Proximate and Amino Acids Composition of Powdered Over Fermented *Mlanding* Tempeh

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**Abstract.** Over fermented *Mlanding* tempeh is widely used by people in southern Java, Indonesia as main ingredient and seasoning due its umami taste. However, this kind of tempeh is very perishable. Alternatively, drying was conducted to prolong the shelf life of the over fermented *Mlanding* tempeh. This research aimed to investigate drying temperature effect on over fermented *Mlanding* tempeh powder proximate and amino acids composition. *Mlanding* tempeh was over fermented for 60 hours, dried at three different temperature (55, 60, 65°C), powdered and analyzed. The result shows drying temperature affect moisture content of the powder but not affect ash, lipid, total N, and dissolved protein content. Drying at 60 and 65°C produce powder with moisture content below 12%. The main amino acids in powdered over fermented *Mlanding* tempeh are glutamic and aspartic acid which are the highest in powder dried at 60°C.

1. Introduction

*Mlanding* tempeh, also called as lamtoro tempeh, is tempeh that made from ripe seed of Leucaena leucochepala. ‘Mlanding’ refers to Leucaena leucochepala in Javanese language [1] whether ‘lamtoro’ refers the same thing in Bahasa Indonesia. This kind of tempeh could be found easily in southern Java [2], especially along Sewu mountains; including Gunungkidul (Yogyakarta), Wonogiri (Central Java), Pacitan and Trenggalek (East Java) [1]. *Mlanding* tempeh processing uses usar as inoculum [3] which contains diverse types of molds, such as Rhizopus sp., and other microorganism [4].

*Mlanding* tempeh processing is similar to soybean tempeh processing. It needs 36 hours fermentation to gain acceptable fresh *Mlanding* tempeh. Prolonged fermentation, usually 1-5 days, will shift the fresh *Mlanding* tempeh into over fermented *Mlanding* tempeh that usually consumed as main ingredient and also seasoning in Javanese culinary [5]. As seasoning, over fermented *Mlanding* tempeh is more preferable than the soybean one due to its higher umami intensity. However, the characteristics of over fermented *Mlanding* tempeh tend to change rapidly toward spoilage due to the microbiology activity in the tempeh.

In order to maintain the over fermented *Mlanding* tempeh quality with desirable characteristic for seasoning, a drying operation with appropriate temperature is needed. Many literatures had shown that temperature affect chemical and sensory characteristic of food product. Vegetables and spices usually dried at 50-70°C. In this research, *Mlanding* tempeh was prolonged fermented up to 5 days and then a sensory analysis was conduct to choose fermentation time that produce the most preferable tempeh. The over fermented *Mlanding* tempeh was then dried at three level temperature, powdered and chemically...
analysed. The aim of this research was to investigate the effect of drying temperature on proximate and amino acids composition of powdered over fermented Mlanding tempeh.

2. Research Method

2.1. Materials and Tools

Some text. Pre-fermented Mlanding tempeh was supplied by a local Mlanding tempeh producer located at Wonogiri region, Central Java. All chemical being used for proximate and amino acids analysis was pro analysis grade. Incubator was used to ferment Mlanding tempeh whether cabinet dryer, dry mill blender and sieving device were used to powder the over fermented tempeh. UPLC Waters H Class with Photodiode Array Detector, Spectrophotometer Shimadzu UV Vis 1240 and glassware were used to analyse Mlanding tempeh powder chemical characteristic.

2.2. Determination of Over Fermented Mlanding Tempeh Fermentation Time

Procedure for preparing Mlanding tempeh followed the previous research method [3]. Pre-fermented Mlanding tempeh (100 g) was inoculated using traditional inoculum (5 g) and then incubated at 30±2°C. The fermentation was conducted for 42 hours, 48 hours, 54 hours, 60 hours, 66 hours and 72 hours to produce over fermented Mlanding tempeh. Over fermented Mlanding tempeh fermentation time was determined using sensory analysis which involved 52 panels to score their preference. Over fermented Mlanding tempeh was crushed using mortar to produce paste sample that served to panels. Fermentation time of Mlanding tempeh that produce most preferred over fermented Mlanding tempeh paste was chosen as standard for over fermenting the tempeh.

2.3. Drying and Powdering of Over Fermented Mlanding Tempeh

Over fermented Mlanding tempeh was dried using cabinet dryer for eight hours at 55°C, 60°C, and 65°C. Further, the powder obtained was proximate analysed using AOAC methods [6] except dissolved protein [7] and amino acids analysed using UPLC [8,9]. Proximate data obtained was analysed using One Way Analysis of Variance which was further analysed using Duncan’s Multiple Range Test at α = 0.05 if there was a significant different between samples. SPSS 16.0 program was used for running the proximate data processing.

3. Results and Discussion

3.1. Over Fermented Mlanding Tempeh Fermentation Time Determination

Fermentation time is a crucial factor which affect fermented food product quality, including for Mlanding tempeh. Fermentation time that produce Mlanding tempeh with appropriate sensory quality and also safe phytic acids and mimosine level is 36 hours [3]. Further fermentation will cause the Mlanding tempeh being over fermented that undergo sensory characteristics change. In this research, fermentation was conduct for 36 hours, 60 hours, 84 hours, 108 hours, and 132 hours. Based on panels opinion, fermentation above 132 hours caused the over fermented Mlanding tempeh became unacceptable for consumption.

| Fermentation Time (hours) | Color  | Aroma  | Taste  | Overall |
|---------------------------|--------|--------|--------|---------|
| 36                        | 4.84b  | 4.06ab | 3.94a  | 4.08a   |
| 60                        | 4.94b  | 4.60b  | 4.60b  | 4.77b   |
| 84                        | 4.38ab | 4.40b  | 4.21ab | 4.44ab  |
| 108                       | 4.46b  | 4.12ab | 3.75a  | 4.06a   |
| 132                       | 3.85a  | 3.65a  | 3.85a  | 3.96a   |

Different letter in the same column shows a significant difference at α = 0.05.

Score: 1= very dislike; 2=dislike; 3=slightly dislike; 4=neither like nor dislike; 5=slightly like; 6=like; 7=very like
Table 1 shows fermentation time did influence panels preference on the over fermented Mlanding tempeh. Highest preference score was gained by Mlanding tempeh that fermented for 60 hours and 84 hours. However, 60 was more efficient due its shorter fermentation time. Therefore, standard time for producing over fermented Mlanding tempeh in this research was 60 hours. Based on fermentation time, this kind of over fermented Mlanding tempeh is categorized as ‘tempe semangit’ (slightly over ripe or over fermented tempeh) [2]. The appearance of fresh and most preferred over fermented Mlanding tempeh is shown in Figure 1.

![Mlanding tempeh](image)

(a) Fresh Mlanding tempeh  (b) Most preferred over fermented Mlanding tempeh

**Figure 1. Mlanding tempeh**

3.2. Proximate Composition

Drying temperature affects the moisture content of the over fermented Mlanding tempeh powder, but does not affect ash, lipid, total N, and dissolved protein (Table 2). Higher drying temperature decreased the moisture content of the powder. Over fermented Mlanding tempeh dried at 60°C and 65°C have met the moisture standar for seasoning flour. The Moisture content of over fermented Mlanding tempeh in this research is higher than moisture content of over fermented soybean tempeh which being fermented for 120 hours [10].

| Drying Temperature | Moisture (%wb) | Ash (%db) | Lipid (%db) | Total N (%db) | Dissolved Protein (%db) |
|--------------------|----------------|-----------|-------------|---------------|-----------------------|
| 55°C               | 12.45±0.707    | 3.69±0.349| 9.93±0.561  | 41.23±1.978   | 0.441±0.243           |
| 60°C               | 9.49±0.347     | 3.96±0.479| 10.69±1.433 | 41.30±2.815   | 0.621±0.064           |
| 65°C               | 9.52±0.568     | 4.13±0.359| 10.17±1.889 | 42.35±3.250   | 0.632±0.125           |

Different letter in the same row shows a significant difference at α = 0.05.

3.3. Amino Acids Composition

Amino acids and peptides have contribution to the fermented food taste[11]. Table 3 shows glutamic acid and aspartic acid are the main compound of the amino acids of the over fermented Mlanding tempeh powder. This is similar with amino acids profile of the over fermented soybean tempeh[5,12]. Both amino acids contribute to the umami taste of food products [13]. Drying at 60°C resulted higher proportion umami amino acid in over fermented Mlanding tempeh.

4. Conclusion

Moisture content of the over fermented Mlanding tempeh powder is affected by drying temperature whether ash, lipid, total N, and dissolved protein content are not affected. Moisture content below 12%
is reached by drying at 60 and 65°C. Highest glutamic and aspartic acids is found in the powder dried at 60°C. A further study on the development of instant seasoning based on over fermented *Mlanding tempeh* is challenging and need to be executed.

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