Study of the effect of wrapping type and yeast concentration on the making of peanuts tempeh

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Abstract. In recent years, soybean production has continued to decline, while the need for soybeans is still relatively high, domestic soybean needs or 86.95% must be fulfilled from imports. Tempeh has white color, compact texture and specific flavor. White color is caused by the fungus mycelia that grow on the surface of soybean seeds. The compact texture is caused by the fungus mycelia attached on the soybean seeds. Degradation of components in soybeans can cause the formation of specific flavors after fermentation. This research aims to make tempeh by using conventional method. In order to get best result, various variables were investigated such as: type of wrapping media (banana leaf, greaseproof paper, non-perforated plastic sheet), type of raw material (soybeans and peanuts) and yeast concentration (1% and 2%). The results show that on the 0th to the 1st day the weight of the tempeh has increased, while on the 2nd day it decreased. From the organoleptic questionnaire assessed by respondents, the good quality tempeh was made of peanuts compared to soybeans. The best wrapping media for tempeh was banana leaf because it had better aeration than the other wrappers and the more yeast that use, the better the results will be. The more yeast is used, the protein content will decrease because the addition of inoculum concentration allows more numbers of mold Rhizopus sp. grow.

1. Introduction

Indonesia is the largest tempeh producer country in the world and is the largest soybean market in Asia. As much as 50% of Indonesian soybean is consumed in the form of tempeh, 40% tofu, and 10% as other products such as: tauco, soy sauce, ketchup and others. In Indonesia, the average tempeh consumption in person per year is estimated to be approximately 6.45 kg [1]. National soybean consumption has increased every year. The reason is the high public demand for soybeans as a food source of vegetable protein and an increase in public awareness of health levels. This indicates that the gap between soybean production and consumption which is quite significant influences the fulfillment of national soybean availability where domestic soybean production is only able to fulfill domestic needs of no more than 15%. Therefore, 86.95% domestic soybean needs must be fulfilled from imports [2].

Tempeh as a source of vegetable protein is generally made by traditional way. Tempeh contains various nutrients needed by human body such as protein, fat, carbohydrates, and minerals. Several studies have shown that nutrients are more easily digested, absorbed, and utilized. This is because molds which grow on soybeans hydrolyze complex compositions into simple compounds that are easily digested [3].

It is expected that modification raw materials of soybean will reduce consumption of soybeans without removing the benefits of the tempeh properties. Tempeh is made of soybeans by using the
Fermentation on solid media with the assist of *Rhizopus oryzae*. Fermentation of soybeans into tempeh is divided into three phases: fast growth phase, transition phase and decay phase. During the fermentation process, the raw materials will be incubated for three days for the purpose of sterilization.

The advantage of fermented foods, the protein, fat, and polysaccharides can be hydrolyzed so that the food has higher digestibility. Fermentation causes changes in flavor that are considered to be preferable than raw materials that are not fermented. Similarly, vitamins such as vitamin B can be increased in the amount of fermented food due to the presence of ingredients produced by microorganisms [4].

Phytic acid contained in fat has the advantage that is as an anti-nutritional compound. High levels of phytic acid in fat can bind to metals and proteins to form complexes of insoluble compounds and cause the availability of minerals and proteins for the body decreasing. The longer the fermentation time, the fungal mycelium is getting thicker due to increasing of yeast’s growth. With the growth of yeast and the thickness of the fungus mycelium, the phytase enzymes produced are increasingly shown to decrease phytic acid levels [5].

This research aims to make tempeh by using conventional method. In order to get best result, various variables were investigated such as: type of wrapping media (banana leaf, greaseproof paper, non-perforated plastic sheet), type of raw material (soybeans and peanuts) and yeast concentration (1% and 2%).

2. Materials and methods

Tempeh quality requirements that generally apply in Indonesia is based on the Indonesian National Standard (SNI 3144: 2015), as listed in the table 1.

| Test criteria                  | Unit | Requirements                                      |
|-------------------------------|------|---------------------------------------------------|
| Condition:                    |      |                                                   |
| Texture                       | -    | Compact, if sliced intact (not easy to fall)      |
| Color                         | -    | White evenly on the entire surface               |
| Aroma                         | -    | Typical smell of tempeh without smell of ammonia |
| Water content                 | %    | Max. 65                                           |
| Fat content                   | %    | Min. 7                                            |
| Protein content               | %    | Min. 15                                           |
| Crude fiber content           | %    | Max. 2.5                                          |
| Metal contamination:          |      |                                                   |
| Cadmium (Cd)                  | mg/kg| Max. 0.2                                          |
| Lead (Pb)                     | mg/kg| Max. 0.25                                         |
| Tin (Sn)                      | mg/kg| Max. 40                                           |
| Mercury (Hg)                  | mg/kg| Max. 0.03                                         |
| Arsenic contamination (As)    | mg/kg| Max. 0.25                                         |
| Microbes contamination:       |      |                                                   |
| *Coliform*                    | APM/g| Max. 10                                           |
| *Salmonella sp.*              | -    | Negative/25 g                                     |

This research was conducted in Industrial Microbiology Laboratory, Department of Chemical Engineering, Universitas Diponegoro. Operation variable of this experiment is shown in table 2. Soybeans and peanuts were used as raw materials. The base weight of beans before fermentation was 70 grams. The weight of tempeh were recorded in 0, 24 and 48 hours.
Table 2. Operation variable

| Variables   | Raw materials | Wrapping media     | Yeast |
|-------------|---------------|--------------------|-------|
| Variable 1  | Soybeans      | Banana leaf        | 0.7 gram |
| Variable 2  | Soybeans      | Banana leaf        | 1.4 gram |
| Variable 3  | Soybeans      | Non-perforated plastic | 0.7 gram |
| Variable 4  | Soybeans      | Non-perforated plastic | 1.4 gram |
| Variable 5  | Soybeans      | Greaseproof paper  | 0.7 gram |
| Variable 6  | Soybeans      | Greaseproof paper  | 1.4 gram |
| Variable 7  | Peanuts       | Banana leaf        | 0.7 gram |
| Variable 8  | Peanuts       | Banana leaf        | 1.4 gram |
| Variable 9  | Peanuts       | Non-perforated plastic | 0.7 gram |
| Variable 10 | Peanuts       | Non-perforated plastic | 1.4 gram |
| Variable 11 | Peanuts       | Greaseproof paper  | 0.7 gram |
| Variable 12 | Peanuts       | Greaseproof paper  | 1.4 gram |

This experiment was started by cleaning 420 grams of soybeans and 420 grams of peanuts from gravel and other impurities then soak for 8-10 hours and drained. The skin of soybeans and peanuts were peeled, then washed until the skin is completely gone. Soybeans and peanuts were steamed for approximately 30 minutes. The soybeans and peanuts were dried then spread to thickness (1-2 cm) to speed up drying. Soybeans and peanuts will cool down if they appear dry (no longer wet) then it can be inoculated. The soybean and peanut be weighed respectively 70 grams then 12.6 grams of yeast was added according to the variable (1% w and 2% w). Soybeans and peanuts were mixed with yeast according to variables (1% w and 2% w) until blended. The mixture was wrapped in banana leaf, non-perforated plastic and greaseproof paper and each variable was tightly attached to the wrapper with a masking tape to keep it intact and label each variable to distinguish each variable when incubated. For three days the mixture was incubated by keeping all the variables in the box. The changes were observed in tempeh every day: weight, color, aroma, texture of mycelium in tempeh. This respondent analysis test was conducted by taking samples from the three people to test fermented tempeh in terms of sight (color), aroma and texture. They will provide an assessment on a scale 1 to 5 namely:

- Scale 1 = very bad or the results of observations approaching raw materials
- Scale 2 = bad
- Scale 3 = enough
- Scale 4 = good
- Scale 5 = very good or the results of observations approaching the commercial tempeh

3. Discussion

3.1. Effect of fermentation time on the weight of tempeh produced

Data obtained in the form of tempeh weight on the n<sup>th</sup> day divided by the initial weight of tempeh during the fermentation process were grouped based on how many hours the experiment was conducted. Figure 1 shows the graph of effect of time on tempeh weight ratio.
From figure 1, it can be seen that for all variables, the weight is increasing from day 0 to day 1 and experiencing a weight loss from day 1st to 2nd day. The increase in weight on all variables on day 0 occurred because the number of soybean seeds and peanuts that are used more and more, resulting the levels of tempeh water increasing. Tempeh processing such as soaking and steaming caused the water content increased because it undergoes a process of absorption of water (hydration). Soaking and boiling will provide an opportunity to absorb water (hydration) so that the weight is doubled [8]. On the day 0 to day 1 experiment, the tempeh fermentation was kept in a closed place in humid conditions. The optimum tempeh storage condition is at low humidity (0%) [9]. If the tempeh fermentation storage is in humid conditions, the production of water content will be formed again and also cause contaminants that produce byproducts thereby increasing the mass of the tempeh. Because it is stored in a humid place, the water contained in the tempeh does not evaporate so that the tempeh weight increases.

The decrease in tempeh on day 1 to day 2 is due to protein degradation. Fungi that grow on tempeh (Rhizopus sp.) produce enzymes that break down complex compounds. During the fermentation process there are a number of proteins used by the fungus Rhizopus oligosporus as a source of nitrogen for its growth [10]. Rhizopus oligosporus is a proteolytic fungus and important in the breakdown of proteins into simpler compounds. This fungus will degrade proteins during fermentation into dipeptides and so on into NH₃ or NH₂ compounds that are lost through evaporation. The longer fermentation means the longer the chance of fungus to degrade protein, so that even more degraded protein [11]. As a result of more protein being degraded, more compounds will be lost through evaporation, so the weight of tempeh will decrease further.

3.2. Effect of differences in raw materials on the quality of tempeh that produced

The effect of differences in raw materials on the quality of tempeh was compared between variable 6 to variable 8. The visualization of tempeh can be seen in figure 2.
Figure 2. The visualization of a) peanuts tempeh and b) soybeans tempeh

Based on figure 2, it can be seen that the physical quality of tempeh with raw material of peanuts ingredients is better than tempeh made of soybeans. The quality of tempeh on variable 6 with raw material of peanut has a more compact texture than the texture of tempeh on variable 8 with raw material of soybeans. In variable 6 the compactness of the texture can be seen from the composition of the white mycelium, and tightly attaches the beans together. Whereas in variable 8 the compactness of the texture is marked from the mycelium arrangement only occurs at the edges.

From the respondent analysis test, the obtained results are in the form of an assessment of the color, aroma test and texture of mycelium in tempeh on scale 1 to 5, as shown on figure 3.

Figure 3. Graph of differences in raw materials against sensory tests on tempeh

Table 3. Average sensory test values in tempeh

| Variable | Color | Mycelium texture | Aroma |
|----------|-------|------------------|-------|
| 6        | 3.2   | 3.3              | 3.1   |
| 8        | 1     | 1.4              | 1     |

From figure 2 and 3, it can be seen that variable 6 has a scale of 3.2 on the color assessment, which is already close to the color of the finished tempeh, the texture of mycelium on a scale of 3.3 which has covered the entire raw material and white due to mycelium but there is a black spot on the tip of the tempeh and the aroma on the scale of 3.1 is a slightly sharp smell of tempeh. The variable 8 has a scale
of 1 on the color assessment that is still the color of the raw material, white color is not visible, the texture of the mycelium scale 1.4 is the mycelium structure that is seen at the end and the shape is not compact and the scent on scale 1 is still smelled of peanut.

Good tempeh is a compact tempeh, the whole tempeh’s surface is covered with white mold mycelium, not stained with black due to spores, not slimy, easily sliced, not rotten and does not smell ammonia [11][12]. The new yeast will have the opportunity to produce good tempeh which will influence the formation of the flavor and aroma of the resulting tempeh [13]. The materials contained in peanuts are 40–48% oil, 25-30% fat, 25% protein, and 18% carbohydrates and vitamin B complex [14] and the materials contained in soybeans are 40.5% protein, 20.5% fat, 22.2% carbohydrate, 4.3% crude fiber, 4.5% ash, and 6.6% water [15]. The fat content in raw materials is used by mold tempeh as a source of nutrition. Rhizopus oligosporus and Rhizopus oryzae produce lipase enzymes that will convert fat to triglycerides and free fatty acids during fermentation [16]. Mold uses these free fatty acids as a carbon source [17].

3.3. Effect of difference wrapping on quality of tempeh produced

For the effect of different packaging on the quality of tempeh that produced comparing variable 1 with variable 9. The visualization of product tempeh can be seen in figure 4.

![Figure 4](image-url)

**Figure 4.** The visualization of a) tempeh with non-perforated plastic wrapper and b) tempeh with banana leaf wrapper

Based on figure 4, it can be seen that the physical quality of tempeh with a banana leaf wrapper is better than a non-perforated plastic wrapper. The quality of tempeh in variable 9 with the raw material of peanut wrapped in banana leaf has a more compact texture than the texture of tempeh in variable 1 with raw material wrapped in peanuts which is non-perforated. In variable 1 there is no cohesiveness of the texture marked by the absence of white color because of the composition of the mycelium in the texture of tempeh. Whereas in variable 9 the compactness of the texture can be seen from the composition of the white mycelium, and tightly attaches the beans together.

From the respondent analysis test, the results were obtained in the form of an assessment of the color, aroma and texture test of mycelium on tempeh on a scale 1 to 5, as shown in figure 5. From figure 4 and figure 5, it can be seen that variable 1 has a scale of 1.6 on the color assessment that is still the color of raw materials, the texture of mycelium on a scale of 1.9 that is the structure of the mycelium has not been seen and the shape has not been compactly and aroma on a scale of 1.6 that is still smelled of peanut. On variable 9 it has a scale of 2.7 on the color assessment that is close to the color of tempeh, scale 3.1 on the structure of mycelium that is the entire surface of tempeh has been covered by mycelium and aroma on a scale of 2.9 that is close to the smell of tempeh in general.
Plastic bags are airtight, so the surface of the plastic bag must be perforated small by using a stick so that aeration can occur [18]. It is better if in the process of fermentation there is a lot of oxygen, it can cause metabolic processes too fast so that the temperature rises and mold growth is inhibited [19].

3.4. Effect of yeast addition differences on the quality of tempeh that produced
For the effect of difference in the addition of yeast on the quality of tempeh produced comparing to variable 1 with variable 2. The visualization of product tempeh can be seen in figure 6. Based on figure 6, it can be seen that the physical quality of tempeh with 1% w (0.7 g) yeast is better than the 2% w (1.4 g) yeast in peanuts wrapped with non-perforated plastic. Tempeh’s quality in variable 1 with 0.7 grams yeast did not have cohesiveness of the texture which is marked by the absence of white color due to the composition of mycelium in the texture of the tempeh. Whereas in variable 2 with 1.4 grams of yeast the mycelium structure is present although it is still very thin.

**Figure 5.** Graph of differences in wrapping of sensory tests on tempeh

**Table 4.** Average sensory test values on tempeh

| Variable | Colour | Mycelium Texture | Aroma |
|----------|--------|------------------|-------|
| 1        | 1.6    | 1.9              | 1.6   |
| 9        | 2.7    | 3.1              | 2.9   |

**Figure 6.** The visualization of a) tempeh with yeast of 1% w (0.7 gr) and b) tempeh with yeast of 2% w (1.4 gr)
From the respondent analysis test, the results were obtained in the form of an assessment of the color, aroma and texture test of mycelium on tempeh with the scale 1 to 5, as shown on figure 7.

![Graph of differences in the addition of yeast to the sensory tests in tempeh](image)

Figure 7. Graph of differences in the addition of yeast to the sensory tests in tempeh.

| Variable | Colour | Mycelium texture | Aroma |
|----------|--------|------------------|-------|
| 1        | 1.6    | 1.9              | 1.6   |
| 2        | 1.9    | 2                | 1.7   |

From the figure 6 and figure 7, it can be seen that variable 1 has a scale of 1.6 on the color value of the material’s color, the texture of the mycelium on a scale of 1.9 that is the structure of the mycelium has not been seen and the shape is not yet complex and the scent on a scale of 1.6, still smells of peanut. The variable 2 has a scale of 1.9 on the color of the raw material color, scale 2 of the mycelium structure, namely the tempeh’s surface is still slightly covered by mycelium and the form is not compact and the aroma on a scale of 1.7 that still smells of peanut.

Differences in the concentration of yeast giving variations to the decrease in tempeh protein levels. This is because it is related to the concentration of inoculum allows a greater number of molds *Rhizopus* sp. which grows and after the fermentation process the total content of amino acids will decrease due to *Rhizopus* sp. use amino acids as a source of N (nitrogen) for its growth while tempeh’s protein is expressed as N-total. So, the more yeast is needed, the protein’s content will decrease [20].

4. Conclusion
1. Tempeh can be made from raw soybeans or peanuts. The longer the fermentation time, the smaller the weight of tempeh produced.
2. Tempeh from peanuts has better color, texture, and aroma assessment of tempeh than soybeans.
3. Peanut tempeh with banana leaf wrappers has a better quality and the worst quality in peanut tempeh with a plastic wrap that is non-perforated.
4. The effect of adding 2% w of yeast has a color, texture of mycelium and aroma that is better than tempeh that uses 1% w of yeast.

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