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Title: Rice Flour - A Functional Ingredient for Premium Crabstick

Abstract approved:

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Rice flour possesses functional properties in enhancing texture and whiteness. This study was carried out to evaluate rice flour as a functional ingredient for premium crabstick and to develop a commercially viable recipe for premium crabstick. Crabstick pastes were formulated with constant surimi (42%) and combined rice flour and starch (8%) concentrations. Rice flour concentration was varied (0, 1, 3, and 5%) in order to evaluate changes in physicochemical properties of crabstick pastes. The physical properties of cooked gels were measured during refrigerated and frozen storage. Rice pastes with various concentrations of rice flour (5 to 40%) mixed in water, demonstrated similar patterns for differential scanning calorimetry (DSC) with

endothermic peaks at around 63.5^{o}C. During refrigerated storage up to 21 days, gel strength increased gradually, while cohesiveness was not significantly changed (P>0.05). At 1% rice flour addition, fracture gel properties during 21 days of refrigerated storage showed optimum results. During frozen storage, water retention ability (WRA) gradually decreased as freeze-thaw (F/T) cycles were extended. However, the water loss was minimized as rice flour concentration increased. Two different crabstick samples (control and 1% rice flour) demonstrated no significant difference (P>0.05) in gel hardness and cohesiveness, respectively. Rice flour (1%) can, therefore, be used to replace various starches as a functional ingredient in premium crabstick formulations.

Rice Flour – A Functional Ingredient for Premium Crabstick

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I understand that my thesis will become part of the permanent collection of Oregon State University libraries. My signature below authorizes release of my thesis to any reader upon request.

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Sungik Hur, Author

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CHAPTER 1

INTRODUCTION

Surimi-based crabstick has become the most popular surimi seafood outside Japan since its development by Sugino and Osaki independently in Japan in 1975 (Okada, 1992). Currently, approximately 35% of surimi produced globally is used for crabstick production, predominantly in the U.S.A. and Europe. South Korea (Korea hereafter) also played an important role in developing the U.S. surimi seafood market during the mid 1980s. Korea is the second largest surimi seafood producing country (Park, 2005a). Korea uses about 15% of the total surimi produced in the world, second in consumption after Japan, which uses approximately 50%. Crabstick became popular in Korea when it was introduced in the late 1970s. It was predominantly used for *Gimbap* (Sea Vegetable Roll) as a meat ingredient. Korea's traditional surimi seafood (*Ahmook*) makes up the majority of surimi seafood sales at 260 billion South Korean won (KRW) (Hwang, 2011).

Due to fluctuation of surimi price during the last 20 years, however, the quality of some crabsticks has changed significantly. This was largely due to fluctuations in the price of surimi. Crabstick quality began to suffer as manufacturers decreased the concentration of surimi in the final product or used lower quality surimi in an attempt to cut costs. Consumers started to show

their displeasure with the low quality crabstick and the market became flat in the late 1990s and early 2000s. The market size of low quality crabstick is currently, about 90 billion KRW.

Hansung Enterprise (Seoul, Korea) introduced the first premium quality crabstick (Crami) in Korea in 2002 and it was joyously received by Korean markets. Other companies began to follow the Hansung Enterprise business plan of producing premium quality crabstick and together played a major role in revitalizing the Korean crabstick market. The premium crabstick market in Korea has grown by 60% over the last 4 years (Hwang, 2011).

However, to prevent a cycle of quality reduction–market crash and to promote a positive perception of crabstick, it would be beneficial to research new functional and healthy ingredients to add value to the crabstick product line. As in many Asian cultures, rice remains a major part of the daily diet of Koreans. Recently, it has received increased attention from Western consumers due to its healthy nutritional values as a great source of vitamins (thiamine, niacin, folic acid, pantothenic acid, and riboflavin) and minerals (magnesium, phosphorus, copper, manganese, selenium, calcium, and potassium) (Busch, 2009). Rice is low in fat, but contains essential omega-3 and omega-6 fatty acids. Rice protein consists of four fractions with different solvent solubility: albumin (water-soluble), globulin (salt-soluble), glutelin (alkali-soluble), and

prolamin (alcohol-soluble) (Ju et al., 2001). Rice is also gluten free. Therefore, rice can be used as alternative for those suffering from Celiac disease.

Since rice is primarily consumed in its whole grain form, other uses for rice, as a functional food ingredient, have not been evaluated (Oh et al., 2010). Currently there has been little research done in muscle foods using rice flour as a gel enhancing ingredient. Based on the textural properties of traditional foods made using rice (i.e., rice cake) in Korea and Japan, rice flour can be an effective functional ingredient in enhancing textural properties of crabstick. Unlike wheat flour, rice flour can provide white chalky color to crabstick, where white color is considered as premium quality. In addition, Korean rice, which is round and short grain, has lower amylose content (12–19%) compared to long grain rice containing 19–23% amylose (Parkinson, 2011). Korean short grain rice, therefore, contains higher (81–89%) amylopectin that can show better freeze-thaw stability or minimize the severity of retrogradation.

Our objectives were to evaluate rice flour as a functional ingredient for premium crabstick and further develop a commercially viable recipe for premium crabstick.

CHAPTER 2

LITERATURE REVIEW

2.1 Fish muscle protein

Muscles of animal are formed of bands, sheets, or parallel arrangement of elongated cells called myofibers or muscle fibers (MacIntosh et al., 2006; Strasburg et al., 2008). The fibers of muscle are comprised of a number of different proteins. According to Strasburg et al. (2008), muscle proteins are divided into two categories: biological function and solubility. Biological function refers to a protein's contribution to muscle structure and contraction, while the solubility category is based on differential solubilization of protein at different salt concentrations. Based on solubility, the proteins are then classified as either sarcoplasmic, myofibrillar, or stroma proteins (connective tissue).

Myofibrillar proteins constitute the largest portion of muscle protein. Myosin (thick filament) and actin (thin filament) are the primary components of myofibrillar protein. Unlike mammalian or avian muscle, fish muscle contains a higher percentage of myofibrillar proteins and a lower percentage of stroma proteins (Lanier et al., 2005) (Table 2.1). Postmortem myosin binds to actin and forms actomyosin. Thus, actomyosin is the predominant protein component in surimi. Myosin and actomyosin can be extracted from fish

muscle at high ionic strength (0.5 M KCl) (Lanier et al., 2005; Strasburg et al., 2008). Unlike myofibrillar proteins of mammal muscle, myofibrillar proteins of fish are partially soluble in water. Excessive washing during surimi production will result in a significantly reduced yield.

Sarcoplasmic proteins comprise several proteins, including enzymes and heme (iron-containing) proteins (Lanier et al., 2005; Strasburg et al., 2008). These proteins consist of water soluble components and can be dissolved at low ionic strength (>0.3 mM) (Scopes, 1970). In surimi manufacturing, these sarcoplasmic proteins are largely removed through washing and dewatering in order to concentrate myofibrillar proteins (Lanier et al., 2005). However, some sarcoplasmic proteins such as transglutaminase (TGase), which induces a cross-link between two amino acids, actually improve the gelling ability of surimi.

Stroma proteins, primarily collagen, are insoluble in water or saline solution. Collagen can be converted to gelatin by heating. However, this thermally irreversible gelatin interferes with the gelation of the myofibrillar proteins. In surimi gelation, however, the presence of collagen does not significantly affect the gelling ability of surimi because fish has only a small percentage of stroma protein (Lanier et al., 2005) and almost all connective tissues are removed through a refining process.

Table 2.1 Comparison of protein composition of fish and animal meats (Lanier et al., 2005)

Animal Species	(% of Total Proteins)		
	Sarcoplasmic	Myofibrillar	Stroma
Cod	21	76	3
Carp	23-25	70-72	5
Flatfish	18-24	73-79	3
Beef	16-28	39-68	16-28

2.2 Surimi and surimi gelation

The word “surimi” was first used in Japan. Surimi is made by myofibrillar proteins which are obtained from fish flesh that has been mechanically deboned, gutted, headed, and washed with water. The next step

the proteins are mixed with cryoprotectants and then frozen. Generally the surimi is packed in 10-kg polyethylene bags. The surimi bags are frozen to -25 ℃ for 2.5 hours (Okada, 1992; Park, 2005b). Two 10-kg blocks are placed in a cardboard box ready for sale.

It is necessary to distinguish "surimi" from "minced fish" because these terms are often used with a similar meaning. Minced fish becomes raw surimi after a washing, dewatering, and refining process, which remove fat, water-soluble components, and connective tissue. This raw surimi consists of myofibrillar proteins of fish. Before 1960, surimi processing was performed shore side where fish were caught and used within a few days as a refrigerated raw material. Quantity of production was limited due to limited shelf-life and the undesirable effects from freezing and frozen storage. During frozen storage, myofibillar proteins of surimi undergo denaturation and aggregation, which results in poor functionality (Shenouda, 1980). However, with the discovery of cryoprotectants in 1960, the surimi industry was able to freeze surimi. The cryoprotectant consists of low molecular weight molecules, such as sugars, polyols, many amino acids, carboxylic acids and polyphosphates (Zhou et al., 2006). Commonly used cryoprotectants in surimi are sucrose and sorbitol alone or in a mixture (1:1) at approximately 9% w/w to raw surimi for cold water species, and 6% sucrose for tropical species (Park and Lin, 2005; Zhou et al., 2006). Cryoprotectants are preferentially excluded from contact with the

surface of proteins in water. This exclusion causes re-orientation of the water molecules. As a result, the hydrophobic core proteins interact strongly leading to the stabilization of proteins. Raw surimi is mixed with cryoprotectants and quickly frozen to produce “frozen surimi”. The industry prefers to handle frozen surimi because it has a longer shelf life than raw surimi, it is easier to transport, store, and handle, therefore, the term “surimi” generally refers to frozen surimi (Okada, 1992).

Surimi is unique with its gel-forming ability and water-holding capacity. It is an intermediate material for surimi-based products such as kamaboko and crabsticks (Park and Lin, 2005). Frozen surimi blocks are partially thawed and cut into small pieces (2 to 3 cm). These small pieces are chopped and then mixed with salt. Salt breaks the ionic linkages among the myofibrillar proteins and enhances protein solubilization and dispersion because of their increased affinity for water. This reaction is necessary for the development of a gel network formed by denaturing the proteins and exposing the reactive sites from the interior of the protein (Lanier et al., 2005). After mixing the surimi with salt, additional ingredients such as water, starch, and egg white are mixed for the development and modification of textural characteristics (Park and Lin, 2005). During heating of salted surimi paste at 80-90 ℃, the myofibrillar proteins unfold (denature) and then interact to form intermolecular bonds, which are ionic linkages, hydrophobic interactions, and covalent bonds. These

bonds form a three-dimensional network resulting in gels (Lanier et al., 2005). In addition, hydrogen bonds are developed when gels are chilled after heating or during chilled storage. This is why surimi gels become firmer at colder temperatures

2.3 Rice flour

Rice is a major caloric source as a staple food for a large portion of the world's human population, especially in East and South Asia, the Middle East, Latin America, and the West Indies (Oh et al., 2010). It is believed that rice was first cultivated in central India, but was quickly put into large cultivation by the Chinese. This happened as long as 5,500 years ago with rice quickly spreading throughout Asia. It took rice over 4,500 years to reach Europe in the 12th Century, AD. Then rice was brought to the United States in 1694 (Chang, 2003). Today, more than 100 countries of the world cultivate rice. World production of rice has risen steadily from about 200 million tons in 1960 to over 678 million tons in 2009 (FAO, 2009). China produced 29% of the world's production in 2009, followed by India at 19%, Indonesia at 9%, and the United States produced about 1.5% of the world's total annual production during 2009. However, the United States which produced about 15% of the annual world exports of rice (FAO, 2009). The continuous growth of global rice production has further elevated and rice has become as important as wheat.

The major components of rice are protein and starch, with approximately 8 and 80%, respectively. Rice protein is composed of four different proteins, which are albumin, globulin, glutelin, and prolamin. Glutelin is a high molecular weight protein ($6 \times 10^4 \sim 6 \times 10^5$) and covers approximately 80% of the protein of the rice endosperm (Tecson et al., 1971). Rice starch is composed of amylose and amylopectin. The amylose content is classified as waxy (0-2%), very low (2-10%), low (10-20%), intermediate (20-25%), and high (25-33%) (Villareal et al., 1994). The amylose content correlates positively with water absorption and volume expansion, hardness, and whiteness of cooked rice, whereas it correlates negatively with stickiness and glossiness of cooked rice (Villareal et al., 1994).

Rice is classified into two cultigens (cultivated species): African rice (*Oryza glaberrima*) and Asian or common rice (*Oryza sativa*). Compared to *Oryza sativa, Oryza glaberrima*, which is typically grown in West Africa, shows poor milling quality. For example, grains are brittle and shatter easily. *Oryza sativa* contains two major subspecies, which are sticky, short and medium grained *japonica* or *sinica* variety, and non-sticky, long-grained *indica*. According to FAO classification, long grain milled rice is 6.0 to 7.0 mm in length while medium and short milled grains are 5.0 to 5.9 mm and <5.0 mm, respectively (Table 2.2) (Smith, 1995). In addition, long grain rice has 23 to 27% amylose, while medium and short grain rice have 15 to 20% and

18 to 20%, respectively. In other words, long grain rice has the most amylose and the least amylopectin, so it tends to be the fluffiest and least sticky when cooked. Medium and short grain rice have more amylopectin and less amylose, so these are sticky when cooked (Chang, 2003; Smith, 1995).

In surimi-based products where various starches are added, the expressible moisture and compressive force of surimi gels are increased during refrigerated storage when starches with higher amylose content are added. This was attributed to retrogradation of the gelatinized starch. During frozen storage, high amylose starches undergo severe retrogradation, resulting in gels with higher expressible moisture as well as increased brittleness (Park, 2005b). In other words, the short and medium grain rice is sticky and retrograde slowly, whereas long grain rice is firm, dry and retrograde rapidly (Bao and Bergman, 2004; Hizukuri et al., 1989).

Even though the setting response is decreased because of the dilution of myosin and decreased heavy chain cross-linking, the gel strength of surimi gel is increased when flour is used instead of starch (Park, 2005b). Rice flour is a form of flour made from finely milled rice. Rice flour includes a small amount of water (about 8%), fat (about 1%) and ash (about 1%) (Chun and Yoo, 2004). The rheological properties of rice flour can play an important role in processing control, estimating texture of foods, and heat processing design. The effect of temperature on rheological properties is also important because a

wide temperature range is encountered during processing and storage of foods containing starches (Chun and Yoo, 2004; Ju et al., 2001).

Table 2.2 Typical kernel lengths by class as recognized by the USDA and the Food and Agriculture Organization of the United Nations (Smith, 1995).

	Typical U.S. Rice			FAO Classification
Grain	Rough (mm)	Brown (mm)	Milled (mm)	Milled (mm)
Extra long grain	-	-	-	> 7.0
Long grain	8.9-9.6	7.0-7.5	6.7-7.0	6.0-7.0
Medium grain	7.9-8.2	5.9-6.1	5.5-5.8	5.0-5.9
Short grain	7.4-7.5	5.4-5.5	5.2-5.4	< 5.9

2.4 Starch

Starch is a major ingredient in surimi-based products because of its gelling properties, freeze-thaw stability and economic reasons (Lee, 1984). Rice, corn, wheat, and potato are the main sources of starches and differ

significantly in composition, thermal, rheological properties, respectively (Singh et al., 2003).

Starch, which is in the form of granules, is the major polysaccharide and primarily an energy storage organelle in plants (Singh et al., 2003). Starch granules are made up of amylose and amylopectin molecules arranged radially. Amylose is a linear chain of (1→4)-linked α-D-glucopyranosyl units. Many amylose molecules contain a few branches connected by α-D-(1→6) linkages at the branch points, constituting 0.3-0.5% of the linkage. In contrast, amylopectin is a very large, highly branched polymer with α-D-(1→6) branch point linkages that constitute 4-5% of the total linkages (Figure 2.1) (BeMiller and Huber, 2008; Park, 2005b; Singh et al., 2003).

Starch is semi-crystalline, which is a partially crystalline polymer, in nature with varying levels of crystallinity (Ahmed et al., 2008). Due to the semi-crystalline structure of starch, two phase transitions (glass transition and melting) occur during heating in the presence of excess water. While glass transition is concerned with the amorphous phase (mainly amylose), the melting of crystallites is exclusively associated with amylopectin (Liu et al., 2010). The melting of crystallites can be detected by X-ray diffraction patterns, and differential scanning calorimetry (DSC) (Liu et al., 2010).

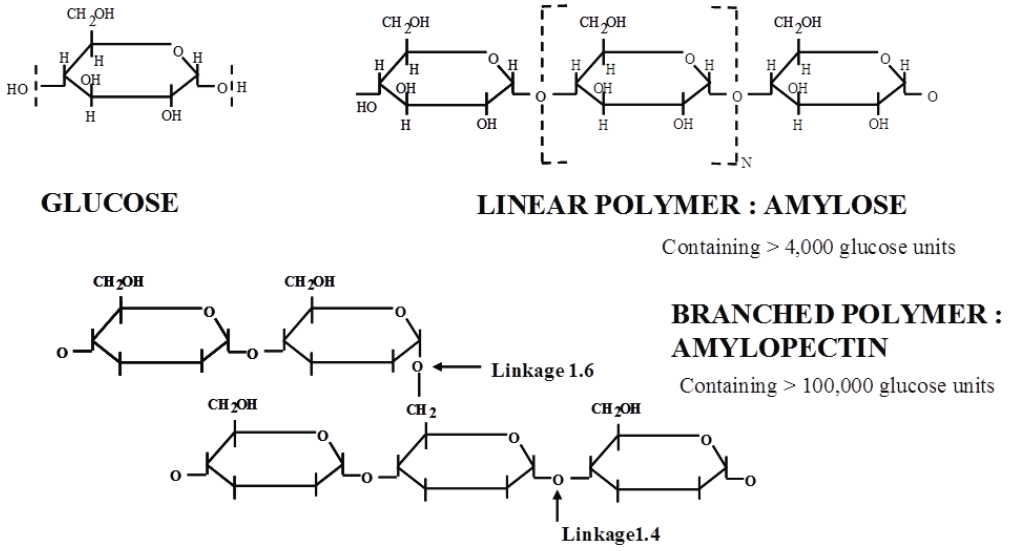


Figure 2.1 Two basic components of starch: amylose and amylopectin (Park, 2005b)

Starch granules absorb water resulting in swelling and lose their crystallinity during heating. This process is known as gelatinization (Parker and Ring, 2001) and the onset temperature depends on the type of starch, method of measurement, starch-water ratio, and pH (BeMiller and Huber, 2008). Continued heating of starch in excess water with stirring causes the granules to further swell, the amylose to leach more, and the granules to

disintegrate, forming a viscous material called paste (BeMiller and Huber, 2008). During cooling the gelatinized starch paste undergoes retrogradation or recrystallization. This is the reason for the increased firmness of cooked food after cooling or storage. The rate of retrogradation depends on the ratio of amylose and amylopectin in the starch because amylose undergoes retrogradation much more rapidly than amylopectin (BeMiller and Huber, 2008).

Starch is typically added to surimi seafood at a concentration between 4-12% (Park, 2005b). However, the effect of starch addition is more pronounced at low concentration (<3%) than at high concentration (6 to 9%) (Yang and Park, 1998). When over 9% starch is added, shear stress values are significantly reduced, indicating that starch inhibited the gelation of fish proteins by competing for the available water. In addition, as starch concentration increases, the concentration of fish protein is subsequently reduced. Therefore, addition of the optimum concentration of starch to the formulation is very important to obtain maximal gel strength. Wheat and potato are the most widely used starches in surimi seafood products. Potato starch increases gel strength more than any other native starches, but has poor freeze/thaw stability during frozen storage (Okada, 1986). Pre-gelatinized starch is a modified starch and swells in cold water. If used in a large quantity (>2%) in surimi seafood, it can inhibit the gel formation of fish proteins due to

its ability to bind water quickly, leaving limited water available for fish proteins (Park, 2005b). While modified starches are used to improve freeze/thaw stability, some modified starches reduce gel elasticity and firmness (Mauro, 1996) depending on the modification method. Therefore, the optimum level of starch must be determined with all possible considerations in product development such as texture, color, flavor, surimi content, water content, salt or sugar content, refrigerated or frozen storage, and expected cost (Park, 2005b).

2.5 Methodology to determine texture properties

Texture is the most important property in measuring the functional characteristics of surimi gel as surimi seafood products (Kim et al., 2005) and rheological measurements are very important to control the chemical interactions of food components, which produce particular food structure with desired textural attributes (Kim et al., 2005).

Rheology is defined as the science of material deformation and flow. This term ‘Rheology’ was invented by Professor Bingham of Lafayette College, Indiana, on the advice of his Professor of Classics and the definition was accepted in 1929 when the American Society of Rheology was founded (Barnes et al., 1989a). All materials have the following three categories of rheological properties: viscosity, elasticity, and viscoelasticity.

Viscosity is synonymous with internal friction and is a measure of resistance to flow. Viscosity (η) is denoted by shear stress (τ) and shear rate ($\dot{\gamma}$). Shear stress is the applied shear force (F) divided by area of the surface (A) and shear rate (or velocity gradient) is defined as shear strain (γ) divided by time (t) or the speed of the machine (v) divided by the initial height (H). Shear strain is the ratio of the deformation (ΔL) to the initial height or thickness (H) of the material (Kim et al., 2005).

$$\gamma = \Delta L/H,$$

$$\dot{\gamma} = \gamma/t = v/H,$$

$$\tau = F/A,$$

$$\eta = \tau/\dot{\gamma}$$

In 1678, Robert Hooke defined the properties of elasticity and proposed that "the power of any spring is in the same proportion with the tension thereof". In other words, elasticity is defined as the property of material that returns to its original shape after the stress has been removed (Barnes et al., 1989a).

The word "viscoelastic" means the simultaneous existence of viscous and elastic properties in a material. All real materials have these properties and

exhibit time dependent strain (Barnes et al., 1989b; Kim et al., 2005). The particular response of a sample in a given experiment depends on the time-scale of the experiment in relation to natural time of the material. Thus, if the experiment is relatively slow, the sample will appear to be viscous rather than elastic, whereas, if the experiment is relatively fast, it will appear to be elastic rather than viscous (Barnes et al., 1989b).

To measure the textural properties of surimi gel by instrumental methods, there are two main classes of rheological tests; fracture and non-fracture gel analysis (Park, 2008).

2.5.1 Fracture analysis

Even though the punch (penetration) test is an empirical test, many studies have been reported that correlate puncture methods with the sensory properties of surimi gel. The most popular fracture gel measurement technique for evaluating gel strength in the surimi industry as well as academia is the punch test (Kim et al., 2005). In this test, a punch probe of a specific diameter (5.0 mm) and a constant deformation rate (60 mm/min) is utilized. The result of the punch test is denoted as force (F) and the depth of penetration (cm) at the rupture point of the gel (Hamann and MacDonald, 1992). Force indicates the strength of gel measured as force values at rupture of the sample during probe penetration and depth of penetration indicates the displacement between

when the probe contacts the sample and when the sample ruptures. In surimi studies, this force and penetration is called breaking force and deformation, respectively (Kim et al., 2005; Park, 2008). However, considering the fundamental rheology, the use of "penetration distance" instead of "deformation" is more appropriate in describing the cohesiveness. Often these two values are multiplied together and used to calculate gel strength, which is also referred to as jelly strength in Japan. But, this equation is rheologically wrong because it can result in different surimi gels having the same gel strength although protein quality of the surimi gels is significantly different. Therefore, breaking force and penetration distance values must be used individually (Kim et al., 2005).

2.5.2 Non-fracture analysis

Oscillatory dynamic measurements are used to obtain the information of the non-fracture gel properties including phase transition. The experimental results from oscillatory shear are often presented as three main rheometer terms for simplicity: Storage modulus (G'; cos-δ), which is associated with the storage of energy; loss modulus (G"; sin-δ), which is associated with the loss of energy; and phase angle (δ), which is often expressed as tan-δ (G"/G') (Kim et al., 2005; Malkin and Isayev, 2006). A perfectly elastic material would exhibit $\delta = 0$ °, whereas a perfectly viscous material would exhibit $\delta = 90$ °.

In fish protein gelation, an oscillatory test can be done with holding frequency (often <1 Hz) and strain amplitude while varying temperatures through a gel-forming history (Hamann and MacDonald, 1992).

In the study of surimi, myosin of fish proteins were typically unfolded at two major stages during heating. α-helices in the tail portion of myosin unfold at around 35 ℃ and the unfolded hydrophobic regions interact with each other above 50 ℃ (Kim et al., 2005).

CHAPTER 3

MATERIALS AND METHODS

3.1 Materials

Two forms of rice flour were manufactured using medium grain rice by Daesun Flour Mills Inc. (Hampyeong, Korea). Rice flour (for cake) was ground using a roll mill and sieved using a 40-mesh screen. Rice flour (for noodle) was ground using a jet mill and sieved using a 180-mesh screen. Based on our preliminary evaluation of rice flour, rice flour for cake was eliminated due to its large particle size. Particles were too large to be uniformly mixed with surimi paste. Therefore fine rice flour (for noodle) was used for the entire study. Proximate analysis for rice flour was conducted in triplicate using AOAC method (AOAC, 1995). Moisture concentration was 12.64 ± 0.28%, protein concentration was 5.74 ± 0.03%, and pH was 6.29 ± 0.02.

Alaska pollock (*Theragra chalcogramma*) surimi (A grade) was obtained from American Seafoods (Seattle, WA, USA). The surimi was stored frozen at -18 ℃ until used in surimi crabstick paste preparation.

3.2 Paste preparation

Surimi crabstick pastes were made using four different rice flour concentrations (0, 1, 3, and 5%) while maintaining a final moisture concentration of approximately 76.5% as shown in Table 3.1. The crabstick paste of 0% rice flour concentration was used as the control sample. With our interest in finding a better utilization of rice flour, we attempted to replace a premixed starch with rice flour while keeping the content of surimi equal to that used for premium crabstick. Total addition of starch and rice flour was maintained at 8%. As rice flour replaced total starch concentration the amount of starch added was subsequently reduced. The premixed starch consisted of potato starch: wheat starch: modified starch at a ratio of 3:4:1.

Surimi chopping and mixing were done according to the method of Park (2005c). The frozen surimi blocks were thawed at room temperature (about 23 ℃) for 1 h and cut into small pieces (2 to 3 cm cubes). The pieces were chopped at low speed (1,800 rpm) for 1 min in a vertical vacuum cutter (UM5; Stephan Machinery Corp., Columbus, OH, USA). Salt (2%) was then added and chopped at low speed for another 1 min. Then one of four different rice flour concentrations (0, 1, 3, or 5%) combined with the appropriate amount of premixed starch to total 8% was sprinkled into the bowl, respectively. Additional ingredients, 5% liquid egg white, 2% sugar, 1% seasoning, and 40%

ice/water, were added and then chopped on low speed for 1 min. The crabstick paste was then chopped at high speed (3,600 rpm) with a vacuum of 40 - 60 kPa for 3 min.

Table 3.1 Formulation for crabstick paste prepared with premixed starch and rice flour. CON, control; LEW, liquid egg white; P starch, potato starch; W starch, wheat starch; M starch, modified starch

	CON	1%	3%	5%
Surimi	42	42	42	42
Water	40	40	40	40
Salt	2	2	2	2
LEW	5	5	5	5
P starch	3	2.6	1.9	1.1
W starch	4	3.5	2.5	1.5
M starch	1	0.9	0.6	0.4
Sugar	2	2	2	2
Seasonings	1	1	1	1
Rice Flour	0	1	3	5
Total (%)	100	100	100	100
Moisture (%)	76.4	76.4	76.6	76.8

3.3 Gel Preparation and storage

The paste was then packed into a polyethylene vacuum bag and subjected to a vacuum machine (Reiser VM-4142; Roescher Werke GMBH, Osnabrueck, Germany) to remove air pockets that might have been introduced while removing paste from the bowl and placing it into a bag.

3.3.1 Ohmic heating

The paste was stuffed into nylon tubes (inner diameter = 30 mm, length = 220 mm) using a sausage stuffer (Model 14208; The Sausage Maker, Buffalo, NY, USA) and subjected to ohmic heating. This fast ohmic cooking was used to mimic the initial cooking of commercial crabstick, which is done fast in a thin sheet. Stuffed tubes were placed between two stainless steel electrodes (i.e. electrodes were inserted into each end of the tube) and tightened by the center tube aluminum clamp. The needle thermocouple (needle thermocouple 1.6 mm x 260 mm; Ecklund-Harrison Technologies, Inc., Fort Meyers, FL, USA) was then inserted into the nylon tube and used to control cooking time along with temperature controller (Cni 3254-C24; Omega Engineering, Inc., Stamford, CT, USA). One of two electrodes was connected to an air cylinder (SR-242-Q; Bimba Manufacturing Company, Monee, IL, USA) providing 3.5 bar and the valve of the cylinder was then opened to provide a solid contact between the

electrodes and the sample paste. The sample was then heated using 250V. When the sample temperature reached 90 ℃, the sample was held for 1 min. Total cooking time was approximately 90 seconds.

3.3.2 Water bath heating

Ohmically cooked surimi gels, which were rolled in a plastic bag and sealed, were heated at 90 ℃ for 40 min. This water bath cooking was used to simulate the pasteurization of commercial crabstick products. Pasteurized samples were chilled in ice water for 15 min. The chilled gels were subjected to either refrigerated (4 ℃) or frozen (-18 ℃) storage.

3.3.3 Gel storages

The quality of refrigerated samples was measured for texture, color, and water retention ability during refrigerated storage (4 ℃) at Day 1, 7, 14, and 21. As for the measurement of frozen stability (-18 ℃), frozen samples were subjected to a number of freeze-thaw (F/T) cycles, which are commonly used to mimic long term frozen storage. Frozen samples were treated with freezing and thawing for 0, 3, 6, and 9 cycles. All samples were partially vacuum sealed to maintain the shape of samples and to minimize sample spoilage. One F/T cycle was defined as freezing samples at least 16 h in the freezer (-18 ℃) and

then thawing at refrigerated condition (4 ℃) for 7 h. The 0 cycle samples were measured before freezing.

3.4 Micro differential scanning calorimetry (DSC)

The thermal properties of rice flour were measured by DSC using six different rice flour paste (a mixture of flour to water at ratios of 5:95, 10:90, 15:85, 25:75, 40:60, and 55:45, respectively) and crabstick paste with four different rice flour concentrations (0, 1, 3, and 5%) as prepared above. DSC was performed using a micro DSC III (Setaram, Inc., Lyon, France). The instrument was calibrated for temperature accuracy using naphthalene. Sample, accurately weighed at approximately 500 ± 5 mg, was sealed in a hastelloy sample vessel. Another calibration with sample was performed along with an empty reference vessel to determine the amount of distilled and deionized (DDI) water required as a reference. Samples were scanned with a reference vessel containing DDI water at a heating rate of 1.0 ℃/min over a temperature range of 20-90 ℃. All of the samples were conducted at least in duplicate or until two uniform thermograms were obtained consecutively.

3.5 Oscillatory dynamic measurement

The six different rice flour slurries and four different crabstick pastes, both prepared above, were subjected to a dynamic rheometer (CVO-100; Malvern, Inc., Worcestershire, UK) to measure gel network development as a function of temperature. The samples were loaded between a cone and plate geometry (40 mm diameter, 4 ° angle) with a gap of 150 μm. A solvent trap with a moistened sponge inside was used to prevent moisture loss. The elastic modulus (G') of the crabstick paste was measured with 0.1 Hz frequency and shear stress of 40 Pa after determining the linear viscoelastic region at a heating rate of 1.0 ℃/min during the temperature sweep from 20 to 90 ℃. All of the samples were subjected to temperature sweep at least twice or until two uniform rheograms were obtained consecutively.

3.6 Fracture gel texture analysis

Gels cooled in ice/water and stored overnight in refrigerated storage (4 ℃) were then set at room temperature for 2h before subjecting to fracture textural properties (breaking force, g and penetration distance, mm) using a TA texture analyzer (TA-XT plus; Texture Technologies Corp., New York, NY, USA) (Kim et al., 2005). Gels were cut into 30 mm long cylinders. A 5 mm

diameter spherical probe was used with a crosshead speed set at 1 mm/s. For each sample, 10 measurements were made.

3.7 Water retention ability (WRA)

Water retention ability (WRA) was measured according to the method developed by Kocher and Foegeding (1993). A microcentrifuge filtration unit consisting of a 1.5 mL-filtrate receiver tube and a sample reservoir with an encapsulated membrane (0.45 μm) was used. Surimi gels, after cutting into fine particles (0.3 ± 0.05 cm), were weighed (0.4 ± 0.05 g), placed in the sample reservoir and then the filtration unit was spun in a microcentrifuge (5415C; Eppendorf, Hamburg, Germany) at 5,500 rpm for 10 min. All of the samples were conducted in triplicate. WRA was determined as:

WRA = [total water (g) in surimi gel - water (g) released]/ total surimi gel (g),

where, total water (g) = % moisture of surimi gel x surimi gel weight (g) and water (g) released = [(microcentrifuge tube weight (g) + water(g)]– microcentrifuge tube weight (g).

3.8 Color properties

Color properties (L*, a*, b*) of gels were measured in triplicate using a Minolta colorimeter (CR-310; Minolta Camera Co. Ltd., Osaka, Japan) before gels were used for fracture textural analysis. The instrument was standardized using a Minolta calibration plate and a Hunter Lab standard hitching tile according to the method of Park (1994). The whiteness was calculated using the equation L*-3b*.

3.9 Crabstick paste preparation and evaluation

Based on texture and color evaluation of rice flour at various concentrations, 1% rice flour was determined as the best. Therefore, crabstick gels were prepared manually using a sheet mold and roller using two formulae (control and 1% rice flour). Crabstick paste (about 40 g) was placed inside the molding sheet frame (7.5 cm wide, 25.5 cm long, 0.15 cm thick) and the paste was evenly filled using a roller according to Poowakanjana and Park (2009). Crabstick paste sheet (1.5 mm thick) on aluminum foil was cooked in steam (92 ℃) for 1 min. Then this partially cooked sheet was manually rolled into a crabstick shape. Color paste, which was prepared mixing 2% carmine and 98% surimi paste prepared above, was applied on the surface before cooking in steam (92 ℃) for 1 min. Then the crabstick was wrapped using a plastic film

and cooked in steam (92 ℃) for 40 min to mimic the commercial pasteurization step, which also completed the gelation of egg white, starches, and rice flour. Crabsticks were then cooled in ice water for 30 min and equilibrated to room temperature (about 23 ℃) before texture measurement.

Gel texture of crabstick was measured using a wire cutter attached to a TA texture analyzer (TA-XT plus; Texture Technologies Corp., New York, NY, USA). Gel breaking force (g) and penetration distance (mm) were reported. For each sample, 10 measurements were made.

3.10 Statistical analysis

The results were presented as the average and standard deviation of each experiment conducted in triplicate and evaluated using SPSS (version 13) software package (SPSS Inc., Chicago, IL, USA). ANOVA with Tukey's test was used to determine statistical significance ($P<0.05$).

CHAPTER 4

RESULTS AND DISCUSSION

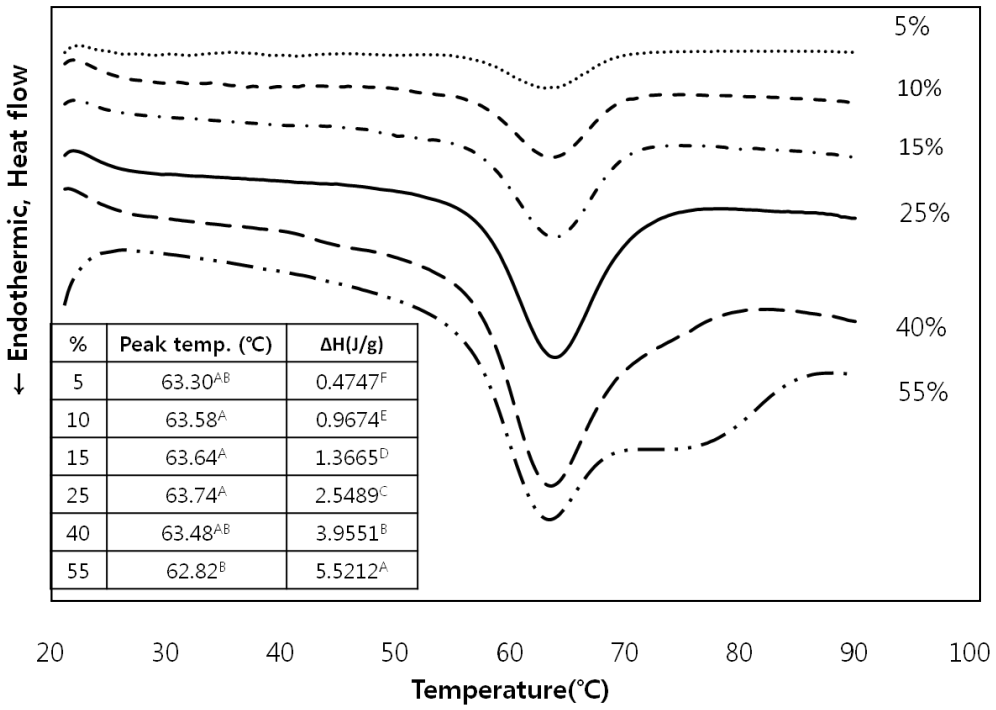
4.1 Effect of rice flour concentrations on micro DSC

Differential scanning calorimetric properties of rice flour slurry were evaluated using a concentration from 5 to 55% (Figure 4.1). At a concentration of 5 to 40%, similar DSC patterns were obtained with an endothermic peak at around 63.5 ± 0.2 ℃ possibly due to the gelatinization of starch and gelation of albumin (water-soluble) (Juliano, 1994). Enthalpy values increased as flour concentration increased. This indicated more thermal energy was required to swell rice starch in the presence of excessive water before aggregation as flour concentration increased (Ahmed et al., 2008; Marruf et al., 2001).

When rice flour was mixed with surimi and other ingredients, two peaks were developed at around 36.5 ℃ and around 71-72 ℃ and the peak temperature was shifted to higher temperatures as the rice flour concentration increased from 0 to 5%, indicating gelation was slightly delayed (Figure 4.2). According to several reports, Alaska pollock myosin and actin started unfolding and aggregated at around 30-35 ℃ and 65-75 ℃, respectively (Beas et al., 2006; Fukushima et al., 2003; Kim et al., 2005).

At 55% flour concentration, two endothermic peaks were obtained at 62.8 ℃ and 78.0 ℃. According to Tsutsui et al. (2005), the gelatinization temperature of medium grain rice starch is 62 ℃ when measured at 10% starch suspension. Therefore, the first peak (62.8 ℃) is possibly related to the melted starch crystallites with water and denaturation of rice protein (Gorinstein et al., 1996; Ju, 2001) and the second peak linked to the melting of remaining starch crystallites (BeMiller and Huber, 2008; Marruf et al., 2001).

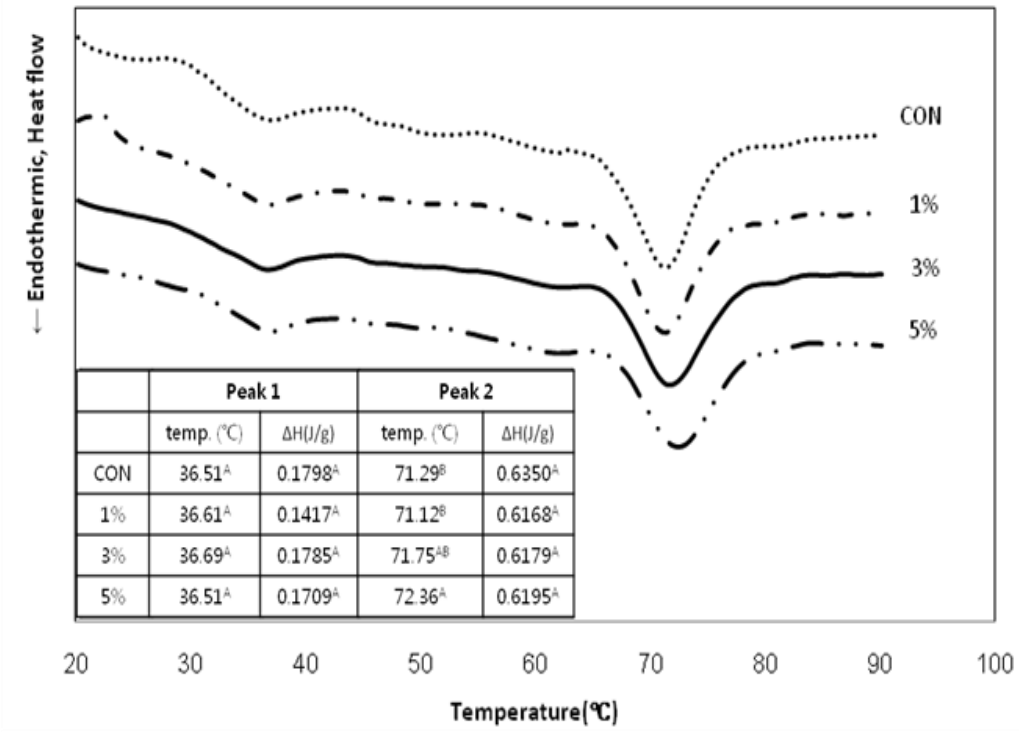
Park (2005b) reported that starch gelatinization is delayed by the presence of myofibrillar proteins, salt, and sugar in the surimi-starch system. Potato starch, which has a gelatinization temperature around 60 ℃ in water, is gelatinized at 70 ℃ with surimi. On the other hand, the gelatinization temperature of rice flour is 63 ℃ in water (Figure 4.1), which further suggests that this rice flour possibly gelatinized at a temperature higher than 70 ℃. Therefore, Peak 2 was delayed as rice flour concentration increased.



%	Peak temp. (℃)	ΔH(J/g)
5	63.30^{AB}	0.4747^{F}
10	63.58^{A}	0.9674^{E}
15	63.64^{A}	1.3665^{D}
25	63.74^{A}	2.5489^{C}
40	63.48^{AB}	3.9551^{B}
55	62.82^{B}	5.5212^{A}

Figure 4.1 Differential scanning calorimetry (DSC) for rice flour paste.

Different superscript letters in the same column of the table denote significant differences (P<0.05).



	Peak 1		Peak 2	
	temp. (℃)	ΔH(J/g)	temp. (℃)	ΔH(J/g)
CON	36.51^{A}	0.1798^{A}	71.29^{B}	0.6350^{A}
1%	36.61^{A}	0.1417^{A}	71.12^{B}	0.6168^{A}
3%	36.69^{A}	0.1785^{A}	71.75^{AB}	0.6179^{A}
5%	36.51^{A}	0.1709^{A}	72.36^{A}	0.6195^{A}

Figure 4.2 Differential scanning calorimetry of surimi gels as affected by rice flour. Different superscript letters in the same column of the table denote significant differences (P<0.05).

4.2 Oscillatory dynamic measurement

Dynamic rheology is often used to measure gel formation and phase transition during temperature sweep from 20 to 90 ℃. While fracture gel analysis measures the texture upon rupture of gels, dynamic rheology measures non-fracture gel properties through monitoring the development of heat-induced gelation. Dynamic rheogram for storage modulus (G'), exhibiting the nature of elastic properties for four treatments was generally in the same trend except between 45–85 ℃, where covalent bonds and hydrophobic interactions are completed for proteins (Lanier et al., 2005; Li-Chan et al., 1985) and gelatinization of starch was completed (Ahmed et al., 2008; Lii et al., 1995). The significant increase in G' of four treatments was due to the formation of a 3-dimensional network from amylose which leached out and was reinforced by strong interaction among the swollen starch particles (Eliasson, 1986; Hsu et al., 2000). Once heating was completed, all showed equal G' and G", indicating four treatments have the same elastic and viscous properties, respectively (Figure 4.3). An increased G' peak at near 36 ℃ (Figure 4.5) was probably due to the formation of a semi-gel like structure of myosin tail. As heating continued, this semi-gel like structure ruptured and released some fluidity and resulted in a decreased G' (Yoon et al., 2004).

Transition of phase angles for the four treatments was somewhat similar except between 20–32 ℃ and 45–57 ℃ (Figure 4.4). A phase angle of 90 ° indicates perfect viscous properties, while 0° denotes perfect elastic properties. The transition of the phase angle from 40 to 10 ° during temperature sweep explains that the sample paste was quite elastic (less viscous) at the beginning and became more elastic once heating was completed.

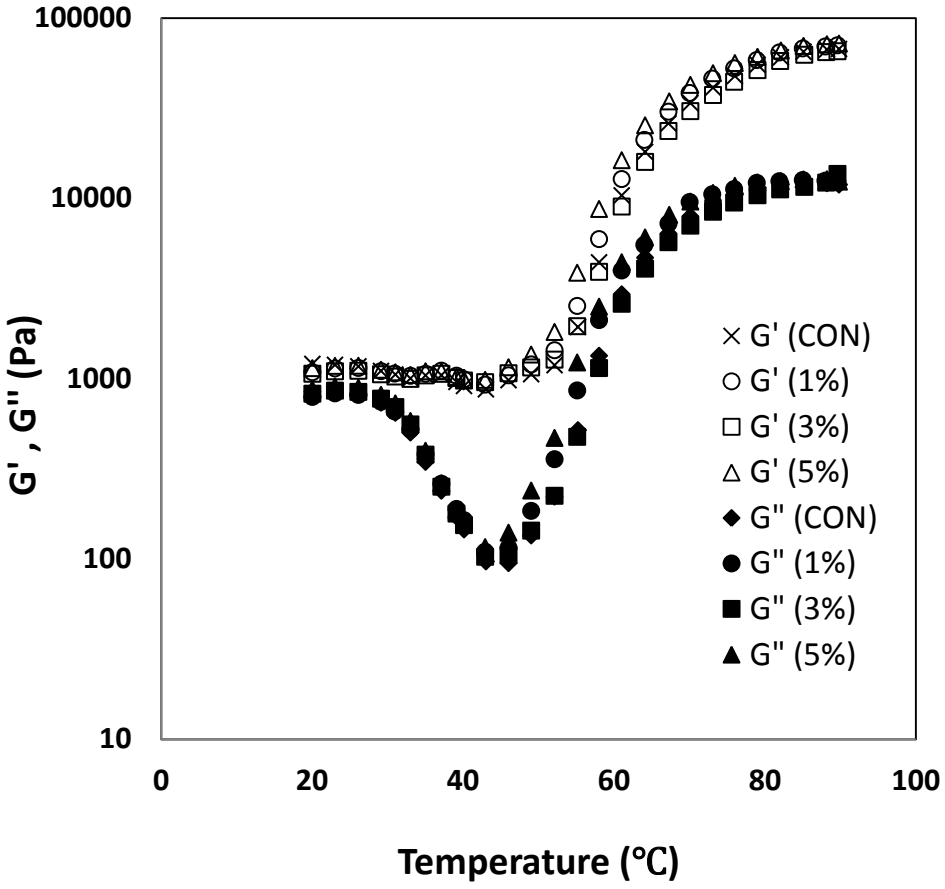


Figure 4.3 Effect of rice flour on the thermal gelation of surimi paste. Storage modulus (G') and loss modulus (G") of surimi paste during temperature sweep.

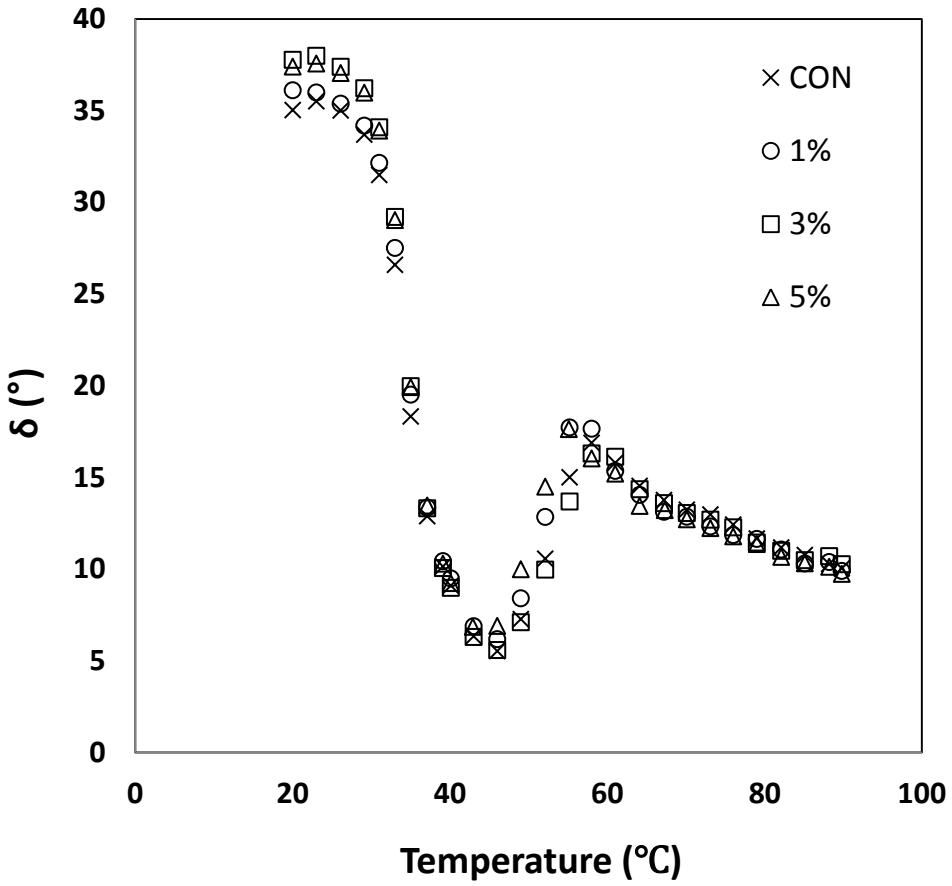


Figure 4.4 Effect of rice flour on the thermal gelation of surimi paste. Phase angle (δ) of surimi paste during temperature sweep.

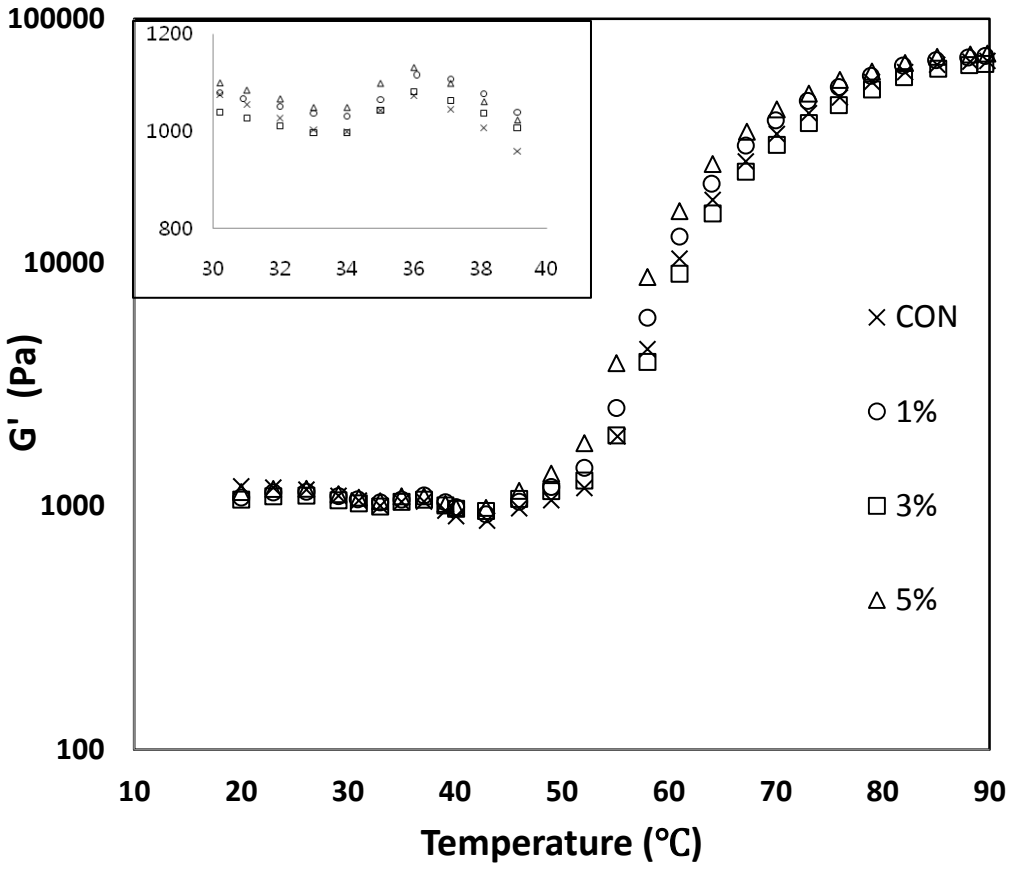


Figure 4.5 Effect of rice flour on the thermal gelation of surimi paste. Storage modulus (G') of surimi paste during temperature sweep. The insert figure shows the local maxima observed at approximately 36 °C.

4.3 Fracture gel texture

Fracture properties of gel texture measured using a penetration probe is highly correlated with sensory evaluation (Smewing, 1999). Breaking force (g) indicates the gel strength, while penetration distance (mm) denotes the gel cohesiveness. Gel strength decreased significantly ($P<0.05$) as rice flour concentration increased (Figure 4.6) indicating the reinforcing effect of rice flour on the strength of surimi gel was not as strong as that of starches. During refrigerated storage of 21 days, gel strength gradually increased (Figure 4.6), while cohesiveness remained unchanged ($P>0.05$) (Figure 4.7).

Among several major forces that stabilize protein-starch gels, hydrogen bonds become stronger as temperature decreases (Lanier et al., 2005). According to Lanier et al. (2005) and Howe et al. (1994), gel hardness is strongly influenced by hydrogen bonds during refrigerated temperatures and gel cohesiveness, which increases at elevated temperatures (up to 60 ^{o}C) by hydrophobic interactions, is not affected at refrigerated storage. In addition, 1% rice flour showed almost identical fracture gel properties compared to the control after 21 days of refrigerated storage (Figure 4.6; Figure 4.7).

For the frozen storage effect, which was evaluated using freeze/thaw (F/T) cycles, gel strength significantly increased ($P<0.05$) (Figure 4.8), while gel cohesiveness significantly decreased ($P<0.05$) as F/T cycles were extended

(Figure 4.9). These changes were minimized as the concentration of rice flour increased. A high concentration of amylopectin in Korean rice flour possibly minimized the changes of texture during frozen storages. On the other hand, the surimi-starch gels become brittle and rigid because high amylose starches undergo severe retrogradation during storage (Park, 2005b). A treatment with 1% rice flour demonstrated similar gel strength and cohesiveness compared to the control after 9 F/T cycles.

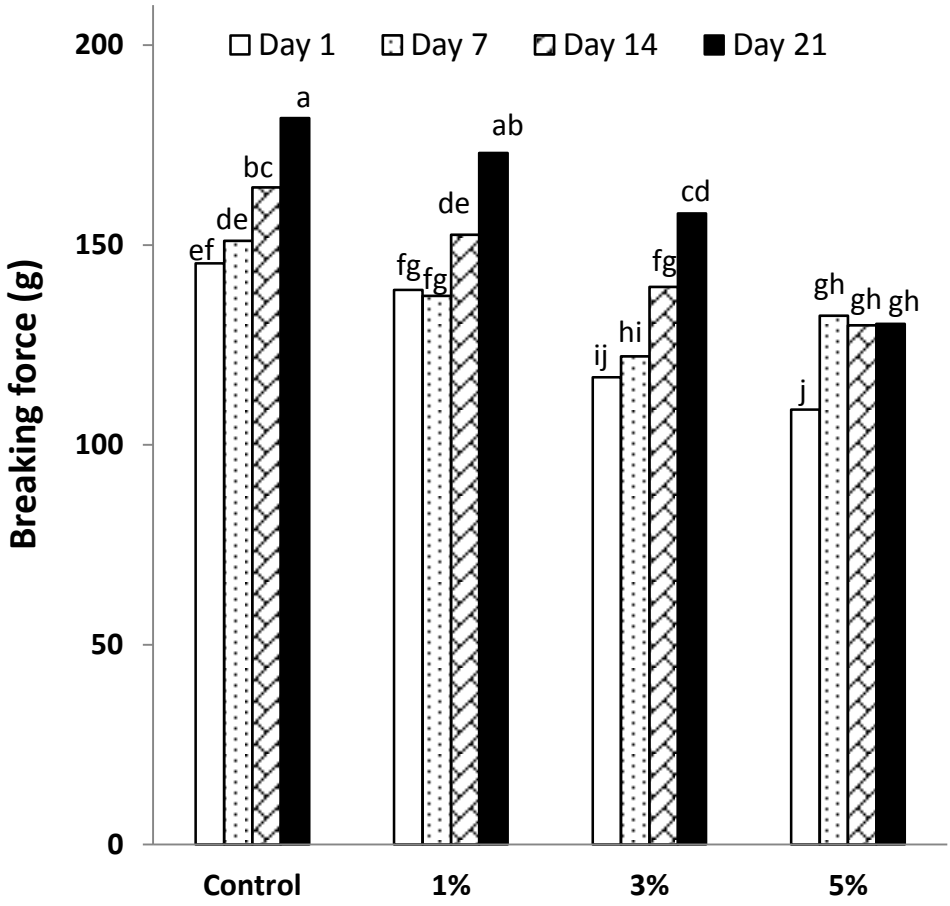


Figure 4.6 Breaking force of various surimi gels during refrigerated storage. Different letters denote significant differences (P<0.05).

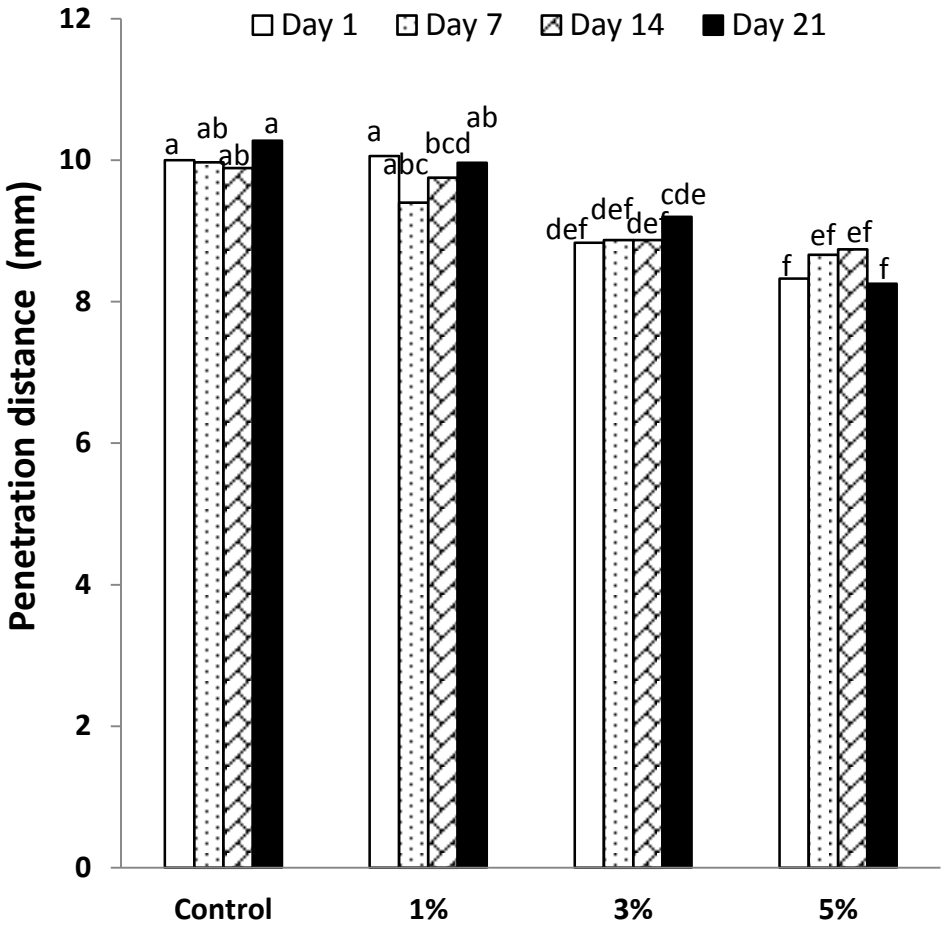


Figure 4.7 Penetration distance of various surimi gels during refrigerated storage. Different letters denote significant differences (P<0.05).

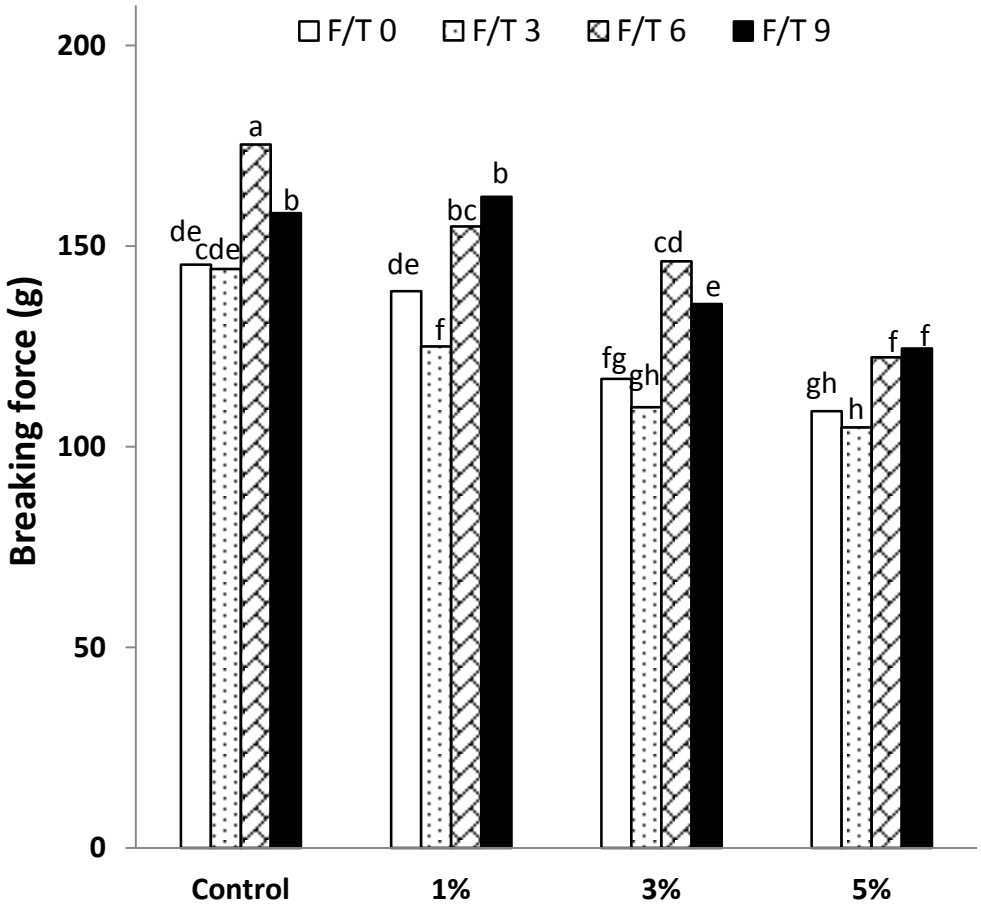


Figure 4.8 Breaking force of various surimi gels during freeze/thaw cycles. Different letters denote significant differences (P<0.05).

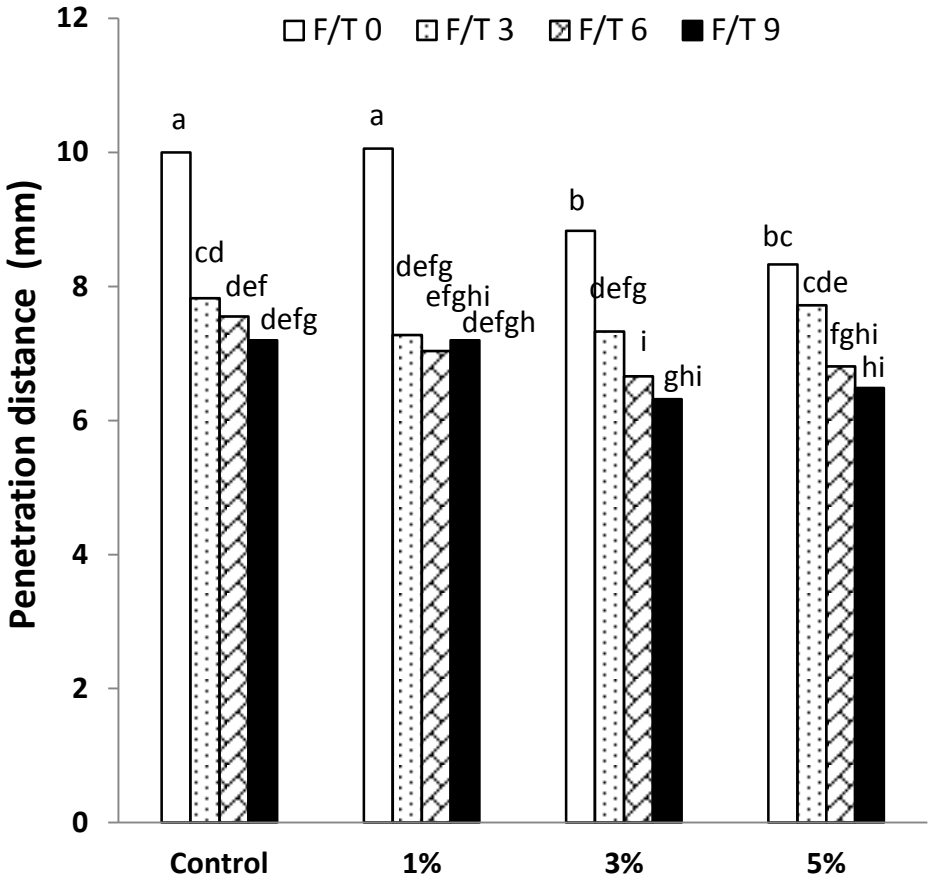


Figure 4.9 Penetration distance of various surimi gels during freeze/thaw cycles. Different letters denote significant differences (P<0.05).

4.4 Water retention ability (WRA)

Water molecules behave as either free water or bound water in the gel matrix. As external force is given to the gel matrix, free water is released while bound water is retained in the gel matrix. WRA measures how much water is retained as bound water in the gel matrix. WRA during refrigerated storage for 21 days did not show a visual difference while a statistical difference was noted (Figure 4.10). However, there was an interesting trend that overall WRA increased as the concentration of rice flour increased possibly due to the high amylopectin content in Korean rice flour. During frozen storage, it was noted that WRA gradually decreased as F/T cycles were extended (Figure 4.11). However, the reduction was minimized as rice flour concentration increased. With regard to WRA, 5% rice flour gave the best performance. Starch retrogradation and ice recrystallization affect the deterioration of the frozen paste during storage (Ferrero et al., 1993) and high-amylose starches easily undergo retrogradation during frozen storage. In other words, high-amylopectin starch would release the least amount of free water during frozen storage (Park and Lanier, 2005). Therefore, due to its higher amylopectin content compared to starches used, the amount of free water released from the gel samples was effectively reduced.

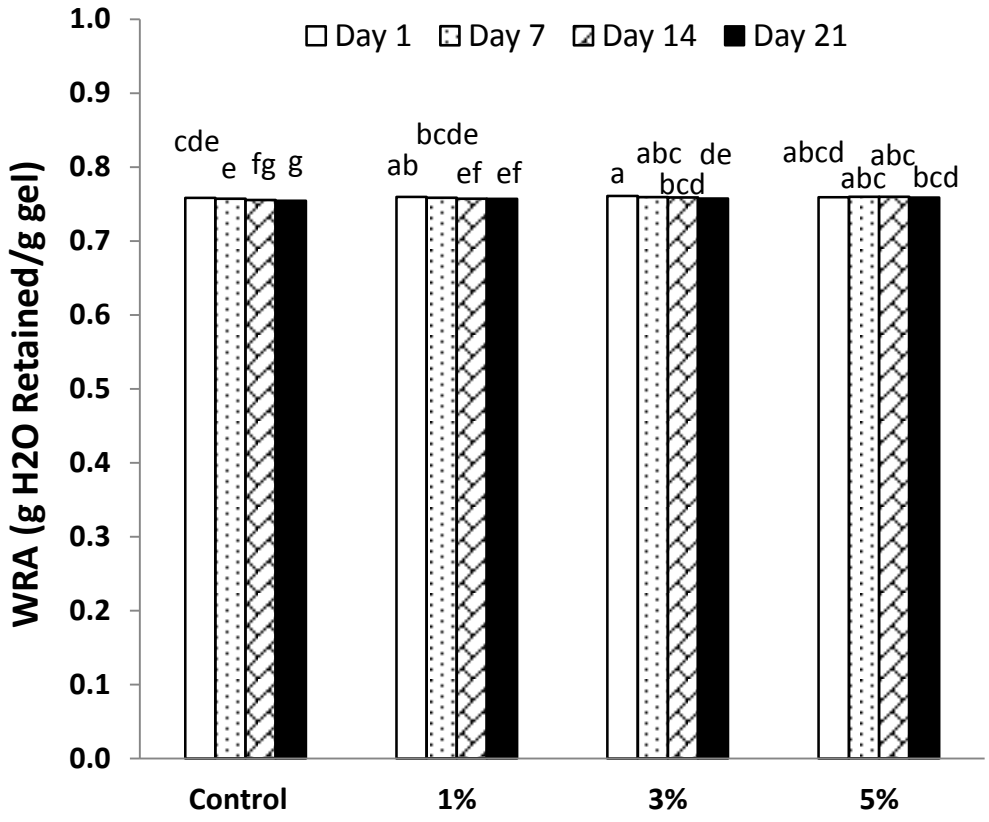


Figure 4.10 Water retention ability (WRA) of various gels during refrigerated storage. Different letters denote significant differences (P<0.05).

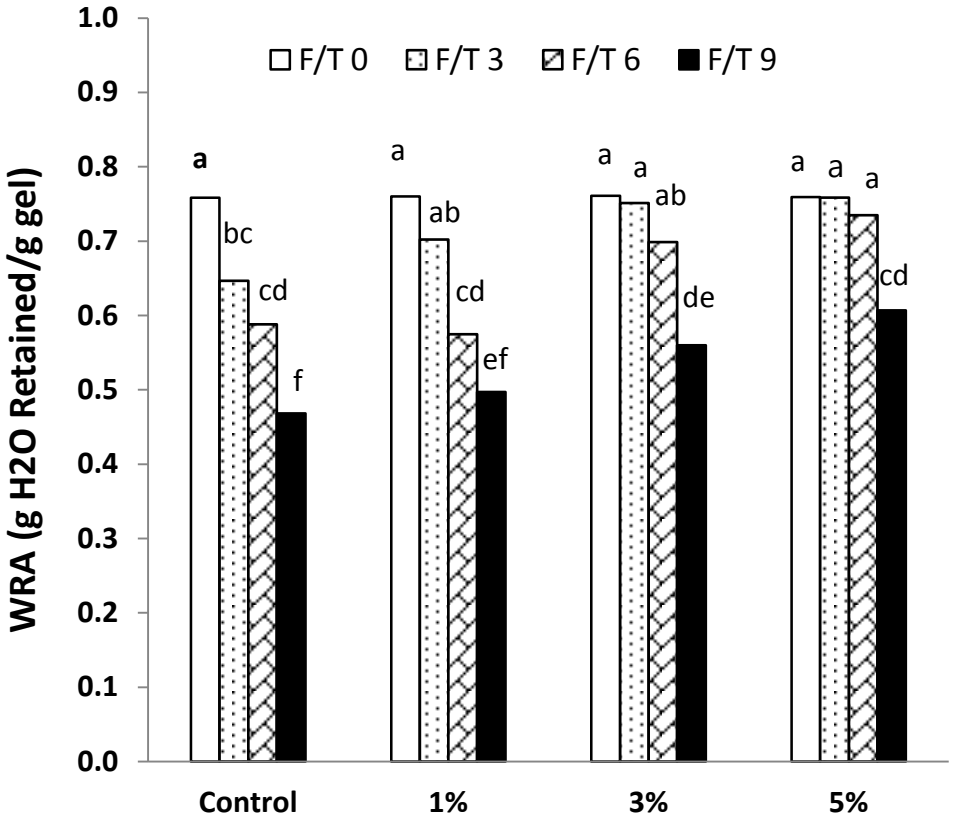


Figure 4.11 Water retention ability (WRA) of various gels during freeze/thaw cycles. Different letters denote significant differences (P<0.05).

4.5 Gel color

Appearance and color most affect consumer's decision making for purchase choice. Color of surimi gels is affected by color quality of surimi and the properties and concentration of starch used. Typically, the higher the L* value or the lighter a surimi gel is, the higher its color quality. When starch granules are fully swollen, translucent gels with lower L* values are obtained. Starch gels with larger granules (e.g., potato) exhibit more translucence while those with smaller granules (e.g., wheat or rice) are more opaque. For b* values, when starch granules are not fully swollen, the gels are more yellow in hue (higher b*) compared to when starch granules are fully swollen and are slightly blue in hue (negative b*) (Park, 2005b). Whiteness value increased as refrigerated storage extended (Figure 4.12). As rice flour concentration increased from 0 to 5%, the proportion of added starch content decreased and the proportion of rice protein increased. This proportional change significantly affected gel color from translucence to opaqueness during refrigerated storage.

However, the whiteness of frozen samples decreased as F/T cycles were extended (Figure 4.13). Ice crystal formation during frozen storage and the possible growth of ice crystals with repeated F/T cycles probably derived water molecules out of gel matrices, resulting in reduced L* values. The highest whiteness after 9 F/T cycles was found with a gel made with 5% rice flour. For both cases, the change was minimized as the concentration of rice

flour increased. Among three samples containing rice flour, 1% treatment showed the closest whiteness compared to the control.

4.6 Crabstick preparation and texture evaluation

Based on the results above, a formula containing 1% rice flour demonstrated comparable texture and color compared to the control. Two different crabstick samples (control and 1% rice flour) were prepared for textural analysis (Table 3.1). Two crabstick samples demonstrated 259.35 g and 277.64 g for breaking force as well as 8.06 mm and 8.90 mm for penetration distance, respectively. They were not statistically different ($P>0.05$). This result indicates 1% rice flour can effectively replace various starches without affecting textural quality (Kim and Lee, 1987; Park, 2005b).

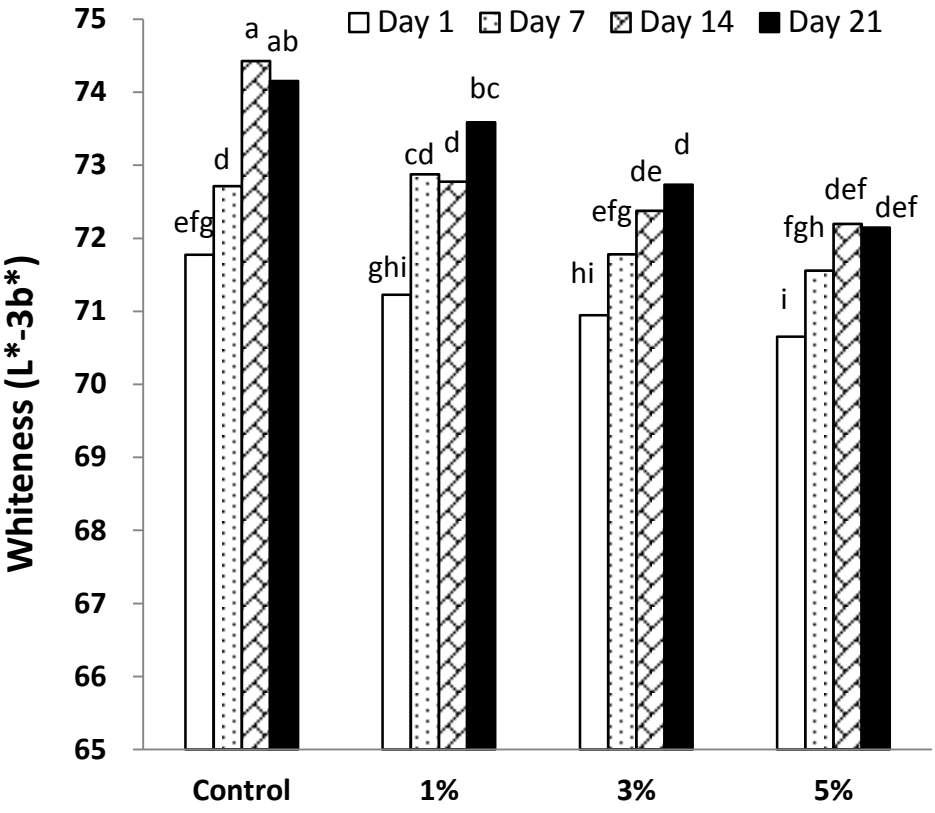


Figure 4.12 Whiteness values of various crabstick gels during refrigerated storage. Different letters denote significant differences (P<0.05).

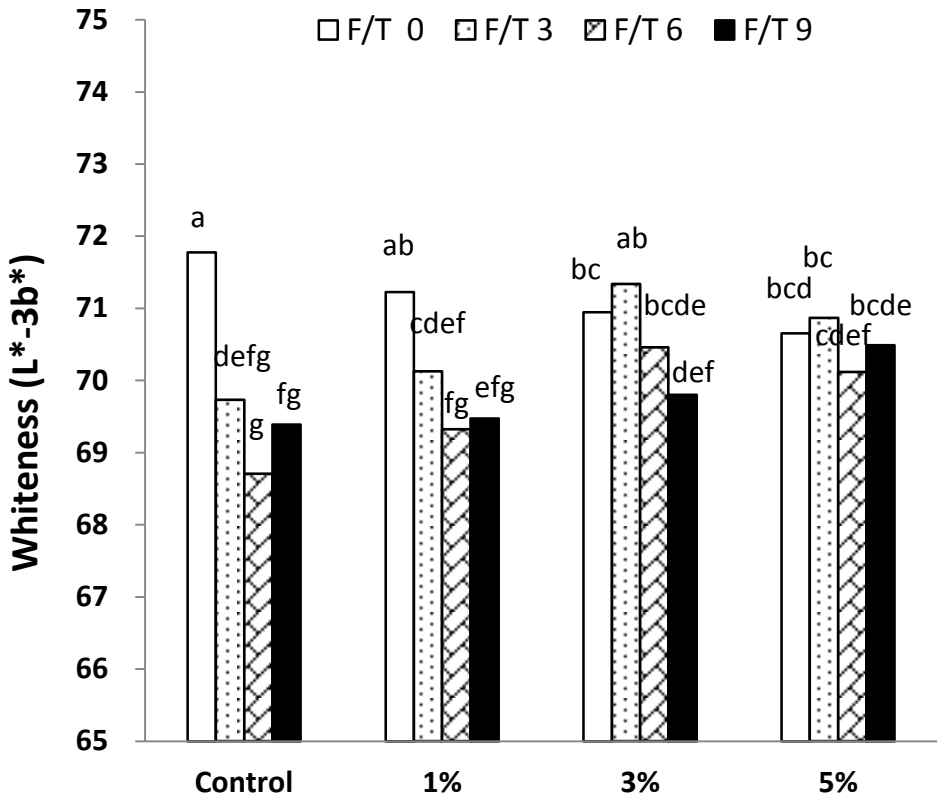


Figure 4.13 Whiteness values of various crabstick gels during freeze/thaw cycles. Different letters denote significant differences (P<0.05).

CHAPTER 5

CONCLUSION

Rice flour was evaluated as a functional ingredient for premium crabstick. With our interest in premium crabstick, formulation development was not intended to reduce surimi concentration but to replace various starches with rice flour. Total concentration of starches and rice flour remained equal at 8% in four different test formulae. Dynamic rheology and micro differential scanning calorimetry were not able to differentiate the viscoelastic properties and thermal properties as affected by rice flour addition (Ahmed et al., 2008). However, fracture gel analysis demonstrated textural changes of gels containing various rice flour concentrations during refrigerated and frozen storage, respectively. It was probably due to different thermal treatments of the samples: dynamic rheology was done using continuous temperature sweep from 20-90 ℃ while fracture gels were prepared using a 2-step heating process (fast ohmic heating followed by slow steam heating). Considering the texture, whiteness, and water retention ability of surimi gels as well as prepared crabstick samples, 1% rice flour can be functionally used for the production of premium crabstick. Rice flour at 3% and 5% addition showed slightly reduced gel texture. However, if the addition of 3-5% rice flour is desired, it must be verified with sensory panel to determine the level of acceptance by consumers.

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