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# Development and Nutritional Characterization of Biscuits Enriched with Kalakai (*Stenochlaena palustris*) and Snakehead Fish (*Channa striata*) Flours

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***ABSTRACT**: Stunting remains a public health concern in Central Kalimantan, where the prevalence (22.1% in 2024) is still above the national average (19.8%). This study aimed to develop a high-nutrient functional biscuit for complementary feeding (MP-ASI) from local resources, using kalakai (Stenochlaena palustris) flour, a source of iron and vitamins, and haruan fish (Channa striata) flour, a source of protein and albumin, as partial substitutes for wheat flour. The experiment consisted of four formulations: P0 (control), P1, P2, and P3, which contained different proportions of wheat flour, kalakai flour, and haruan fish flour. Each treatment was performed in three independent replications, resulting in a total of 12 experimental units. The experimental data were analyzed using analysis of variance (ANOVA), followed by Duncan's Multiple Range Test (DMRT) at a 5% significance level ($\alpha = 0.05$) to determine significant differences among treatment means. The biscuits were analyzed for moisture content, water activity (aw), protein, carbohydrate, iron, vitamin C, microbiological quality (total plate count, E. coli, and Salmonella), and sensory acceptance. Substitution significantly affected ($P \leq 0.05$) moisture content (3.1-4.2%), aw (0.3-0.7), protein (9.2-19.2%), and carbohydrate (69.6-60.4%). Iron increased from 2.4 mg/100 g in the control to 7.1-11.9 mg/100 g in the substituted biscuits, and vitamin C from 1.9 to 10.9-12.4 mg/100 g. All formulations met the maximum moisture limit of SNI 2973:2018 (5%), and E. coli and Salmonella were negative in 25 g. The best formulation was determined using the De Garmo effectiveness index method by integrating selected nutritional, physicochemical, and sensory evaluation parameters. P2 was selected as the best formulation because protein content was about 73% higher and its iron content about 3.6 times higher than the control; its moisture content (3.6%) was well below the 5% limit of SNI 2973:2018; its water activity (0.5) was the same as P1; its total plate count was low ($1 \times 10^2$ CFU/g); and it obtained the highest sensory score ("like very much").*

***Keywords:** Biscuit, Haruan Fish, Kalakai, Stunting.*

## 1. INTRODUCTION

Nowadays, stunting is one of the concerns in Indonesia because the prevalence of stunting tends to fluctuate. According to the Indonesian Nutrition Status Survey, the prevalence of stunting was 24.4% in 2021, decreasing to 21.6% in 2022. Despite this decline, the prevalence remained above the national target of 19.8%. However, globally, Indonesia's stunting prevalence rate ranks 27th highest out of 154 countries that have stunting data. The Indonesian Health Survey (SKI) shows that in 2022 the achievement of stunting prevalence in Central Kalimantan Province (Central Kalimantan) ranks 11th highest in Indonesia.

Stunting is a condition where children are too short for their age due to chronic malnutrition, especially in the first 1,000 days of life. Many factors that cause stunting, that were inadequate feeding of complementary food (MP-ASI), poor quality of micronutrients, lack of food diversity, low energy levels from complementary foods dan inadequate feeding (Fitriani et al., 2020). Feeding patterns have a great effect on infant weight growth (Hardiningsih et al., 2020).

Despite Indonesia's abundant natural resources, particularly diverse plant- and animal-based food resources, stunting remains a significant nutritional challenge, highlighting the need to optimize these local resources into nutritious and accessible food products.

Central Kalimantan as an area rich in local wisdom generally utilizes natural resources derived from agricultural products and wild plants in the middle of the forest and around swampy areas. Central Kalimantan's local wisdom such as kalakai and haruan fish can be used as raw materials for making high-nutrition biscuits, complementary foods for MP-ASI. Improving the nutritional quality of food products represents one of the strategies that can support efforts to address stunting and other forms of malnutrition. One potential approach is the utilization of locally available food resources with high nutritional value.

Kalakai is a plant that is commonly processed by the community as a vegetable. Kalakai has some benefits as a functional food (Pandingan, Oslo, Josephine, & Anwar, 2022). According (Maharani, Haidah, & Haiyinah, 2005) to the kalakai plant, it has an Fe content of 291.32 mg/100g, calcium, vitamins A and C. Due to the high Fe content, kalakai is a fortification ingredient in the formulation of baby porridge flour. The results of the study show that the formulation has a Fe mineral content close to the SNI MP-ASI standard of instant powder (Sholihah et al., 2018).

Kalakai, particularly the red variety, has been reported to contain several important nutrients and micronutrients, including iron (41.53 ppm), copper (4.52 ppm), vitamin C (15.41 mg/100 g), protein (2.36%), β-carotene (66.99 ppm), and folic acid (11.30 ppm). In its dried form, kelakai contains 7.28% moisture, 9.15% ash, 1.37% fat, 11.43% protein, 70.77% carbohydrates, and an energy value of 341.13 kcal. The use of kalakai as a substitute material for making high-nutrient biscuits is an alternative to providing food that has good nutritional content (Qamariah & Yanti, 2018; Wijinindnyah et al., 2022) .

To complement the content in high-nutrient biscuits, the nutritional component that needs to be added is protein. The provision of protein sources is by combining kalakai flour with haruan fish flour. Haruan fish (*Chana striata*) is one of the most produced freshwater fish in Kalimantan. Haruan fish has a high albumin content even when compared to seawater fish (Listyanto & Andriyanto, 2009). Albumin in Haruan fish is expected to be an alternative to providing nutrients to improve the nutritional quality of the community. All stakeholders must take a role in supporting the government's efforts to reduce stunting rates. The Central Kalimantan Provincial Government in November 2023 held a seminar on the results of a survey study on the acceleration of stunting reduction, stating that the reduction in stunting rates in Central Kalimantan is targeted at 15.38% in 2024. To support

efforts to accelerate this reduction, research was carried out on making high-nutrition biscuits based on the use of local wisdom which is available in abundant quantities and easy to obtain.

Consuming foods rich in essential nutrients, particularly protein, is important for supporting growth, development, and overall health. One potential approach is to develop alternative food products that combine different nutrient sources to improve their nutritional value. In this regard, the use of kalakai flour and haruan fish flour as partial substitutes for wheat flour in biscuit production offers an opportunity to diversify food ingredients while enhancing the protein and micronutrient content of the product. Therefore, this study evaluates the nutritional characteristics of biscuits formulated with different proportions of kalakai flour and haruan fish flour as a potential nutrient-rich food alternative to support improved nutritional intake (Mananda et al., 2025) .

Based on the explanation of the nutritional content found in kalakai and haruan fish, the researcher is interested in utilizing kalakai plants and haruan fish which are ingredients based on local wisdom as raw materials for making high-nutrition biscuits to support the government in reducing stunting rates.

The purpose of this study is to determine the best formulation for making high-nutrition biscuit products from kalakai flour and haruan fish flour. To find out the characteristics of high-nutrition biscuit products by conducting physicochemical, microbiological and organoleptic tests and to determine the effectiveness of high-nutrient biscuits produced as a companion food product for MP-ASI.

## 2. MATERIALS AND METHODS

### 2.1 Materials and Equipment

The raw materials used in this study are kalakai and haruan fish. Both ingredients are processed into flour before being used in making biscuits. The stages of the flour manufacturing process are as follows.

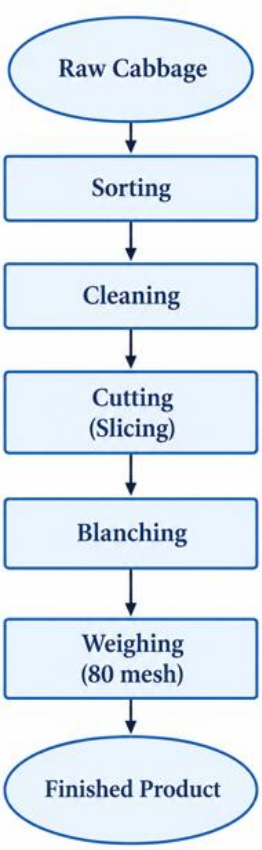



**Figure 1.** Flow Chart Producing of Kalakai Flour

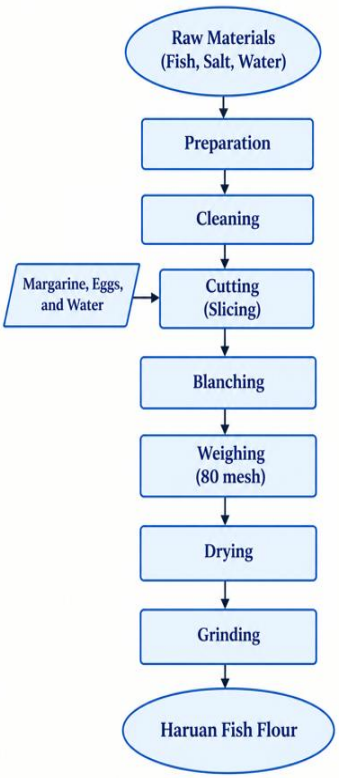



**Figure 2.** Flow Chart Producing of Haruan Fish Flour

## 2.2 Biscuit Making

The biscuit production process starts with the preparation of flour, salt, sugar, and milk, which are mixed thoroughly to obtain a homogeneous mixture. Margarine, eggs, and water are subsequently incorporated during the kneading stage. The mixture is kneaded until a uniform dough with suitable consistency is obtained. The dough is then rested to allow the ingredients to become properly integrated and to improve its handling properties. After resting, the dough is molded into the desired shape and size. The molded dough is subsequently baked under controlled temperature conditions until the biscuits are fully cooked and develop the desired physical characteristics. The process results in the final biscuit product (modified from (Capraz, Capraz, Turan, & Isik, 2025)

To determine the best formulation of biscuits in this study, differences in the concentration of wheat flour, kalakai flour and haruan fish flour were carried out using 4 levels of treatment, namely P0 (control), P1 (50% : 20% : 30%), P2 (50% : 15% : 35%) and P3 (50% : 10% : 40%). The formulation is presented in Table 1.

**Table 1.** Biscuit Formulation

| Raw materials | Treatment | | | |
|---|---|---|---|---|
| | P0 | P1 | P2 | P3 |
| Wheat flour | 100% | 50% | 50% | 50% |
| Kalakai flour | - | 20% | 15% | 10% |
| Haruan flour | - | 30% | 35% | 40% |
| Salt (g) | 1 | 1 | 1 | 1 |
| Refined sugar (g) | 79 | 79 | 79 | 79 |
| Milk powder (g) | 20 | 20 | 20 | 20 |
| egg (g) | 50 | 50 | 50 | 50 |
| Margarine (g) | 25 | 25 | 25 | 25 |
| Water (ml) | 25 | 25 | 25 | 25 |

Note: % treatment of the total weight of the dough

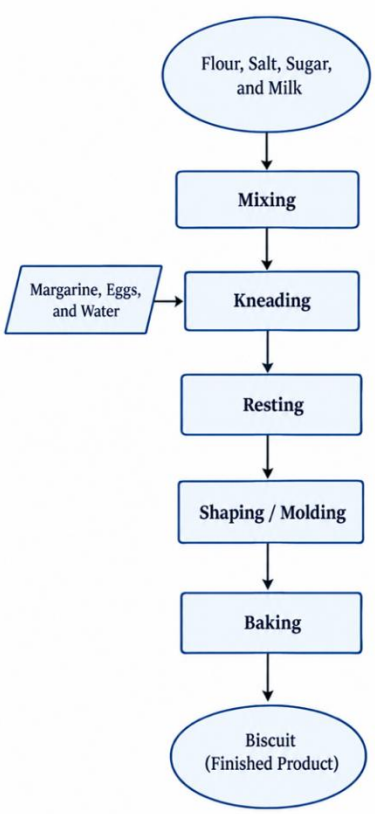



**Figure 3**. Flow Chart of Making Biscuit

The formulation of high-nutrition biscuits is based on the adequacy of energy and protein of toddlers, namely for energy of 1550 kcal and protein of 39 g. Kalakai flour and snakehead fish flour are substitutes for wheat flour in making biscuits.

## 2.3 Physicochemical Analysis of Biscuits

The physicochemical properties of the biscuits were evaluated by analysing moisture content, water activity, protein content, carbohydrate content, iron content, and vitamin C content. Moisture content was determined using the oven-drying method and expressed on a wet-weight basis (w/w). Water activity (aw) was measured using a water activity meter. Protein content was analyzed using the Kjeldahl method, while carbohydrate content was calculated using the by-difference method. Iron content was determined using Atomic Absorption Spectrophotometry (AAS). Vitamin C content was analyzed using the iodometric titration method (Fatmawati & Mardiana, 2014) (Fatmawati et al., 2018) (Maharani, Haidah, & Haiyinah, 2005). All analyses

were conducted to evaluate the nutritional composition and physicochemical characteristics of the biscuits produced from different proportions of kalakai flour and haruan fish flour.

### 2.4 Microbiological Testing

Microbiological analysis was conducted to determine the Total Plate Count (TPC) and detect the presence of *Salmonella* in the biscuit samples. TPC was determined using the standard plate count method, and the results were expressed as colony-forming units per gram (CFU/g). *Salmonella* detection was performed using an appropriate microbiological detection method, with the results reported as positive or negative per 25 g of sample.

### 2.5 Organoleptic Testing

Organoleptic testing using 25 semi-trained panelists to find out the level of preference for taste, color, texture, aroma. Testing of consumer preference/hedonic using a questionnaire using a scale of 1-5 (very likes – dislikes).

### 2.6 Research Desain

The experimental design in the study was to use factorial Completely Randomized Design (CRD) with four treatments (P0, P1, P2, and P3) each with three replications. The experimental data were analyzed using Microsoft Excel 2019 and SPSS version 26.0. Analysis of variance (ANOVA) was performed to determine the effects of the treatments on the observed parameters. When significant differences were found, further comparisons among treatment means were conducted using Duncan's Multiple Range Test (DMRT) at a 5% significance level ($\alpha = 0.05$), corresponding to a 95% confidence level. The results are presented as means ± standard deviations.

### 2.7 Determination of Best Formulation

The best treatment was determined using the De Garmo effectiveness index method. Eight parameters were evaluated, namely moisture content, water activity, protein content, carbohydrate content, iron content, vitamin C content, total plate count (TPC), and sensory acceptance. Each parameter was assigned an equal weight of 0.125. The effectiveness value (Ne) was calculated based on the higher-is-better or lower-is-better criteria. The weighted value (Nh) was obtained by multiplying the effectiveness value by its respective weight. The treatment with the highest total Nh value was selected as the best formulation.

## 3. RESULTS AND DISCUSSION

### 3.1. Proximate Analysis of Result Moisture Content and Water Activity (Aw)

The results of the analysis of the moisture content of biscuits from the substitution of kalakai flour and haruan fish flour can be seen in Table 2.

**Table 2.** Moisture content and water activity

| Treatment | Moisture content (%) (SD ±) | Water Activity (Aw) (SD ±) |
|---|---|---|
| P0 | 3,1 ± 0,06 a | 0,3 ± 0,02 a |
| P1 | 3,4 ± 0,07 b | 0,5 ± 0,05 b |
| P2 | 3,6 ± 0,09 c | 0,5 ± 0,08 b |
| P3 | 4,2 ± 0,09 d | 0,7 ± 0,06 c |
| Average | 3,57 | 0,5 |

Note: The numbers in the same column are followed by lowercase letters that are not the same, different significantly according to DNMRT at the level of α = 5%.

Based on the analysis of the fingerprints, it was shown that the substitution of kalakai flour and haruan fish meal had a statistically different effect ($P \leq 0.05$) on the moisture content and water activity (aw) of biscuits. The highest moisture content was in the P3 treatment biscuits with a value of 4.2% and the biscuits with the lowest moisture content were in the P0 treatment of 3.1%. The higher the addition of kalakai flour and haruan fish meal, the higher the moisture content of the biscuits. The moisture content in an ingredient can determine the shelf life of a product (Meikapasa et al., 2025) .

Moisture content is an important factor influencing the shelf life and storage stability

of food products. Generally, products with lower moisture content tend to exhibit a longer shelf life, as reduced moisture availability can limit deterioration during storage. Conversely, high moisture content may accelerate product deterioration and shorten its storage life. Furthermore, elevated water activity (aw) can adversely affect the textural properties of food products, potentially resulting in a softer texture and reduced crispness (Supriyanto et al., 2024).

The moisture content in biscuits affects the water activity, and texture. In the activity of water (Aw), which increased from 0.3 ± 0.02 (P0) to 0.7 ± 0.06 (P3), although the DNMRT test showed that P1 and P2 were not significantly different, while P0 and P3 were significantly different from other treatments. The moisture content and water activity are relatively low. This condition is beneficial for the shelf life because it is below the growth threshold of most microbes and the moisture content is the maximum limit of SNI biscuit. Biscuits have good quality standards based on SNI 2973:2018 so they are good for consumption. Based on the SNI standards for biscuits, the maximum moisture content is 5% (Badan Standardisasi Nasional, 2018).

The increase in moisture content as the proportion of kalakai flour and haruan fish flour increases is most likely related to the hygroscopic properties and water holding capacity of the two ingredients (Fatmawati et al., 2018). The fiber in kalakai flour has a hydroxyl group that can bond hydrogen with water molecules, while the protein in haruan fish meal has the ability to bind water through interaction with polar groups in its amino acid chain. The higher the proportion of these two ingredients replacing starch-dominated wheat flour, the greater the capacity of the dough matrix to hold water during and after the baking process, so that there is relatively less water evaporated at the same temperature and baking time (Khasanah & Mumpuni, 2021; Sundari et al., 2024). Based on solubility, proteins can be classified into albumins, globulins, glutelins, prolamins, histones, and protamines (the Osborne classification). Albumins are soluble in water and coagulate upon heating (Fatmawati & Mardiana, 2014).

An important advantage of this study is the conversion of fresh kelakai leaves into a powdered form through a controlled drying process. The removal of moisture during drying may increase the concentration of nutrients and bioactive compounds on a dry-weight basis compared with fresh kelakai leaves. Furthermore, kelakai leaf powder offers greater practicality and flexibility for food applications, as its reduced moisture content facilitates handling, storage, transportation, and incorporation into various food formulations. This processing approach therefore provides a more efficient and versatile form of kelakai that can potentially enhance its utilization as a functional food ingredient (Wijinindnyah et al., 2022).

### 3.2. Protein Content and Carbohydrate Content

The results of the analysis of biscuit protein content from the substitution of kalakai flour and haruan fish flour can be seen in table 3.

**Table 3.** Protein content and carbohydrate

| Treatment | Protein Content (%) (SD ±) | Carbohydrate Content (%) (SD ±) |
|---|---|---|
| P0 | 9,2 ± 0,08 a | 69,6 ± 0,08 a |
| P1 | 12,6 ± 0,12 b | 65,8 ± 0,15 b |
| P2 | 15,9 ± 0,06 c | 65,1 ± 0,04 b |
| P3 | 19,2 ± 0,18 d | 60,4 ± 0,06 c |
| Average | 14,22 | 68,02 |

Note: The numbers in the same column are followed by lowercase letters that are not the same, different significantly according to DNMRT at the level of α = 5%.

Based on the analysis of the fingerprints, it was shown that the substitution of kalakai flour and haruan fish meal had a statistically different effect (P≤0.05) on the protein content and carbohydrate content of biscuits. The substitution of wheat flour with kalakai flour and haruan fish meal changed the nutritional composition of biscuits significantly. The increase in protein in the P1 to P3 treatment was mainly sourced from

haruan fish flour, because haruan fish (Channa striata) is known to be high in protein and rich in albumin (C. K. Sari & Sari, 2023; Utomo et al., 2011). The higher the proportion of haruan flour, the greater the protein contribution to the product, while the proportion of carbohydrates tends to decrease because wheat as a source of starch decreases. The energy content of dried kelakai leaves is comparable to that of dried moringa leaves, which has been reported at approximately 344.8 kcal. In addition to the contribution of fish flour, kalakai flour also contribute plant-derived protein and dietary fiber to the biscuit formulation.

Haruan fish is one of the good sources of nutrients. According to (Rahman et al., 2018) , the protein content in Haruan fish ranges from 78.32% fat to 2.08% and vitamin A 0.265mg. Haruan fish has a high content of arachidonic acid (AA) and docosahexaenoic acid (DHA). Proximate analysis of Haruan fish showed the amount of crude protein at 23%, crude fat at 5.7% and ash at 1.8%. These results show that Haruan fish is a low-fat source of nutrition, because the fat content contained is less than 10% (Agustin et al., 2016; Khasanah & Mumpuni, 2021; Mabrur et al., 2018).

Nutritional needs are adjusted based on age and the type of food consumed. What affects is the quality and quantity of food consumption. Quality can be seen from the diversity of types of food available, while quantity is assessed based on the amount of energy and protein in the food intake given. Stunting prevention is carried out starting from the first 1000 days, from pregnancy to 2 years of age. Stunting not only inhibits growth and development, but also permanent brain damage to adulthood. Consuming foods that have good nutrients will make the body maximize the use of nutrients for the body's metabolic function (Nooragni, 2020) .

Based on the Minister of Health Regulation No. 28 of 2019 concerning Nutritional Adequacy Figures (AKG), the average energy requirement of children aged 1-3 years is 1,350 kcal, while the need for protein is 20 g and carbohydrates are 215 g/day. Thus, theoretically, around 410 g of substituted biscuits for kalakai flour and haruan fishmeal are needed to get 1,350 kcal if energy is only calculated from proteins and carbohydrates. The calculation is based on using the approach of the contribution of biscuits to AKG.

### 3.3. Iron (Fe) Content and Vitamin C Content.

The results of the analysis of biscuit iron content from the substitution of kalakai flour and haruan fish meal can be seen in table 4.

**Table 4.** Iron (Fe) Content and Vitamin C Content.

| Treatment | Fe Content (mg/100g) | Vitamin C (mg/100g) |
|---|---|---|
| P0 | 2,4 | 1,9 |
| P1 | 7,1 | 12,4 |
| P2 | 8,6 | 10,9 |
| P3 | 11,9 | 11,3 |
| Average | 7,5 | 9,12 |

Based on the results of the study, a significant increase was obtained in P1 biscuits with P2 to P3. P0 which is a control treatment, biscuits without the addition of fish meal substitution and flour have the lowest iron levels. The same thing happens with significantly increased vitamin C levels.

Kalakai contains antioxidants that can act as a hemoglobin booster. In addition, Fe in kalakai also plays a role in increasing hemoglobin levels. The flavonoid content in kalakai can protect red blood cells from oxidative stress and cell damage due to free radicals (Oksal et al., 2023). The results of (Petricka et al., 2018) the study prove that consuming kalakai for a week significantly increases hemoglobin levels. (Mawaddah, 2019), It also proves that the administration of kalakai extract increases hemoglobin levels in students who experience anemia due to iron deficiency.

Vitamin C in kalakai vegetables also plays a role in increasing iron absorption. This compound reduces $Fe^{3+}$ to $Fe^{2+}$, a form that is easier to absorb so that vegetable iron can be better utilized by the body (Erlina &

Husna, 2024). The efficacy of kalakai against anemia is supported by findings, which report that the consumption of kalakai (*Stenochlaena palustris*) increases hemoglobin levels in pregnant women with anemia, especially in the at-risk age group, namely under 20 years and over 35 years old. Most of the 34 respondents experienced significant improvement after consuming it (Sari et al., 2023).

The calcium content in kalakai is also expected to be useful if consumed to meet iron needs and can reduce stunting if used as a high-nutrient biscuit product to accompany complementary food (MP-ASI). The availability of kalakai in abundant quantities can make kalakai a potential ingredient in the manufacture of value-added functional food products. Substitution using haruan fish meal also affects the iron content in biscuits. This is because Haruan fish also contains important mineral compounds such as albumin, zinc (Zn), copper (Cu), and iron (Fe) as well as unsaturated fatty acids. The minerals contained in Haruan fish also help the body's metabolic process (Aliyyah et al., 2024; Fahriza et al., 2021).

Biscuits that contain high nutrients such as protein, iron and vitamins can be categorized as functional biscuits. Omega-3 fatty acids and essential amino acids in haruan fish are thought to underlie its biological effects, acting together to strengthen antioxidant defenses, attenuate inflammatory responses, and support tissue repair (Yulizal et al., 2025).

### 3.4. Microbiological Testing Result

The results of the microbiological testing from the substitution of kalakai flour and haruan fish meal can be seen in table 5.

**Table 5.** Microbiological analysis result

| Treatment | ALT | E. coli and Salmonella |
|---|---|---|
| P0 | $1x10^4$ | Negative/25 g |
| P1 | $1x10^2$ | Negative/25 g |
| P2 | $1x10^2$ | Negative/25 g |
| P3 | $1x10^3$ | Negative/25 g |

The microbiological quality of products intended for toddlers and pregnant women must meet applicable food safety regulations. The total plate number is $1x10^2$ - $1\times10^4$ colonies/g, E. coli and Salmonella are negative/25 g indicating that the product meets the SNI limits for biscuits and BPOM's provisions on microbial contamination. The low contamination is supported by the baking process that inactivates most vegetative microbes, as well as the low water activity that limits the growth of post-processing microbes. The values of ALT at or near the upper limit indicate the need for hygienic control in the handling of flour, especially fishmeal as a raw material that is susceptible to contamination, as well as in packaging.

The types and prevalence of spoilage microorganisms in food products vary considerably and are influenced by several factors, including processing conditions, formulation, packaging, storage, distribution, and handling practices. During storage, microorganisms may proliferate when favorable environmental conditions are present, particularly in food matrices containing sufficient nutrients to support microbial growth. Although biscuits generally have a relatively low moisture content, changes in moisture during storage can affect their microbiological stability. Moisture migration may occur when packaging materials have inadequate barrier properties, allowing water vapor from the surrounding environment to permeate the package and subsequently be absorbed by the biscuit matrix. An increase in moisture content can alter the physicochemical characteristics of biscuits and potentially create more favorable conditions for microbial growth (Hernawati et al., 2018)

### 3.5. Sensory Evaluation

The results of the sensory evaluation from the substitution of kalakai flour and haruan fish meal can be seen in table 6.

**Table 6**. Sensory evaluation result

| Treatment | Average of Sensory Evaluation | Interpretation |
|---|---|---|
| P0 | 5 | Like very much |
| P1 | 4 | Like |
| P2 | 5 | Like very much |
| P3 | 3 | Like slightly |

The sensory evaluation results showed that all biscuit formulations received positive acceptance scores from the panelists, with mean scores ranging from 3 to 5. Treatments P0 and P1 obtained a score of 4, corresponding to the category of *like*, indicating that these formulations were generally well accepted by the panelists. The highest sensory score was observed in P2, which achieved a score of 5 and was categorized as *like very much*. This result suggests that the P2 formulation had sensory characteristics that were highly acceptable to the panelists.

In contrast, P3 received a score of 3, corresponding to the category of *like slightly*, indicating a relatively lower level of acceptance compared with the other formulations. The lower sensory acceptance of P3 may be associated with changes in the sensory characteristics of the biscuit resulting from differences in formulation. An increased proportion of functional ingredients may influence attributes such as flavor, aroma, color, texture, or aftertaste, which in turn can affect consumer preference. However, the score of 3 still indicates that the P3 formulation remained within the acceptable range of the hedonic scale.

Overall, the sensory evaluation indicates that the incorporation of the tested ingredients into the biscuit formulation did not result in rejection by the panelists. Among the evaluated treatments, P2 demonstrated the highest degree of sensory acceptance based on the mean hedonic score. Nevertheless, sensory preference should be interpreted together with the physicochemical and nutritional characteristics of the formulations to determine the overall suitability of the developed biscuit.

### 3.6. Determination of the Best Formulation

The determination of the best biscuit formulation was conducted using the De Garmo method (effectiveness index). Eight parameters were considered, including moisture content, water activity, protein content, carbohydrate content, iron content, vitamin C content, total plate count, and sensory evaluation. Each parameter was assigned an equal weight of 0.125

The effectiveness value (Ne) was calculated by normalizing the experimental results based on whether each parameter was classified as higher-is-better or lower-is-better. Subsequently, the effectiveness value was multiplied by its respective weight to obtain the weighted effectiveness value (Nh). The total effectiveness score was determined by summing the weighted values of all parameters for each treatment. Comparison of the biscuit formulations shown at Table 7.

**Table 7**. Comparison of the biscuit formulations

| Parameter | P0 | P1 | P2 | P3 |
|---|---|---|---|---|
| Moisture (%) | 3.1 | 3.4 | 3.6 | 4.2 |
| Water activity | 0.3 | 0.5 | 0.5 | 0.7 |
| Protein (%) | 9.2 | 12.6 | 15.9 | 19.2 |
| Carbohydrate (%) | 69.6 | 65.8 | 65.1 | 60.4 |
| Iron (mg/100 g) | 2.4 | 7.1 | 8.6 | 11.9 |
| Vitamin C (mg/100 g) | 1.9 | 12.4 | 10.9 | 11.3 |
| Total plate count | $1\times10^4$ | $1\times10^2$ | $1\times10^2$ | $1\times10^3$ |
| Sensory score | 5 | 4 | 5 | 3 |

The results showed that treatment P0 obtained a total effectiveness score of 0.375, P1 obtained 0.622, P2 obtained 0.714, and P3 obtained 0.601. Among the four treatments, P2 achieved the highest effectiveness score of 0.714, indicating that it was the most favourable formulation based on the selected evaluation parameters.

Treatment P2 contained 15.9% protein, 8.6 mg/100 g iron, and 10.9 mg/100 g vitamin C, with a moisture content of 3.6% and water activity of 0.5. Furthermore, P2 demonstrated a high sensory acceptance score of 5 (like very much) and a total plate count of $1 \times 10^2$

CFU/g. Therefore, based on the De Garmo effectiveness index, P2 (50% wheat flour, 25% kalakai flour, and 25% haruan fish flour) was selected as the best biscuit formulation.

P2 offered the best balance. Its protein content was about 73% higher and its iron content about 3.6 times higher than the control; its moisture content (3.6%) was well below the 5% limit of SNI 2973:2018; its water activity (0.5) was the same as P1; its total plate count was low ($1\times10^2$ CFU/g); and it obtained the highest sensory score ("like very much"). P2 was therefore selected as the best formulation.

## 4. CONCLUSIONS AND RECOMMENDATIONS

### Conclusion

P2 was identified as the best biscuit formulation, consisting of 50% wheat flour, 25% kalakai flour, and 25% haruan fish flour, with the highest effectiveness score of 0.714. This formulation demonstrated a favourable balance of nutritional value, physicochemical characteristics, microbiological quality, and sensory acceptance. P2 contained 15.9% protein, 8.6 mg/100 g iron, and 10.9 mg/100 g vitamin C, with a moisture content of 3.6% and water activity of 0.5. It also achieved the highest sensory acceptance score of 5 (like very much) and a total plate count of $1 \times 10^2$ CFU/g. Therefore, P2 represents the most favourable formulation among the treatments evaluated and has potential for development as a nutrient-enriched functional biscuit for MP-ASI using local ingredients.

### Recommendation

Further research should determine the shelf life and the most suitable packaging for the P2 biscuit, because moisture uptake during storage may increase water activity and the risk of microbial growth. Analyses of energy, fat, calcium, zinc, and albumin are recommended, together with an assessment of the nutrient contribution per serving against the Indonesian Nutrient Adequacy Standard (AKG) for toddlers. Acceptance should be tested with the actual target group (toddlers and their mothers), and flavor and aroma should be improved, since fish flour may affect taste. Clinical or intervention studies (e.g., hemoglobin and growth outcomes) are needed to prove the effectiveness of the biscuit against stunting, and hygienic control of raw materials, especially fish flour, should be maintained throughout production.

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