: Fish floss product from the whole frozen Blue whiting hot dried for 60 minutes; BWM: fish floss product from the beheaded and gutted frozen Blue whiting hot dried for 70 minutes; AM: mackerel floss product

4.4 Effects of storage time on the quality of fish floss products.

Three groups of fish floss products were produced according to the optimized processing and quality changes during storage were studied including:

BWC group: smoked fish floss product made from the whole frozen Blue whiting raw material and being hot dried for 60 minutes.
BWM group: smoked fish floss product made from the beheaded and gutted Blue whiting and being hot dried for 70 minutes.

AM group: smoked fish floss product made from the beheaded and gutted Atlantic mackerel.

4.4.1 Microbial changes

The Total viable count (TVC) and H_2S-producing bacteria count (cfu/g) of the fish floss products during stored time are shown in Table 10. In general, the number of microorganisms of fish floss products was little during prolonged chilled time, almost less than 20 cfu/g. The TVC of BWM increased after storage time of 2 weeks and the amount of microbiology in AM sample also grew at week 4 in storage. However, those growths were so slight.

Table 10: Total viable count (TVC) (cfu/g) and H_2S-producing bacteria count (cfu/g) of the fish floss products during stored time.

	BWC		BWM		AM	
Time (weeks)	TVC (cfu/g)	H_2S-Producing bacteria (cfu/g)	TVC (cfu/g)	H_2S-Producing bacteria (cfu/g)	TVC (cfu/g)	H_2S-Producing bacteria (cfu/g)
0	< 20	< 20	< 20	< 20	< 20	< 20
2	< 20	< 20	80	< 20	< 20	< 20
4	< 20	< 20	< 20	< 20	40	< 20

4.4.2 Color changes

<u>Lightness (L-Value)</u>

The L-Value measurement of the fish floss products are shown in Figure 5. Generally, the lightness (L-Value) decreased for all three groups of products as storage time increased. The significant negative correlation ($p<0.05$) between in lightness and storage time was observed for AM group ($r= -0.8$), BWM ($r= -0.8$) and BWC ($r= -0.69$). However, the decrease of lightness was not significant from week 2 to week 4 for BWM and BWC. Also there was not a significant change in lightness for AM between the initial value and after storage for 2 weeks ($p>0.05$). Moreover, the lightness of mackerel floss product was always significant lower compared to Blue whiting floss products over the storage time ($p<0.01$).

For the Blue whiting group, there was no significant difference of lightness in the final products after hot drying ($p>0.05$). However, the brightness in BWM was higher than in BWC at week 2 and week 4 ($p<0.0005$).

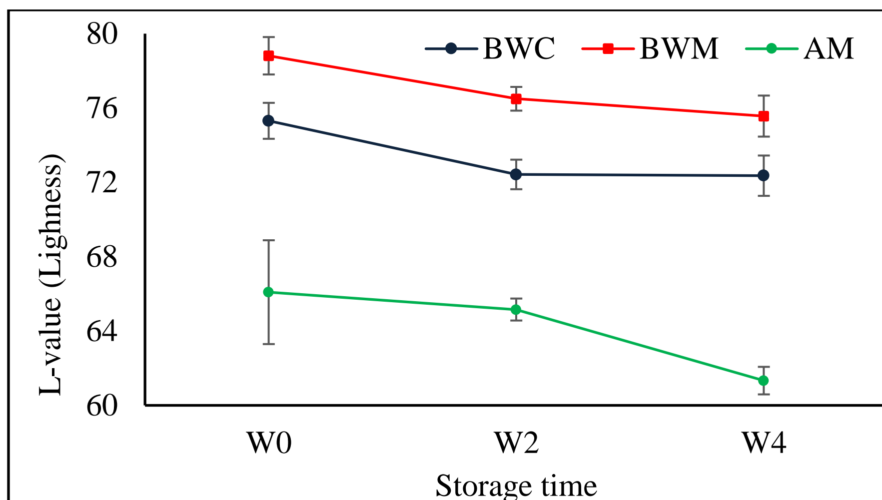

Figure 5: The lightness (L-Value) of the fish floss products during storage time.

a-value

The a-value (redness) represents the component of green color (a<0) and red color (a>0) of the products as shown in Figure 6. In general, the redness of Atlantic mackerel floss product was always higher compared to both of Blue whiting floss products as the chilled storage progressed ($p<0.05$). However, the difference was not significant at the initial products after processing among them ($p>0.05$). On the other hand, the significant differences of redness among the fish floss products were observed at week 2 and week 4 ($p<0.0005$).

For AM group, a value was rather stable the first 2 weeks after hot-drying ($p>0.05$) but increased significantly from week 2 to week 4 ($p<0.005$). Likewise, with the BWM group, there was no significant difference of redness at week 2 compared to the arrival product after hot-drying but the a-value increased significantly from week 2 to week 4 ($p<0.05$). Meanwhile, the redness of BWC group was reduced after stored for 2 weeks and rather stable until week 4 ($p>0.05$).

Besides, the significant ($p<0.005$) linear regression was contributed for AM group ($r=0.73$) and BWM group ($r=0.73$).

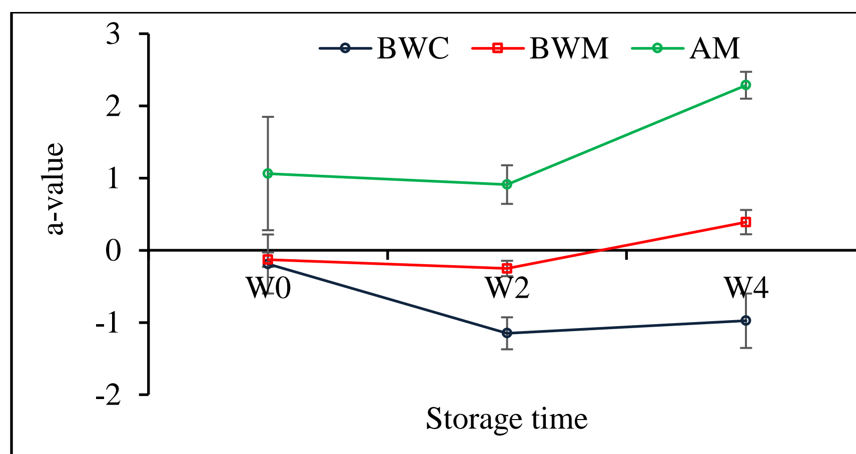

Figure 6: a-value of the fish floss products after stored time of 0 week, 2 and 4 weeks.

<u>b-value</u>

The b value (yellowness) stands for intensity of blue (b<0) and yellow (b>0) color of the fish floss products and provided in Figure 7. According to the figure, the yellowness of all three groups of fish floss products grew when the chilled storage prolonged. Nevertheless, the differences were insignificant at week 0 and week 2 for BWM group and week 2 compared to week 4 for BWC group (p>0.05). Additionally, there was no significant difference of yellowness of the final product appeared at week 0 comparing BWM with AM group (p>0.05). By contrast, a higher significantly b-value of AM group compared to Blue whiting groups was investigated at week 2 and week 4 (p<0.0005).

For Blue whiting groups, b-value of BWC was normally less than in the BWM although the difference was not significant at week 2 (p>0.05).

There was a significant (p<0.01) positive correlation between yellowness and stored time was generated for all samples, in which, AM group (r=0.93), BWC (r=0.7) and BWM (r=0.78)

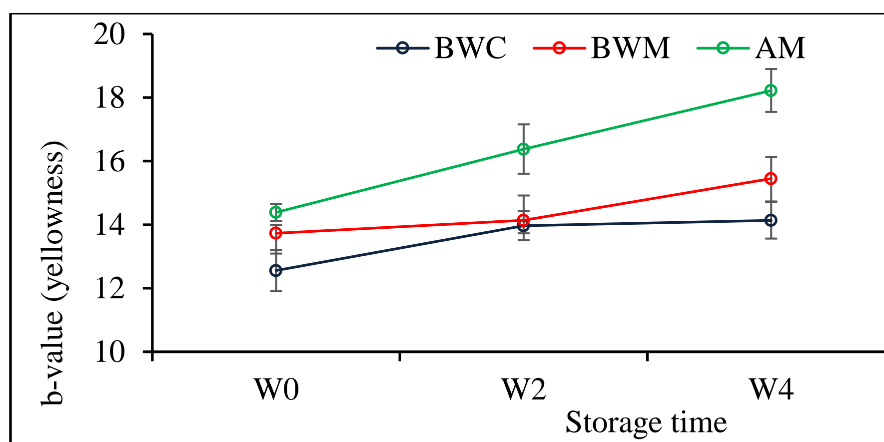

Figure 7: b-value (yellowness) of the fish floss products during chilled storage time.

4.4.3 Lipid quality

Lipid content (%), free fatty acids content (FFA, g/100g lipid), peroxide value (PV) and thiobarbituric acid reactive substance (TBARS) concentrations ((µmol/kg) are presented Figure 8a, 8b, 9 and 10, respectively.

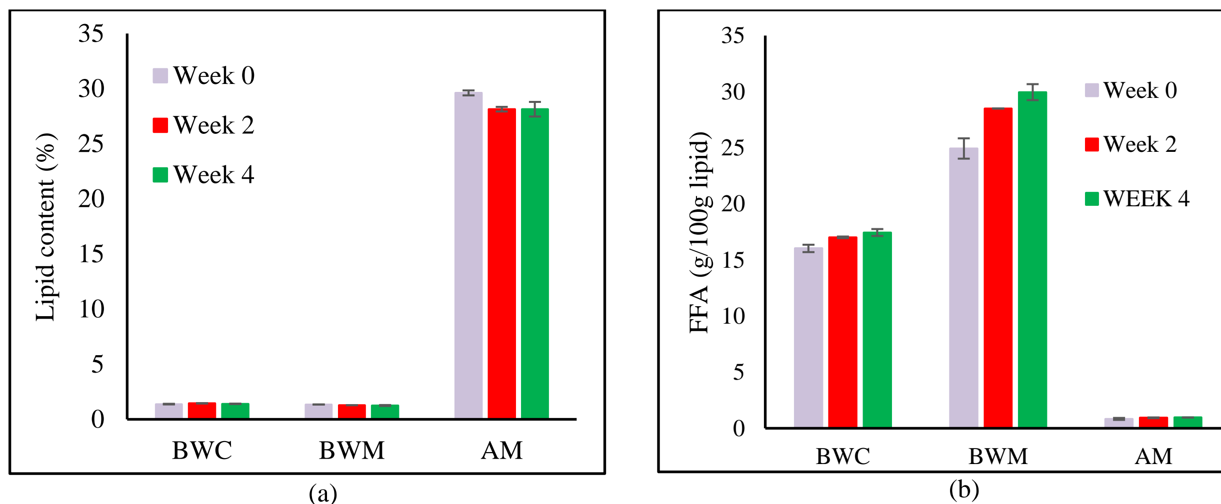

Figure 8: The lipid content (%) (a) and free fatty acids (g FFA/100g lipid) (b) in the fish floss products during storage.

Lipid content and FFA

It was evident that the lipid content of the final products was always higher than in the raw materials because of the decrease of water content after processing, the raw fish lipid contents were 0.69%, 0.41% and 15.01% compared to the final products lipid content of 1.38%, 1.35% and 29.61% in BWC, BWM, and AM group, respectively. By contrast, the FFA concentrations were rather stable after processing compared to the initial raw materials for AM group ($p>0.05$), with 0.86 (g/100g lipid) in raw fish and 0.92 (g/100g lipid). The FFA in the final products made from Blue whiting rose after the processing, was indicated clearly for BWC group ($p<0.05$). However, the increase was not significant for BWM group ($p>0.05$).

In additional, the lipid content of fish floss product remained rather stable during prolonged chilled time for BWM ($p>0.1$) and AM ($p>0.05$). Meanwhile, the lipid content of BWM group at week 4 showed significantly lower concentration compared to the final product right after hot drying ($p<0.05$). Moreover, the FFA was also stable during storage time for AM and BWC group ($p>0.05$). Nonetheless, the FFA of BWM rose significantly after stored for 4 weeks compared to the initial product ($p<0.0005$) and a strongly significant positive correlation between increase FFA and chilled storage time was generated for this group ($r=0.94$).

Within Blue whiting fish floss product groups, no significant difference of lipid concentration was indicated when the storage time progressed but the FFA concentrations in BWM group were always significant higher than BWC group in the storage($p<0.05$).

PV and TBARS

Lipid hydroperoxide (PV) and thiobarbituric acid reactive substance (TBARS) are used to indicate the lipid oxidation in food products. In general, after processing, the PV of arrival fish floss products increased compared to the initial raw materials. This presented clearly for AM group

(p<0.0005) with an increase of approximately 4 times, from 54.22 (µmol/kg) in the initial material to 201.29 (µmol/kg) in the final product. However, the growths were insignificant in both Blue whiting groups (p>0.05). For the Blue whiting groups, the PV was rather stable during prolonged storage time. Moreover, the peroxide concentration of the BWC was always higher than the BWM over the storage time but the differences was not significant at week 0 and week 4 (p>0.1). For the PV of AM group, there was a slight decrease at week 2 (p>0.05) but a rapid increase after that 2 weeks (p<0.0005), from 166.77 to 336.71 (µmol/kg). A significant (p<0.05) positive correlation between the PV and storage time was also observed in this group (r=0.79)

Likewise, the TBARS also grew significantly after processing in BWM group (p<0.0005) and AM group (p<0.05). Nevertheless, the TBARS of BWC arrival product was significantly lower than in the initial material (p<0.001). The TBARS concentrations remained rather stable during storage time for Blue whiting fish floss products groups. However, the TABRS of AM group increased considerably over storage and a strongly significant (p<0.0005) positive linear correlation with chilled storage time was indicated in this group (r=0.94).

For Blue whiting groups, The TBARS of BWM was always significant less than the BWM group at the same storage time (p<0.01).

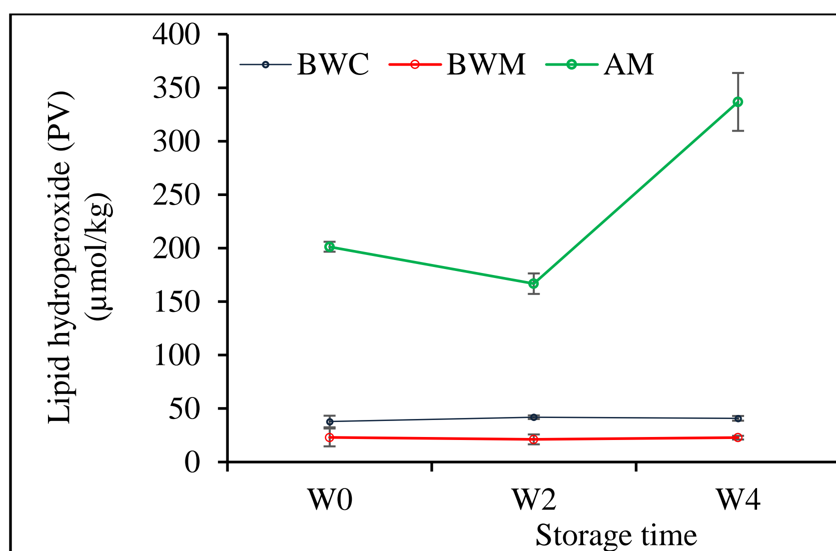

Figure 9: Lipid hydroperoxide formation (µmol/kg) in the fish floss products at week 0, 2 and 4.

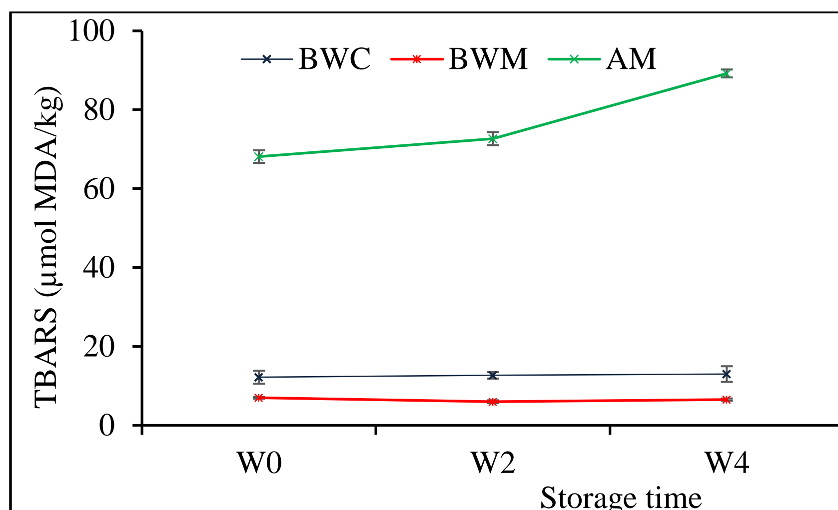

Figure 10: TBARS formation (µmol MDA/kg) in the fish floss products during storage time.

4.4.4 Water content and water activity

Water content (%) and water activity of the fish floss products were showed in Table 11 and 12. As we can see, the water content of BWC and AM group were rather stable during storage time. Likewise, the water activities were no significant change after stored time of 4 weeks for both BWM and AM group. On the other hand, the water content of BWM was significantly lower at week 4 compared to week 2 ($p<0.05$) and also with water activity of BWC ($p<0.04$).

Table 11: Mean of water content (%) of the fish floss products during stored time*.

Time (weeks)	BWC	BWM	AM
0	60.44±0.40a	52.86±0.32a	36.59±0.25a
2	60.00±0.01a	52.93±0.12a	36.79±0.37a
4	59.53±0.30a	51.89±0.13b	36.17±0.27a

a, b Means followed by different letters within same column are significant different ($p<0.05$)
*: The results are expressed as Mean ± Standard Deviation.

Table 12: Mean of water activity of the fish floss products after stored time of 0 week, 2 and 4 weeks *.

Time (weeks)	BWC	BWM	AM
0	0.946±0.001a	0.920±0.001a	0.920±0.000a
2	0.946±0.001a	0.921±0.002a	0.921±0.001a
4	0.942±0.002b	0.919±0.000a	0.920±0.002a

a, b Means followed by different letters within same column are significant different ($p<0.05$)
*: The results are expressed as Mean ± Standard Deviation.

4.4.5 Sensory quality changes

The fish floss products were evaluated sensory quality, including specific characteristics (smoke odor and smoke flavor) and spoilage characteristics: butiric odor, TMA odor, sour odor, rancid odor, spoilage odor, sour flavor, TMA flavor, rancid flavor, bitter flavor and spoilage flavor according to the Generic Descriptive Analysis method. The sensory score of odor and flavor attributes of the fish floss products are shown in Table 13 and 14.

After processing, all the final products had specifically smoke odor and flavor quite strong (more than 50 GDA scores). The butyric odor, bitter flavor, rancid odor and flavor were not significantly difference compared to in the initial raw material for AM group ($p>0.05$). Likewise, there was not significant changes for TMA, sour, rancid odors and TMA, sour, rancid flavor in the Blue whiting final products after processing compared to the raw materials ($p>0.05$). Within the Blue whiting groups stored for same time, no any significant differences were observed for each sensory attribute between two samples ($p<0.05$).

Generally, smoke odor and smoke flavor were slight decreased in storage for all fish floss products. Meanwhile, the attributes showing spoilage of the product went up at week 4, such as TMA odor (AM group), sour odor (all samples), rancid odor (all samples), and TMA flavor (AM). However, those changes were insignificant statistically ($p>0.05$).

Additionally, smoke odor and flavor of all samples during were rather strong, with the score ranges of 50-59 and 49-57, respectively. On the other hand, the spoilage characteristics were detected so slightly for all products during chilled storage (almost those less than 10 scores). So, it could conclusion that the quality of fish floss products was rather stable and being not spoilage during chilled storage.

Table 13: GDA scores of odor attributes of various fish floss products during storage.

Sample	Storage time	O-smoke	O-butiric	O-TMA	O-sour	O-rancid	O-spoilage
	0	56	9	2	1	3	1
	2	51	5	2	1	3	0
AM	4	50	8	6	5	6	1
p-value		*0.513*	*0.416*	*0.213*	*0.406*	*0.634*	*0.518*
	0	59	6	1	0	1	1
	2	52	4	5	1	4	4
BWC	4	51	12	3	3	5	1
p-value		*0.264*	*0.451*	*0.301*	*0.312*	*0.351*	*0.093*
	2	56	3	2	0	0	0
BWM	4	51	6	1	0	2	0
p-value		*0.272*	*0.465*	*0.068*	*0.54*	*0.67*	*0.685*

Table 14: Means of GDA score of flavor attributes of various fish floss products during storage.

Sample	Storage time	F-smoke	F-sour	F-TMA	F-rancid	F-bitter	F-spoilage
	0	53	2	2	4	11	1
	2	57	2	5	1	8	1
AM	4	52	5	8	7	12	1
p-value		*0.706*	*0.6*	*0.359*	*0.459*	*0.896*	*0.737*
	0	53	1	1	0	5	0
	2	53	1	7	3	9	3
BWC	4	49	2	3	3	7	1
p-value		*0.817*	*0.424*	*0.117*	*0.648*	*0.692*	*0.347*
	2	57	1	7	1	9	3
BWM	4	49	1	3	2	6	0
p-value		*0.105*	*0.923*	*0.385*	*0.369*	*0.604*	*0.291*

5 DISCUSSION

5.1 The optimization of steaming time and hot-drying time

The research indicated that the optimal steaming time was 10 minute for both fillet Atlantic mackerel and beheaded, gutted and skinless Blue whiting at 100ºC. This is an importance step in the fish floss processing. The bond between muscles with bone, skin is broken under heat, so the muscle can be separated easily. Some off-odors are removed in this step. Moreover, under high temperature at this stage, enzymes and microorganisms are inactivated. This can prevent further quality changes which can adversely affect the quality of final product. The steaming time varies according to raw material species, steaming temperature and previous treatments.

The study results were similar to another fish floss processed from Greater amberjack (*Seriola dumerili*) (Nguyen and Truong 2010), Tilapia (*Oreochromis niloticus*) (Nguyen *et al.* 2010). However, this steaming time was much less than compared to in the pork floss processing which took 3 hours for steaming (Liao *et al*. 2009).

The optimized hot-drying time at 85ºC was 40 minutes for Atlantic mackerel and 60-70 minutes for the Blue whiting floss processing. The drying time depend on the drying method, fish species and the desirable water content of final product. In processing fish floss from Indian mackerel (water content of 76.44%), the drying condition is 95ºC for 65 minutes and final product had a moisture content of 22% (Tran and Do 2012). According to study the fish floss processing of Greater amberjack (*Seriola dumerili*) (Nguyen and Truong 2010), the fish shreds is dried at 100ºC for 70 minutes.

5.2 Chemical compositions and amount of microorganisms of the final products

The protein content, lipid content in the final products increased significantly comparing to the raw materials, from 21.13±0.2% to 39.57±0.05% (Blue whiting) and 19.44±0.05% to 27.71±0.37% (Atlantic mackerel) for protein, from 0.413 to 1.35% (Blue whiting) and 15.01±0.11% to 29.61±0.23% (Atlantic mackerel) for lipid. This was probably a consequence of the evaporation of water during processing. Water content went from 80.4±4 down to 52.86±0.32 (BWM) 77.42±0.07 down to 60.44±0.40 (BWC) and 63.4±0.13 down to 36.59±0.25 (Atlantic mackerel).

Thermal oxidation is one of the greatest threats to lipid in food. Unsaturated lipids are especially susceptible (Stewart *et al*., 2003; Webera *et al*., 2008). The results indicated a significant amount of lipid hydroperoxide and thiobituric acid reactive substances (TBARS) were formed in the processing for most of fish floss products, especially in AM group. This results are in agreement with (Fu *et al*, 2014). High temperature used accompanying drying process for a long time can be the most important cause for the formations lipid hydroperoxide and TBARS in the fish floss products.

The number of microorganisms went down in the processing. This can be seen clearly for Blue whiting, with amount of microorganisms were $10^{3.4}$ and 10^5 cfu/g in the raw materials and were reduced to below 20 colonies/g in the final products. Those decreases can be explained by the high temperature used and the reduction of water activity because of the smoking and drying in the processing according to (Derrick 2009).

According to a survey performed in commercial meat floss products in Malaysia (Huda *et al*., 2012), the fat content varied from 3.20-31.14%, the protein content was within the range of 19.86-30.15% and the water content from 8.6-13.56%. The protein content of fish floss from Blue whiting and the lipid content in Atlantic mackerel floss were so high. In contrast, its lipid content was small in Blue whiting floss product. It is also clear that water contents of the smoked fish floss made from Atlantic mackerel and Blue whiting were much higher. Also, the water activity was high. Those can be a problem concerning the storage of products. Normally, with the low water content, commercial floss meat can be kept at room temperature for a long time (Huda *et al*. 2012, Li *et al*. 2000). So, chilled storage should be one of suitable methods for storage the fish floss products having high water content made from Atlantic mackerel and Blue whiting. In fact, the study also investigated the shelf life of the product can be at least 4 weeks stored at 2-4ºC.

5.3 Quality changes of the fish floss products during chilled storage time

The brightness decreased while the yellowness increased by storage time for all fish floss products. According (Liao *et al.* 2009), the deep brown color of shredded meat developed at higher cooking temperature correlated with the non-enzymatic browning (such as Maillard reaction) and caramel reaction during meat floss preparation. Those browning reactions can be continued in the storage so the lightness can be decreased while the yellowness raise.

Thermal processing is still one of the most common methods for achieving safe convenience foods with an extended shelf life. The microbial growth can be inactivated or inhibited after applying thermal treatments (Skipnes *et al.* 2011). The low number of total viable counts and spoilage bacteria and the so slow microbial growth during storage of the products were shown in this research. According to the Guideline of Public health laboratory service - London (Gilbert *et al*. 2000), the microbial quality of the fish floss products was classified satisfactory for the consumer. Moreover, epidemiological data indicate that foods involved in listeriosis outbreaks are those in which the organism has multiplied and generally contain levels significantly higher than 100 CFU/g. (Buchanan *et al*. 1997). The sensory results also presented no any spoilage attributed detected over the storage at 2-4ºC for all products. So, these observations suggest that more than 4 weeks of shelf-life for fish floss products made from Atlantic mackerel and Blue whiting may be expected.

Lipid oxidations are catalyzed by light, heat, trace metals or enzymes and involves free radical generation. Free radicals propagate autoxidation by reacting with oxygen to form hydroperoxides, which breakdown to generate other new free radicals and TBARS. For Blue whiting groups, lipid hydroperoxide and TBARS concentration were rather stable by the storage time. It can be explained by the low lipid content of this raw material (lean fish). However, the rancidity was strongly affected by storage time shown in Atlantic mackerel (fatty fish) fish floss product.

6 CONCLUSION

The research results showed that the optimal steaming time was 10 minutes at 100ºC for both Atlantic mackerel and Blue whiting fish floss processing. The appropriate hot drying time of Atlantic mackerel floss was 40 minute while 60-70 minutes was the relevant hot drying time of fish floss processing made from Blue whiting at 85ºC.

Sensory quality, water content, water activity of all fish floss products were rather stable during chilled storage at 2-4ºC for 4 weeks. In additional, the amounts of microorganisms, the concentrations of PV and TBARS in Blue whiting and FFA in Atlantic mackerel floss products did not change significantly. However, lipid oxidation in AM group occurred significantly. Moreover, the lightness went down but the yellowness rose for all products during chilled storage.

The study also indicated that the difference about sensory, lipid quality, the TVC and the number of spoilage bacteria were not significant within Blue whiting fish floss products at the same storage time.

The fish floss products made from Atlantic mackerel and Blue whiting had high nutrient value and can be stored at 2-4ºC at least 4 weeks.

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APPENDICES

Appendix 1: Sensory results

Table 1: Sensory score of cooked Atlantic mackerel fillet*

Sensory Attribute	Score	Sensory Attribute	Score
O-oil	24	F-oil	24
O-metallic	25	F-metallic	26
O-sweet	31	F-sweet	33
O-mouldy	4	F-acidic	4
O-butiric	1	F-mouldy	5
O-rancid	4	F-bitter	8
		F-rancid	5

*: The values are expressed as Mean±Standard Deviation

Table 2: The score of sensory attributes of cooked fillets from the whole frozen and beheaded, gutted frozen Blue whiting raw fish*

Attribute	Whole frozen	Beheaded, gutted frozen	Attribute	Whole frozen	Beheaded, gutted frozen
O-sweet	27	26	F-salt	19	15
O-shellfish	21	18	F-metallic	22	21
O-vanilla	22	14	F-sweet	33	28
O-TMA	4	6	F-pungent	8	10
O-sour	1	3	F-sour	1	1
O-rancid	1	7	F-dryed fish	20	18
O-frozen	10	12	F-TMA	1	3
			F-frozen	9	15
			F-rancid	4	4

*: The values are expressed as Mean±Standard Deviation

Appendix 2: Summary analysed data

Tukey HSD test and linear Regression

M: Mean; RM: raw material; Differences significant ($p<0.05$) are marked red colour

Table 1: Tukey HSD test for L-value of AM group between weeks of storage time

Storage Time (Week)	Tukey HSD test; Variable: AM (L-Value) (Spreadsheet8) Marked differences are significant at p < .05000		
	{1} M=66.090	{2} M=65.158	{3} M=61.332
0 {1}		0.713203	0.005483
2 {2}	0.713203		0.011875
4 {3}	0.005483	0.011875	

Table 2: Linear Regression for L-value of AM sample with storage time

N=12	Regression Summary for Dependent Variable: AM (L-Value) (Spreadsheet8) R= .80302012 R²= .64484132 Adjusted R²= .60932545 F(1,10)=18.156 p<.00166 Std.Error of estimate: 1.6276					
	b*	Std.Err.of b*	b	Std.Err.of b	t(10)	p-value
Time	-0.803020	0.188457	-1.25239	0.293917	-4.26104	0.001660

Table 3: Tukey HSD test for L-value of BWC samples between weeks of storage time

Stored Time (week)	Tukey HSD test; Variable: BWC (L-value) (Spreadsheet8) Marked differences are significant at p < .05000		
	{1} M=75.310	{2} M=72.424	{3} M=72.358
0 {1}		0.005318	0.004604
2 {2}	0.005318		0.993522
4 {3}	0.004604	0.993522	

Table 4: Linear Regression for L-value of BWC sample with storage time

N=13	Regression Summary for Dependent Variable: BWC (L-value) (Spreadsheet8) R= .69016224 R²= .47632392 Adjusted R²= .42871700 F(1,11)=10.005 p<.00903 Std.Error of estimate: 1.1706					
	b*	Std.Err.of b*	b	Std.Err.of b	t(11)	p-value
Time	-0.690162	0.218190	-0.66750	0.211026	-3.1631	0.009029

Table 5: Tukey HSD test for L-value of BWC samples between weeks of storage time

Stored Time (week)	Tukey HSD test; Variable: BWM (L-value) (Spreadsheet8) Marked differences are significant at $p < .05000$		
	{1} M=78.810	{2} M=76.496	{3} M=75.564
0 {1}		0.016288	0.002004
2 {2}	0.016288		0.291903
4 {3}	0.002004	0.291903	

Table 6: Linear Regression for L-value of BWM sample with storage time

N=13	Regression Summary for Dependent Variable: BWM (L-value) (Spreadsheet8) R= .80676860 R^2= .65087558 Adjusted R^2= .61913700 F(1,11)=20.507 p<.00086 Std.Error of estimate: .95169					
	b*	Std.Err.of b*	b	Std.Err.of b	t(11)	p-value
Time	-0.806769	0.178153	-0.77695	0.171569	-4.5285	0.000860

Table 7: Tukey HSD test for L-value of storage time of 0 week between various samples

Sample	Tukey HSD test; Variable: Week 0 (L-value) (Spreadsheet8) Marked differences are significant at $p < .05000$		
	{1} M=75.310	{2} M=78.810	{3} M=66.090
BWC {1}		0.119342	0.002041
BWM {2}	0.119342		0.000511
AM {3}	0.002041	0.000511	

Table 8: Tukey HSD test for L-value of storage time of 2 weeks between various samples

Sample	Tukey HSD test; Variable: Week 2 (L-value) (Spreadsheet8) Marked differences are significant at $p < .05000$		
	{1} M=72.424	{2} M=76.496	{3} M=65.158
BWC {1}		0.000209	0.000198
BWM {2}	0.000209		0.000198
AM {3}	0.000198	0.000198	

Table 9: Tukey HSD test for L-value of storage time of 4 weeks between various samples

Sample	Tukey HSD test; Variable: Week 4 (Value) (Spreadsheet8) Marked differences are significant at $p < .05000$		
	{1} M=72.358	{2} M=75.564	{3} M=61.332
BWC {1}		0.000836	0.000190
BWM {2}	0.000836		0.000190
AM {3}	0.000190	0.000190	

Table 10: Tukey HSD test of a-value of AM samples between various storage time

Stored Time (week)	Tukey HSD test; Variable: AM (a-value) (Spreadsheet10) Marked differences are significant at $p < .05000$		
	{1} M=1.0633	{2} M=.91000	{3} M=2.2860
0 {1}		0.880593	0.007579
2 {2}	0.880593		0.002193
4 {3}	0.007579	0.002193	

Table 11: Linear Regression for a-value of AM sample with storage time

N=12	Regression Summary for Dependent Variable: AM(a-value) (Spreadsheet14) R= .73075059 R^2= .53399643 Adjusted R^2= .48739607 F(1,10)=11.459 p<.00694 Std.Error of estimate: .55443					
	b*	Std.Err.of b*	b	Std.Err.of b	t(10)	p-value
Time	0.730751	0.215871	0.338913	0.100118	3.385124	0.006943

Table 12: Tukey HSD test for a-value of BWC sample between weeks of storage time

Stored Time (week)	Tukey HSD test; Variable: BWC(a-value) (Spreadsheet10) Marked differences are significant at $p < .05000$		
	{1} M=-.1900	{2} M=-1.150	{3} M=-.9760
0 {1}		0.006979	0.021942
2 {2}	0.006979		0.693974
4 {3}	0.021942	0.693974	

Table 13: Tukey HSD test for a-value of BWM sample between various storage weeks

Stored Time (week)	Tukey HSD test; Variable: BWM (a-value) (Spreadsheet10) Marked differences are significant at $p < .05000$		
	{1} M=-.1300	{2} M=-.2520	{3} M=.39000
0 {1}		0.453780	0.001045
2 {2}	0.453780		0.000240
4 {3}	0.001045	0.000240	

Table 14: Linear Regression for a-value of BWM sample with storage time

N=12	Regression Summary for Dependent Variable: BWM (a-value) (Spreadsheet14) R= .72513331 R^2= .52581832 Adjusted R^2= .48271090 F(1,11)=12.198 p<.00504 Std.Error of estimate: .23681					
	b*	Std.Err.of b*	b	Std.Err.of b	t(11)	p-value
Time	0.725133	0.207623	0.149100	0.042691	3.49254	0.005036

Table 15: Tukey HSD test for a-value of storage time of week 0 between various samples

Sample	Tukey HSD test; Variable: Week 0 (a-value) (Spreadsheet10) Marked differences are significant at p < .05000		
	{1} M=-.1900	{2} M=-.1300	{3} M=1.0633
BWC {1}		0.988897	0.055512
BWM {2}	0.988897		0.066311
AM {3}	0.055512	0.066311	

Table 16: Tukey HSD test for a-value of storage time of week 2 between various samples

Sample	Tukey HSD test; Variable: Week 2 (a-value) (Spreadsheet10) Marked differences are significant at p < .05000		
	{1} M=-1.150	{2} M= -0.2520	{3} M=0.91000
BWC {1}		0.000226	0.000198
BWM {2}	0.000226		0.000200
AM {3}	0.000198	0.000200	

Table 17: Tukey HSD test for a-value of storage time of week 4 between various samples

Sample	Tukey HSD test; Variable: Week 4 (a-value) (Spreadsheet10) Marked differences are significant at p < .05000		
	{1} M=-.9760	{2} M=.39000	{3} M=2.2860
BWC {1}		0.000193	0.000190
BWM {2}	0.000193		0.000190
AM {3}	0.000190	0.000190	

Table 18: Tukey HSD test for b-value of AM sample between various weeks of storage

Stored Time (week)	Tukey HSD test; Variable: AM (b-value) (Spreadsheet21) Marked differences are significant at p < .05000		
	{1} M=14.387	{2} M=16.380	{3} M=18.220
0 {1}		0.010581	0.000249
2 {2}	0.010581		0.007960
4 {3}	0.000249	0.007960	

Table 19: Linear Regression for b-value of AM sample with storage time

N=12	Regression Summary for Dependent Variable: AM (b-value) (Spreadsheet24) R= .93181722 R²= .86828334 Adjusted R²= .85511167 F(1,10)=65.921 p<.00001 Std.Error of estimate: .65137					
	b*	Std.Err.of b*	b	Std.Err.of b	t(10)	p-value
Time	0.931817	0.114768	0.95500	0.117623	8.11915	0.000010

Table 20: Tukey HSD test for b-value of BWC sample between various storage weeks

Stored Time (week)	Tukey HSD test; Variable: BWC (b-value) (Spreadsheet21) Marked differences are significant at p < .05000		
	{1} M=12.557	{2} M=13.966	{3} M=14.134
0 {1}		0.013620	0.006997
2 {2}	0.013620		0.878819
4 {3}	0.006997	0.878819	

Table 21: Linear Regression for b-value of BWC sample with storage time

N=13	Regression Summary for Dependent Variable: BWC (b-value) (Spreadsheet24) R= .70435222 R²= .49611205 Adjusted R²= .45030405 F(1,11)=10.830 p<.00719 Std.Error of estimate: .61236					
	b*	Std.Err.of b*	b	Std.Err.of b	t(11)	p-value
Time	0.704352	0.214028	0.36330	0.110394	3.29093	0.007193

Table 22: Tukey HSD test for b-value of BWM sample between various storage weeks

Stored Time (week)	Tukey HSD test; Variable: BWM (b-value) (Spreadsheet21) Marked differences are significant at p < .05000		
	{1} M=13.730	{2} M=14.140	{3} M=15.450
0 {1}		0.628820	0.007216
2 {2}	0.628820		0.015329
4 {3}	0.007216	0.015329	

Table 23: Linear Regression for b-value of BWM sample with storage time

N=13	Regression Summary for Dependent Variable: BWM (Spreadsheet24) R= .77556590 R²= .60150246 Adjusted R²= .56527541 F(1,11)=16.604 p<.00184 Std.Error of estimate: .61599					
	b*	Std.Err.of b*	b	Std.Err.of b	t(11)	p-value
Time	0.775566	0.190334	0.45250	0.111049	4.07476	0.001836

Table 24: Tukey HSD test for b-value between various samples at week 0

Sample	Tukey HSD test; Variable: week 0 (b-value) (Spreadsheet21) Marked differences are significant at p < .05000		
	{1}	{2}	{3}
	M=12.557	M=13.730	M=14.387
BWC {1}		0.086376	0.015018
BWM {2}	0.086376		0.366725
AM {3}	0.015018	0.366725	

Table 25: Tukey HSD test for b-value between various samples at week 2

Sample	Tukey HSD test; Variable: week 2 (Spreadsheet21) Marked differences are significant at p < .05000		
	{1}	{2}	{3}
	M=13.966	M=14.140	M=16.380
BWC {1}		0.886832	0.000375
BWM {2}	0.886832		0.000539
AM {3}	0.000375	0.000539	

Table 26: Tukey HSD test for b-value between various samples at week 2

Sample	Tukey HSD test; Variable: week 4 (Spreadsheet21) Marked differences are significant at p < .05000		
	{1}	{2}	{3}
	M=14.134	M=15.450	M=18.220
BWC {1}		0.020881	0.000190
BWM {2}	0.020881		0.000244
AM {3}	0.000190	0.000244	

Table 27: Tukey HSD test for lipid content of AM sample between various storage weeks

Stored Time (week)	Tukey HSD test; Variable: AM (lipid content) (Spreadsheet2) Marked differences are significant at p < .05000		
	{1} M=29.614	{2} M=28.138	{3} M=28.133
0 {1}		0.053457	0.052857
2 {2}	0.053457		0.999999
4 {3}	0.052857	0.999999	

Table 28: Tukey HSD test for lipid content of BWC sample between various storage weeks

Stored Time (week)	Tukey HSD test; Variable: BWC (Lipid content) (Spreadsheet2) Marked differences are significant at p < .05000		
	{1} M=1.3800	{2} M=1.4522	{3} M=1.4055
0 {1}		0.942211	0.997123
2 {2}	0.942211		0.982821

4 {3}	0.997123	0.982821	

Table 29: Tukey HSD test for lipid content of BWM sample between various storage weeks

Stored Time (week)	Tukey HSD test; Variable: BWM (lipid content) (Spreadsheet2) Marked differences are significant at p < .05000		
	{1} M=1.3471	{2} M=1.2845	{3} M=1.2531
0 {1}		0.145844	0.043844
2 {2}	0.145844		0.553043
4 {3}	0.043844	0.553043	

Table 30: Tukey HSD test for FFA of AM sample between various storage weeks and the raw material

Stored Time (week)	Tukey HSD test; Variable: AM (FFA) (Spreadsheet2) Marked differences are significant at p < .05000			
	{1} M=.83524	{2} M=.93559	{3} M=.96175	{4} M=.89379
0 {1}		0.493435	0.324393	0.750934
2 {2}	0.493435		0.978091	0.885778
4 {3}	0.324393	0.978091		0.664571
RM {4}	0.750934	0.885778	0.664571	

Table 31: Tukey HSD test for FFA of BWC sample between various storage weeks and the raw material

Stored Time (week)	Tukey HSD test; Variable: BWC (FFA) (Spreadsheet2) Marked differences are significant at p < .05000			
	{1}	{2}	{3}	{4}
	M=16.031	M=17.019	M=17.450	M=14.127
0 {1}		0.662631	0.405116	0.136778
2 {2}	0.662631		0.953165	0.028278
4 {3}	0.405116	0.953165		0.015179
RM {4}	0.136778	0.028278	0.015179	

Table 32: Linear Regression of FFA of BWC sample with the storage time

N=6	Regression Summary for Dependent Variable: BWC (FFA) (Spreadsheet22) R= .93061799 R^2= .86604985 Adjusted R^2= .83256231 F(1,4)=25.862 p<.00705 Std.Error of estimate: .27889					
	b*	Std.Err.of b*	b	Std.Err.of b	t(4)	p-value
Time	0.930618	0.182996	0.35457	0.069723	5.08546	0.007054

Table 33: Tukey HSD test for FFA of BWM sample between various storage weeks and the raw material

Stored Time (week)	Tukey HSD test; Variable: BWM (FFA) (Spreadsheet2) Marked differences are significant at $p < .05000$			
	{1} M=24.938	{2} M=28.500	{3} M=29.961	{4} M=28.545
0 {1}		0.170769	0.044870	0.078216
2 {2}	0.170769		0.840810	0.999991
4 {3}	0.044870	0.840810		0.793108
RM {4}	0.078216	0.999991	0.793108	

Table 34: Linear Regression of FFA of BWM sample with the storage time

N=6	Regression Summary for Dependent Variable: BWM (FFA) (Spreadsheet22) R= .94391095 R^2= .89096787 Adjusted R^2= .87279585 F(1,6)=49.030 p<.00042 Std.Error of estimate: .87324					
	b*	Std.Err.of b*	b	Std.Err.of b	t(4)	p-value
Time	0.943911	0.134804	1.30363	0.186176	7.00212	0.000423

Table 35: Tukey HSD test for PV of AM sample between various storage weeks and the raw material

Stored Time (week)	Tukey HSD test; Variable: AM (PV) (Spreadsheet4) Marked differences are significant at $p < .05000$			
	{1} M=193.45	{2} M=161.27	{3} M=336.71	{4} M=54.218
0 {1}		0.224216	0.000287	0.000296
2 {2}	0.224216		0.000271	0.000967
4 {3}	0.000287	0.000271		0.000252
RM {4}	0.000296	0.000967	0.000252	

Table 36: Linear Regression of PV of AM sample with the storage time

N=9	Regression Summary for Dependent Variable: AM (PV) (Spreadsheet4a) R= .79417271 R^2= .63071029 Adjusted R^2= .56916201 F(1,6)=10.247 p<.01857 Std.Error of estimate: 54.808					
	b*	Std.Err.of b*	b	Std.Err.of b	t(7)	p-value
Time	0.794173	0.248089	35.8133	11.18760	3.201157	0.018573

Table 37: Tukey HSD test for PV of BWC sample between various storage weeks and the raw material

Stored Time (week)	Tukey HSD test; Variable: BWC (PV) (Spreadsheet4) Marked differences are significant at p < .05000			
	{1} M=27.408	{2} M=21.199	{3} M=22.788	{4} M=21.752
0 {1}		0.480684	0.691045	0.551751
2 {2}	0.480684		0.979517	0.999135
4 {3}	0.691045	0.979517		0.994118
RM {4}	0.551751	0.999135	0.994118	

Table 38: Tukey HSD test for PV of BWM sample between various storage weeks and the raw material

Stored Time (week)	Tukey HSD test; Variable: BWM (BV) (Spreadsheet4) Marked differences are significant at p < .05000			
	{1} M=37.907	{2} M=41.904	{3} M=40.852	{4} M=36.000
0 {1}		0.563327	0.762779	0.881689
2 {2}	0.563327		0.984491	0.164284
4 {3}	0.762779	0.984491		0.295872
RM {4}	0.881689	0.164284	0.295872	

Table 39: Tukey HSD test for TB of AM sample between various storage weeks and the raw material

Stored Time (week)	Tukey HSD test; Variable: AM (TB) (Spreadsheet16) Marked differences are significant at p < .05000			
	{1} M=68.121	{2} M=72.678	{3} M=89.204	{4} M=7.4214
0 {1}		0.014842	0.000231	0.000231
2 {2}	0.014842		0.000231	0.000231
4 {3}	0.000231	0.000231		0.000231
RM {4}	0.000231	0.000231	0.000231	

Table 40: Linear Regression of TB of AM sample with the storage time

N=9	Regression Summary for Dependent Variable: AM (TBARS) (REG.) R= .94227242 R²= .88787731 Adjusted R²= .87185978 F(1,7)=55.432 p<.00014 Std.Error of estimate: 3.4681					
	b*	Std.Err.of b*	b	Std.Err.of b	t(7)	p-value
Time	0.942272	0.126560	5.27068	0.707926	7.44524	0.000144

Table 41: Tukey HSD test for TB of BWC sample between various storage weeks and the raw material

Stored Time (week)	Tukey HSD test; Variable: BWC (tb) (Spreadsheet16) Marked differences are significant at p < .05000			
	{1} M=12.206	{2} M=12.665	{3} M=12.997	{4} M=21.511
0 {1}		0.987574	0.942488	0.000988
2 {2}	0.987574		0.995263	0.001322
4 {3}	0.942488	0.995263		0.001645
RM {4}	0.000988	0.001322	0.001645	

Table 42: Tukey HSD test for TB of BWM sample between various storage weeks and the raw material

Stored Time (week)	Tukey HSD test; Variable: BWM(TB) (Spreadsheet16) Marked differences are significant at p < .05000			
	{1} M=6.9929	{2} M=5.9899	{3} M=6.5209	{4} M=2.9864
0 {1}		0.269680	0.788479	0.000402
2 {2}	0.269680		0.726776	0.001705
4 {3}	0.788479	0.726776		0.000696
RM {4}	0.000402	0.001705	0.000696	