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Master's Thesis of Science in Agricultural Biotechnology

**Effect of curdlan and resting process
on the physicochemical properties of
whole tofu noodle**

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전두부 면의 이화학적 특성에 미치는 영향

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Effect of curdlan and resting process on the physicochemical properties of whole tofu noodle

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이 논문을 석사학위 논문으로 제출함

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ABSTRACT

In the first chapter of this study, dried noodle containing high-content whole tofu was developed. Whole tofu (60 wt%) which has more protein and dietary fiber than tofu, potato starch (34 wt%), and curdlan (2-4 w/w%) were used as the main materials. The elasticity (G') and viscosity (G'') of the dough increased as the curdlan was added. The high temperature (45 °C) resting process was applied to control the moisture of the dough. After 45 minutes of the resting process at 45°C, the dough's elasticity (G') increased more than 50% compared to the not rested sample. Therefore, curdlan 4% and 45 min of the resting process at 45 °C were set to make the optimized dough and dried noodle. The noodle's texture properties including hardness and gumminess increased more than 10%. In addition, cooking time and cooking loss decreased presenting improved cooking quality. Consequently, as a result of the addition of curdlan and the resting process, the dried noodle with improved quality and convenience was developed.

In the second chapter, a new strategy that maximizes the effect of curdlan gelation in making the whole tofu dough was proposed. In order to set the optimal temperature for gelation and gelatinization, the temperature of the resting process was set to 55 °C. It means that curdlan gelation occurs and wheat starch does not gelatinize (60 °C). As a result of the resting process at

55 °C, the value of water absorption doubled due to the gelation of curdlan. Besides, both elasticity (G') and viscosity (G'') increased by approximately 50%. The hardness and gumminess of the 55 °C rested sample increased by 100% and 15%, respectively than other samples. Even though the gelation of curdlan slightly disrupted gluten structure, however, curdlan gel strengthened the internal structure of the dough. As a result, the use of wheat starch, curdlan, and the 55 °C resting process contributed significantly to improving the whole tofu dough's quality.

Through these studies, the dried whole tofu noodle that highly nutritious, convenient, and enhanced quality was developed. Furthermore, a method to improve the quality of dough through curdlan gelation was proposed. These results are expected to be applied to manufacture various dough-based products such as cookies or bread.

Keywords: whole tofu, curdlan, resting process, dried noodle, dough, rheological properties, texture properties

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I. CHAPTER 1

Development of dried whole tofu noodle: The effect of curdlan and resting process on noodle quality

전두부를 첨가한 건면의 개발:
커들란과 레스팅 공정이 면의 품질에
미치는 영향

I.1. INTRODUCTION

The high awareness of healthy diets worldwide is increasing the demand for foods that contain ingredients with potential health benefits. Generally, noodles are eaten in the form of long pieces based on unleavened dough, and they are a convenient and inexpensive food that is consumed throughout the world as a staple food as well as a snack. To date, there have been efforts to make nutritionally improved noodles that by adding various ingredients, including beetroot (Chhikara et al., 2019), banana flour (Choo & Aziz, 2010), and okara (Kang et al., 2018). In particular, soy protein is added to noodles to produce low-fat, low-cholesterol, and high-protein noodles (Collins & Pangloli, 1997; Rani et al., 2019; Shi et al., 2016). However, it is still a challenge that soy protein inhibits the activity of gluten, making it difficult to form the dough (Dekkers et al., 2018; Guo et al., 2018; He et al., 2019; Wang et al., 2017).

Tofu, which has antioxidant activity, digestibility, and protein bioavailability (Preinerstorfer & Sontag, 2004), is attracting attention in both academic and industrial studies as an additive in processed foods, i.e., foods such as noodles (Xin et al., 2018), sausage (Didi Ismanto & Surya Murtius, 2013), hamburger (Kraemer et al., 2016), and ice cream (Joseph & Houghton, 2014) in order to improve the nutritional value and functionality. Whole tofu

is made from finely-ground soybeans by a heating and coagulating process, but, unlike the general tofu process, it does not remove the soybean curd. Therefore, the whole tofu has a relatively high protein and dietary fiber content compared to general tofu, which results in its higher nutritional value. Curdlan is a natural polymer that is insoluble in water, and it consists of β -(1,3)-linked glucose residues. Curdlan forms an elastic gel the properties of which differ depending on the temperature. In addition, curdlan is known to make both a cold-set gel and a heat-set gel, so it has a wide range of applications in the food industry (Cai & Zhang, 2017). A resting process that ensures that the water content is distributed evenly and extensively is used extensively in baking and making noodles. To improve the quality of the dough that has a high content of whole tofu, a resting process is conducted at high temperature so the starch will not be gelatinized.

In this study, whole tofu noodles were made by adding curdlan followed by high-temperature resting process as a strategy to manufacture dried noodles with high whole tofu content. In addition, we investigated the physicochemical properties of the proposed whole tofu dough and the dried noodles.

I.2. MATERIALS AND METHODS

I.2.1. Materials

Whole tofu was acquired from Agricultural Jandari Village Gongdongche Co., Ltd. (Gyeonggi, Korea). Commercial potato starch was purchased from the Cheiljedang Co. (Seoul, Korea). Purified salt (>99% sodium chloride) and baking soda (99.9% sodium bicarbonate) were obtained from Hanju Co. (Ulsan, Korea) and Bread Garden (Seoul, Korea), respectively. Purified wheat gluten was purchased from Roquette Freres SA (Lestrem, France). Curdlan (Cur) was obtained from the Zion Food Additives Company (Gyeonggi, Korea).

I.2.2. Preparation of the dried whole tofu noodle

Salts (1:1 NaCl:NaHCO₃) were added to the whole tofu and the mixture was blended. Commercial potato starch, vital wheat gluten, and curdlan (2, 3, and 4 w/w%, respectively) were mixed thoroughly. Blended whole tofu paste (60 wt%) was added to the flour and kneaded for 5 min using a KitchenAid mixer (Model 5KSM, KitchenAid, St. Joseph, MI, USA) to make the dough. The dough that was prepared was rested at 45 °C for 0 to 120 min. After resting, the dough was pressed to 2 mm sheets using a noodle maker

(BE-8500, Bethel Industry, Uijeongbu, Korea) and cut into noodles that were 3 mm wide. Fresh noodles were dried at room temperature for 20 hours. The detailed manufacturing process of dried whole tofu noodles and the sample design of the whole tofu dough are provided in Figure I.1 and Table I.1, respectively.

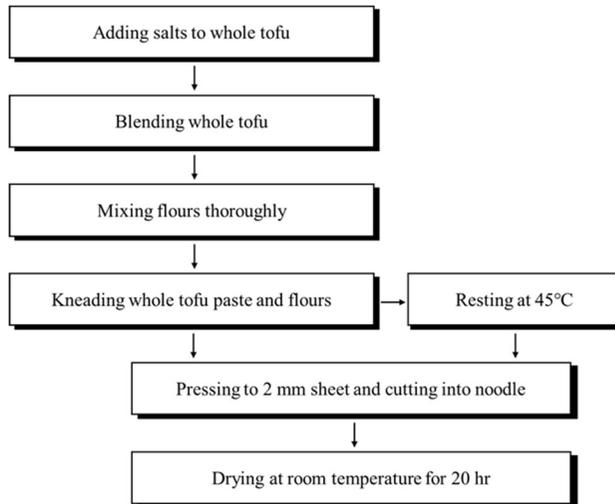


Figure I.1. Process of manufacturing dried whole tofu noodles.

Table I.1. Sample design of the whole tofu dough

Sample (ID)	Ingredients (g)					Resting process
	Whole tofu	Potato starch	Salts	Gluten	Curdlan	
Cur 2 w/w% (C2)	180	99	4.5	16.5	6	-
Cur 3w/w% (C3)	180	99	4.5	16.5	9	-
Cur 4w/w% (C4)	180	99	4.5	16.5	12	-
Cur 4w/w% 45 °C (C4R45)	180	99	4.5	16.5	12	45 °C 45 min

I.2.3. Rheological properties of the whole tofu dough

The oscillation frequency sweep test was conducted from 0.1 to 10 Hz at 30 °C using a controlled stress rheometer (RheoStress 1, Thermo Scientific™ HAAKE, Karlsruhe, Germany) to determine the storage modulus (G'), the loss modulus (G''), and $\tan \delta$. The plate diameter was 20 mm, and the gap was set to 5 mm. The rims of the samples were coated with petroleum jelly to prevent the evaporation of water.

I.2.4. Measurement of the color of the whole tofu dough

The color of the whole tofu dough samples was measured with a chromameter (CR-400, Konica Minolta Sensing, Inc., Chiyoda City, Tokyo, Japan) using the L^* , a^* , b^* color scale. L^* is a measure of brightness, and a^* and b^* indicate the red-green and yellow-blue chromaticity, respectively. An increase in positive values of a^* indicates an increase in redness, and a decrease in the positive value of b^* indicates a decrease in yellowness. The whole tofu dough samples were rested from 0 min to 120 min, respectively, and the color was measured after resting. Measurements were made in triplicate at three locations on the surface of the dough.

I.2.5. Analysis of the nutrition contents of the dried whole tofu noodle

Nutritional analysis was conducted by SGS Korea Co., Ltd. (Gyeonggi, Korea). Carbohydrate, crude protein, crude fat, sodium, moisture, ash, dietary fiber contents, and calories were measured based on the Food Code (Korea). The crude protein content was determined using Kjeldahl, and Soxhlet was used to measure the crude fat content. Energy conversion coefficients of carbohydrate, crude protein, and crude fat were based on the classification of the Food and Agriculture Organization of the United Nations (FAO). The Ottogi noodle (middle thinness) was used as a control sample.

I.2.6. Cooking properties of the dried whole tofu noodle

Cooking properties were measured by the following procedures. Optimal cooking time was determined according to AACC approved method 66–50 (AACC, 2000) with slight modifications. The cooked noodle was pressed using a transparent glass and the white core of the noodle was observed. The optimal cooking time was set when the white core of the noodle disappeared. To determine cooking loss, 25 g of noodle samples were cooked using 300 ml of distilled water. After cooking, the noodles were rinsed with 200 ml of distilled rinsing water. Cooking water and rinsing water were

collected in a beaker, dried at 105 °C for 24 h, and the residue was weighed. The water absorption of the whole tofu noodles was determined by subtracting the weight of the dried noodles from the weight of the cooked noodles. All of the cooking properties were evaluated after cooking for the optimal cooking time.

I.2.7. Analysis of the texture of the dried whole tofu noodle

All samples of the dried noodles (10 g) were put into 300 mL of boiling distilled water and cooked for each optimal cooking time. The cooked noodles were rinsed with cold water, and three strands of cooked noodles were selected, cut into lengths of 2 cm, and placed parallel to each other. The Ottogi noodles (middle thinness) were used as the control sample. The analysis of the texture of the noodle samples was conducted using a texture analyzer (Model CT3-10kg, Brookfield, MA, USA), and TA4/1000 cylinder probe was used. Trigger force was set to 5 g and the pre-test, test, and post-test speeds were 2.0 mm/s. The hardness, adhesiveness, resilience, cohesiveness, springiness, gumminess, and chewiness of the noodle samples were evaluated. All measurements were conducted 7 times.

I.2.8. Scanning electron microscopy (SEM)

The noodle samples were freeze-dried and cut into thin slices, and the sliced samples were coated with platinum that was 25 nm thick. Scanning electron micrographs were taken using a field-emission scanning electron microscope (SUPRA 55VP, Carl Zeiss, Oberkochen, Germany) using an SE2 detector. The electron high tension value was 5.0 kV. Magnification of the noodle samples' microstructures was observed at 200 x and 700 x.

I.2.9. Statistical analysis

The data were compared statistically with a one-way analysis of variance (ANOVA) followed by the Duncan post-hoc test. The analysis was performed using IBM SPSS Statistics version 25 for Windows (IBM Inc., Armonk, New York, USA). A value of $p \leq 0.05$ was used to indicate significant differences. More than triplicate measurements were performed in each experiment.

I.3. RESULTS AND DISCUSSION

I.3.1. Determination of curdlan content in making the whole tofu dough

To develop noodles that contain high-protein, preliminary experiments were conducted to measure the maximum amount of whole tofu that can be added. Consequently, the tofu content was set at 60% of the weight of the whole dough, and curdlan was introduced into the dough to control the excess moisture in the tofu. Figure I.2 shows the appearance of the dough samples with curdlan ranging from 0 w/w% to 5 w/w%. It was difficult to obtain noodles with 0% curdlan because the dough was too sticky. However, as the curdlan content increased, the stickiness decreased because the curdlan absorbed water and formed a gel network. However, the dough that contained 5 w/w% curdlan was difficult to make into noodles because the dough separated into several chunks. It seemed to be because of the overabundance of the curdlan, which impeded the interaction of the starch granules and water, thereby preventing the amylose from aggregating (Xin et al., 2018). Therefore, 2 w/w% to 4 w/w% of curdlan addition was suitable to make the tofu dough, and we finally decided to use a curdlan content of 4 w/w% to make the dough.

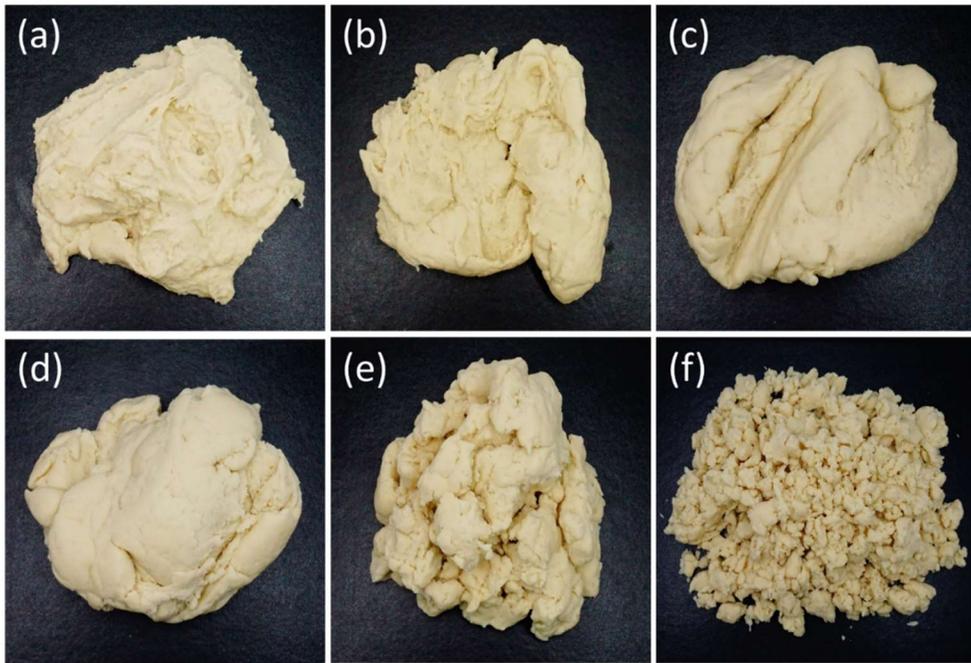


Figure I.2. Appearance of whole tofu dough with the addition of curdlan: (a) curdlan 0 w/w%; (b) 1 w/w%; (c) 2 w/w%; (d) 3 w/w%; (e) 4 w/w%; (f) 5 w/w%.

I.3.2. Rheological properties of the whole tofu dough

In order to identify the quality of the dough, we measured the rheological properties of the dough samples with the added curdlan amounting from 2 w/w% to 4 w/w%. As the concentration of curdlan increased from 2 w/w% to 4 w/w%, the G' and G'' values also increased (Figures I.3 (a) and (b)). $\text{Tan } \delta$, which is known as the dissipation factor, represents the ratio of the viscosity to elasticity. The $\text{Tan } \delta$ value of the dough samples that contained whole tofu decreased as the curdlan increased (Figure I.3 (c)), which means that curdlan has a greater effect on the increase in the G' than in the G'' of the dough. Furthermore, to increase the quality of the noodles, we introduced a resting process to make the whole tofu dough. The resting process, which usually is accompanied by making bread and which is common in the process of manufacturing noodles (Morris et al., 2000), helps the water to be distributed evenly and promotes the formation of the internal matrix. We introduced a high-temperature resting process to improve the quality of the dough. High-temperature resting helps the starch granules to swell, thereby increasing the firmness of the dough as the gel strength of the curdlan increases (Nakao, 1991). However, resting at a temperature that is too high may lead to the deterioration of quality of the dough due to the gelatinization of the starch. To control this, a resting process was conducted at 45 °C for 0 to 120 min. As

shown in Figure I.4, the resting process influenced the rheological properties of the whole tofu dough. The G' value, which represents the strength of the dough, showed a tendency to increase as the high-temperature resting time passed, and after 90 minutes, the G' value showed no significant difference ($p < 0.05$). The G' value decreased a little from 45 min to 60 min, which was thought to have imparted some heterogeneity to the samples because the ingredients of the dough were not fully hydrated in the mixing process (Barrera et al., 2007). The resting process increased overall G' , G'' and decreased $\text{Tan } \delta$. It is known that at or above 55 °C, curdlan forms a firm, resilient gel, and the strength of the gel of curdlan is independent of the incubation time (Maeda et al., 1967). As shown in Figure I.5, the 45 °C resting process increased significantly ($p < 0.05$) the G' value, which represents the elasticity of the sample. These results were thought to be related to the research results that the resting process helped the gluten network to bind with water tightly and polymerize (Kim et al., 2007; Liu et al., 2021). The rheological properties result of the rested samples seemed to be due to the interaction of various factors, such as the firmer curdlan and the swelling of the potato starch. Thus, the curdlan and the high-temperature resting process made the property of the dough more desirable for noodles.

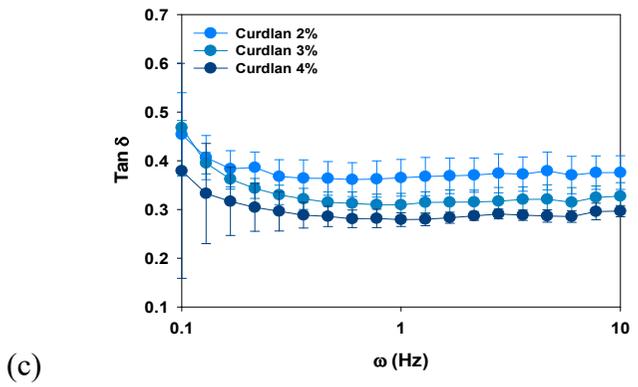
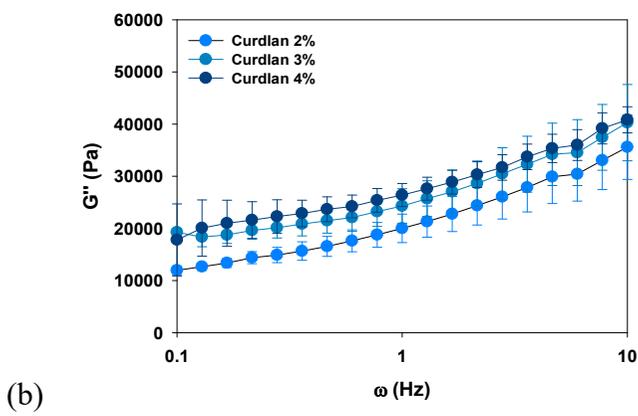
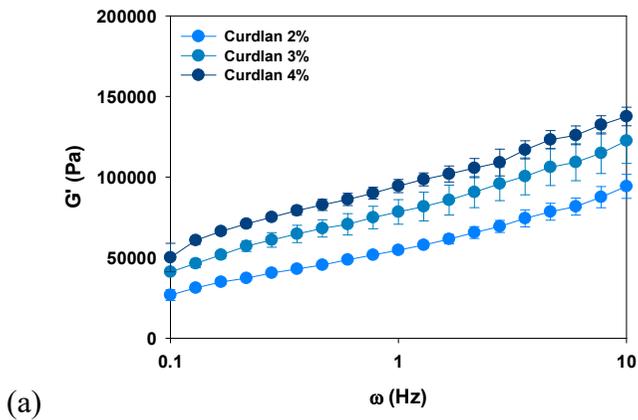


Figure I.3. Oscillation frequency sweep test results of whole tofu dough with the addition of curdlan 2, 3, 4 w/w%. (a) G' ; (b) G'' ; (c) $\text{Tan } \delta$.

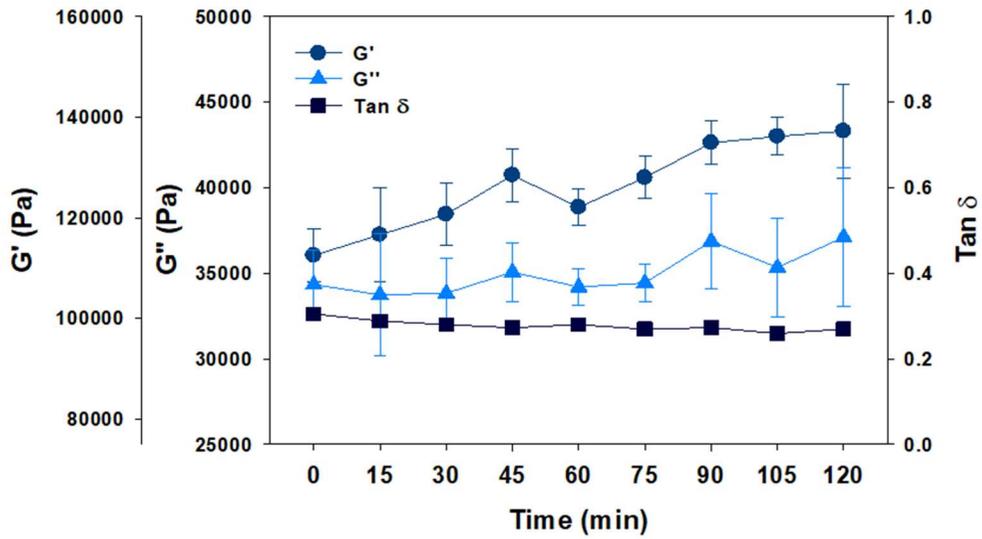


Figure I.4. Oscillation frequency sweep test results of 45 °C rested (from 0 min to 120 min, 10 Pa, 1 Hz) whole tofu dough with the addition of curdlan 4 w/w%. (a) G'; (b) G''; (c) Tan δ.

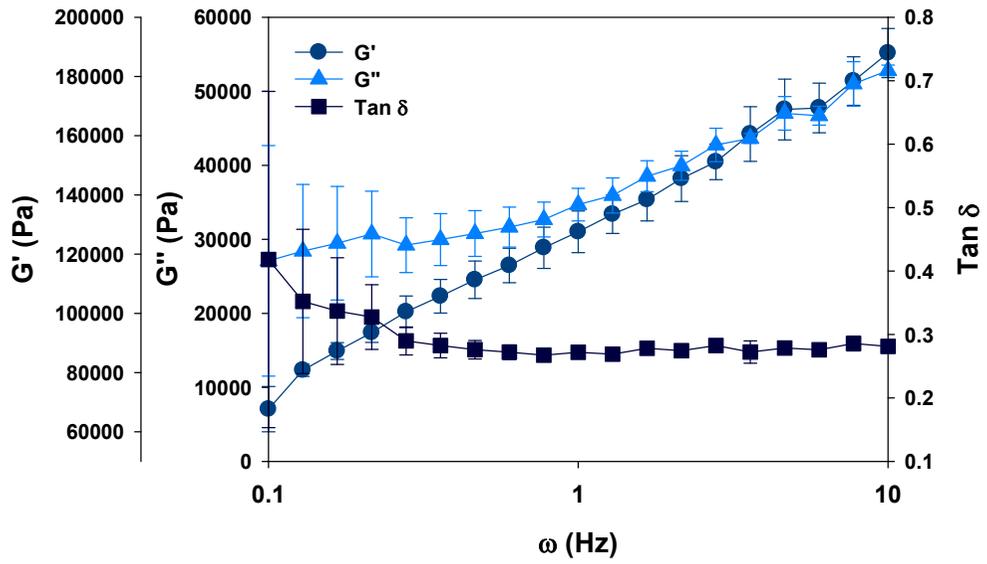


Figure I.5. Oscillation frequency sweep test results of whole tofu dough with the addition of curdlan 4 w/w% rested at 45 °C for 45 min.

I.3.3. Color measurement of the whole tofu dough

To optimize the duration of the resting process, we checked the color change of the dough. Generally, color is regarded as an important factor that influences consumers' preferences. Several conditions are associated with discoloration, such as increased protein content, absorption of water by the dough, damage in the starch, and increased storage time (Morris, 2018). In particular, since the dark color of food is an indicator of its decomposition and deterioration, it is important to select the appropriate processing method to achieve the desired color of the noodles (Siah & Quail, 2018). Since tofu contains a lot of moisture (almost 80 wt%) and protein, the color of the samples of tofu dough is darker than the colors of other noodles, such as pasta. In particular, when the resting time gets longer, the color of all of the tofu dough becomes darker. The results of the color changes of whole tofu dough during the resting time are shown in Table I.2. As the resting process progressed, the L^* and b^* values of the dough samples decreased, and, conversely, the a^* value increased. These results exhibited darker discoloration of the sample when the resting process proceeded. According to (EFI), a ΔE value higher than 5 indicates a very obvious difference in color. The results in Table I.2 indicate that resting less than 60 min was proposed to be suitable for making noodles. In conclusion, considering color, G' , and time

efficiency (Morris et al., 2000), resting the dough for 45 min was selected.

Table I.2. Colorimeter results (L^* , a^* , b^*) of 45 °C rested (from 0 min to 120 min) curdlan 4 w/w% whole tofu dough

	L^*	a^*	b^*	ΔE
0 min	84.04 ^a	-1.33 ^{ab}	20.62 ^a	-
15 min	82.01 ^b	-1.45 ^a	20.42 ^a	2.08
30 min	80.76 ^c	-1.26 ^b	20.31 ^{ab}	3.35
45 min	80.06 ^{cd}	-1.05 ^c	20.12 ^{abc}	4.14
60 min	79.32 ^{de}	-0.87 ^d	19.23 ^{bcd}	4.97
75 min	78.79 ^{ef}	-0.76 ^d	19.24 ^{bcd}	5.48
90 min	78.37 ^f	-0.58 ^e	18.92 ^d	6.01
105 min	78.39 ^f	-0.45 ^{ef}	18.49 ^d	6.14
120 min	77.33 ^g	-0.36 ^f	19.08 ^{cd}	6.98

Data are presented as means \pm standard deviations (n = 9). The values with superscript letters in the same row indicate significant difference ($p < 0.05$).

I.3.4. Nutrient composition of the dried whole tofu noodle

The nutritional components of whole tofu noodles were investigated, and the results are shown in Table I.3. Compared to the commercial noodles used as a control (Ottogi noodles), the calorie value of the whole tofu noodle was about 350 Kcal/100 g, and it was similar to the calorie value of the control. However, the carbohydrate and protein values were 10 g lower and 5 g higher than the control, respectively. The crude fat content was higher than the control, but the fat is from tofu, so it is mostly unsaturated fat, which is good for health. The salt content was lower than the control, and this result shows that the whole tofu noodle can be served as a low-salt food. In addition, since the whole tofu was not filtered, the whole tofu noodle had high dietary fiber and isoflavone, which is contained in the okara. Consequently, the dietary fiber value of the whole tofu noodle with a curdlan concentration of 4 w/w% was about 3 times higher than that of the sweet potato (3 g/100 g, USDA). Thus, it was confirmed that the whole tofu noodles had higher nutritional value than the commercially available noodles.

Table I.3. Nutrient compositions of the dried whole tofu noodle

Sample	Calorie (kcal/100 g)	Carbohydrate (g/100 g)	Crude protein (g/100 g)	Crude fat (g/100 g)	Salt (mg/100 g)	Moisture (g/100 g)	Ash (g/100 g)	Dietary fiber (g/100 g)
Control	350	76	10	0.7	1300	-	-	-
Curdlan 4%	348.58±4.12	64.29±1.65	15.24±0.76	5.36±0.50	1087.67±7.64	11.60±0.38	3.50±0.02	28.91±2.48

The control was a commercial noodle (Ottogi noodle). Data are presented as means ± standard deviations (n = 3).

I.3.5. Textural profile analysis (TPA) of the cooked whole tofu noodle

Since textural properties are directly correlated with mouthfeel, textural profile analysis (TPA) is an important factor for verifying noodles' performance. Table I.4 provides the TPA results of whole tofu noodles. As curdlan content increased, the overall texture indicators increased. In semi-solid foods, including noodles, gumminess would be an influential factor regarding the mouthfeel. The addition of curdlan increased the hardness and gumminess values of the samples. This was judged to result from the gelation of the curdlan and the swelling of the starch granules, which facilitated the firmness of the structure of the noodles. This was similar to the previous result, in which starch swelled through the resting process, which clearly was influenced by the internal structure of the noodles (Seetharaman et al., 2004). However, the application of the resting process decreased the texture properties slightly, including hardness, gumminess, and adhesiveness. A decrease in adhesiveness due to the resting process had a positive effect on the quality of the noodles. Adhesiveness shows the degree of stickiness, and high adhesiveness is not preferred for the texture of the noodles (Guo et al., 2003). Therefore, the whole tofu noodle showed better texture qualities than the control, and the C4 and C4R45 had an especially noticeable improvements in the texture properties.

Table I.4. Texture profile analysis results of cooked whole tofu noodles

Sample	Hardness (g)	Adhesiveness (mJ)	Resilience	Cohesiveness	Springiness	Gumminess (g)	Chewiness (g)
Control	2420.14±284.27 ^a	0.43±0.13 ^a	0.27±0.11 ^a	0.52±0.06 ^a	0.69±0.02 ^a	1244.43±58.62 ^a	856.57±38.34 ^a
C2	8009.71±257.94 ^b	1.81±0.45 ^{bc}	0.10±0.03 ^b	0.37±0.02 ^c	0.57±0.02 ^c	2971.71±98.45 ^b	1698.57±97.47 ^b
C3	8288.29±323.57 ^b	2.13±0.25 ^{cd}	0.12±0.06 ^b	0.41±0.03 ^{bc}	0.61±0.02 ^b	3352.29±163.51 ^c	2058.86±106.77 ^c
C4	8793.43±442.82 ^c	2.21±0.33 ^d	0.12±0.04 ^b	0.41±0.02 ^{bc}	0.59±0.03 ^{bc}	3554.14±87.57 ^d	2106.29±93.80 ^c
C4R45	8020.29±429.96 ^b	1.77±0.18 ^b	0.17±0.08 ^b	0.43±0.02 ^b	0.60±0.03 ^b	3467.14±255.80 ^{cd}	2093.43±153.11 ^c

The control was a commercial noodle (Ottogi noodle). Data are presented as means ± standard deviations (n = 7).

Different superscript letters in the same row stand for significant difference ($p < 0.05$)

I.3.6. Cooking properties of the dried whole tofu noodle

To evaluate the physical characteristics of whole tofu noodles, cooking properties were measured, including of noodles is judged to be better with less cooking time and less cooking loss (Kang et al., 2018). Figure I.6 (a) shows that cooking time tended to decrease as curdlan increased and the resting process applied. However, the resting process did not significantly influence the cooking time. An increase of the curdlan reduced the time required for the starch gelatinization of the starch, resulting in reduced cooking time (Xin et al., 2018). As shown in Figures I.6 (b) and (c), as curdlan increased, water absorption and cooked weight tended to increase. This was thought to be because the curdlan gelled, thereby absorbing water and strengthening the internal structure of the dough, and these results were consistent with the TPA results. Generally, high losses during cooking are related to high turbidity and the low palatability of consumers. As shown in Figure I.6 (d), cooking loss decreased as curdlan was added. Through the reduction of cooking loss, we observed that the yield rate of noodles increased and the turbidity of the cooking water decreased. However, the resting process slightly increased the cooking loss. To figure out the reason, the residue after cooking was analyzed using a microscope, and it was determined that most of the residue consisted of starch granules (data not shown). It was thought that

the gelation of the curdlan interfered with the starch granules, preventing them from being embedded tightly in the gluten network due to the inactivation of the gluten network (Liang et al., 2020). In addition, it was thought that the result might be correlated with the finding that the disturbance of the protein-starch matrix could increase cooking loss (Aydin & Gocmen, 2011). However, even though cooking loss increased, there was no significant difference between the weights of the cooked C4 and C4R45 samples. Therefore, these results showed that adding curdlan and using the resting process contributed to the cooking qualities of the noodles.

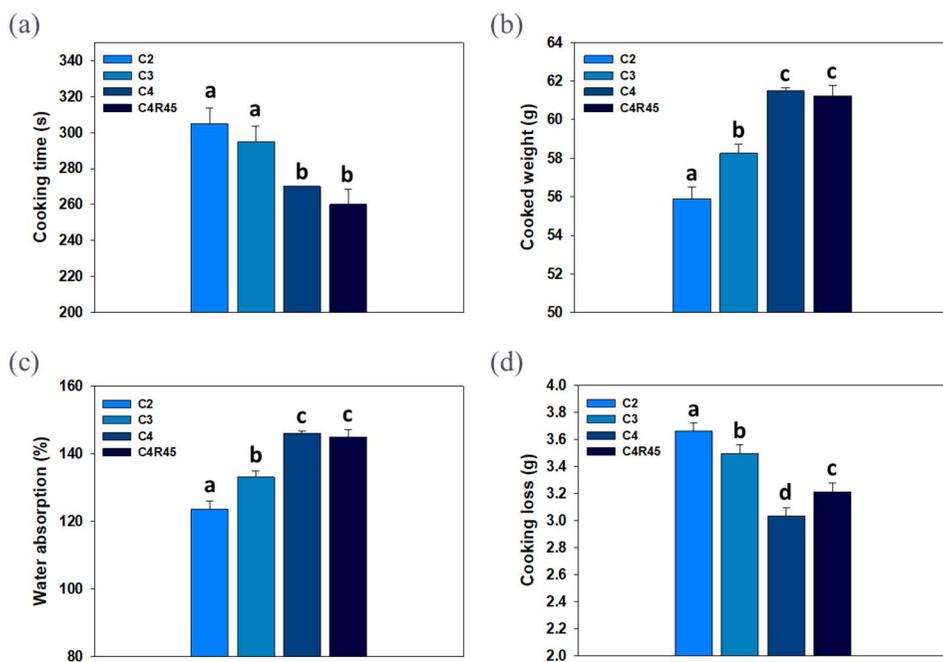


Figure I.6. Cooking properties of dried whole tofu noodles with the addition of curdlan 2, 3, and 4 w/w% and 4 w/w% rested at 45 °C for 45 min: (a) Cooking time (s); (b) Cooked weight (g); (c) Water absorption (%); (d) Cooking loss (%). Different superscript letters in the figure denote significant difference ($p < 0.05$).

I.3.7. Microstructure of the cooked whole tofu noodle

To observe the microstructure of a whole tofu noodle, scanning electron microscope images of the surface of the cooked whole tofu noodle were examined. As shown in Figure I.7, according to the difference in the amount of curdlan and the resting process, the microstructure surface of the whole tofu noodles was different. As curdlan increased, the surfaces of the noodles seemed to be smooth in the 200 x SEM images (Figures I.7 (a) - (c)), and the size of the hole was decreased in the 700 x SEM images (Figures I.7 (e) - (g)). This corresponded with the results of a previous study (Xin et al., 2018). The increase in curdlan showed a smooth surface as the starch granules decreased on the surfaces of the noodles. However, the C4R45 200 x image (Figure I.7 (d)) subjected to the resting process showed a rough surface. This was considered to have resulted from the leaking out of swelled starch granules, thereby making big holes on the surface of the noodle (Figure I.7 (h)). However, since there was no significant difference in the cooked weight of C4R45 treated with resting process and C4 without the resting process, it was judged that there would be a high possibility that noodles and a sauce could be mixed better in C4R45 with a relatively rough surface. In conclusion, it was confirmed that the microstructure of the whole tofu noodles was correlated with the results of the rheological and cooking properties.

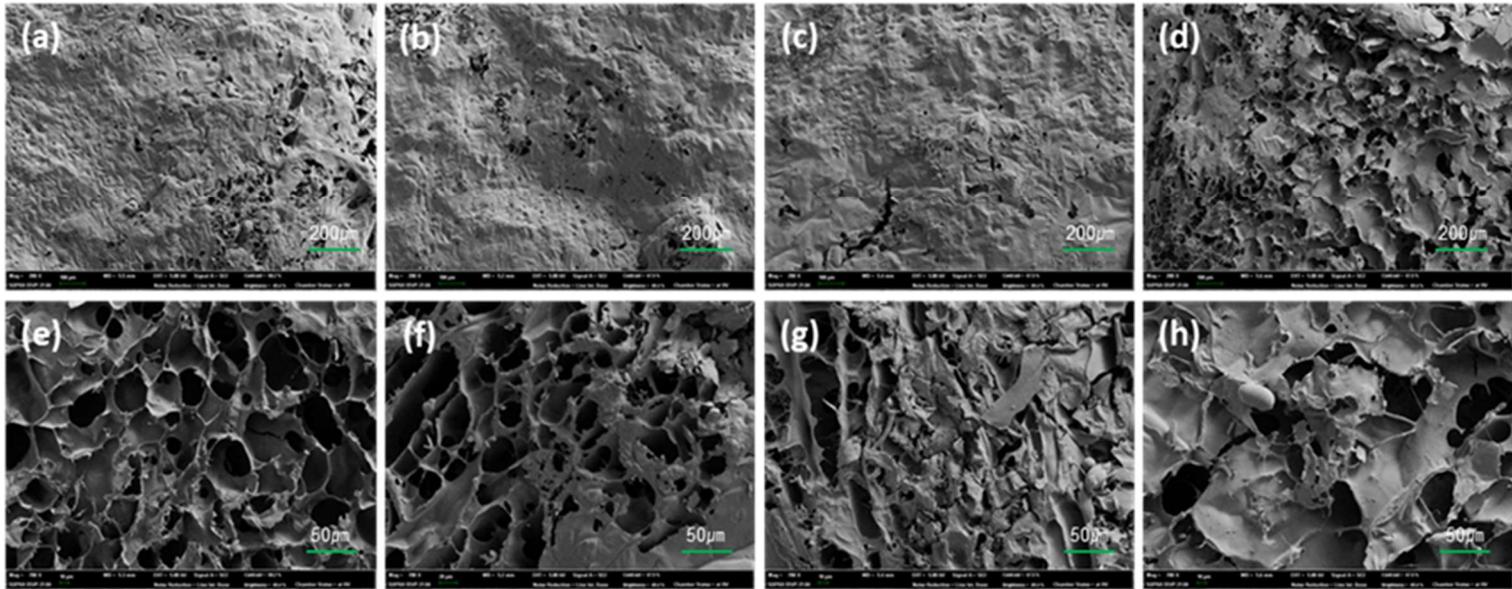


Figure I.7. SEM images of a cooked whole tofu noodle (surface). 200 x (a) C2; (b) C3; (c) C4; (d) C4R45. 700 x (e) C2; (f) C3; (g) C4; (h) C4R45.

I.4. CONCLUSION

Dried noodles that contain 60 wt% of whole tofu were manufactured by adding curdlan and applying the high-temperature resting process. The optimal ratio of curdlan (2 to 4 w/w%) to produce a whole tofu dough was proposed. As a result of analyzing the rheological properties, the storage modulus (G') increased as the curdlan was added. It could be judged that the curdlan's gel-forming characteristic increased the elasticity of the dough. In addition, as a result of high-temperature resting (45 °C), G' increased about 25%. Therefore, it was thought that the high-temperature resting process helped to form a compact internal structure in the dough, which might have been induced by the swelling of the starch and the polymerization of the gluten polymer. Cooking time was less than 5 min in all samples, and it was much shorter than commercial noodles, such as pasta. Cooking loss tended to decrease as the amount of curdlan increased because curdlan has the property of absorbing moisture. Furthermore, adding curdlan increased the hardness and gumminess by enhancing the internal structure of the whole tofu noodle. It was confirmed that the addition of curdlan and the resting process improved the quality of the noodles, resulting in increasing the texture and cooking properties. Therefore, whole tofu noodles could be substituted for commercial

noodles. Based on these results, it is expected that other products with high nutrition and improved textural properties can be developed.

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EFI. DELTA E, DELTA H, DELTA T: WHAT DOES IT MEAN?

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II. CHAPTER 2

Enhanced quality of the whole tofu dough using curdlan gelation

커들란 젤화를 이용한
전두부 반죽의 품질 향상 연구

II.1. INTRODUCTION

Tofu, which is made by heating soymilk with a coagulant causing protein dissociation, is widely consumed as a healthy and nutritious animal protein replacer. In particular, whole tofu made without removing insoluble protein is increasing in consumption because it can provide more nutritional value and unique pudding texture than normal tofu. Many research and product development attempts have been made to increase the functionality of noodles by adding soy protein in the form of soybean flour, okara, and tofu (Cheigh et al., 1976; Lu et al., 2013; Xin et al., 2018). However, tofu has 70 wt% of water content and soy globulins including glycinin (11S) and β -conglycinin (7S), which account for about 70% of the total seed protein, could disturb the gluten network (Guo et al., 2018; Thanh & Shibasaki, 1976). For these reasons, the noodle production containing high content of tofu is still challenging.

Curdlan, an FDA-approved polysaccharide produced by glucose fermentation of *Alcaligenes faecalis*, provides two types of heated edible gels: an irreversible high-setting gel formed at approximately 80 °C and a reversible low-setting gel formed at approximately 55 °C. In this study, in order to figure out the effect of curdlan gelation on the formation of the whole tofu dough,

we investigated the dough made by controlling the resting temperature of curdlan gelling properties. In particular, we tried to predict the dough formation mechanism according to the curdlan gelation effect (55 °C reversible gel formation) based on the physicochemical properties and microstructure analysis of dough prepared through the different resting processes.

II.2. MATERIALS AND METHODS

II.2.1. Materials

Whole tofu was purchased from Agricultural Jandari Village Gongdongche Co., Ltd. (Gyeonggi, Korea). Commercial wheat starch was made from Tereos Syral Co. (Aalst, Belgium). Purified salt (>99% sodium chloride) and baking soda (99.9% sodium bicarbonate) were obtained from Hanju Co. (Ulsan, Korea) and Bread Garden (Seoul, Korea), respectively. Purified wheat gluten was purchased from Roquette Freres SA (Lestrem, France). Curdlan (Cur) was obtained from Zion food additives Co. (Gyeonggi, Korea).

II.2.2. Preparation of the whole tofu dough

Salts (1:1 NaCl:NaHCO₃) were added to the whole tofu and the mixture was blended. Commercial wheat starch, vital wheat gluten, and curdlan (2 w/w%) were mixed thoroughly. Blended whole tofu paste (60 wt%) was added to the flour and kneaded for 10 min using KitchenAid mixer (Model 5KSM, KitchenAid, St Joseph, MI, USA) to make the dough. Prepared dough was rested for 30 min at three different temperatures (4 °C, 25 °C, and 55 °C).

After resting, the dough was pressed to 4 mm sheets using a noodle maker (Atlas 150, Marcato, Campodarsego, Italy). Control samples were made without addition of curdlan and resting process (Control A) and with curdlan and without resting process (Control B). The detailed sample design of the whole tofu dough is provided in Table II.1.

Table II.1. Sample design of the whole tofu dough

Sample (ID)	Ingredients (g)					Resting process
	Whole tofu	Wheat starch	Salts	Gluten	Curdlan	
Control A (A)	180	102	4.5	13.5	-	-
Control B (B)	180	102	4.5	13.5	6	-
Cur 2 w/w% 4 °C (C2R04)	180	102	4.5	13.5	6	4 °C 30 min
Cur 2 w/w% 25 °C (C2R25)	180	102	4.5	13.5	6	25 °C 30 min
Cur 2 w/w% 55 °C (C2R55)	180	102	4.5	13.5	6	55 °C 30 min

II.2.3. Analysis of the nutrition contents of the whole tofu dough

Nutritional analysis was conducted by SGS Korea Co., Ltd (Gyeonggi, Korea). Carbohydrate, crude protein, crude fat, moisture, ash, dietary fiber contents, and calories were measured based on Food Code (Korea). The crude protein content was determined using Kjeldahl and Soxhlet was used to measure the crude fat content. Energy conversion coefficients of carbohydrate, crude protein and crude fat were based on classification of Food and Agriculture Organization of the United Nations (FAO).

II.2.4. Water hydration properties of the whole tofu dough

Water hydration parameters of wheat starch and curdlan mixture were measured based on the method described by Soojung Heo (Heo et al., 2014) with slight modifications. Wheat starch (2.125 g) and curdlan (0.125 g) was mixed with distilled water (36 mL). For control samples, wheat starch (2.125 g) and not rested wheat starch (2.125 g) and curdlan (0.125 g) mixture were used. The solution was vortexed and agitated at 180 rpm for 30 min at different temperatures (4 °C, 25 °C, and 55 °C). Rested samples were centrifuged at 14000 g for 45 min at 20 °C. Supernatant of samples were dried at 105 °C for 48 hours. Wet sediment and dried supernatant were used to calculate water

hydration parameters such as water absorption index (WAI), water solubility (WS), and swelling power (SP).

$$\text{Water absorption index (WAI, \%)} = \frac{\text{Wet sediment weight}}{\text{Dry sample weight}} \times 100$$

$$\text{Water solubility index (WSI, \%)} = \frac{\text{Dry supernatant weight}}{\text{Dry sample weight}} \times 100$$

$$\text{Swelling power (SP)} = \frac{\text{Wet sediment weight}}{\text{Dry sample weight} \times (1 - \text{WSI} (\%)/100)}$$

II.2.5. GMP content of the whole tofu dough

The whole tofu dough was freeze-dried and grinded into particle size of 0.25 mm. The sample (6 g) was suspended in 1.5% (w/v) sodium dodecyl sulfate (SDS) solution (120 ml). The mixture was stirred at room temperature for 30 min and centrifuged at 13500 g for 20 min at 20°C. The supernatant was removed and the sediment was used to measure GMP content. Kjeldahl method was used to calculate GMP content of the dough.

II.2.6. Rheological properties of the whole tofu dough

The oscillation frequency sweep test was conducted from 0.1 to 10 Hz at 30 °C using a controlled stress rheometer (RheoStress 1, Thermo Scientific™ HAAKE, Karlsruhe, Germany) to determine the storage modulus (G'), the loss modulus (G''), and $\tan \delta$. The plate diameter was 20 mm, and the gap was set to 5 mm. The rims of the samples were coated with petroleum jelly to prevent the evaporation of water.

II.2.7. Analysis of the texture of the whole tofu dough

The analysis of the texture of the whole tofu dough samples was conducted using a texture analyzer (Model CT3-10kg, Brookfield, MA, USA), and TA25/1000 cylinder probe was used. The sheeted dough (4 mm) was cut into round shape (diameter: 2.5 cm). Trigger force was set to 5 g and the pre-test, test and post-test speed were 2.0 mm/s. The hardness, adhesiveness, resilience, cohesiveness, springiness, gumminess, and chewiness of the dough samples were evaluated. All measurements were conducted 9 times.

II.2.8. Scanning electron microscopy (SEM)

The whole tofu dough samples were freeze-dried and cut into thin slices. The sliced samples were coated with platinum that was 25 nm thick. Scanning electron micrographs were taken using a field-emission scanning electron microscope (SUPRA 55VP, Carl Zeiss, Oberkochen, Germany) using an SE2 detector. The electron high tension value was 5.0 kV. Magnification of the samples' microstructures were observed at 700 x.

II.2.9. Statistical analysis

The data were compared statistically with a one-way analysis of variance (ANOVA) followed by Duncan post-hoc test. The analysis was performed using IBM SPSS Statistics version 25 for Windows (IBM Inc., Armonk, New York, USA). A value of $p \leq 0.05$ was used to indicate significant differences. More than triplicate measurements were performed in each experiment.

II.3. RESULTS AND DISCUSSION

II.3.1. Nutrient composition of the whole tofu dough

Nutrient composition of the whole tofu dough is shown in Table II.2. The whole tofu dough's calorie is about 180 kcal. The dough has 35 g of carbohydrate and 7 g of crude protein. Taking account of egg, which is best source of protein, the whole tofu dough contains same amount of protein as an egg (6 to 7 g). Crude fat content is 3.14 g, moisture content is approximately 50%, and ash content is 1.68 g. In particular, dietary fiber removes harmful substances, promotes intestinal motility, and lowers blood sugar to help prevent lifestyle diseases and cancer. The dietary fiber content of the whole tofu dough is more than 5 g, and considering the recommended daily intake of dietary fiber for adults (20 to 25 g, MOHW, Korea), it can be seen that the dough has a high fiber content. Overall, the whole tofu dough has advantage of being rich in nutrients.

Table II.2. Nutrient compositions of the whole tofu dough

Sample	Calorie (kcal/100 g)	Carbohydrate (g/100 g)	Crude protein (g/100 g)	Crude fat (g/100 g)	Moisture (g/100 g)	Ash (g/100 g)	Dietary fiber (g/100 g)
Cur 2%	183.97±1.13	34.78±0.20	6.95±0.03	3.14±0.25	53.44±0.10	1.68±0.02	5.62±0.19

Data are presented as means ± standard deviations (n=3).

II.3.2. Water hydration properties of the whole tofu dough

The mixture of wheat starch and curdlan was rested at different temperatures (4 °C, 25 °C, 55 °C) for 30 min and water hydration properties were measured. Table II.3 shows the water hydration properties of mixture samples. Wheat starch granules swell as temperature increases and it is shown in the water absorption index, however, it did not dramatically affect the water hydration properties. The addition of curdlan increased the water absorption index apparently. However, there is no significant difference between control B, 4 °C, and 25 °C rested samples. While water absorption index of 55 °C rested sample increased by 100% compared to other samples. This result is mainly affected by curdlan gelation. The water solubility index decreased as resting temperature increase, and control A showed the lowest water solubility index. Low water solubility index means low affinity to water molecules. Curdlan has extensive intra and intermolecular hydrogen bonds and when curdlan is heated to 55 °C, high hydration occurs (Zhang et al., 2002). Besides, heat treatment causes molecular arrangement of curdlan polymer suspension (Maeda et al., 1967). Since curdlan is well dispersed in the solution, curdlan added samples without resting and 4 °C rested showed high value in water solubility index. While 25 °C and 55 °C rested sample showed low water solubility index due to curdlan gelation and decrease in hydrophilicity. Ruoran

Zhang's result (Zhang & Edgar, 2014) that the curdlan aqueous solution maintains a mixture of single helices and loose intertwined triple helices at room temperature and when temperature increases, it forms more aggregated rod-like triple helices by noncovalent association supports this result. Curdlan gel holds together with starch granules and fewer wheat starch granules and curdlan disperse in the supernatant. No rested, 4 °C and 25 °C rested samples' swelling power were slightly higher than the sample without curdlan, while 55 °C rested sample's swelling power was doubled than others. It is due to gelation of curdlan. Curdlan is known to absorb more than twice of water after gelation. Consequentially gelation of curdlan is a major factor that affects water hydration qualities.

Table II.3. Water hydration properties of the whole tofu dough

Sample			WAI (%)	WSI (%)	SP
Wheat starch	Curdlan	Resting			
2.125 g	-	-	171.99±2.56 ^a	0.28±0.11 ^a	1.72±0.03 ^a
2.125 g	0.125 g	-	201.73±2.30 ^b	1.26±0.33 ^{ab}	2.04±0.02 ^b
2.125 g	0.125 g	4 °C 30 min	203.16±3.08 ^b	3.69±1.16 ^c	2.11±0.05 ^c
2.125 g	0.125 g	25 °C30 min	214.61±0.27 ^c	2.19±0.58 ^b	2.19±0.01 ^d
2.125 g	0.125 g	55 °C30 min	448.01±4.24 ^d	0.96±0.14 ^a	4.52±0.04 ^e

Different superscript letters in the same row denote significant difference ($p < 0.05$).

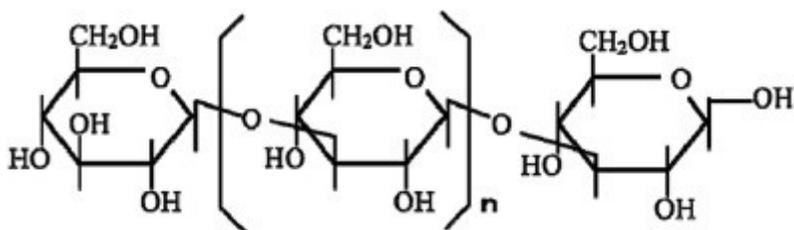


Figure II.1. Chemical structure of curdlan (Tilottoma Saha, 2020).

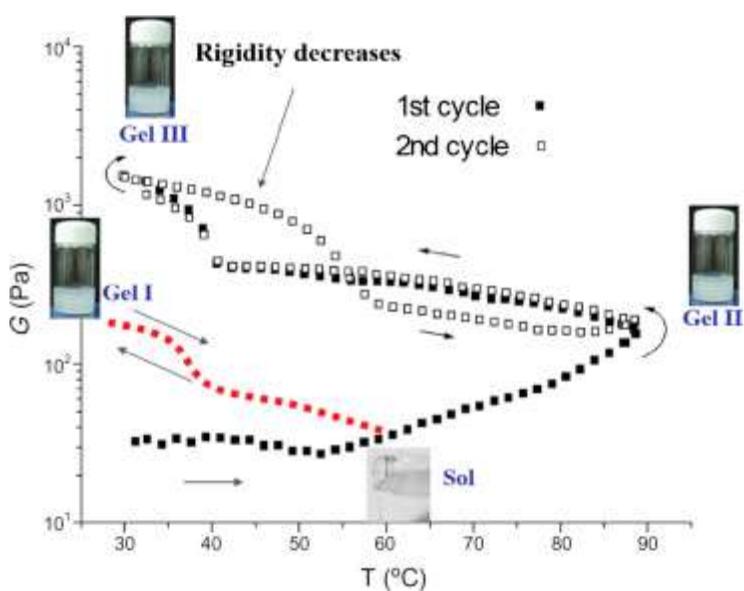


Figure II.2. Typical temperature dependence of storage modulus G' for 2% suspension of curdlan during two heating and cooling cycles at $1^\circ\text{C}/\text{min}$ (Cai & Zhang, 2017).

II.3.3. GMP content of the whole tofu dough

In the dough system, disulfide bridges established between the glutenin chains provide stability and strength to the dough, improving bread quality (Barrera et al., 2007). Glutenin macropolymer (GMP) content is a key factor of dough property that demonstrates gluten activation, furthermore, the amount of GMP is highly associated with the strength of dough (Wieser, 2007). High GMP content indicates the polymeric structure of gluten proteins (Wang et al., 2007). According to Figure II.3, the GMP content of the dough without curdlan was similar. It means that the resting process at different temperatures did not influence gluten structure itself. However, the GMP content of the curdlan added whole tofu dough samples displayed different consequences. As a result of the gelation of curdlan, the gluten frame structure of the dough disrupted and GMP content decreased. This is because of curdlan gelation, which hinders the vital wheat gluten from fully combining with water so that the gluten could not stretch and form gluten network (Liu et al., 2020). After all, while gelation of curdlan disrupted gluten structure, the hindrance did not significantly affect the dough's quality.

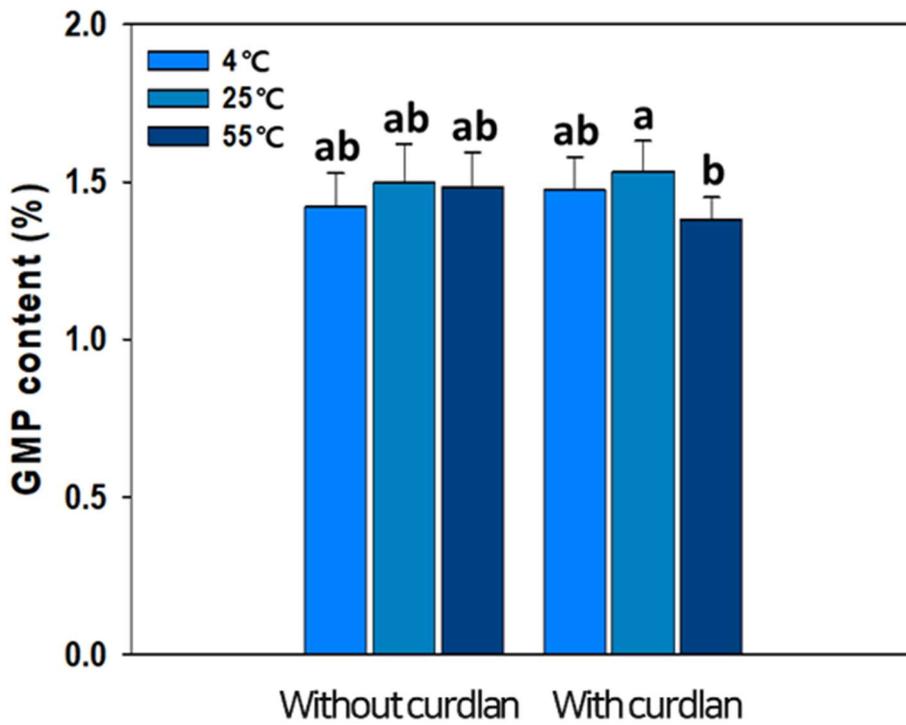
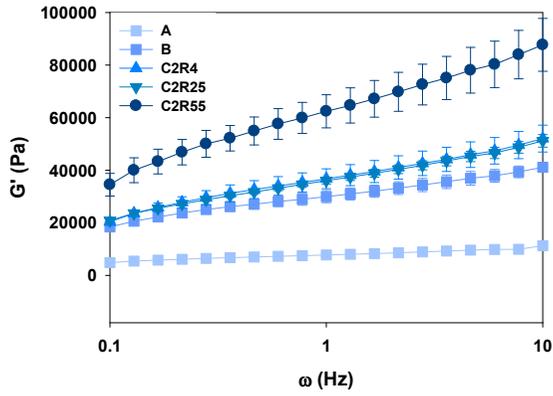


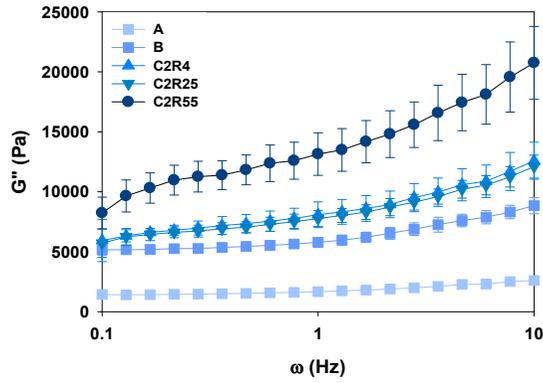
Figure II.3. GMP content of the whole tofu dough (Without and with curdlan) rested at different temperatures. Different superscript letters stand for significant difference ($p < 0.05$).

II.3.4. Rheological properties of the whole tofu dough

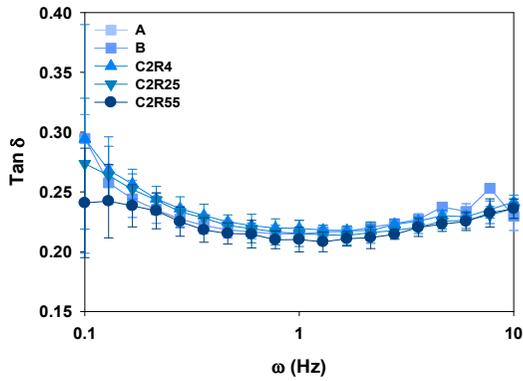
Figure II.4 exhibits the rheological qualities of the whole tofu dough. Compared to control A which is made without curdlan, curdlan added samples showed higher G' and G'' . There were no significant differences in the G' and G'' values between 4 °C and 25 °C rested samples. However, the 55 °C rested sample showed significantly higher G' and G'' values than those of the other samples, which is related to curdlan gelation. The colloidal curdlan can form a thermo-reversible gel at about 55 °C, and when the temperature is increased to 80 °C or higher, the curdlan can form a thermo-irreversible gel, which is highly stable and solid. Typically, thermo-reversible low set gel's storage modulus is more than twice as high as curdlan sol (Zhang et al., 2002). High G' and G'' value contribute to making better dough. Therefore, it was judged that the sample with a resting process at 55 °C formed the better quality dough than the other samples.



(a)



(b)



(c)

Figure II.4. Oscillation frequency sweep test results of whole tofu dough with the addition of curdlan 2 w/w% at the different temperatures. (a) G' ; (b) G'' ; (c) $\text{Tan } \delta$.

II.3.5. Texture profile analysis (TPA) of the whole tofu dough

There are three factors that affect the dough properties: gluten structure, swelling of starch, and curdlan gelation. Gluten structure mainly builds the frame structure of the dough and closely related to the springiness of the sample. Starch swelling and curdlan gelation affect the density of the inner structure which regulates hardness and cohesiveness. Texture analysis results of the whole tofu dough samples are shown in Table II.4. As mentioned in previous study, curdlan enhanced the hardness of the dough, indicating that curdlan made firmer structure. Adhesiveness and resilience showed similar values between samples, while cohesiveness and springiness decreased as curdlan added and resting temperature increase. The decline of GMP content at 55 °C rested sample supports interruption of gluten network by curdlan gelation and decrease in cohesiveness and springiness. This result is in coincidence with Ying Liang's (Liang et al., 2020), that curdlan absorbs water from starch and gluten. In semi-solid food, gumminess serves as crucial index which indicates the texture of the product. Gumminess of 55 °C rested sample is highest among samples and it is due to gelation of curdlan. That is, gluten disruption by curdlan gelation in dough quality has minor effect. Hardness increases even when cohesiveness decreases, significantly increasing gumminess. Overall, the influence of curdlan is most dominant and the

application of the resting process positively contributed to the whole tofu dough structure by reinforcing inner structure which is mainly caused by curdlan gelation.

Table II.4. Texture profile analysis results of the whole tofu dough

Sample	Hardness (g)	Adhesiveness (mJ)	Resilience	Cohesiveness	Springiness	Gumminess (g)	Chewiness (g)
A	69.11±12.72 ^a	2.71±0.33 ^a	0.05±0.01 ^{ab}	0.94±0.09 ^a	0.98±0.05 ^a	65.33±15.53 ^a	64.67±16.70 ^a
B	982.78±185.83 ^b	6.03±0.78 ^b	0.03±0.02 ^a	0.89±0.08 ^a	0.93±0.05 ^b	870.56±137.96 ^b	810.22±158.06 ^d
C2R04	2340.67±110.71 ^c	6.93±0.71 ^c	0.07±0.04 ^b	0.57±0.06 ^b	0.62±0.06 ^c	1329.33±119.74 ^d	822.78±126.63 ^d
C2R25	2038.67±221.17 ^d	6.12±0.66 ^b	0.04±0.03 ^a	0.57±0.06 ^b	0.57±0.04 ^d	1162.33±119.27 ^c	664.00±96.74 ^c
C2R55	5853.78±581.82 ^c	5.94±0.84 ^b	0.04±0.03 ^a	0.24±0.03 ^c	0.39±0.02 ^c	1376.00±62.82 ^d	543.00±43.69 ^b

The data with different superscript letters in the same row is significantly different ($p < 0.05$) for every texture profile.

Data are presented as means ± standard deviations (n=9)

II.3.6. Microstructures of the whole tofu dough

To get a better understanding of the internal structure of the whole tofu dough, SEM images of the dough's surface and cross-section were observed. As shown in Figure II.5, the curdlan added samples were able to observe starch which is enclosed in curdlan gel on both surface and cross-section. Especially, 55 °C rested sample's surface showed most smooth surface among samples. The microstructure of the whole tofu dough was influenced by gelation of curdlan at 55 °C, giving a tighter and packed structure. This demonstrate that the addition of curdlan and curdlan gelation contributed positively to strengthening the internal structure of the whole tofu dough.

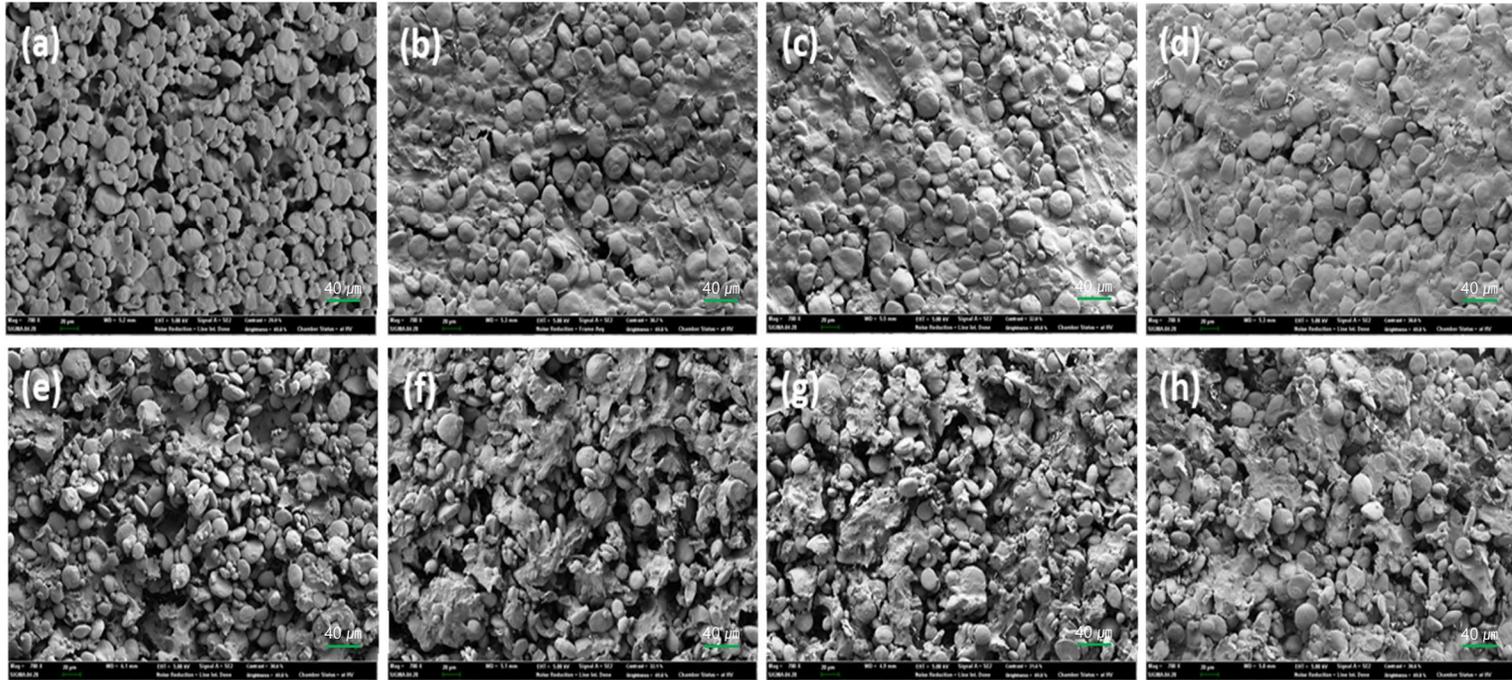


Figure II.5. SEM images of the whole tofu dough (700 x). Surface (a) A; (b) C2R04; (c) C2R25; (d) C2R55. Cross section (e) A; (f) C2R04; (g) C2R25; (h) C2R55.

II.4. CONCLUSION

In order to figure out the impact of curdlan gelation on the whole tofu dough, the resting process at different temperatures was conducted. The physicochemical characteristics of the whole tofu dough were evaluated. Water absorption properties doubled and rheological properties increased approximately 50% after resting at 55 °C. GMP content slightly decreased, and hardness and gumminess increased significantly in texture profile analysis. The microstructure of the whole tofu dough rested at 55 °C became more condensed and smooth. Taken together, the resting process at 55 °C improved the dough quality by gelation of curdlan. Even though curdlan gelation disrupted the gluten network, curdlan gelation positively and mainly contributed to the internal structure of the dough. In addition, the curdlan gel structure could hold excessive water resulting in a solid dough structure. This investigation may provide information on the influence of curdlan gelation in the food processing industry and expand the use of curdlan in food products such as bread and cookies.

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국 문 초 록

최근 두부는 단백질 함량이 높고 칼로리가 낮아 아시아를 넘어 전세계적으로 소비되고 있다. 특히, 두부를 다양한 형태로 가공한 제품들, 대표적으로 압착성형한 두부면이나 푸주가 인기를 끌고 있다. 하지만 압착성형 된 두부면은 식감이 딱딱하고, 상대적으로 전분의 함량이 낮아 소스를 잘 흡수하지 못하는 등 관능적 특성이 좋지 못하다는 단점이 있다. 또한 건조 시 기름 분리 및 품질 저하 등의 어려움으로 인해 건조면으로 제공되지 못하고 보존수에 의존하는 등 편리성 개선의 문제가 존재한다.

이에 본 연구의 첫 장에서는 물성이 향상되고 편리하면서도 고함량의 전두부를 함유한 건면을 개발하고자 하였다. 일반 두부보다 단백질 및 식이섬유의 함량이 높은 전두부(60 w/w%)와 수분을 조절하기 위한 감자전분(34 w/w%) 및 커들란(2~4 w/w%)을 주재료로 선택하였으며, 반죽의 수분 조절을 위한 전략으로 고온 레스팅(45°C) 공정을 도입하였다. 제조된 반죽의 탄성(G')과 점성(G'')은 커들란 첨가량이 높아질수록 증가하였고 45°C에서 45분간 레스팅 된 후에는 레스팅 전보다 탄성(G')이 50% 이상

증가하였다. 커들란이 4% 첨가된 전두부 건면의 조리 시간은 20% 짧아졌으며 조리 손실 또한 약 15% 감소하였다. 경도와 검성 또한 10% 이상 증가하여 결론적으로 품질이 향상된 전두부 건면을 제조할 수 있었다.

첫 장의 연구에서는 감자전분의 호화온도(50°C)와 검의 젤화 온도(55°C)간의 차이로 인해 최적의 반죽 특성을 부여하지 못하였다. 이에 두번째 장에서는 커들란 젤화의 효과를 최대화하여 물성이 향상된 전두부 반죽을 만들기 위한 새로운 전략을 제시하고자 했다. 젤화와 호화 기작의 최적 온도를 설정하기 위해서 고온 레스팅 온도를 커들란 젤화가 일어나는 55°C로 설정하고, 호화 온도가 60°C인 밀 전분을 선택하여 반죽을 제조했다. 다양한 온도(4°C, 25°C, 55°C)에서 전두부 반죽을 레스팅 해본 결과 55°C에서 커들란의 젤화로 인해 수분 흡수 정도가 2배 이상 증가하고 탄성(G')과 점성(G'') 모두 50% 가까이 높아졌다. 씹는 느낌과 직결된 경도는 55°C 샘플이 25°C 샘플보다 2배 이상 향상되고 검성은 15% 강화되었다. 한편, 글루테닌 고분자 함량분석을 통하여 커들란의 젤화가 글루텐 활성을 미미하게 저해함을 확인하였지만, 커들란 젤화가 반죽의 내부 구조를

강화하는 특성이 커 종합적으로 커들란의 고온 레스팅이 반죽 특성을 향상시킴을 확인하였다. 결과적으로 밀가루 전분과 커들란 및 55°C의 레스팅 공정의 활용은 전두부가 함유된 반죽의 물성 향상에 크게 기여하는 것을 확인할 수 있었다.

앞선 두 실험을 통해 단백질과 식이섬유 함량이 높으면서 간편하고 건조 후에도 물성이 좋은 전두부 건면을 개발하였고, 커들란 젤화를 통해 반죽의 품질을 향상시키는 방법을 제안하였다. 이러한 결과는 전두부 및 커들란을 활용한 쿠키 등의 반죽 기반의 다양한 제품 개발에 도움이 될 것으로 사료된다.

주요어: 전두부, 커들란, 레스팅 공정, 건면, 반죽, 물성 특성, 질감 특성

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