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The Organoleptic Test and Nutrient Analysis of Buffalo Milk-Based Nuggets with The Addition of Bilih Fish (Mystacoleucus Padangensis) as Additional Foods Growing Toddlers

Uji Organoleptik dan Analisis Zat Gizi Nugget Berbasis Susu Kerbau dengan Penambahan Ikan Bilih (Mystacoleucus Padangensis) sebagai Makanan Tambahan Tumbuh Kembang Balita

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ABSTRACT

Background: Bilih fish (mystacoleucus padangensis bleeker) is a local food source in West Sumatra with the potential to be one of the processed food products that can contribute to reducing stunting. Bilih fish contains complete nutrients for the growth and development of toddlers.

Objectives: This research aims to (1) develop the formulas of buffalo milk-based nuggets with the addition of bilih fish, (2) select the best nugget formula based on an organoleptic test, and (3) analyse the nutritional content through tests (proximate, zinc, and calcium) in the formulas of buffalo milk-based nuggets with the addition of bilih fish as supplementary food for the growth and development of children aged 1-3 years (toddlers).

Methods: The research design employed was experimental studies. The experimental design for the nugget formula was a Completely Randomised Design (CRD). The nugget formula was created with four treatment levels: F0 = standard formula (buffalo milk nugget + 0 g bilih fish), F1 (buffalo milk nugget + 10 g bilih fish), F2 (buffalo milk nugget + 30 g bilih fish), and F3 (buffalo milk nugget + 50 g bilih fish). The organoleptic test was conducted with 30 semi-trained panelists and 2 replications. The data collected were analysed by applying the Kruskal-Wallis and Mann-Whitney tests.

Results: The best formula was F1, indicated by a bright yellow colour, savoury taste, aromatic smell, and tender texture. There were no significant differences among F0, F1, F2, and F3. The nutritional analysis results for F1 include water (46.90%), ash (2.32%), protein (11.77%), fat (7.60%), calcium (5.52 mg), and zinc (16.63 mg) per 100 g of nuggets.

Conclusions: The best formula is F1 (buffalo milk nugget + 10 g bilih fish).

INTRODUCTION

The 2022 Global Nutrition Report revealed that Indonesia remains "on course" to meet one target for maternal, infant, and young child nutrition (MIYCN), where Indonesia is among 194 countries struggling with nutritional issues in toddlers, including stunting, wasting (body weight falls below standard), and overweight (excessive body weight)¹. According to Basic Health Research (2013), the prevalence of undernutrition in Indonesia has increased by 1.7%.² As much as 16% of toddlers experience developmental disorders, and 1 in 100 toddlers has low intelligence and a speech delay. Zinc deficiency occurring globally in 2016 reached 17%. Based on a survey conducted across 12 provinces in Indonesia, the average prevalence of zinc deficiency amounted to 36.1%, with 11.7% in West Sumatra².Toddlers with inadequate zinc intake have the potential to experience

stunting at a rate 7.8 times higher than those with adequate zinc intake $^{3}\!\!$

The direct cause of undernutrition (stunting) is the insufficient intake of nutrients and infections that typically occur in toddlers. The insufficient intake of nutrients such as carbohydrates, fat, protein, omega-3, zinc, and calcium may result in stunting and low intellectual ability in children. Fulfilling the daily nutritional needs becomes absolutely necessary to help children grow, stay healthy, and have high intellectual ability, particularly between the ages of one and three (toddler stage)^{4,5}. The growth and development period is critical and occurs rapidly when children are under five years old and subsequently decelerates as they reach the age of starting school. For that reason, the nutritional fulfilment of toddlers must be a priority^{6,7}.

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Amerta

Nutritional fulfillment can be achieved by consuming local foods, such as bilih fish and buffalo milk. Bilih fish is a local food in West Sumatra⁸. The nutritional content of fresh bilih fish per 100 grams includes energy ranging from 85 to 132 Kcal, protein ranging from 13.02% to 17.33%, fat ranging from 0.2% to 4.62%, carbohydrates at 3.48%, ash content at 1.52, and water content ranging from 76.44% to 77.47%. Many researchers have examined the micronutrients contained in bilih fish. Per 100 grams, bilih fish contains zinc ranging from 4.76 to 17.33 milligrams, calcium ranging from 22 to 1128.05 milligrams, iron at 34.7 milligrams, vitamin A at 129.3 RE, vitamin E at 0.88 milligrams, phosphorus at 1.20 milligrams, omega-3 fatty acids at 0.553 grams, DHA at 0.237 grams, EPA at 0.207 grams, omega-6 fatty acids at 0.252 grams, and AA at 0.096 $grams^{9,10,11,12}.$ Bilih fish contains higher levels of zinc and calcium than other fish, such as mackerel, tuna, carp, tilapia, catfish, anchovies, walking catfish, snakehead, and bullet tuna9. Additionally, according to the research conducted by Han et al. (2012), pure/fresh milk produced from buffalo contains various nutrients that are beneficial for humans. The nutritional content of buffalo milk consists of 74.2-4.6% protein, 8% fat, 4.64% lactose, 92% calcium, 38% iron, 118% phosphorus, strongly high vitamin A, and low cholesterol (0.65 mg/g)¹³.

The utilisation of the potential of omega-3 fatty acids, calcium, and zinc in bilih fish (Mystacoleucus padangensis) can take the form of direct consumption or through product development. The development of fish flour-based processed food has been conducted by Widodo et al. (2015) and Nugraha (2012). Their studies indicated that the nutritional status of toddlers who were given biscuits made from snakehead fish flour and catfish flour had an impact on improving the nutritional status of the children ^{14,15}. The research conducted by Diana et al. (2019) suggested that the development of a local product with the addition of 20 grams of bilih fish flour to one formula of chocolate-flavoured biscuit dough significantly influenced the increase in tail length, Y-Maze score, brain volume, and the number of hippocampal neuron cells (CA4 and DG) in experimental rats. Rats are the animals used for samples for toddlers^{11,12,13,14,15}.

Bilih fish contains nutrients such as omega-3, calcium, and zinc, which are essential for the growth and development of children. Omega-3 plays a vital role in children's brain development and intelligence4. Meanwhile, calcium can help in bone growth and the development of children. Calcium deficiency can inhibit a child's growth. Zinc is an essential mineral for the and breakdown of macronutrients synthesis (carbohydrates, fat, protein) and cell components (nucleic acids). Additionally, zinc functions in cell regeneration, metabolism, the repair of body cells, and growth¹⁵. The deficiency of these nutrients impacts growth and developmental disorders in children, such as stunting and others¹⁶.

Various alternative methods can be implemented to improve the growth and nutritional status of children through interventions using local food ingredients with supplementary feeding (PMT)⁹. There are various products to be used as supplementary food for children, such as nuggets. Basically, nuggets are made from ground meat (chicken, beef, etc.) and frozen to maintain their quality during storage¹⁵. Many innovations have been made to create various nugget variants¹⁷. Milk nuggets are made from buffalo milk as a substitute for sardines, meat, and other small fish consumed whole, and it is also a good source of calcium¹⁸.

Fish has organoleptic properties that are less favoured by children. Therefore, to enhance children's preference for fish, it is essential to produce local fishbased food products, i.e., nuggets. Nuggets have several advantages: they are easy to make, have a longer shelf life, are easy to store, are affordable, safe, and preferred and accepted by children. Bilih fish can be potentially made into various processed products to address growth problems and stunting. The developed fish products may include bilih fish biscuits, sticks, and syrup 9,10,11. Thus, it is crucial to evaluate their agro-socioeconomic aspects, focusing on the production of buffalo milk-based nugget products with the addition of bilih fish as a local food addition to support the nutrition of toddlers. The research objectives include (1) the production of buffalo milk-based nugget products with bilih fish, (2) the selection of the best formula based on the organoleptic test, and (3) the analysis of nutritional substances (proximate, zinc, and calcium) in the buffalo milk-based nugget formula with bilih fish as a supplementary food to support the growth and development of children aged 1-3 years. The study aims to provide valuable information, insights, and innovations regarding the utilisation of local food ingredients with the potential to support the growth and development of toddlers.

METHODS

Design, Location, and Time

The study, which employed an experimental design, was conducted from April to September 2022. The nugget production and organoleptic tests were undertaken at the Culinary Laboratory of the Department of Nutritional Science, Faculty of Public Health, Universitas Andalas. The nutritional content of the nuggets was analysed at the Laboratory of Animal Product Processing Technology, Faculty of Animal Science, and the Laboratory of the Faculty of Engineering, Universitas Andalas.

Ingredients and Tools

The research materials included bilih fish, buffalo milk, wheat flour, tapioca flour, bread flour, cooking oil, eggs, and margarine. The tools consisted of stainless steel bowls, a saucepan, a cutting board, a knife, a blender, a strainer, a plastic spatula, a measuring spoon, a scale, a measuring cup, tissues, an oil filter, a wok, frying spatula, thermometer, packaging, and molds. The organoleptic tests (hedonic and quality hedonic tests) used questionnaires, pens, serving plates, and tissues. Meanwhile, for nutritional analysis, the following tools were used: blender, porcelain cup, desiccator, clamps, spoon, oven, analytical balance, sieve, combustion furnace, Kjeldahl flask, soxhlet, electric heater, filter paper, fat thimble, and Inductively Coupled Plasma (ICP).

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Formulas for Buffalo Milk-Based Nuggets with the Addition of Bilih Fish

These formulas involved the addition of four levels of Bilih Fish: F0 = standard formula (buffalo milkbased nuggets without bilih fish), F1 (buffalo milk-based nuggets+10 g bilih fish), F2 (buffalo milk-based nuggets+30 g bilih fish), and F3 (buffalo milk-based nuggets+50 g bilih fish). The nuggets' characteristics were identified by conducting the organoleptic test (hedonic and quality hedonic tests) with two repetitions. The research formula is presented in Table 1.

Table 1. Formulas for Buffalo Milk-Based Nuggets with the Addition	າ of Bilih Fish
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	Weight of Ingredients per Formula					
Type of Ingredients	FO	F1	F2	F3		
	(0%)	(5%)	(15%)	(25%)		
Bilih Fish (g)	0	10	30	50		
Buffalo milk (g)	200	200	200	200		
Margarine	3	3	3	3		
Wheat flour (g)	50	50	50	50		
Tapioca Flour (g)	50	50	50	50		
Bread Flour (g)	20	50	50	50		
Chicken Eggs (g)	11	11	11	11		

F0 = The standard formula (buffalo milk-based nuggets + 0 g of bilih fish), F1 (buffalo milk-based nuggets + 10 g of bilih fish), F2 (buffalo milk-based nuggets + 30 g of bilih fish), and F3 (buffalo milk-based nuggets + 50 g of bilih fish).

The organoleptic test involved 30 semi-trained panellists to test the organoleptic characteristics of the buffalo milk-based nuggets. The panellists were students of the Nutritional Science study program, Faculty of Public Health, Universitas Andalas, who had learned the materials and/or attended the training on the organoleptic test before. To be eligible as the panellist, the following criteria should be satisfied: healthy, willing to be the panellist, having no colour vision deficiency, and being committed to not being hungry during the organoleptic test undertaken. Before undertaking the test, equalising perception with the panellists was conducted.

The panellists assessed the smell, colour, texture, and taste of the four levels of nugget formula (Table 1). To evaluate their overall liking, each panellist adopted the 7-point hedonic scale to assess the colour, smell, texture, and taste, stated as 1 = dislike very much, 2 = dislike moderately, 3 = dislike slightly, 4 = neither like nor dislike, 5 = like slightly, 6 = like moderately, and 7 = like very much. Likewise, the nuggets' hedonic quality (colour, smell, taste, and texture) was also evaluated using the 7point hedonic scale, where for the colour attribute, 1 = extremely greyish-white - 7 = golden yellow; for the smell attribute, 1 = extremely rancid - 7 = extremely aromatic; for the taste attribute, 1 = extremely tasteless - 7 = extremely savoury and; for the texture attribute, 1 = extremely tough - 7 = extremely tender. Meanwhile, the nugget formula was assessed based on the score resulting from the organoleptic test to analyse its nutrition afterwards. The best nugget formula was selected following the highest score generated by other formulas after F0 (standard formula)¹⁹.

Nutrient Analysis

The nutrient analysis was conducted by testing the proximate, calcium, and zinc. The proximate analysis, which included water, ash, protein, and fat, and the calcium and zinc analysis were executed by adopting methods introduced by AOAC. In this case, the analysis of protein, fat, carbohydrates, energy, water, ash, as well as calcium and zinc was carried out, respectively, by employing the kjeldahl, soxhlet, by different, by calculation, oven, gravimetry, and Inductively Couple Plasma (ICP) methods²⁰.

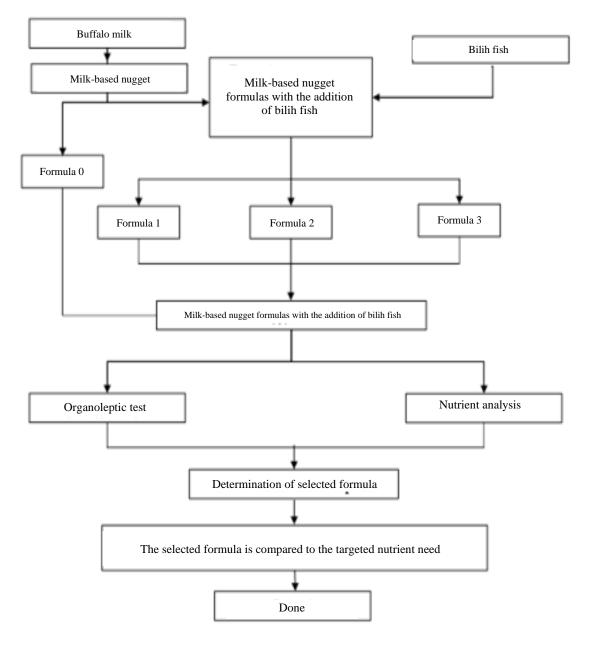
Data Analysis

The experimental design of the nugget formulas utilised the Completely Randomised Design (CRD). To analyse the organoleptic test, we employed the Kruskal-Wallis test with the Mann-Whitney test with a 95% confidence interval (α =0.05)²¹. The research flow is provided in the following figure.

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RESULTS AND DISCUSSION

The Formula Development of the Buffalo Milk-based Nuggets with the Addition of Bilih Fish

Fish nuggets are a fishery product processed using at least 30% crushed fish meat, flour, and other ingredients and then cooked²². In this research, the

nuggets were made in four levels of the bilih fish addition, including 0 g, 10 g, 30 g, and 50 g. Table 1 and Figure 1 present the results of producing buffalo milk-based nuggets with the addition of bilih fish made in several formulas.

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Figure 2. Buffalo milk-based nuggets with the addition of bilih fish

The Organoleptic Test of Buffalo Milk-Based Nuggets with the Addition of Bilih Fish

The organoleptic test involves a sensory evaluation of taste, colour, smell, texture, and overall quality utilising the five senses. There are two organoleptic test results, i.e., hedonic test and hedonic quality. The organoleptic test results of this study are presented in Tables 2 and 3.

Taste

The key factor that determines the acceptance of products is taste (Awashi et al. 2012). The hedonic test result demonstrated that the average score of the panellists' preference for the nuggets' taste was within the range of 5.00–6.00 (neither like nor dislike–like moderately). The highest score for the taste (like moderately) was given to standard nuggets F0, F1, and

F2, and the lowest score (neither like nor dislike) was for F3. The Kruskal-Wallis test result suggested that the difference in the addition of bilih fish had a significant effect (p<0.05) on the panellists' level of preference for the nuggets' taste. The result of the Mann-Whitney posthoc test indicated that the preference level for the taste of FO nuggets was significantly different from F2 and F3 but not significantly different from F1. The highest level of bilih fish addition reached 25% (50 grams), decreasing the panellists' preference for the nuggets. The more bilih fish was added, the stronger the fish taste in the nuggets was. It is in line with a study conducted by Restu (2012), suggesting that the taste of nuggets was affected by the level of fish added.²⁴ Moreover, the taste of nuggets was influenced by the use of various raw materials, the making process, and the additional main ingredients, such as salt and so on²⁵.

Table 2. Average hedonic test results of buffalo milk-based nuggets with the addition of bilih fish among the students of the Department of Nutritional Science, Faculty of Public Health, Universitas Andalas.

Treatment	Param	eter for Hedo	Tatal		
Treatment	Colour	Smell	Taste	Texture	Total
FO	7.00ª	6.00ª	6.00ª	6.00 ^a	25.00 ^a
F1	6.00 ^b	6.00ª	6.00ª	6.00 ^a	24.00ª
F2	5.00 ^c	6.00 ^b	6.00 ^b	6.00 ^a	23.00 ^a
F3	4.00 ^d	6.00 ^b	5.00 ^b	6.00 ^a	21.00ª
p-value	0.000	0.002	0.015	0.092	0.392

Numbers in different letters in the same column indicate a significant difference (p<0.05) with the Mann-Whitney test. Sensory scores (1–7) include the colour scores (1 for extremely greyish-white to 7 for golden yellow), smell (1 for extremely rancid to 7 for extremely aromatic), taste (1 for extremely tasteless to 7 for extremely savoury), texture (1 for extremely tough to 7 for extremely tender). F0 = the standard formula (Buffalo Milk-Based Nuggets + 0 g of Bilih Fish), F1 (Buffalo Milk-Based Nuggets + 10 g of Bilih Fish), F2 (Buffalo Milk-Based Nuggets + 30 g of Bilih Fish), and F3 (Buffalo Milk-Based Nuggets + 50 g of Bilih Fish).

Colour

Colour was the first sensory element seen directly by the panellists²⁶. The hedonic test results showed that the average score of the panellists' preference for the nuggets' colour ranged between 4.00 and 7.00 (neither like nor dislike–like very much). The highest score for the colour (like moderately) was given to F1 nuggets, and the lowest score (dislike slightly) was for F3 nuggets. The Kruskal-Wallis test results demonstrated that the difference in the addition of bilih fish significantly affected (p<0.05) the panellists' preference scores for the colour of nuggets containing bilih fish. The results of the Mann-Whitney test suggested that the panellists' preference scores for the F0 nuggets' colour differed from those of F1, F2, and F3 nuggets. The addition of bilih fish changed the product colour, ranging from extremely greyish-white to golden yellow, thereby increasing the panellists' preference for the selected product.

Smell

Smell is a sensory element crucial for food selection. The hedonic test results indicated that the average score of the panellists' preference for the smell of the buffalo milk-based nuggets with the addition of bilih fish ranged within 6.00 (like moderately). The highest score for the smell (like slightly) was given to standard nuggets F0. F1, F2, and F3. The Kruskal-Wallis test results revealed that the difference in the addition of

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bilih fish flour significantly affected (p<0.05) the panellists' preference scores for the nuggets' smell. The results of the Mann-Whitney test suggested that the preference level for the smell of F0 nuggets was significantly different from F2 and F3 nuggets but not significantly different from F1 nuggets.

Texture

The determinant of the taste of a product is its texture²⁷. The hedonic test results indicated that average scores of the panellists' preference for the texture of buffalo milk-based nuggets with the addition of bilih fish ranged within 6.00 (like moderately). The highest score for the texture was given to all nugget formulas: F0, F1, F2, and F3. The Kruskal-Wallis test results demonstrated that the difference in the addition of bilih fish had no

significant effect (p<0.05) on the panellists' preference level for the nuggets' texture. The results of the *Mann Whitney* test indicated that the preference level for the texture of F0, F1, F2, and F3 nuggets was not significantly different. More addition of bilih fish flour, which makes the texture crispier, did not increase the preference level for the texture.

Quality Characteristics of the Buffalo Milk-based Nuggets with the Addition of Bilih Fish

Quality characteristics are the physical properties of the nugget formula. The quality of the nugget was obtained through sensory evaluation by observing the colour, bite texture, taste (savoury, sweet, flavorful), and rancid smell. The results of the hedonic quality test of nuggets are presented in Table 3.

Table 3. Average hedonic quality test results of buffalo milk-based nuggets with the addition of bilih fish among the students of the Department of Nutritional Science, Faculty of Public Health, Universitas Andalas.

	Parame				
Treatment	Golden Yellow Colour	Rancid Smell	Savoury Taste	Bite Texture	Total
FO	6.00ª	6.00ª	6.00ª	6.00 ^a	24.00ª
F1	7.00ª	6.00 ^b	6.00ª	6.00ª	25.00ª
F2	4.00 ^b	6.00c	6.00ª	6.00ª	22.00ª
F3	3.00 ^c	5.00 ^{bd}	6.00ª	6.00 ^a	20.00ª
p-value	0.000	0.018	0.920	0.587	0.392

Numbers in different letters in the same column indicate a significant difference (p<0.05) with the Mann-Whitney test. Sensory scores (1–7) include the colour scores (1 for extremely greyish-white to 7 for golden yellow), smell (1 for extremely rancid to 7 for extremely aromatic), taste (1 for extremely tasteless to 7 for extremely savoury), texture (1 for extremely tough to 7 for extremely tender). F0 = the standard formula (Buffalo Milk-Based Nuggets + 0 g of Bilih Fish), F1 (Buffalo Milk-Based Nuggets + 10 g of Bilih Fish), F2 (Buffalo Milk-Based Nuggets + 30 g of Bilih Fish), and F3 (Buffalo Milk-Based Nuggets + 50 g of Bilih Fish).

Colour

The average nugget's colour score ranged between 3.00 (pale grey) and 7.00 (golden yellow). The higher the colour quality value was, the more golden yellow the colour of the product was. This was attributed to the addition of 10 grams of bilih fish. Conversely, with a lesser addition of Bilih fish in the product, the colour appeared brighter. The Kruskal-Wallis test results indicated a significant effect of bilih fish addition on the quality of the nugget's colour (p<0.05). The results of Mann Whitney tests suggested that the quality level of F0 nugget colour was not significantly different from F1 nuggets. The increasing addition of bilih fish in the product affected the quality level of the nugget's colour, making it more greyish.

Smell

The average score of the nugget's smell ranged between 5.00–6.00 (slightly aromatic). The Kruskal-Wallis test results demonstrated that there was a significant effect of adding bilih fish on the quality level of the nugget's smell (p<0.05). The results of the Mann-Whitney tests showed a significant difference in the smell levels between F0 nuggets and F1, F2, and F3 nuggets. The increasing addition of bilih fish affects the product's smell quality, making it more rancid. This finding aligns with research conducted by Nadhifah (2021), where a higher percentage of fish addition to nuggets results in a

stronger or more concentrated smell.²⁸ It is supported by Cahyadi et al. (2018), stating that an increased concentration of fish in nuggets intensifies their smell²⁹.

The intensified smell in nuggets containing bilih fish may also be attributed to the degradation of food substances due to heat or an increase in temperature during the nugget frying process. In this process, the protein in the food ingredients is broken down into glutamic acid, increasing a distinct acidic smell and taste. Furthermore, adding garlic to the nugget batter can result in a distinctive smell because of its sulfur content³⁰. The Maillard reaction, which leads to the formation of compounds like furfural and maltol, is another reason for the intense smell in nuggets. This finding aligns with the study by Pomalingo and Misnati (2021), which suggests that adding more fish flour can diminish the product's smell because the smell of the fish masks it, thereby making the panellists tend to dislike it³¹.

Taste

The average quality level of the nugget's taste was 6.00 (savoury). Adding Bilih fish did not have an effect on the nugget's taste quality (p>0.05), as indicated by the Kruskal-Wallis test. Further, the Mann-Whitney test indicated no significant difference between F0 nuggets and F1, F2, and F3 nuggets. Increasing the addition of bilih fish does not affect the savoury taste quality of the product.

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Bite Texture

The average quality level of the nugget's texture was 6.00 (soft). Adding Bilih fish did not have an effect on the nugget's bite texture quality (p>0.05), as indicated by the Kruskal-Wallis test. Further, the Mann-Whitney test indicated no significant difference between F0 nuggets and F1, F2, and F3 nuggets. Increasing the addition of bilih fish does not affect the bite texture quality of the product. According to Amalia and Kusharto (2013), hardness texture measurement is determined by a higher hardness score, indicating a decrease in the product's tenderness level³².

Nutrient Content of Buffalo Milk-Based Nugget Formulas with the Addition of Bilih Fish

Table 4. Results of Proximate, Calcium, and Zinc Analysis per 100 Grams of Buffalo Milk-Based Nugget with the Addition of Bilih Fish Compared to the Nutritional Adequacy Rate for Children Aged 1–3 Years Old in 2019

Parameter	Treatment				Nutritional Adequacy Rate for Children	
	FO	F1	F2	F3	Aged 1–3 Years Old	
Water Content (%)	43.97	46.80	46.90	48.03	-	
Ash Content (%)	1.80	2.32	2.54	3.07	-	
Protein (g)	10.85	11.77	12.13	13.08	Protein (g)	20
Fat (g)	6.97	7.60	7.94	8.52	Fat (g)	44
Carbohydrates (g)	36.41	31.51	30.49	27.300	Carbohydrates (g)	215
Calcium (mg)	5.25	5.52	5.97	6.20	Calcium (mg)	650
Zinc (mg)	16.18	16.63	17.15	17.48	Zinc (mg)	3
Energy (kcal)	251.77	241.52	241.94	238.2	Energy (kcal)	1,350

Nutritional Adequacy Rate. F0 = The standard formula (Buffalo Milk-Based Nuggets + 0 g of Bilih Fish), F1 (Buffalo Milk-Based Nuggets + 10 g of Bilih Fish), F2 (Buffalo Milk-Based Nuggets + 30 g of Bilih Fish), and F3 (Buffalo Milk-Based Nuggets + 50 g of Bilih Fish).

Source: The Ministry of Health, 2019

Water Content

Based on the study results, the water content in nuggets tended to increase in F0 to F3 nuggets. The lowest water content was observed in F1 nuggets (43.97%), while the highest was in F3 nuggets (48.03%). The water content increased with the increasing proportion of bilih fish added to the nuggets. Stirring the batter while blending the nugget ingredients also affected the water content. Buffalo milk (200 grams), one of the main ingredients in the nuggets, contained water (82.8%), serving as a dispersing agent for solid components present in milk. It also acted as a solvent for milk constituents and remaining solid materials such as macro- and micronutrients, as well as other compounds²⁵. Cahyadi et al., 2018 revealed that the water content of nuggets tends to increase with the proportion of fish added to the nuggets.²⁹ Based on SNI 7758: 2013 concerning fish nuggets, the maximum water content should be 60%, and the fish nuggets studied in this research have satisfied this standard²².

Ash Content

The increase in ash content in this research was indicated in F0 and F3 nuggets. Compared to F2 and F3 nuggets, F1 nuggets contained the lowest ash content (1.80%). Meanwhile, F3 nuggets had the highest ash content (3.07%). Ash refers to the residue from burning organic matter, which produces inorganic matter³⁴. The ash content is an indicator determining the mineral quality and content in an ingredient. Food ingredients with a high ash content will have a high mineral content.²⁹ The increase in the ash content of the nuggets in this research was due to the addition of bilih fish and other ingredients, such as wheat flour, chicken eggs, and sugar, which contributed to the ash content due to the minerals contained therein³⁴.

Protein

The protein contained in F0 to F3 nuggets was observed to increase. The highest protein content was 13.08% in F3 nuggets, while the lowest protein was 10.85%. This result aligns with research by Cahyadi et al. (2018), suggesting that the addition of freshwater fish to nuggets increases the protein content²⁶. The protein content of bilih fish is relatively high; thus, it can be an alternative source of protein, mainly for the growth and development of children and teenagers. Based on the 2019 Nutritional Adequacy Rate, the protein needs for toddlers, or children aged 1-3, is 20 grams. Meanwhile, one serving of nuggets (±25 grams) is equivalent to 14.7% of protein needs. Protein, unlike carbohydrates and fat, is a macronutrient containing nitrogen elements, and it is also the source of essential amino acids required by the body³⁵.

Fat

The highest fat content of nuggets in this research was observed in F3 nuggets (8.52%), while F1 nuggets indicated the lowest fat content (6.79%). Bilih fish is known as a source of unsaturated fatty acids that have a positive effect on degenerative diseases.¹⁸ Frying nuggets with cooking oil may increase their fat content. A statistical test proved that the addition of bilih fish did not indicate a significant effect on the fat content of the produced nuggets. According to SNI 7758:2013, the maximum fat content for fish nugget products cooked by frying is 15%, while the fat content identified in the nuggets of this research ranged from 6.79% to 8.52%. Therefore, the fat content of all nugget formulas met the SNI requirement²².

Calcium

In this research, F0 nuggets were observed to contain the lowest calcium content (5.25 milligrams),

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Nutrition

while F3 nuggets had the highest calcium content (6.20 milligrams). In other words, the calcium content would increase in line with the increase in bilh fish content in each formula in this research. This finding is in harmony with that reported by Siswanti et al. (2017), suggesting that the higher the concentration of fish flour added to a product, the higher the calcium content of the product³⁶. In this research, apart from the buffalo milk, the basic ingredients used that contained calcium were eggs and margarine. A statistical test on this research proved that the addition of bilh fish did not have a significant effect on the calcium content of the nuggets.

Zinc

The zinc content observed in all nugget formulas in this research indicated an increase. F3 nuggets were found to contain the highest zinc (17.48 mg/100 g of nuggets). The greater the percentage of bilih fish, the higher the zinc content in the nuggets. This finding aligns with research conducted by Steward (2015), which demonstrates the essential role of zinc in human growth, DNA synthesis, IGF-1 receptors, IGF-1 activators, and bone growth.³⁷ Foods rich in protein often contain substantial zinc levels (Table 4).

Based on the aforementioned finding, the most optimal formula was F1 (buffalo milk-based nuggets + 10 g bilih fish), with the highest overall hedonic test score of 24.00%. The nutrient analysis confirmed that this formula has complied with the relevant SNI standard. A 100-gram serving of F1 nuggets comprised four nuggets, each weighing 25 grams. When consumed as an alternative supplementary food to meet the daily energy requirements of toddlers based on the 2019 Nutritional Adequacy Rate, it is recommended to consume six nuggets per day, equivalent to 1350 kcal, to fulfil their energy needs.

CONCLUSIONS

The formulas of the buffalo milk-based nuggets were developed through four different treatments: FO (buffalo milk-based nuggets with 0 g bilih fish), F1 (buffalo milk-based nuggets + 10 g bilih fish), F2 (buffalo milkbased nuggets + 30 g bilih fish), and F3 (buffalo milkbased nuggets + 50 g bilih fish). The most optimal formula identified in this study was F1, characterised by a golden yellow colour, a subtle fish aroma, a savoury taste, and a soft texture. F1 nuggets contained water content (46.90%), ash content (2.32%), protein (11.77%), fat (7.60%), calcium (5.52 milligrams), and zinc (16.63 milligrams) per 100 grams of nuggets. Future research is recommended to develop this formula through baking to reduce water content without elevating fat content, thereby following the relevant SNI standard and allowing household-scale production. Further development of this nugget formula should address water and fat content in F1 to comply with quality standards for nuggets as supplementary food, ensuring adherence to the criteria stipulated in the Regulation of the Minister of Health of the Republic of Indonesia No. 51 of 2016 on standards for nutritional supplementation products. Additionally, it is advisable to analyse other nutrients, such as carbohydrates and other micronutrients, to facilitate further preclinical and clinical research aiming to

determine the potential impact of buffalo milk-based nuggets with the addition of bilih fish on toddlers' growth and development.

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Conflict of Interest and Funding Disclosure

The authors declare that they have no conflicts of interest related to this article.

Author Contributions

HY: writing-original draft; FMD dan HH: supervision, writing-review and editing.

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