



Formulation and organoleptic test of edamame-mackerel tuna sprinkle powder as a high protein supplementation for stunting

Formulasi dan uji organoleptik bubuk tabur (sprinkle powder) edamame-tongkol sebagai suplementasi tinggi protein untuk stunting

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Abstract

Stunting can also reduce the quality of human resources. Developing regional foods with high protein content, such as edamame and mackerel tuna, can help reduce the prevalence of stunting. Edamame and mackerel tuna were processed into a sprinkle powder. This study aimed to determine the best formulation of sprinkle powder for use as a high-protein supplement. This research was conducted at the University of Jember from January to April 2024 using a completely randomized design at three levels. The experiments were carried out on 25 semi-trained panelists, and proximate tests were analyzed in the laboratory. The results were analyzed using SPSS v24. The results of the statistical analysis of the hedonic test showed that there were no differences in all parameters (color, aroma, taste, and overall) ($p > 0,05$). The results of the proximate test showed that F3 had the highest water content ($9,79 \pm 0,1\%$), ash ($9,21 \pm 0,09\%$), and protein ($43,18 \pm 0,14\%$), while the highest fat and carbohydrate contents were possessed by F1, $13,21 \pm 0,05\%$ and $31,39 \pm 0,1\%$, respectively). In conclusion, the most preferred formulation was F3, and proximate analysis also showed that the highest protein content was found in F3.

Keywords: Edamame flour, fish protein hydrolysate, sprinkle powder, mackerel tuna, stunting

Abstrak

Salah satu upaya dalam menurunkan prevalensi *stunting* adalah pengembangan produk pangan lokal tinggi protein, seperti edamame dan ikan tongkol. Edamame dan ikan tongkol diolah menjadi bubuk tabur. Penelitian ini bertujuan untuk mengetahui formulasi terbaik bubuk tabur edamame tongkol (tameko) sebagai suplementasi tinggi protein. Penelitian ini dilakukan di Universitas Jember pada Januari - April 2024 dengan menggunakan Rancangan Acak Lengkap faktor tunggal tiga taraf. Uji organoleptik dilakukan pada 25 panelis semi terlatih dan uji proksimat dilakukan di laboratorium. Hasil uji dianalisis menggunakan SPSS versi 24. Hasil analisis statistik uji hedonik menunjukkan tidak terdapat perbedaan yang signifikan pada semua parameter (warna, aroma, rasa, dan keseluruhan) dengan $p > 0,05$. Hasil uji proksimat menunjukkan bahwa F3 memiliki kadar air ($9,79 \pm 0,1\%$), abu ($9,21 \pm 0,09\%$), dan protein ($43,18 \pm 0,14\%$) tertinggi, sedangkan kadar lemak dan karbohidrat tertinggi dimiliki oleh F1, masing-masing $13,21 \pm 0,05\%$ dan $31,39 \pm 0,1\%$. Kesimpulan, formulasi yang paling disukai adalah F3 dan hasil uji proksimat juga menunjukkan bahwa kadar protein tertinggi terdapat pada F3.

Kata Kunci: Bubuk tabur, hidrolisat protein ikan, *stunting*, tepung edamame, tongkol

Introduction

Nutritional challenges persist throughout life. Poor nutrition often starts in utero and extends

to adolescent and adult life, particularly for girls and women. It also spans several generations. Undernutrition experienced during childhood,

adolescence, and pregnancy has a cumulative negative effect on the birth weight infants (Marshall et al., 2022). Low Birth Weight (LBW) is defined as a birth weight of less than 2,500 grams and is a global public health issue associated with both short-term and long-term complications (WHO, 2024).

Infants with LBW experience intrauterine growth retardation (IUGR), while women are born malnourished and face a significantly increased risk of mortality during the neonatal period or in later stages of infancy. If they survive, they are unlikely to significantly catch up on this lost growth and are more likely to experience a variety of developmental deficits. Therefore, infants with low birth weight are at a higher risk of being underweight or experiencing stunting during early development. The repercussions of undernourishment persist into adulthood. Epidemiological studies from both developing and industrialized nations increasingly indicate a correlation between fetal undernutrition and a heightened risk of various chronic diseases in adulthood (Bianchi & Restrepo, 2022).

In the Asian region, the prevalence of LBW is reported at 17,3%, whereas in Southeast Asia, it varies between 7% and 21% (WHO, 2014). According to the World Health Assembly Resolution, the World Health Organization (WH) has set a target to reduce the incidence of LBW by 30% by 2025, acknowledging that an estimated 15-20% of the 20 million births worldwide each year involve LBW. The risk of LBW is higher in developing countries, including Indonesia (Mediani, 2020).

Birth weight is one of the most critical determinants of infant survival, growth, and future development. Risk factors associated with the incidence of LBW include the family's socioeconomic characteristics and the mother's health history. Socioeconomic factors are related to the mother's education, type of residence, family size, and economic status (Utami & Wulandari, 2020). LBW is a birth outcome indicator with a long-term impact and is a significant risk factor associated with maternal and infant mortality. LBW is more prevalent among infants born to young mothers under the age of 20 (Falcão et al., 2020; Trerotoli et al., 2020) Pregnancy in mothers under the age of 20 carries a higher risk of complications such as preeclampsia (high blood pressure during pregnancy), eclampsia (seizures during pregnancy), and infections. These conditions

significantly increase the likelihood of preterm birth and delivering a LBW baby (WHO, 2023).

This situation is also linked to rising rates of early marriage. The impact of early marriage includes adolescent pregnancies, which are high-risk owing to the physical and emotional immaturity of young mothers. The risks for pregnant adolescents include miscarriage, pregnancy-related anemia, bleeding, inadequate nutrition, and maternal mortality. For the infants, risks include preterm birth, LBW, congenital abnormalities, infections, and infant mortality (Letari & Fitrianti, 2017).

Food security is a condition where sufficient food is available for every person at all times, and each individual has access to it, both physically and economically (BKP, 2021). Food insecurity, the major predictor of LBW associated with poor-quality dietary intake and decreased nutritional status among women, and poor nutritional status in pregnancy and pre-pregnancy, is linked to poor birth outcomes (Desalegn et al., 2021).

The prevalence of LBW in Indonesia increased from 11,37% in 2020 to 12,72% in 2021 (MoH, 2021). West Java is among the provinces with a rising trend in LBW cases, with rates increasing from 6,3% in 2018 to 11,96% in 2021 (Dinas Kesehatan Jawa Barat, 2018, 2021). West Java has a population exceeding 48 million, and its high birth rate significantly contributes to the incidence of LBW among infants. Various factors, including limited access to quality healthcare, insufficient awareness regarding prenatal nutrition, and socioeconomic challenges, further exacerbate this issue. The rising prevalence of LBW presents substantial risks for long-term health outcomes, including stunting and chronic diseases (Arsyi, 2021). Consequently, West Java has been prioritized in accelerated stunting reduction programs in Indonesia.

Disparities between regions in West Java at the district/city level require specific interventions, particularly in areas with a high prevalence of LBW. Ecological study is a type of research design that focuses on population characteristics. This study incorporated spatial mapping to examine the prevalence of LBW and its related factors across different regions in West Java. This mapping technique helps identify geographic patterns and essential for health policy markers to increase attention to low birth weight causes (Rahman et al., 2023).

Although numerous studies have explored LBW at the individual level, research at the macro level in Indonesia remains limited. Most existing studies have predominantly focused on personal and household factors, creating a gap in understanding the broader systemic determinants of LBW (Tarigan et al., 2023). This research is important because it highlights the need for more comprehensive investigations that consider larger-scale influences on LBW. Expanding the research to include macro-level factors could provide a more holistic understanding of LBW in the Indonesian context. This study aimed to determine the prevalence of LBW in West Java and identify risk factors associated with LBW. These findings will inform the design of targeted interventions to prevent perinatal issues related to LBW, thereby improving maternal and child health outcomes in the region.

Methods

This study used a single-factor Completely Randomized Design (CRD) with three sample treatments, namely, a comparison of edamame flour formulation with tuna fish protein hydrolysate (FPH). The RAL was used because the experimental unit was considered homogeneous. Each treatment was repeated 3 times. The formulation of edamame flour with HPI cobs is as follows:

Table 1. Tameko powder formulation

Formulation	Edamame Flour (%)	FPH Mackarel Tuna (%)
F1	70	30
F2	60	40
F3	50	50

This research was conducted in five stages: making edamame flour, making FPH cob powder, making edamame cob powder (Tameko), and proximate testing at the Agricultural Biochemical Chemistry Laboratory and Agricultural Product Engineering Laboratory, Faculty of Agricultural Technology, University of Jember. The gravimetric method from AOAC 2005 was used for water and ash content tests. The fat content test was performed using the Soxhlet method from the AOAC 2005.

The protein content test used the Kjeldahl method from BSN 1992, and the carbohydrate test used the difference method by Winarno (2008). The acceptance criteria for water and ash content were the INS 1995 thresholds. The fifth stage was an organoleptic test conducted at Jember University, Bondowoso Campus, involving 25 semi-panelists. trained. This study was conducted between January and April 2024.

Tools and materials

The tools used included a freeze dryer, digital analytical balance, Philips brand mixer, food processor, Philips brand blender, 80 mesh Tyler sieve, centrifuge, pan, stove, knife, measuring cup, baking sheet, cake oven, bowl and spoon. At the proximate analysis stage, various tools were used, including glassware, Soxhlet extraction tools, Kjeldahl flasks, burettes, ovens, ashing furnaces, desiccators, porcelain cups, iron spatulas, measuring pipettes (Pyrex), Erlenmeyer flasks, glass funnels, and dropper pipettes.

The materials used included fresh tuna from fishermen in Puger District, Jember, East Java, Indonesia and edamame soybeans obtained from Mitra Tani 27. The chemicals used were distilled water, petroleum benzene (p. a.), papain enzyme, HCl, selenium, NaOH, H₂SO₄, H₃BO₃, mmb indicator (methyl red methyl blue), aluminum foil, and filter paper.

Making Edamame Flour

Edamame flour was prepared as described by Siregar et al. (2023), with modifications. Edamame soybeans were soaked for 8 h until the epidermis could be easily removed. The mixture was boiled for 20 min and dried in the sun for 4 h. Further drying was performed in an oven at 60 °C for 24 h. Edamame that had been completely dried was then ground into a powder and sifted using an 80 mesh Tyler sieve to obtain edamame flour (Siregar et al., 2023).

Making FPH Cob Powder

A total of 50 g of tuna meat was added with distilled water (2:1) and crushed using a food processor. Next, 5% papain enzyme was added to tuna meat. Hydrolysis was carried out in a water bath for 3 h at 55°C. After 3 h, the enzyme was inactivated by incubation at 85°C for 20 min. The mixture was centrifuged for 30 min at 10°C at a speed of 3500 rpm. The results of

centrifugation were the supernatant and the residue. The supernatant was dried using a freeze-dryer. The final result is a dry form of tuna fish protein hydrolyzate (Witono et al., 2020).

Making Cob Edamame Powder (Tameko)

Edamame flour was mixed with the FPH cob until it was homogeneous with the formulation according to the treatment. In the manufacturing process, 1% garlic powder, 1% salt, and 25% powdered sugar were added to Tameko powder.

Data Analysis

The data were analyzed using SPSS version 24.0. The organoleptic test data seen in this research are a hedonic test that includes the panelists' level of liking for color, aroma, taste, and the overall product. The assessment scale for this hedonic test ranges from 1 to 7 with details of 1 very dislike, 2 dislike, 3 somewhat dislike, 4 neutral, 5 somewhat like, 6 like, and 7 very like. The proximate tests in this study included ash, water, protein, fat, fat, and carbohydrate contents. Next, the hedonic and proximate tests were analyzed using one-way ANOVA. Data were considered to have a significant difference (p -value $< 0,05$).

Result and Discussion

Tameko Powder Hedonic Test Results

The results of statistical analysis of the hedonic test showed that there were no significant differences in all test parameters, including

color, aroma, taste, or overall between F1, F2, and F3 (p -value $> 0,05$). Although it did not show a significant difference based on aroma, taste, and overall parameters, F3 was the formula most preferred by the panelists. Formula 3 (50% Edamame Flour : 50% FPH Mackarel Tuna) has a more neutral aroma because the fishy smell of the tuna fish protein hydrolyzate is neutralized by the aroma of the edamame. This is similar to taste parameters.

Based on taste parameters, the formula most preferred by the panelists was F3. If the concentration of edamame flour used is high, the taste of edamame flour will also be more dominant. Likewise with FPH cobs. If the FPH concentration of the cob is higher, the savory or umami taste will be more dominant. In the F3 formulation, there was an equal ratio of edamame flour to FPH cob so that the unpleasant aroma and taste of edamame flour was not very prominent, and the fishy aroma of FPH cob was slightly reduced. In the F3 formula, the dominant taste was umami or savory, with a slightly fishy aroma. The combination of all the raw materials in making loose powder produced a slightly greenish-white color for all treatments, causing the panelists to be unable to differentiate the color of Tameko loose powder for all treatments. Overall, the Tameko powder product most liked by panelists was F3. Formula F3 was the selected formula based on the hedonic test because the aroma, taste and overall product was the formula most liked by the panelists.

Table 2. Tameko powder hedonic test results

Parameter	Average \pm SD			p-value
	F1	F2	F3	
Color	4,92 \pm 1,22	4,92 \pm 1,11	4,92 \pm 1,51	1,000
Aroma	3,4 \pm 1,19	3,52 \pm 1,45	3,88 \pm 1,48	0,64
Flavor	4,12 \pm 1,27	4,16 \pm 1,49	4,48 \pm 1,64	0,444
Overall	4,4 \pm 1,08	4,44 \pm 1,08	4,76 \pm 1,45	0,522

In terms of color parameters, formulas F1, F2, and F3 had the same assessment by the panelists. Based on the organoleptic tests of color parameters, no significant differences were found. This is because the F1, F2, and F3 formulations had almost the same color, namely, predominantly greenish yellow. The more cob FPH is given, the darker the color becomes, making it difficult for the panelists to distinguish the color of each formulation.

The FPH cobs have a greenish-yellow color. When FPH cobs and edamame flour were mixed, the dominant color was greenish-yellow. The greater the proportion of cob FPH, the darker was the tameko powder produced. The dark color produced by the FPH powder can occur because the manufacturing process uses an enzymatic hydrolysis method. This dark color originates from the auto-oxidation of Hb and the conversion of Fe²⁺ to Fe³⁺ to form metHb. Fish

contains byproducts that contain large amounts of blood. The blood contains hemoglobin (Hb) and iron as pro-oxidants. These components can react with each other during enzymatic reactions (Nikoo et al., 2023). The brownish color of the FPH tuna is caused by the red pigment in the flesh of the fish. The higher the pigment contained in red meat, the higher the browning intensity on the FPH (Parvathy et al., 2018). On the other hand, if the proportion of edamame flour is greater, the color will be brighter. The green color in edamame flour comes from the chlorophyll pigment content, but due to the heating process during flour production, the chlorophyll concentration can decrease and eventually fade to whitish (Kurniawan et al., 2020).

In terms of aroma parameters, the formula most preferred by the panelists was F3. F3 provided a more savory aroma than the other formulas. F3 has the highest proportion of cob FPH among the other treatments, so F3 has the most savory aroma. The more FPH added, the more delicious the aroma. Fish protein hydrolysate is the final product of the partial hydrolysis of proteins containing a mixture of various amino acids, oligopeptides, and peptides. The shorter the peptide chain produced by the hydrolysate, the more savory the aroma it produces. Various natural ingredients that have high protein content can produce a delicious taste and aroma (Ghassani & Agustini, 2022). 100 grams of tuna contains 22 grams of protein (Hidayat et al., 2020). The savory aroma of FPH cobs comes from amino acids, such as glutamic acid, alanine and glycine (Oko et al., 2019; Wang et al., 2020). Apart from its savory aroma, tameko powder also has a pleasant aroma. However, in F3, savory aroma was more dominant than unpleasant aroma. This delicious aroma originates from edamame flour. There is a pleasant smell or what is usually called beany flavor which is the result of the oxidation of linoleic acid by the enzyme lipoxygenase (Yang et al., 2023).

In terms of taste parameters, the formula that panelists liked the most was F3. F3 (FPH cob 50%: edamame flour 50%) is a tameko powder formulation with the tastiest taste among all formulas. The higher the FPH of the cobs, the stronger the savory taste. The savory taste originates from the amino acids contained in the FPH cob. Of the 17 types of amino acids in FPH cobs, glutamate is the highest amino acid with an amount of 6,38%. The glutamate content in FPH cobs can be used as an alternative natural food flavoring (Ramandhani et al., 2022). Apart from that, edamame flour also has a little glutamic acid so it can add a savory taste to tameko powder (Barikah et al., 2021). The savory taste can be attributed to various natural ingredients that are high in protein. Protein hydrolysis can be performed using various methods, one of which is enzymatic. One of the advantages of using this enzymatic method is that it produces a specific amino sequence. In addition, the enzymatic method does not damage amino acids because it can be performed under conditions that are not extreme and produce short peptide chains with a delicious taste. The resulting protein hydrolyzate using the enzymatic method has the potential to be used as a flavoring ingredient because of its savory taste (Restiani, 2017; Wicaksono & Winarti, 2021).

Overall, the Tameko loose powder product most preferred by the panelists was F3. F3 has the color, aroma, and taste that are most accepted by the panelists. Overall, F3 had a greenish-yellow product with the most delicious aroma and taste. The strong savory taste of Tameko powder makes it a natural flavoring agent that can be added to various foods.

Tameko Powder Proximate Test Results

The proximate tests on tameko powder carried out in this study included ash, water, protein, fat, and carbohydrate contents. The results of the statistical analysis showed significant differences in all parameters.

Table 3. Tameko powder proximate test results

Proximate	Amount (%)			p-value
	F1	F2	F3	
Water	9,36±0,01	9,51±0,1	9,79±0,1	0,002*
Ash	7,01±0,14	7,60±0,09	9,21±0,09	<0,001*
Fat	13,21±0,05	11,37±0,07	9,50±0,06	<0,001*
Proteins	39,03±0,44	41,15±0,11	43,18±0,14	<0,001*
Carbohydrate	31,39±0,1	30,37±0,38	28,32±0,34	<0,001*

*p-value <0,05 indicates there is a significant difference

Based on the proximate test results, the highest levels of ash, water, and protein were found in F3, with the largest proportion of cob FPH, while the highest fat and carbohydrate contents were found in F1, with the largest proportion of edamame flour. The higher the proportion of cob FPH, the higher the water, ash, and protein contents. Meanwhile, if the proportion of edamame flour is given, the fat and carbohydrate contents of the tameko powder are higher.

The highest water content in tameko powder was found in F3 (9,79 %). The greater the proportion of FPH cobs used, the higher is the water content of the tameko powder. Siregar et al.'s research. (2023) stated that the FPH cob has a higher water content than the edamame flour. The water content in FPH cob reaches 12,95%, while in edamame flour it is only 9,35% (Parvathy et al., 2018; Siregar et al., 2023). This shows that the high water content in the tameko powder was caused by the high HPI of the added cob. Even though the water content in F3 reached 9,79%, this figure was still below the requirements for flavoring quality. Based on Indonesian National Standards (INS), the water content in flavorings is a maximum of 12%.

The water content of a food ingredient can affect the shelf life of the product, thereby causing changes in its chemical characteristics. Apart from that, water content can also affect the texture, appearance and taste of a food ingredient (Ghassani & Agustini, 2022). Food products can be categorized as safe if they have a water content of <14% so that they can prevent damage due to microbes (Agustina et al., 2024)

The ash content of the tameko powder increased with increasing FPH content of the added cobs. The ash content in F1 was the lowest of all treatments at 7,01%, whereas the highest ash content was found in F3 (9,21 %). Based on INS, the quality standard for ash content is a maximum of 7% for flavoring, whereas all tameko powder treatments have an ash content of >7%; therefore, this tameko powder still does not meet the INS standard quality. The ash content of food ingredients can be influenced by various factors, including the method used, the ashing temperature and time, and the type of material being ashed. If you use a higher temperature for a longer time, the amount of water that evaporates will also increase.

In this study, the highest fat contents were observed in F1 (13,21 %) and F3 (9,5 %). This shows that the lower the edamame flour content, the lower is the fat content. The fat content of tameko powder is largely influenced by edamame flour, because edamame flour has a higher fat content than cob FPH. Based on previous research, powdered cob FPH has a fat content of 4% (Parvathy et al., 2018), while the fat content in edamame flour is 18,65% (Siregar et al., 2023). The fat content of food can be influenced by various factors including the processing method used. If processing uses heating or drying principles, it can cause the fat content to decrease because some of the fat will melt out (Ghassani & Agustini, 2022).

Fat is the source of energy. Each gram of fat provided 9 kcal. Fat is the largest source of energy reserves in the body and can be stored in the subcutaneous tissue. Toddlers with low fat intake have a higher risk of becoming stunted than toddlers with sufficient fat intake. Stunting is an important nutritional problem and exists in almost all areas, including coastal areas which have easy access to fish (Yuliantini et al., 2022). Previous research has shown a correlation between fat intake and the incidence of stunting. Toddlers who consume insufficient fat have an 8,529 times higher risk of becoming stunted than toddlers who consume sufficient fat. An inadequate fat intake can reduce the nutritional status of toddlers.

Protein is one of the most important nutrients for the body and is the most important component of hydrolyzed products. A food ingredient that is made into a protein hydrolysate aims to fulfill the need for animal protein, especially fish protein, to prevent stunting in toddlers. The protein content measured in the hydrolysate product was the dissolved protein. Based on the results, F3 formula had the highest protein content of 43,18%. The higher the cob FPH powder, the higher the protein content in the tameko powder. Based on previous research, FPH cobs have a higher protein content than edamame flour. FPH cob contains 75,17% protein (Parvathy et al., 2018), while edamame flour contains 37,41% protein (Siregar et al., 2023). The high levels of protein in F3 are expected to increase protein intake in toddlers so that they can achieve optimal nutritional status, because proteins play an important role in the growth and development of toddlers (Karlina et al., 2023).

Protein is a nutritional component that the body needs, especially for the growth and development of children. Proteins can be obtained from vegetable- and animal-side dishes. Most people prefer animal protein to vegetable protein. Adequate protein intake is important for the growth and development of children. Optimal protein intake can help children achieve optimal nutritional status. On the other hand, low protein intake can increase the risk of stunting in children (Yuliantini et al., 2022). Previous research has shown a correlation between protein intake and the incidence of stunting. Toddlers who consume less protein than they need have an 8.8 times higher risk of becoming stunted compared to toddlers whose protein intake is sufficient (Karlina et al., 2023).

Carbohydrates are macronutrients that the body also needs, in addition to protein and fat. The highest carbohydrate content was observed in F1 (31,39%). The higher the amount of edamame flour, the higher is the carbohydrate content. The carbohydrate content in tameko powder is predominantly derived from edamame flour. Edamame flour contains 31,76% carbohydrates (Siregar et al., 2023). Apart from being the main source of energy, carbohydrates also function as contributors of energy to the brain and nerves and as regulators of metabolism. In toddlers, carbohydrates play a role in supporting brain development and activity. If a toddler has a low carbohydrate intake, the toddler has a 6,5 times risk of becoming stunted (Yuliantini et al., 2022). If carbohydrate consumption is less than required, fat stores will continue to decrease, which can reduce the nutritional status of toddlers. Low consumption of nutrients can affect a child's growth and development process (Karlina et al., 2023).

Compared with Taburia (multivitamin and multimineral powder for toddlers) powder, Tameko powder has many differences, such as color, taste, and aroma. Tameko powder has a greenish-yellow color with a savory taste and delicious aroma typical of sea fish; therefore, it is more suitable for use as a flavoring ingredient in food. Compared with existing food flavorings, tameko powder has the advantage of being high in protein, so it can increase protein intake in toddlers.

Conclusion

Based on the hedonic test, the taste, aroma, and overall parameters that the panelists liked the most were F3 with a ratio of edamame flour and cob FPH of 50:50. Proximate test results also showed that the highest protein content was found in F3 (43,18 %). Even though it has a high protein content, there needs to be improvements in the tameko powder processing process because the ash content parameters in tameko powder are still not in accordance with the INS standards.

It is necessary to conduct further research to obtain the best results according to INS and test the right amount of administration so that tameko powder can be implemented to overcome stunting.

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