ISSN: 2347-467X, Vol. 07, No. (2) 2019, Pg. 504-516

Current Research in Nutrition and Food Science

www.foodandnutritionjournal.org

Development of Foxtail Millet and Flying Fish Flour-Based Cookies as Functional Food

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Abstract

Foxtail millet (*Setaria italica*) and flying fish (*Hyrundicthys oxycephalus*) are the most widely available food in West Sulawesi province, potentially to be developed into functional food as they contain many nutrients and bioactive components which may be beneficial for health. This study aimed to explore the nutrients content of foxtail millet and flying fish cookies as a functional food and to evaluate organoleptic aspects of the product. Design of this study was pre-experimental. Wheat, foxtail millet and flying fish were used as the component of the flour. There are five treatments were employed in this study, which were presented in %: F1=70:25:5, F2=70:20:10, F3=70:15:15, F4=70:10:20, and F5=70:5:25. The organoleptic test performed to girl students from three senior high schools in Mamuju, West Sulawesi province by a hedonic method used five scores, and analyzed by Kruskall-Wallis test (ά=0.05). The panelists were considered to accept the product when the score was in between 3 to 5. The findings indicated that the best combination of wheat flour: foxtail millet flour: flying fish flour ratio was F3 cookie. The score for color, taste, and texture was above 3 (3.7 ± 0.9, 3.7 ± 0.9, and 3.5 ± 0.8, respectively). A proximate test showed that F3 cookies contained protein (11.89%), carbohydrate (45.19%), crude fiber (4.51%), and fat (21.30%), water (5.94%), and ash (1.26%). In conclusion, the use of foxtail millet and flying fish for cookies was acceptable and high nutrition content. A further study is needed to examine the health benefit of the cookies.

Article History

Received: 19 January 2019
Accepted: 16 May 2019

Keywords

Cookies;
Foxtail Millet Flour;
Flying Fish Flour.

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Doi: http://dx.doi.org/10.12944/CRNFSJ.7.2.20

Background

Functional food has been an alternative for those who want to maintain healthy and get disease-free life,[1] thus the interest in getting information about diet-health interaction, particularly about functional foods, has been increased.[2] The use of functional food now begins as a lifestyle in modern society. However, many factors associated with the use of functional food in the communities, such as type of product, compatibility, stability, consumer acceptance, and regional preference.[3]

Functional food can bridge the traditional gap between food and medicine, thus ensuring the potential benefit of a new developed functional food is important.[4] As a healthy diet, it needs to be consumed adequately in order to meet nutritional needs and other health benefits, such as reduce the risk of disease.[5,6] However, although functional food can contribute to human health, it needs to be supported by other healthy lifestyles, such as exercise and keep personal hygiene.[1]

There are three different group of functional foods; 1) natural functional foods that do not require processing and can be eaten immediately (e.g. vegetables and fruit); (2) traditional functional foods are functional foods that are processed in traditional ways which are inherited from generation to generation (e.g. herbs, tempeh and tofu); (3) modern functional food is functional food made with special recipes with new recipes.[7] A functional food has to meet sensory, nutritional and physiological requirements. The physiological properties of functional food are determined by the bioactive components contained in it, such as, dietary fiber, inulin, antioxidants, PUFAs, prebiotics, and probiotics.[8]

Indonesian local food, such as foxtail millet and flying fish are potential that can be developed into functional food as they contain many nutrients and bioactive components which may be beneficial for health.[9] Cereals including foxtail millet have been accepted as functional and nutraceutical foods because they provide dietary fiber, protein, energy, minerals, vitamins, antioxidants, and phytochemicals needed for human health,[10,11] while, flying fish has omega-3, protein, vitamins, minerals, and taurine.[9] However, flying fish (*Hyrundicthys oxycephalus*) have a low commercial value consumed by low-income people, so formulation is needed as an alternative to adding value to flying fish, so that there is an increase in consumption by society at large.[12]

Despite the fact that foxtail millet and flying fish are abundant in West Sulawesi province, people remain under-utilized as a food source. It is only traditionally managed and did not extend to the entire community. Especially for flying fish which has a short shelf life and the quality of this food may be decreased depending on water content.[13] In addition, the production of flying fish does not exist at any time due to the influence of seasonal changes. As a results, people in the communities are rarely consumed this food. Therefore, a powdering process for this food may be a good way to maintain

Fig. 1: Foxtail Millet Flour

the existing of the product and keep it as ready-to-eat-food products which contains 65% of protein and high-quality amino acid.[14]

The product innovation of flying fish and foxtail millet-based, in the form of healthy snacks, is essential with the aim of producing functional foods. This product then can be used in an effort to overcome the problem of nutrition-health, especially in communities in the West Sulawesi province. Besides containing calories, healthy snacks have dietary fiber, antioxidants, vitamins, and minerals which are important for the human body. However, the development of functional food from foxtail millet and flying fish, in the form of snacks, is less studied. Therefore, this study aimed to explore the nutrient composition – protein, carbohydrate, fat, fiber, water, and ash - of foxtail millet and flying fish cookies as a functional food and to evaluate organoleptic aspects of the product.

Materials and Methods

Design

This research was pre-experimental design. Sample cookies were divided into 5 types (formula), based on comparison of foxtail millet flour and fish flying flour to wheat flour (%) (Table 1). This research has three stages of work, namely the process of making millet flour (foxtail millet), making flying fish flour, and making cookies.

Materials and Tools

I. Fluor making

a. Material: foxtail millet, fish flying obtained from West Sulawesi province, clean water

b. Tools: knives, gloves, pans, cutting boards, spoons, 80 mesh flour sieves, analytical scales, basins, bowls, mills/blenders, oven burns, baking pans, bowls, baskets and molds.

c. Flour preparation process

Millet Flour

The procedure for making millet flour began with the separation of the bark and millet seeds, then filtered, washed and soaked for 4 hours to produce the best physical, chemical and organoleptic characteristics,[15] then dried in the sun until it is correct dry really. Furthermore, dried foxtail millet seed are blended and sifted using an 80 mesh stainless sieve to produce millet flour (Fig.1). The millet flour was then roasted for 2 hours at 130 °C to reduce water content, so that it can last a long time. After that, millet flour be examined in the laboratory for proximate testing (water, ash, fat, carbohydrate, fiber and protein content).

Table 1: Formulation of biscuit according to foxtail millet and flying fish flour concentrations

Material	Concentration				
	F1 (70:25:5)	F2 (70:20:10)	F3 (70:15:15)	F4 (70:10:20)	F5 (70:5:25)
Wheat Flour (g)	175	175	175	175	175
Millet Flour (g)	62.5	50	37.5	25	12.5
Flying Fish Flour (g)	12.5	25	37.5	50	62.5
Corn starch Flour (g)	10	10	10	10	10
Chicken Egg Yolk (egg)	2	2	2	2	2
Refined Sugar (g)	125	125	125	125	125
Baking powder (tsp)#	½	½	½	½	½
Salt (tsp) #	½	½	½	½	½
Margarine (g)	100	100	100	100	100
Powder milk (g)	25	25	25	25	25
Water (ml)	35	35	35	35	35
Orange essence (ml)	1.5	1.5	1.5	1.5	1.5

#tsp = teaspoon; F1 = Formula 1; F2 = Formula 2; F3= Formula 3; F4= Formula 4; F5 = Formula 5

Flying Fish Flour

The process of flying fish powdering started from the stage of cleaning and weeding of fresh flying fish to remove scales, head fins, viscera and gills of fish, then wash with clean water for up to 3 repetitions. Furthermore, soaking was done using lime juice and ginger juice for 15 minutes to remove the fishy odor, and then washed using clean water. The next stage is steaming for 30 minutes to activate the enzyme and kill the decaying microbes which are pathogenic and do not form spores. After steaming, the fish, skin and bones of the fish are separated. The obtained fish meat was then smoothly finished to form like coconut pulp, and then put into the oven at 60 °C for 12 hours. After being put in the oven, it was blended and sieved using a stainless sieve with an 80 mesh size. After being flour (Fig.2), oven cooking was continued at 130 °C for 2 hours, then brought to the laboratory for proximate tests (moisture, ash, fat, carbohydrate, fiber and protein)

Preparation for Making Cookies

The main ingredients for the product formulation cookies are foxtail millet and fish flying. Supporting materials include wheat flour, white sugar, egg yolks, cornstarch (maize), orange essence as a flavor to minimize the smell of fish (Table 1).

The chemicals used for proximate analysis were aquadest (H_2O), sodium hydroxide, selenium, cuprum sulfate ($CuSO_4$), red methyl indicator, sodium sulfate, indicator, hydrochloric acid 001 N, hydrochloric acid 3%, acid nitrate concentrated, sodium tetra borate, selenium catalyst, iodide kalium solution (KI 20%), sulfuric acid solution, boric acid solution 2%, SeO_2, 25% K_2SO_4, NaOH 30%, H_2SO_4 25%, H_2SO_4 1.25%, sodium thiosulfate solution ($Na_2S_2O_7$) 0.1 N, starch solution 0.5%, Na_2CO_3, hexane, sulfuric acid 1.25%, sodium hydroxide (NaOH) 3.25%, ethanol. 96%

Fig. 2: Flying Fish Flour

The tools used for the proximate analysis are : analytical balance, Erlenmeyer (500 ml), stand cooler, volumetric flask (500 ml), funnel (10 ml), goiter pipette (25 ml), electric heater, stopper, porcelain/ platinum cup, burette, dropper pipette, litmus paper, luff solution, indicator of phenolphthalein, filter paper, fat pumpkin, Soxhlet tool, electric heater, oven, fat free cotton, weighing bottle covered, exicator, electric furnace, desiccator, vacuum pump, krustang, Whatman 41 filter paper, horn spoon, pipette (5 ml and 50 ml), volumetric flask (100 ml).

Formula Cookies Development

All ingredients were first extracted except margarine, egg yolks and orange essence. Orange essence is a mixture of liquid food additives produced from the processing of orange juice that is used as coloring, flavoring, and giving aroma to minimize the smell of fish in cookies. After sieving, mixing flour, foxtail millet flour and flying fish flour is then mixed and then sifted again. The next stage is mixing margarine and sugar using a mixer, then adding essence orange, egg yolks, and mixing until it is well mixed. Next, add corn flour, baking powder, salt, milk powder to the mixture, and mix again until well mixed, then the flour mixture (wheat, millet, and flying fish) was put into the mixture, then stir until evenly distributed, then pour a water little by little then stirred again. After the mixture was well mixed, making cookies using biscuit maker and producing 50 pieces of biscuits, then baking with the oven at a temperature of 160-1700 C for 15 minutes.

Organoleptic Test

Testing of the organoleptic properties of biscuits as supplementary food products was carried out to panelists (female adolescents) as many as 100 young women from 3 senior high schools in Mamuju district used the hedonic scale scoring method with 5 scales namely (1) very dislike, (2) like, (3) ordinary, (4) like, and (5) really like. The organoleptic properties data tested included the color, aroma, taste, texture, and crispness of the products F1, F2, F3, F4, and F5. Hedonic tests to determine preferences for cookies, panelists were asked to taste product samples and among each tasting

biscuit cookies samples, it was required to consume water as a neutralizer. Then one formula was chosen as the best product based on the organoleptic test and Kruskall- Wallis test.

Proximate Analysis from Foxtail Millet Flour, Fish Flying Flour and Selected Cookies

Determination of the proximate composition (protein, carbohydrates, crude fibre, fat, water, ash) of the foxtail millet flour, fish flying flour, and selected formulas (F3) was carried out in laboratory for testing of industrial and plantation centers Ministry of Industry of the Republic of Indonesia, and refers to Indonesian National Standard (Standard Nasional Indonesia = SNI) 01-2891-1992[16].

Determination of Protein Level

Measurement of protein levels was carried out by the Kjeldahl method. The principle of measuring protein content is that the nitrogen compound is converted to ammonium sulfate by concentrated H_2SO_4. Ammonium sulfate formed is broken down with NaOH. The released ammonia is tied with boric acid and then entangled with an acidic solution. The measurement process begins with weighing 0.51 g of samples, then put it in a 100 ml Kjeldahl flask, then adding 2 g of selen mixture and 25 ml of concentrated H_2SO_4. Furthermore, heated above the electric heater until boiling and the solution becomes clear green (about 2 hours), left to cool then diluted and put in a 100 ml volumetric flask, and adjusted to

Table 2: Organoleptic Test Results for Biscuit-Cookies

Evaluated Aspects	Formula	Very Like		Like		Ordinary		Less Like		Dislike	
		n	%	n	%	n	%	n	%	n	%
Color	F1	11	11	45	45	33	33	10	10	1	1
	F2	9	9	55	55	22	22	11	11	3	3
	F3	19	19	45	45	27	27	8	8	1	1
	F4	12	12	41	41	33	33	11	11	3	3
	F5	19	19	39	39	32	32	7	7	3	3
Taste	F1	18	18	39	39	29	29	8	8	6	6
	F2	17	17	37	37	23	23	15	15	8	8
	F3	19	19	35	35	21	21	13	13	12	12
	F4	9	9	29	29	15	15	31	31	16	16
	F5	17	17	24	24	15	15	18	18	26	26
Texture	F1	6	6	50	50	25	25	16	16	3	3
	F2	6	6	40	40	39	39	12	12	3	3
	F3	6	6	45	45	41	41	5	5	3	3
	F4	11	11	36	36	37	37	14	14	2	2
	F5	14	14	35	35	32	32	16	16	3	3
Aroma	F1	20	20	30	30	21	21	23	23	6	6
	F2	12	12	28	28	23	23	24	24	13	13
	F3	12	12	18	18	27	27	27	27	16	16
	F4	8	8	16	16	26	26	29	29	21	21
	F5	10	10	17	17	16	16	28	28	29	29
Crispness	F1	14	14	43	43	27	27	9	9	7	7
	F2	14	14	40	40	33	33	11	11	2	2
	F3	8	8	47	47	33	33	9	9	3	3
	F4	15	15	36	36	30	30	16	16	3	3
	F5	15	15	34	34	31	31	14	14	6	6

the line mark, followed by taking the solution using a pipette 5 ml was put into a distiller, then 5 ml of 30% NaOH and a few drops of phenolphthalein indicator were added, then distilled for 10 minutes, 10 ml of the container used 2% boric acid solution mixed with the indicator, then rinsed the cooling end with distilled water, then drawn with 0.01 N HCl solution, lastly done blank determination. Determination of protein level:

% Nitrogen level= Chloride amonium level× EM Nitrogen / w × 100%

% Protein level = % Nitrogen level × Convection Factor (6,25)

Note: W = weight sample; EM = extraction material

Determination of Carbohydrate Level

The principle of analysis of carbohydrate content assessment using the Luff-Schoorl method is Cu2+ reduction to Cu 1+ by monosaccharide. Free monosaccharides will reduce base solutions from metal salts to their oxide form or free form. The excess of reduced Cu2+ is then quantified by Iodometric titration.

The carbohydrate measurement procedure starts with weighing 5 g of samples and put into 500 ml Erlenmeyer, then added 200 ml of 3% HCl solution then cooled and neutralized with 40% NaOH using litmus or fenoltalein), then transferred into a 500 ml volumetric flask and diluted to the boundary mark, then filtered using Whatman 40 filter paper. The filter in the pipette is 10 ml into the Erlenmeyer and added 25 ml of luff solution, and some boiling stone, then add 15 ml of distilled water and reflux for 10 minutes. After that, the solution was cooled and added 25 mL of 25% sulfuric acid solution (and 15 mL of 20% KI solution. Then titrated with 0.1 N thiosulfate solution until the solution color became light yellow, and added 5 mL of 0.2% starch solution and titrated again until the blue color is gone, the above treatment is also used for blank analysis.

Glucose level = $w_1 \times f_p$ / w × 100%

Carbohydrate level = 0.90 x glucose level

Note: w_1 = sample weight (mg); f_p = dilution factor; w= glucose contained for ml thio is needed in mg from list.

Fig. 3: Cookies Biscuit F3

Determination of Crude Fiber Level

The measurement of crude fiber is carried out by using the principle of strong acid hydrolysis and strong bases. As much as 2 g of fat-free samples are put into the Erlenmeyer and added 50 ml of 1.25% H_2SO_4 solution then simmer for 30 minutes. 50 ml of 3.25% NaOH is added and boil again for 30 minutes. In hot conditions, the solution is filtered with filter paper that has been weighed. The deposits contained in filter paper are washed in succession with 1.25% H_2SO_4 heat, hot water, and 96% ethanol. Filter paper that still has sediment is removed and inserted into the weighing box that has known weight. Then dried at 105ºC, cooled and weighed until the weight remained. If the level of crude fiber is more than 1% broiling the filter paper and its contents, weigh until the weight is constant. Crude fiber content is calculated using the following formula:

- Crude fiber ≤ 1 %

Crude fiber level% = W / W2 × 100%

- Crude fiber > 1 %

Crude fiber level% = W-W1 / W2 × 100

Note: W = sample weight (g); W1 = ash weight (g); W2 = sediment weight on filter paper (g)

Determination of Fat Level

The measurement of the total fat content carried out by the Soxhlet method is the direct extraction of free fat with non-polar solvents using the Soxhlet tool.

The biscuits are weighed as much as 2 g, put in filter paper which is coated with cotton, then clogged with cotton, then dried using an oven at a temperature no more than 80 °C for one hour, then put into a soxhlet device which has been linked to fat pumpkin containing boiling stones which has been dried and known for its weight. Next, extracted with hexane for 6 hours. Then hexane is distilled and the fat extract is dried in an oven at 105 °C. The fat flask is cooled and weighed until the weight is right.

$$\% \text{ Fat} = W - W_1 / W_2 \times 100\ \%$$

Note: W = weight of fat flask after extraction process (g); W_1= fat flask weight before extraction process (g); W_2= sample weight (g).

Determination of Ash Level

Measurement of ash content was carried out by total ash method. Samples of biscuits of 3 g were inserted into a porcelain dish whose weight was known. then placed on a burner, then ignited in an electric furnace at a maximum temperature of 550 °C until the blasting is perfect (occasionally the door of the furnace is opened slightly so that oxygen can enter), then the cup is cooled in a desiccator and weighed until the weight remains. Furthermore, the calculation of ash content uses the formula:

$$\text{Ash level} = W_1 - W_2 / W \times 100\%$$

Note: w = sample weight before broiling (g), w_1 = sample weight + cup after broiling g); w_2 = empty cup weight (g)

Determination of Water Level

Total water content measurements were carried out by thermogravimetric method (oven method). Biscuit samples of 2-3 g were put into aluminum plates with known weight, then dried in an oven at 105o C for 3 hours, then cooled in an exicator and weighed until a fixed weight was obtained. Water content calculation is obtained by comparing the weight of the sample before drying and the weight loss after being dried multiplied by 100%.

Ethical Approval

This study was approved by ethics committee from health research of Medical Faculty of Hasanuddin University, Education hospital of Hasanuddin University, and Public Hospital of Dr. Wahidin Sudiro Husodo (Registration No: 189/H4.8.4.5.31/ PP36-KOMETIK/2018). The researcher provides an explanation of the research procedures before organoleptic testing to the girl adolescents. If they agree to be involved in this study, the research provides an informed consent sheet to be signed and witnessed by the teacher at the school.

Statistical Analysis

Data obtained from organoleptic test results were analyzed descriptively using SPSS version 21 based on the percentage of panelists' acceptance of complementary feeding products. The effect of treatment on panelists' preference level consisting of color, aroma, taste, texture, and crispiness was analyzed using the Kruskall-Wallis test with SPSS 21.0 for Window software.

Result and Discussion

Organoleptic Test

Based on the organoleptic test (color, taste, texture, and aroma) which shows in Table 2, from 5 cookies formulas, Formula 3 was the most preferred choice by the panelist. In color aspect, F3 formula obtained Like from 63% panelists, while F2 and F4 were most panelists disliked it. The panelist consistently gave the “positive” response for F3 in taste, texture, and crispiness aspects (54%, 55%, 55%, respectively). Although there was only 30% of panelist stated “like” and “very like” for the aroma of this formula, it was relative. It can be seen from the distribution for value ”ordinary”, “less like” and “dislike” which were relatively balanced.

Some studies support the results of this study, including Sari’s study regarding development of cookies formula using cork fish flour with various concentrations (0%, 10%, 15% and 20%). The result showed that the substitution of 15% Cork fish meal was became the most preferred taste and texture by panelists.[17] Similarly, the results of the study by Adeleke and Odedeji indicated that the addition of up to 15% of tilapia fish flour on bread dough can be received based on the sensory test by panelists.[18]

The result may be affected by the appearance of F3 which looked brownish yellow and crunchy texture, also flavour of milk, chocolate and fish (Fig. 3). This can be caused by the concentration of both flour substitution ingredients (15% millet flour

and 15% flying fish flour) in a balanced amount. This is supported by Nurul's study that fish cracker formulation with a ratio of starch and fish flour is 1:1 was accepted by panelists.[19] According to Winarno, the criteria for acceptance of a product based on the results of an organoleptic test with the rejection rate must be less than 25%.[20]

Slightly different from the study's Meidyrianto found that biscuit substituted its 20% of flour with 20% skipjack tuna and 50% purple, cassava 30% was the most preferred by panelists in terms of taste, texture and color aspects.[21] Similarly, the findings of Chambo that increasing tilapia flour inclusion rates increase bread firmness and reduce product acceptance (sensory analysis).[22]

Mixing millet flour in the cookies also found that cookies with a concentration of 15% millet flour was the most favorite product by panelists based on sensory analysis which was indicated by the highest value in terms of color, taste, and texture, so that more accepted by the than other formulas.[23] Sensory markers can affect the level of preference and quality of food products. Texture is influenced by soaking time which will reduce particle size because it degrades into simpler compounds through enzymatic reactions that make it softer.[15] Increased levels of millet flour in dough can reduce gluten levels, and cause starch and protein damage.[24] Gluten levels in a product categorized as very low if they contain gluten 20 and 100 mg/kg,[25] and excessive use of gluten-free flour causes the quality of the biscuits to crumble, because gluten plays a role in giving dough strength. But millet flour can be an alternative to produce functional food products because it lacks of gluten content the same as wheat.[26]

Table 3: Analysis of Biscuit Acceptance

Evaluated Aspects	Formula	Mean± SD	P Value#
Color	F1	3.55 ± 0.857	0.367
	F2	3.56 ± 0.914	
	F3	3.73 ± 0.897	
	F4	3.48 ± 0.948	
	F5	3.64 ± 0.969	
Taste	F1	3.55 ± 1.067	<0.001
	F2	3.40 ± 1.172	
	F3	3.73 ± 0.897	
	F4	3.48 ± 0.948	
	F5	2.88 ± 1.465	
Texture	F1	3.40 ± 0.932	0.908
	F2	3.34 ± 0.879	
	F3	3.46 ± 0.809	
	F4	3.40 ± 0.932	
	F5	3.41 ± 1.016	
Aroma	F1	3.35 ± 1.209	<0.001
	F2	3.02 ± 1.239	
	F3	2.83 ± 1.248	
	F4	2.61 ± 1.214	
	F5	2.51 ± 1.337	
Crispness	F1	3,48 ± 1,068	0,895
	F2	3,53 ± 0,937	
	F3	3,48 ± 0,882	
	F4	3,44 ± 1,028	
	F5	3,38 ± 1,090	

#Kruskall Wallis Test

To analyze the difference among formulas, in overall scores, Kruskall-Wallis analysis was done. It can be seen that, in Table 3, taste and aroma aspects differed significantly among formulas ($p<0.001$). In taste aspect, F3 had the highest score while F5 had the lowest (3.73±0.897 vs 2.88 ±1.465). Meanwhile, in aroma aspect, F1 had the highest and F5 consistently had the lowest (3.35±1.209 vs 2.51±1.337). This can be caused by F1 cookies containing millet flour which is as high as 25% (62.5 g) which gives a less pleasant aroma or almost the same as cookies in general and lower concentration of fish meal (5% equivalent to 12.5 g) compared to other formulas. These findings are similar to the study conducted by Shadang and Jaganathan that the addition of 20% and 30% millet flour (pearl millet and foxtail millet) produces a very good aroma.[27]

Based on the results of the panelists' assessment of the 5 cookies formula, Formula 3 (15% concentration of millet flour, 15% of flying fish flour, and 70% of wheat flour) is the most favored by adolescent girls. Therefore, Formula 3 was then continued to the proximate analysis to determine the nutrient compounds. The use of composite flour using vegetable ingredients (including millet flour) to produce food products such as cookies can still be maintained in the same characteristics (physicochemical and functional) with products made from whole wheat flour (full-wheat flour).[28]

Proximate Analysis F3 Compared to the Basic Ingredients (Foxtail Millet and Flying Fish Flour)
Based on the results of proximate analysis of F3 formula, it was found that F3 formula (15% millet flour and 15% flying fish flour) contained 11.89% of protein, 45.19% of carbohydrate, 4.51% of crude fiber, 21.30% of fat, 1.26 of crude ash, and 5.94% of water (Table 4).

This finding is similar to the previous study showed that the level of biscuits protein content with a substitution of 15% puffed meat flour has 11.37 g/100 g.[29] A study found that in 15% tuna-flour biscuits had 12.78% of protein.[30] Nurul states that the protein content in fish crackers is higher with an increase of Dory fish-tapioca flour ratio.[19] A study indicates that the addition of 5 to 15% of tilapia flour in bread was effective in increasing the protein content of bread.[22]

The high levels of protein cookies from F3 cookies were because of the addition of flying fish, milk and eggs which are known as high protein sources, also contain foxtail millet which has a higher protein content compared to other millet like barnyard millet.[31] Based on the quality requirements of SNI 01-2973-1992, in 100 g of biscuits cookies, should be contained 6% of protein,[16] while in SNI 2973-2011, the minimum level of protein in 100g biscuit is 5%.[32] The Indonesian Food and Drug Department suggest that good food source of protein should be filled 20% of Recommended Dietary Allowance (RDA).[33] For example, according to RDA, protein requirements for adolescent girls aged 16-18 years is 56 g, meaning that in 100 g of biscuit must contain 11.2 g of protein.[34] The results of this study found that the protein content of cookies was higher than 20% of RDA. Thus, the cookies are recommended to support protein needs of adolescent girls.

Table 4: The results of proximate analysis of foxtail millet flour, flying fish flour and biscuit-cookies liked by the panellists

Chemical Characteristic	Value		
	Foxtail Millet Flour	Flying Fish Flour	Cookies
Protein (%)	10.69	76.22	11.89
Carbohydrate (%)	54.56	0.64	45.19
Crude Fiber (%)	9.25	1.72	4.51
Fat (%)	3.55	6.38	21.30
Water (%)	8.74	9.42	5.94
Ash (%)	0.87	4.40	1.26

The results of this study also found levels of carbohydrate biscuit cookies was 45.19%, although it was lower than SNI 01-2973-1992 which required at least 70% carbohydrates per 100 g biscuit cookies.[15] It can be due to the replacement of a portion of wheat flour (30%) which is as the main source of carbohydrates. Another reason, the cookie content has high protein. High level of protein in a product potentially reduces the amount of carbohydrate. In Chambo study found that the addition of 5-15% Tilapia fish flour can increase the protein content of bread but reduce carbohydrate levels. Reducing carbohydrate content by adding tilapia flour to bread roll is because the amount of flour is reduced.[22] The study by Mohammed *et. al*. also showed Carbohydrate levels of biscuits are lower when protein concentrations in biscuits fortified with fish protein increase.[35] Higher and lower carbohydrate content is one of the important reasons for substituting starch flour with fish to provide more nutritious food products.[36]

It is important to understand that the changes of carbohydrates content may affect physical characteristics of a food product, such as in color, taste and texture that can influence acceptability, level of preference and shelf life of a food.[23] The results of this study are almost the same as the results of previous studies that used catfish flour in making biscuits and snacks received levels of carbohydrate biscuits (52.3%) and cookies (59.06%).[37]

The content of fiber in cookies produced from this research was 4.51% exceeding the maximum amount recommended by SNI 01-2973-1992 was 0.5%,[16] this is because foxtail millet flour contains high fiber (9.25%). This result is similar to the findings of Widodo and Sirajuddin found the crude fiber content of biscuits fortified with tilapia and red rice (4.23%) and 4.88% on biscuits fortified with sardines and red rice.[38]

The fat content (21.30%) of F3 cookies met the recommendations of SNI 01-2973-1992 which must meet a minimum of 9.5% fat.[16] The high levels of fat cookies from F3 cookies were because of the addition of milk and margarine which are known as high-fat sources. Nurul's study found the fat content of fish crackers elevated with an increase Dory fish flour: tapioca flour ratio. However, it is essential to adjust the ration because it could affect the texture and hardness of fish crackers.[19] Similarly, this study found that the F3 cookies, the ratio of flying fish flour and millet flour was 1:1, has a good texture, thus, becoming the most preferred by young women.

This F3 cookies contains 415.22 kcal per 100 g based on the calculation (protein level x 4 kcal, carbohydrate level x 4 kcal, and fat level x 9 kcal), possible to meet the minimum requirements of the biscuit energy content recommended by SNI 01-2973-1992 (400 kcal/100 g).[16] The result of previous study found that substituted catfish flour biscuits and isolates protein of soybean consist of 480 kcal per 100 g biscuit.[39] It means that biscuit's containing fish flour and vegetable wheat flour can contribute to give considerable energy.

The ash content of the F3 cookies was 1.26%. It was lower than the maximum level recommended by SNI 01-2973-1992 for biscuit products which is 1.6%.[16] The assessment of ash content is important to find out the quality of the product, where the higher the ash content, the worse the quality of the product.[40]

The water content of biscuit cookies produced from this study (5.94%) is greater than the maximum limit recommended by SNI 2973-1992[16] and SNI 2973-2011[32]. These factors have implications for the shelf life of these cookies, however, expire time of the cookies has not been investigated.

Conclusion

Based on the requirements of the cookie quality in SNI 01-2973-1992 and SNI 01-2973-2011, it can be concluded that F3 formula cookies is a good nutritious snack, although the water content is slightly higher and potentially affects shelf life. It is recommended that biscuit cookies produced from this study be tested for their effectiveness in efforts to overcome the nutritional and health problems.

Acknowledgement

Thank you to the Directorate of Research and Community Service, Directorate General of Research and Development Strengthening of the Ministry of Research, Technology and Higher Education which has provided Grant in the form of funds to carry out this research.

Funding

This study is funded by irectorate of Research and Community Service, Directorate General of Research and Development Strengthening of the Ministry of Research, Technology and Higher Education. Grant: 0349.a/B.07/UMI/II/2018

Conflict of interest

The author(s) do not have any conflict of interest.

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