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The effect of extraction method and types of coagulants on the results and physicochemical properties of tofu

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Abstract

Extraction and coagulation techniques are essential stages in tofu production. This research studies the effect of extraction and coagulation techniques on physicochemical properties of soybean extract, soy pulp, and tofu. This study applies three extraction techniques, namely manual heat extraction, manual cold extraction, and cold extraction using mechanical devices. They used nigari coagulant. As a control, the process of making tofu follows the procedure process as in the small industry of tofu with whey coagulant. The measurement of material mass balance is done at each stage of the process, while the evaluation of the product includes water content, ash, protein, fat, magnesium, and texture. The experimental results show that an increase in the ratio of water to soybeans and the increase of extraction temperature can increase the amount of extracted protein. The coagulation using nigari produced 22.18 kg tofu in hot extraction treatment, 19.61 kg in cold extraction and 17.60 kg in machine extraction. The control treatment produced 36.8 kg of tofu. The water content in the control treatment was higher than the other three treatments. Protein levels, magnesium levels, hardness and chewing power for treatment with nigari coagulant were significantly higher than those with whey.

Keywords: extraction; coagulant; nigari; tofu; whey.

Practical Application: get an effective and efficient tofu production process with the right extraction process and coagulant type.

1 Introduction

Tofu is a soy-based food product. The basis of the process of making tofu is by utilizing the soy protein gelation properties. The principle of the process step is that the protein in soybeans is extracted using water into soy milk and then coagulated using coagulant. In general, the process of tofu production is divided into two main stages namely the stage of protein extraction and the stage of coagulation of protein into tofu products. The extraction stage itself generally includes the activities of grinding soybean, cooking soybean porridge, and filtering (Yuwono & Susanto, 2016).

Protein extraction is included in the process of separation through diffusion basis. The steps that determine the rate of protein mass transfer are most likely to be the dissolution of intracellular components and the transfer of solutes to the surface of a solid matrix (Jung et al., 2012). Some variables that affect protein solubility include matrix particle size (grinding efficiency), solvent pH, solvent ionic strength, temperature, time and protein concentration (Preece et al., 2017). The effect of concentration, which is the ratio of the mass of soybeans to the volume of extracting water, was studied (Yuwono & Susanto, 2016). Other studies on the effect of mass ratio of soybean seeds to water volume and extraction temperature on percent protein, fat, and carbohydrates extracted in the process of making vegetable milk have been studied (Ariyanto et al., 2015).

Coagulation of soybean extracted protein in the small tofu industry in the Subang and Sumedang regions, in general, is using whey. Whey is a tofu-coagulating liquid that has been allowed to stand overnight (Darmajana et al., 2015). Besides whey, the types of coagulant in the production of tofu on the market are GDL (*Glucono Delta Lactone*), $CaSO_4$ and $MgCl_2$ (Haqqi, 2011). The use of whey will produce a lot of wastewater from the coagulation process, which is between 110-164 liters per 10 kg of soybean seeds (pH value 4.78-5.29) and smells bad. The whey from the tofu industry is mostly dumped, either into infiltration wells or public channels, so it can pollute the environment. Environmental pollution from tofu wastewater can be reduced by using nigari coagulants (Widaningrum, 2015).

Nigari is a type of chloride coagulant ($MgCl_2$ dan $CaCl_2$) which comes from seawater extract which is a by-product during salt production. Coagulation using $MgCl_2$ creates a more natural tofu flavor and allows maintaining the original taste of soy (Li et al., 2014). At the moment, the use of $MgCl_2$ as coagulant has been popular (Li et al., 2014, 2015; Zhu et al., 2016). The optimum coagulation temperature using nigari is at 85 °C and a concentration of 7.5% (Afifah & Darmajana, 2017) and in general, the coagulation process should be carried out in a temperature ranging 70-85 °C (Ohara et al., 1992). Coagulation below 70 °C produces tofu with a soft and runny texture, while coagulation above 85 °C produces tofu that has a hard and uneven texture.

The protein coagulation process is carried out after the filtering stage (separation of soybean pulp into soy milk and pulp) and produces soybean juice or soy milk. To get soybean juice at a temperature of 70-85 °C, filtering must be done at a high enough temperature (more than 85 °C). In general, filtering

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in the small industry of tofu is done manually using filter cloth and craftsmanship. Filtering at high temperatures can certainly be dangerous and inconvenient for operators/craftsmen. This study aims to study several extraction techniques and the use of nigari coagulants. The results of the research are expected to obtain extraction techniques that are more comfortable and safe for the operator, but can extract protein optimally, as well as reduce the use of the amount of clean water and wastewater produced. In this study, three methods of extraction were carried out, namely manual heat extraction (EPM); manual cold extraction (EDM); and cold extraction using a machine (EDO) with nigari coagulant (seawater extract).

2 Materials and methods

2.1 Material

The study was carried out at CV Stempert Enterprise business unit in Subang. The materials used in this study were imported soybean seeds obtained from soybean distributors in Subang and nigari as coagulants obtained from nigari distributors in Surabaya, whey as a control treatment coagulant was obtained from the business unit. Equipment used include conventional soybean grinding machines, cone-shaped stone grinding machines (extraction machines), filter cloths, tofu molds, digital scales, measuring cups and equipment for chemical analysis as well as gauges for texture parameters (*Texture Analyzer*).

2.2 Method

Experimental design

The design of the experiment used a one-factor randomized design (treatment). The treatments in this study were extraction techniques (milling, cooking, and filtering stages) which consisted of 3 qualitative stages, namely manual heat extraction (EPM); manual cold extraction (EDM); and cold extraction using a cone extraction machine (EDO). These three treatments use nigari coagulant as much as 7.5% by weight of soybean seeds. As a control is manual heat extraction with whey coagulant, which is making tofu according to the work procedures that are normally carried out by small-tofu industries such as CV Stempert Enterprise business unit. Each treatment was repeated 3 times.

Research implementation

Tofu is made in accordance with the steps commonly done in small tofu industries (control treatment, namely manual heat extraction with whey coagulant). The tofu making stage starts with soaking soybean for 4 hours with a ratio of soy: water as much as 1:3, draining, grinding with soybean: water ratio as much as 1:2, cooking at temperature of 90-95 °C for 64 minutes with the ratio of soybean:water of 1:13, filtering (using batis cloth) and room temperature water added with a ratio of soy:water as much as 1:12, coagulation, molding with wood molding and compacting with 8.5 kg ballast for 16 minutes, cutting, and tofu boiling. Soybean raw materials used in each treatment were 10 kg.

In the EPM (Manual Heat Extraction) treatment, the steps carried out only to the cooking activity, the same as the control treatment, then in the filtering stage no water was added. In the EDM treatment (Manual Cold Extraction), which uses stages such as the EPM treatment, with the difference in treatment at the filtering stage carried out before the cooking stage with a ratio of soy: water as much as 1:13. Whereas in the EDO treatment, the grinding and filtering stage uses a machine that functions as a soybean grinder, to make it into soy pulp, and at the same time, filter the soy pulp, so as to produce soy milk and soy pulp (Figure 1). In this process, the addition of water to room temperature is carried out continuously with soybean to water ratio of 1:15.

At each treatment, a mass measurement was done by weighing all ingredients in each stage of the process (soybean, process water, soy porridge, soy pulp, and wastewater). Measurement of water and protein content is carried out on the results of the separation between soybean pulp and soybean juice (milk). Analysis of proximate, magnesium levels and texture of tofu is done on the produced tofu.

Proximate analysis (water, ash, protein, and fat) is carried out according to the procedure in SNI 01-2891-1992 on how to test food and drinks (Badan Standardisasi Nasional, 1992). While carbohydrate content is determined by difference. The magnesium content in tofu and wastewater is determined by the 18-13-1/MUI/SMM-SIG method, ICP-OES, which is carried out by PT Saraswanti Indo Genetech. Tofu texture analysis was performed to obtain data on hardness, cohesiveness, springiness and chewiness using the TA instrument. XTPlus Texture Analyzer (Stable Micro Systems), using the P36 probe with a pre-test speed of 5 mm/s, a test speed of 1 mm/s, and post-test speed of 1 mm/s (Li et al., 2015).

3 Result and discussion

The manual heat extraction (EPM) treatment produces soymilk which has a temperature close to the optimum coagulation temperature of (70-85 °C). The obstacle in this treatment is that filtering is carried out at relatively high temperatures (85-90 °C). In the manual cold extraction (EDM) treatment, the filtering stage is carried out after grinding the soybean seeds and before cooking, so the filtering takes place at room temperature of (28-30 °C), so that the work of the operator or craftsman is safer and more comfortable. After filtering, the soybean juice

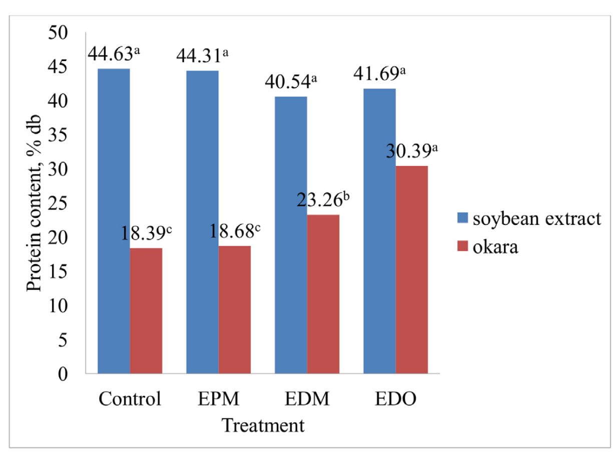

Figure 1. Protein content of soy milk and okara.

is cooked (heated). In the automatic cold extraction (EDO) treatment, the filtering process is carried out using a machine, which can grind the soybean, as well as separating the soybean extract from the soy pulp through the centrifugation process through a filter cloth. After the grinding and filtering process, then the soybean juice is cooked (heated).

The amount of material at each stage of the process during the production of tofu can be seen in the balance sheet as presented in Table 1. Making tofu in the control treatment requires significantly more water than other treatments, namely the ratio of soybeans to water as much as 1 to 30. This is because in the process of making tofu of the control treatment, an addition of water at the filtering stage is as much as 120 liters, while the stage of adding water when filtering is not carried out in other treatments. The addition of water is intended to reduce the temperature of the material when filtering (become 65-70 °C), so that manual squeeze can be performed optimally. It also increases the amount of dissolved protein.

Wastewater produced by each treatment is significantly different. The control treatment produces significantly more wastewater than other treatments. The amount of produced waste is influenced by the amount of water used during the extraction process, the coagulation process, and the tofu molding process. The process of coagulation and molding is the stage that produces the largest wastewater in the tofu production process.

The control treatment produced the highest yield (tofu), which weighed 36.81 kg, and was significantly different at the 95% level from the other treatments. While the heat extraction (EPM) treatment was not significantly different from cold extraction (EDM), but it was significantly different from machine extraction (EDO) (see Table 1). Likewise, cold extraction (EDM) yields no significantly different yield compared to machine extraction (EDO). From Table 1, it is seen that the extraction machine (EDO) produces the lowest yield (tofu), which is 17.60 kg.

Coagulation in acidic conditions with whey coagulant tends to produce soft tofu. Protein coagulation occurs slowly and the tofu bumps formed have a large water-holding capacity. Tofu produced has a high-water content. The heat extraction treatment (control and EPM) results in significantly higher tofu yields than the cold extraction treatment (EDM and EDO), but there is more produced soy pulp because the extraction process at high temperatures limits workers in pressing the soy pulp. The extraction with machine (EDO) produces the least amount of soy pulp because the process of separating the soy pulp is done automatically using a machine.

The effectiveness of the extraction process is shown by the amount of protein produced (Figure 1). This study uses soybean with a water content of 11.93% and a protein content of 36.43%. Protein remaining in the soy pulp in the control treatment and manual heat extraction treatment (EPM) was significantly lower than the cold extraction treatment (EDM and EDO). Protein extracted in soybean extract did not show significant differences among treatments. The control treatment produces pulp with a smaller protein content because the addition of water to the pulp squeeze process helps extract the protein. In the EPM treatment, extraction is carried out at a higher temperature than other treatments. In the method of extraction by machine (EDO treatment) the protein content in the soy pulp is still quite high, because the amount of water used to extract the protein is the least and the temperature of the extraction water is at room temperature.

Ariyanto et al. (2015) reported the effect of the ratio of soybean mass to water volume and extraction temperature on the percentage of extracted protein. Similar results were reported by Yuwono & Susanto (2016) that the more water used, protein extractability and the use of soy: water ratio from 1:10 to 1:30 increased the extracted protein from 73.08% to 83.21%. Increasing the ratio of water volume to soybean mass and extraction temperature increases the percentage of extracted protein. The protein extraction process takes place through a mass transfer process that is influenced by differences in the concentration between soybean and milk (soybean extract) until an equilibrium concentration is reached between pulp and milk. To achieve that equilibrium concentration, the ratio of soybean mass and greater volume of water requires more dissolved protein. The higher the temperature, the greater the diffusivity. With the greater diffusivity, the mass transfer process becomes faster. So that more and more protein is extracted.

One of the advantages of using nigari coagulant is that nigari contains a fairly large amount of magnesium, around 506.1 mg/100 g. For this reason, magnesium content in the results of the experimental process was measured. The magnesium content in tofu and wastewater can be seen in Figure 2. The magnesium (Mg) content in control treatment tofu and wastewater is significantly lower than other treatments. Coagulation with nigari produces tofu with 5 times more magnesium content than tofu from the control treatment (whey coagulant), whereas in wastewater, the magnesium content reaches 11 times more. This is because the nigari coagulant used has a magnesium content of 506.1 mg/100 g. Magnesium is known to be one of the macro minerals that is important for heart health. Magnesium deficiency can cause hypomagnesema with one of its symptoms is irregular heartbeat (Winarno, 2004). Magnesium has a function as a relaxation of vascular smooth muscle so that when magnesium needs are not

Table 1. Material mass balancein various treatments.

Treatments	*Process water, kg*	*Wastewater, kg*	*Okara/ soy pulp, kg*	*Tofu, kg*
Control	294.25^{a}	270.51^{a}	20.33^{c}	36.81^{a}
EPM	171.36^{c}	122.53^{d}	23.45^{a}	22.18^{b}
EDM	168.58^{c}	127.36^{c}	22.04^{b}	19.61^{bc}
EDO	179.11^{b}	146.96^{b}	13.08^{d}	17.60^{c}

Values within each column with different superscript lowercase letters (a-d) represent significant difference ($p < 0.05$).

met. There will be a decrease in blood pressure which causes an abnormal heartbeat.

The nutritional content of tofu produced in various treatments can be seen in Table 2. Soybean juice in the treatment of EPM, EDM and EDO with nigari coagulant ingredients has a temperature in the range of 80-85 °C with coagulation pH of 6.20-6.31. Meanwhile, in making tofu of the control treatment, with whey as the coagulant material, the temperature of coagulation occurs at 62-65 °C with pH of 4.01-4.04.

The water content of tofu of the control treatment was significantly higher compared to other treatments. While the protein content for the control treatment was markedly lower. The high-water content in the control treatment is influenced by the type of coagulant used. Coagulation using whey occurs under acidic conditions. According to Obatolu (2008) coagulation using acidic coagulant will produce tofu with a tenuous matrix so that the water trapped is in high quantities. Instead, the coagulation using nigari ($MgCl_2$) produce tofu with low water holding capacity caused by the tighter tofu matrix.

The level of wet base protein (Table 2) was significantly lower than the other treatments. This is because the water content in the tofu control is significantly higher. Protein levels of control treatment, EPM, EDM, and EDO on a dry basis did not show any significant difference, namely 55.65%, 55.32%, 56.31%, and 57.14%, respectively. Ash content of the control treatment was significantly smaller than other treatments because the EPM, EDM, and EDO treatments used nigari as the coagulant which contains many mineral elements thereby increasing the value of tofu ash produced. Fat content showed no significant difference between the control treatment and other extraction technique treatments with coagulation using nigari. The control treatment had significantly higher carbohydrate levels compared to the EPM and EDM treatments.

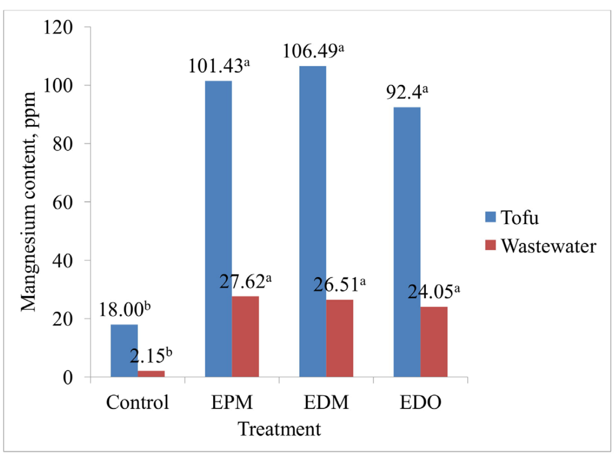

Figure 2. Magnesium content of tofu and wastewater.

SNI 01-3142-1998 regarding quality requirements of tofu requires a minimum protein value of 9%, minimum fat of 0.5%, and maximum ash of 1%. The quality of tofu in various treatments (Table 2) shows that the control protein content does not meet SNI standards while the other treatments have fulfilled the SNI standard. The research result of Apriyati et al. (2017) showed that tofu from imported soybeans with nigari coagulant had a water content of 71.02%, ash content of 0.77%, protein 10.33%, fat 6.34%, and carbohydrate 1.42%. From the aspect of nutrition content and SNI, tofu with nigari coagulant is better and meets SNI requirements.

Tofu texture is one of the parameters that consumers consider. The texture of various tofu treatments can be seen in Table 3. The texture of tofu is determined by the amount of water trapped and the type of coagulant that affects the protein fraction that is coagulated as tofu (Obatolu, 2008; Blazek, 2008). The tofu of the control treatment has significantly lower hardness than other treatments. According to Obatolu (2008), Tofu with high hardness has a low water holding capacity. In other words, tofu with low water content has a tight matrix, causing tofu to become hard so it requires a large deformation force to make tofu change its shape. One of the quality parameters of tofu is determined by the ratio of 7S and 11S protein fractions in soybeans. Corredig & Roesch (2006) reports that the gel produced by the 11S protein fraction (glycinin) is much harder than the gel produced by the 7S protein fraction (β-conglycinin). The same thing was reported by Blazek (2008) that the fraction of 11S protein (glycinin) contributes greatly to increased tofu hardness.

Table 2. Proximate value of tofu in various treatments.

Treatments	*Water, %*	*Protein, %*	*Fat, %*	*Ash, %*	*Carbohydrate, %*
Control	84.29^{a}	8.74^{b}	3.09^{a}	0.36^{c}	3.51^{b}
EPM	79.15^{b}	11.47^{a}	2.39^{a}	0.92^{a}	6.07^{a}
EDM	77.26^{b}	12.80^{a}	2.88^{a}	0.93^{a}	6.14^{a}
EDO	77.78^{b}	12.67^{a}	3.00^{a}	0.83^{b}	5.72^{ab}

Values within each column with different superscript lowercase letters (a-c) represent significant difference ($p < 0.05$).

Table 3. Tofu texture in various treatments.

Treatments	*Hardness, gf*	*Springiness*	*Cohesiveness*	*Chewiness*
Control	393.88^{b}	0.48^{b}	0.41^{a}	77.79^{b}
EPM	825.25^{a}	0.60^{ab}	0.42^{a}	195.13^{a}
EDM	899.34^{a}	0.56^{ab}	0.53^{a}	260.59^{a}
EDT	857.15^{a}	0.63^{a}	0.43^{a}	220.37^{a}

Values within each column with different superscript lowercase letters (a-b) represent significant difference ($p < 0.05$).

Springiness (level of elasticity) of tofu from the control treatment is significantly lower than of the EDT treatment but not significantly different from EPM and EDT treatment. There is no significant correlation between water content and the level of elasticity of tofu (Haqqi, 2011). Protein fraction of 7S (β-*conglycinin*) has a strong influence on the elasticity of soy protein gel, while the protein fraction of 11S (glycinin) contributes greatly to increased hardness (Blazek, 2008). Tofu cohesiveness shows the extent to which tofu changes shape before it breaks which depends on the strength of the protein matrix bond in the tofu (Mhatre et al., 2008). Tofu cohesiveness of all treatments did not show a real difference in the range of 0.41-0.53. Coagulation using $MgCl_2$ results in a compact tofu structure due to the intense interaction between soy protein and magnesium ions (Prabhakaran & Perera, 2006).

The chewiness of the tofu from the control treatment was markedly higher than other treatments. The more compact structure and the harder tofu will make the chewiness higher (Fahmi, 2010). Tofu with low water content has a denser and more compact matrix so that the amount of chew needed to chew tofu is also more than tofu with a matrix that is tenuous (tofu with high water content). Research result of Li et al. (2015) shows a higher texture for tofu that was coagulated using 2.93 g MgCl. It has 918 gf hardness, 0.86 mm springiness, 0.48 cohesiveness, and 365.9 g chewiness.

4 Conclusion

The heat extraction technique produces protein that is not extracted in the soy pulp significantly lower than in the cold extraction technique. An increase in the amount of water to soy ratio and the extraction temperature increases the amount of protein extracted in soybean extract although it is not significantly different. Cold extraction techniques are safer for workers and the waste water produced is less than the extraction techniques commonly used by small tofu industries. Coagulation using nigari produces tofu with higher protein content, magnesium content, hardness and chewing power than coagulation with whey.

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