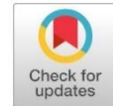




# Development of catfish sausages fortified with duck eggs as a potential nutritional intervention product to reduce stunting rates in toddlers

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## ABSTRACT

**Background:** Stunting among toddlers is a significant public health issue in Indonesia, with a high prevalence in various regions, including Tegal City. One of the efforts to reduce stunting is through nutritional interventions based on animal protein that is easily available and has high nutritional value.

**Objective:** This study aimed to develop catfish sausage fortified with duck eggs as a local nutrition intervention to improve children's nutritional status.

**Materials and Methods:** The research was an experimental study using a completely randomized design with three formulations (S1 had 1 egg, S2 had 3 eggs, and S3 had 5 eggs), each repeated three times. The formulations adhered to SNI 3820:2024 standard requiring minimum 75% meat content. Parameters analyzed included protein, fat, moisture content, pH, texture, and amino acid profile. Statistical analysis using one-way ANOVA was applied at 5% significance level.

**Results:** Sausages with duck egg fortification had protein content of 14.55-15.62% and fat content of 5.27-9.08%. Sample S2 showed the highest protein (15.62%), optimal texture (hardness 8N, chewiness 4N), and stable pH (6.2) meeting SNI standards. Amino acid analysis revealed significant levels of lysine (1.85%), histidine (1.24%), and leucine (2.95%).

**Discussion:** Duck egg fortification significantly improved nutritional quality. A 50g serving provides 60-70% of daily protein needs for children aged 1-3 years. Essential amino acids play crucial roles: lysine supports bone growth and calcium absorption, histidine aids tissue repair and immune function, while leucine activates mTOR pathway for muscle protein synthesis. Sample S2 demonstrated the most balanced formulation with optimal protein enrichment while maintaining desirable texture and microbiological stability during cold storage.

**Conclusion:** Catfish sausage fortified with duck eggs has potential as an effective local wisdom-based nutritional intervention for stunting reduction in Indonesia.

**Keywords:** Sausage; catfish; duck egg; protein; stunting

## BACKGROUND

Various developing countries, including Indonesia, face significant nutritional challenges in young children, particularly stunting. Data from the Indonesian Ministry of Health indicates that in 2018, approximately one-third of the nine million children in Indonesia were affected by stunting. Malnutrition is closely linked to stunting in toddlers in developing countries, as prolonged malnutrition impacts the physical and psychological growth and development of children, which can be measured through height and weight<sup>1</sup>.

In Indonesia, stunting is a serious concern due to its high prevalence, with 21.6% of children under five years old affected, according to the 2021 Indonesian Nutritional Status Survey.<sup>2</sup> In Central Java, the stunting rate reached 20.8%, while in Tegal City, it stood at 16.8%, which is still above the national target of 14% projected in the 2020-2024 National Medium-Term Development Plan (RPJMN). The government has implemented a stunting reduction program through the Supplementary Feeding Program (PMT) at the national level. This program includes the distribution of fortified biscuits for pregnant women and toddlers, with a focus on malnourished children.

Clinical and non-clinical studies have been conducted to examine appropriate supplementation interventions for stunted populations worldwide. Micronutrient and vitamin supplementation has been provided directly to toddlers through tablets, supplement drops, and fortified foods.<sup>3</sup> Other studies indicate that insufficient protein intake, particularly during the first 1,000 days of life, is one of the primary causes of

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stunting<sup>4</sup> Nutritional interventions that include providing protein- and vitamin-rich foods to pregnant women and toddlers are crucial to preventing stunting and its long-term impacts.<sup>5</sup>

Recent data shows that dietary diversity has a significant correlation with the prevalence of stunting. Children from households with low dietary diversity scores are more likely to experience stunting compared to those with more diverse diets.<sup>6</sup> Furthermore, poor complementary feeding practices increase the risk of stunting by up to 1.72 times.<sup>7</sup> Statistics from Indonesia also reveal that the prevalence of stunting is higher in coastal areas compared to other regions, due to economic factors, limited food access, and lower sanitation quality.

One of the abundant local food sources to enhance dietary diversity in coastal areas is catfish (*Clarias spp.*). Catfish contains high protein levels, ranging from 16-18%<sup>8,9</sup> making it a potential ingredient to support nutritional interventions. Processed products such as catfish sausages have high protein content, and fortification with nutrient-dense ingredients like duck eggs offers additional nutritional value. Duck eggs contain higher levels of protein and lipids than chicken eggs and are rich in essential amino acids such as lysine and methionine, which support growth and tissue development.<sup>10</sup> They also provide important vitamins (A, D, and B12) and minerals (iron and selenium) that help prevent anemia and support child development. Supplementation with eggs has been shown to effectively and economically reduce stunting prevalence among children. Therefore, incorporating duck egg fortification into catfish-based products could be a sustainable and cost-effective strategy to enhance nutrition and reduce stunting in coastal communities.<sup>11</sup>

This study aims to develop an innovative, locally sourced catfish (*Clarias spp.*) sausage fortified with duck eggs as a high-protein food product for stunting prevention and nutritional improvement. The product is designed to be a practical and affordable solution for meeting toddlers' nutritional needs while reducing the prevalence of stunting in Indonesia. This innovation integrates high nutritional content, such as protein with complete amino acids to support toddler growth, with flavors tailored to children's preferences. Additionally, the sausages are designed to have good storage stability and balanced fat content. Given these benefits, the product is expected to serve as an effective local wisdom-based nutritional intervention to support the growth and development of toddlers in areas with high stunting rates.

## MATERIALS AND METHODS

### Materials

Catfish that had been filleted and obtained from fish farms in Tegal City, finely ground, SNI-certified salt, ground pepper, nutmeg powder, flavor enhancers, garlic powder, SNI-certified instant coconut milk, cornstarch, wheat flour, skim milk, and duck eggs sourced from farmers in Tegal City. Test materials: aquadest p.a, sulfuric acid (H<sub>2</sub>SO<sub>4</sub>) Hi-Media, copper sulfate (CuSO<sub>4</sub>) Hi-Media, sodium hydroxide (NaOH) Oxoid, boric acid (H<sub>3</sub>BO<sub>3</sub>) Oxoid, diethyl ether Oxoid, buffer solution Hi-Media, dry tissues, and Whatman filter paper.

### Methods

The research was designed as an experimental study using a completely randomized design (CRD) with three treatments and three replications. The treatments consisted of different levels of duck egg fortification in catfish (*Clarias spp.*) sausage formulations: S1 (1 egg), S2 (3 eggs), and S3 (5 eggs). Each treatment represented a distinct proportion of duck egg addition to the base sausage formulation, which followed the SNI 3820:2024 standard requiring at least 75% meat content. All samples were prepared under identical processing conditions, including grinding, mixing, filling, and cooking, to ensure consistency. The study was conducted in several stages: (1) production of catfish sausages according to the specified treatments, (2) analysis of protein, fat, and moisture contents, (3) texture profile analysis to determine product quality, (4) pH measurement as an indicator of shelf-life stability during cold storage at 4°C for seven days, and (5) amino acid profiling of the best-performing sausage. Protein and fat contents were analyzed using the Kjeldahl and Soxhlet methods, respectively, following AOAC (2019) procedures.<sup>12</sup> Texture analysis was performed using a Texture Analyzer (TA.XT Plus) with a two-bite compression test, while pH measurements were taken periodically throughout storage. The best sausage formulation was selected based on the highest protein content, desirable texture parameters, and stable pH values during storage, then subjected to amino acid analysis using High-Performance Liquid Chromatography (HPLC). All quantitative data were analyzed statistically using one-way ANOVA to determine significant differences among treatments at a 5% significance level, followed by Tukey's HSD test for post-hoc comparison. This design enabled a

comprehensive evaluation of the physicochemical and nutritional quality of catfish sausages fortified with varying levels of duck eggs to identify the optimal formulation for stunting intervention.

### **Preparation of Catfish Sausage**

The process of making catfish sausages fortified with duck eggs was based on the SNI 3820:2024 standard,<sup>13</sup> which requires the meat content to be no less than 75% of the total ingredients. The recipe included 250 g of finely ground catfish fillet, 5 g of salt, 5 g of ground pepper, 5 g of nutmeg powder, 11 g of flavor enhancer, 5 g of garlic powder, 65 mL of coconut milk, 25 g of cornstarch, 37.5 g of wheat flour, and 37.5 g of skim milk. Duck eggs were used as a fortification ingredient: 1 egg for sample S1, 3 eggs for sample S2, and 5 eggs for sample S3.

### **Protein Testing**

Protein testing (Kjeldahl method) was conducted according to AOAC (2019).<sup>12</sup> The procedure involved weighing 0.5 g of the sample and placing it into a Kjeldahl flask, followed by the addition of 2 mL of concentrated sulfuric acid (H<sub>2</sub>SO<sub>4</sub>) and 0.9 g of selenium oxychloride (SeOCl<sub>2</sub>) as a catalyst. The solution was then digested for 45 minutes. After digestion, 40 mL of distilled water was added. The solution was distilled, and the distillate was collected in an Erlenmeyer flask containing 15 mL of 4% boric acid (H<sub>3</sub>BO<sub>3</sub>) solution and a few drops of methyl blue (MB) and methyl red (MM) indicators. The solution was titrated with 0.02 N HCl until the color changed to gray. A blank determination was also performed. Statistical analysis using one-way ANOVA was applied to compare protein content across the different samples.

### **Fat Testing**

Fat content was measured using the Soxhlet method AOAC (2019).<sup>12</sup> A 5 g sample was wrapped in filter paper and placed in a Soxhlet extraction apparatus along with diethyl ether as the solvent. The mixture was refluxed for 6 hours until the solvent returning to the fat flask became clear. The solvent in the fat flask was then distilled, and the flask containing the extracted fat was heated in an oven at 105°C until all the solvent evaporated. After cooling in a desiccator, the fat flask was weighed until a constant weight was achieved. Statistical analysis using one-way ANOVA was applied to compare the fat content across the different samples.

### **Texture Testing of the Product**

Texture testing was conducted to determine the desired texture quality of the sausage product using the Texture Profile Analysis method, following the reference with slight modifications.<sup>12</sup> Approximately 5 grams of sausage in cube form were tested, involving two or more compressions (two-bite test) using a TA.XT Plus Texture Analyzer.<sup>14</sup> A 35 mm cylinder probe was utilized, with a testing speed of 1 mm/s (Model TA1 Plus AMETEK-LLOYD, USA). Statistical analysis using one-way ANOVA was applied to compare the texture profile across the different samples.

### **pH Analysis as a Shelf-Life Indicator in Chillers**

pH analysis was conducted to evaluate the shelf-life feasibility of catfish sausages, following the method by AOAC (2019) with slight modifications.<sup>12</sup> The testing was performed at a chiller temperature of 4°C over a period of 7 days, with pH measurements taken periodically.

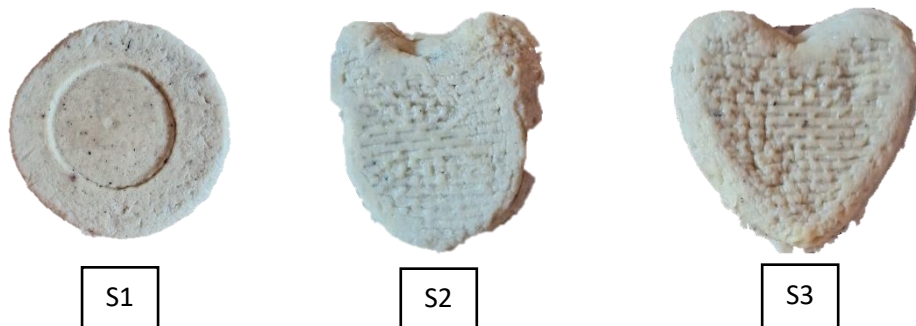
### **Amino acid testing of the best sausages**

The best sausage formulation was determined using a scoring method that integrated three key parameters: protein content, texture quality, and pH stability during storage. Each parameter was assigned a weighted score (protein content = 40%, texture quality = 35%, and pH stability = 25%) to reflect its relative importance in defining the nutritional and physicochemical quality of the product. The treatment with the highest total composite score was selected as the best-performing sausage. Subsequently, the selected sausage sample was subjected to amino acid analysis using a Shimadzu High-Performance Liquid Chromatograph (HPLC), following the method described by reference.<sup>15</sup> The analysis targeted 15 amino acids, including essential and non-essential types, to comprehensively evaluate the amino acid composition and determine the nutritional quality improvement resulting from duck egg fortification.

## **RESULTS**

### **Catfish Sausage Production Process**

The results from the first stage of testing showed that the sausages produced had visually distinct textures and shapes. Sausage S1 appeared darker compared to S2 and S3, which was attributed to its lower egg composition and denser dough structure with minimal moisture content, as shown in Table 2 and supported by texture analysis in Table 3.



**Figure 1. Character Catfish Sausages.**  
Notes: S1 (1 egg), S2 (3 eggs), and S3 (5 eggs)

### Protein and Fat Analysis

The protein and fat contents of catfish sausages fortified with varying levels of duck eggs are presented in Table 1.

**Table 1. Protein and Fat Test Results**

Sample Name	Protein (%w/w)	Fat (%w/w)
Sausage S1	14.55 ± 0.35 <sup>b</sup>	5.27 ± 0.265 <sup>c</sup>
Sausage S2	15.62 ± 0.43 <sup>a</sup>	7.38 ± 0.15 <sup>b</sup>
Sausage S3	15.31 ± 0.23 <sup>a</sup>	9.08 ± 0.085 <sup>a</sup>

Different superscript letters within the same column indicate significant differences ( $p < 0.05$ )

Statistical analysis showed that duck egg addition significantly affected ( $p < 0.05$ ) both protein and fat levels among treatments. Sausage S2 (three eggs) exhibited the highest protein content (15.62 ± 0.43%), followed by S3 (15.31 ± 0.23%) and S1 (14.55 ± 0.35%). The low standard deviation (0.23–0.43) indicates consistent processing and uniform product quality.

The fat content increased significantly ( $p < 0.05$ ) with higher levels of duck egg fortification, ranging from 5.27 ± 0.26% in S1 to 9.08 ± 0.09% in S3. The relatively low standard deviation (0.085–0.265) across treatments indicates good uniformity in fat distribution.

### Moisture Content

The moisture content of the catfish sausages is presented in Table 2.

**Table 2. Moisture Content of Catfish Sausages**

Sample Name	Moisture Content (%w/w)
S1	60.34 ± 0.02 <sup>c</sup>
S2	62.42 ± 0.03 <sup>b</sup>
S3	66.50 ± 0.01 <sup>a</sup>

Different superscript letters within the same column indicate significant differences ( $p < 0.05$ )

The highest moisture content was found in sausage S3 (66.50 ± 0.01%), followed by S2 (62.42 ± 0.03%) and S1 (60.34 ± 0.02%). The increase in moisture content corresponds with the higher proportion of duck eggs in the formulation.

### pH Levels at 4°C Chiller Temperature

The pH values of catfish sausages stored at 4°C are shown in Figure 2. Initial pH values ranged from approximately 6.7 to 7.0, with a gradual decline observed during storage. Sausage S3 reached a final pH value of 6.2, while S1 and S2 maintained pH values above 6.5.

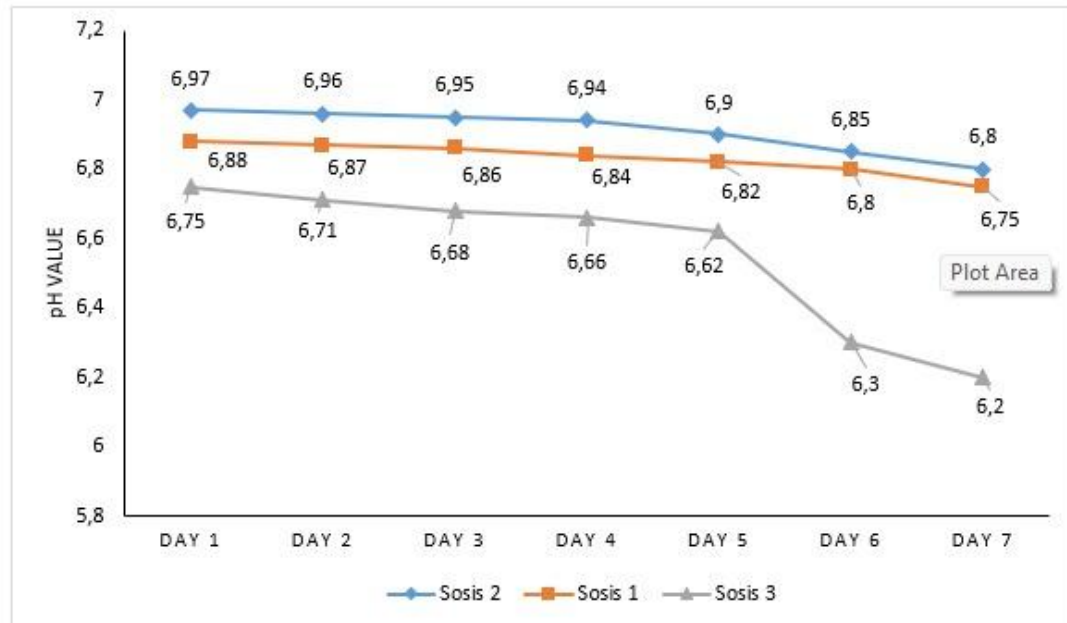


Figure 2. pH Decline in Various Samples of Catfish Sausages

### Texture Analysis of Catfish Sausages

Texture analysis results are presented in Table 3.

Table 3. Texture Test Results of Catfish Sausages

Sample Name	Hardness (N)	Chewiness (N)	Springiness
S1	10 ± 0.01 <sup>a</sup>	6.2 ± 0.01 <sup>a</sup>	0.9 ± 0.01 <sup>a</sup>
S2	8 ± 0.01 <sup>b</sup>	4 ± 0.01 <sup>b</sup>	0.8 ± 0.01 <sup>b</sup>
S3	7 ± 0.2 <sup>c</sup>	3.2 ± 0.3 <sup>c</sup>	0.7 ± 0.4 <sup>c</sup>

Different superscript letters within the same column indicate significant differences ( $p < 0.05$ )

The highest hardness value was observed in sample S1 ( $10 \pm 0.01$ ), followed by S2 ( $8 \pm 0.01$ ) and S3 ( $7 \pm 0.2$ ). The chewiness of sample S1 ( $6.2 \pm 0.01$ ) was higher compared to S2 ( $4 \pm 0.01$ ) and S3 ( $3.2 \pm 0.3$ ).

### Amino Acid Analysis of Catfish Sausages

Amino acid analysis was conducted exclusively on sausage S2. The amino acid composition of S2 is presented in Table 4.

Table 4. Amino Acid Composition of Catfish Sausage S2

Amino Acid	Molecular Weight (kDa)	Concentration (%w/w)
Aspartic Acid	133.1	3.34 ± 0.01
Threonine	119.12	1.68 ± 0.02
Serine	105.09	1.78 ± 0.01
Glutamate	147.1	5.95 ± 0.01
Glycine	75.07	1.55 ± 0.01
Alanine	89.09	1.90 ± 0.02
Valine	117.15	1.87 ± 0.00

**Table 4. Amino Acid Composition of Catfish Sausage S2(continue...)**

Amino Acid	Molecular Weight (kDa)	Concentration (%w/w)
Methionine	149.21	1.07 ± 0.00
Isoleucine	131.17	1.71 ± 0.01
Leucine	131.17	2.95 ± 0.00
Tyrosine	181.19	1.09 ± 0.01
Phenylalanine	165.19	1.83 ± 0.01
Histidine	155.16	1.24 ± 0.00
Lysine	146.19	1.85 ± 0.00
Arginine	174.2	1.90 ± 0.02

## DISCUSSION

### Catfish Sausage Production Process

The darker color in S1 may result from Maillard reactions and lipid oxidation occurring during the cooking process. Limited moisture and higher protein concentration in S1 promote non-enzymatic browning reactions between amino acids and reducing sugars, leading to the formation of melanoidin pigments that darken the product.<sup>16</sup> Additionally, lower egg content means reduced antioxidant capacity from egg yolk components such as lecithin and vitamin E, thereby enhancing lipid oxidation, which can further contribute to color darkening through the formation of oxidized pigments.<sup>8</sup> In contrast, the higher egg levels in S2 and S3 improved the product's lightness due to better emulsification, higher lipid content, and enhanced antioxidant protection from egg-derived phospholipids. Nevertheless, all formulations produced sausages that were easily molded into shapes appealing to children.

### Protein and Fat Analysis

Protein and fat are essential components in processed fish products, including catfish sausages, as they significantly contribute to nutritional value, taste, and texture. The data obtained indicate that the protein and fat content in catfish sausages are sufficient to support daily nutritional requirements, particularly for children at risk of stunting. According to WHO (2020)<sup>17</sup> and FAO (2019)<sup>18</sup>, children aged 1–3 years require approximately 13–14 g of protein and 30–35 g of fat per day to support optimal growth and brain development. Based on the analysis (Table 1), a 50 g serving of the developed catfish sausage provides around 8–9 g of protein and 4–5 g of fat, contributing approximately 60–70% of the recommended daily protein intake and 12–15% of daily fat needs for this age group. This suggests that incorporating the fortified sausage into the daily diet could meaningfully improve protein intake in children with inadequate nutrition. The use of catfish as the main ingredient provides highly digestible protein, while duck egg fortification enhances both protein quality and essential fatty acid content, particularly omega-3, which is vital for neural and physical development in stunted children.<sup>19</sup> Therefore, the developed catfish–duck egg sausage has strong potential as a complementary food to address protein–energy malnutrition and support stunting intervention programs.

The slightly lower protein level in S3 compared to S2 may be attributed to the dilution effect caused by increased egg fat and moisture, reducing the relative protein concentration per unit weight. Nevertheless, all samples demonstrated relatively high protein content, consistent with the characteristics of fish-based products made from high-protein raw materials. Catfish contains approximately 15–24% protein, of which around 42.5% comprises essential amino acids, supporting its role as an excellent protein source for nutritional intervention.<sup>20</sup> The low standard deviation (0.23–0.43) also indicates consistent processing and uniform product quality.

The variation in protein content among treatments may also be influenced by processing parameters such as mixing intensity, temperature, and duration. During heat processing, partial denaturation of fish and egg proteins can alter solubility and water-holding capacity, affecting measurable protein levels. In particular, higher temperatures may reduce extractable protein fractions in samples with more egg addition due to protein aggregation and moisture loss.

In terms of lipid composition, the fat content increased significantly ( $p < 0.05$ ) with higher levels of duck egg fortification, ranging from  $5.27 \pm 0.26\%$  in S1 to  $9.08 \pm 0.09\%$  in S3. This increase reflects the natural lipid contribution of duck egg yolk, which contains 9–10% fat, predominantly unsaturated fatty acids such as omega-3 and omega-6 that provide cardiovascular and anti-inflammatory benefits.<sup>21</sup> The relatively low standard deviation (0.085–0.265) across treatments indicates good uniformity in fat distribution. This

consistency can be attributed to the homogenization and emulsification stages during mixing, which promote stable fat–protein emulsions and prevent lipid separation during cooking. The presence of binders such as skim milk and starch also contributes to emulsion stability and uniform fat dispersion in the sausage matrix.

Overall, both protein and fat analyses confirmed that duck egg fortification significantly improved the nutritional quality of catfish sausages. Moderate egg addition (S2) yielded the most balanced composition, optimizing protein enrichment while maintaining desirable texture and fat levels. These findings highlight the importance of controlled formulation and processing parameters in achieving consistent nutrient profiles and product quality in fortified fish-based functional foods.<sup>22</sup>

### **Moisture Content**

After testing the protein and fat content in catfish sausages, the analysis proceeded with moisture content testing to evaluate the texture consistency of the catfish sausages. Table 2 shows that the highest moisture content was found in sausage S3 ( $66.50 \pm 0.01\%$ ), followed by S2 ( $62.42 \pm 0.03\%$ ) and S1 ( $60.34 \pm 0.02\%$ ). The increase in moisture content corresponds with the higher proportion of duck eggs in the formulation, as eggs contribute additional liquid and fat components to the sausage mixture. Products with higher moisture content tend to be more susceptible to microbial growth because high water activity (*aw*) creates favorable conditions for spoilage and pathogenic microorganisms. Conversely, lower moisture content combined with a lower pH value can exert a synergistic preservative effect, reducing microbial proliferation and extending product shelf life.<sup>23</sup>

Beyond formulation differences, several processing factors can also influence moisture content in sausage production. These include mixing time and intensity, which affect the water-binding capacity of proteins; heating temperature and duration, which determine the extent of water loss through evaporation; and emulsification efficiency, which influences how well water and fat are retained within the protein matrix. Additionally, the type and concentration of binders and fillers (such as starch and skim milk) can significantly affect water retention by forming a gel matrix that traps moisture during cooking. Overheating or insufficient mixing may lead to protein denaturation and emulsion breakdown, reducing the ability of the sausage to retain water. Thus, consistent processing conditions are essential to achieve uniform moisture levels and optimal product stability.<sup>24</sup>

### **pH Levels at 4°C Chiller Temperature**

pH testing in fish sausages is crucial to ensure the quality, safety, and stability of the product during storage. The pH quality testing results for various samples of catfish sausages stored at a chiller temperature of 4°C are presented in Figure 2.

The initial pH values, ranging from approximately 6.7 to 7, indicate that the fish sausages have a slightly neutral to near-alkaline pH. According to the Indonesian National Standard (SNI 3820:2015) for meat sausages, the acceptable pH value during storage ranges between 6.0 and 6.5, ensuring product safety and minimizing microbial spoilage.<sup>25</sup> When compared with this standard, sausage S3, which reached a final pH of 6.2, meets the SNI safety criteria, indicating good quality and microbiological stability. In contrast, sausages S1 and S2, with final pH values above 6.5, are relatively more susceptible to spoilage due to higher microbial activity potential. The decrease in pH during storage is influenced not only by chiller temperature but also by several other factors, such as microbial fermentation activity (especially lactic acid bacteria), oxidation of nitrogenous compounds, and biochemical reactions involving the breakdown of proteins into organic acids. Additionally, the formulation particularly the ratio of egg and fish protein can affect buffering capacity, thereby influencing the rate of pH decline. In conclusion, maintaining a final pH below 6.3 is essential for ensuring product safety and extending shelf life, and the results of this study suggest that the S3 formulation is the most stable and compliant with SNI standards for safe consumption.<sup>26</sup>

### **Texture Analysis of Catfish Sausages**

Texture testing in the production of catfish sausages is crucial, as texture is one of the primary quality indicators desired by consumers. In sausage products, good texture includes optimal levels of hardness, tenderness, elasticity, and chewability, particularly for consumption by toddlers.

Hardness refers to the maximum force required to compress a sample to a certain limit. Hardness values are often an essential indicator for evaluating the chewiness and density of materials, which are closely related to the water content and protein network within the sample. Based on the data, the highest hardness value was observed in sample S1 ( $10 \pm 0.01$ ), followed by S2 ( $8 \pm 0.01$ ) and S3 ( $7 \pm 0.2$ ). The higher hardness in sample

S1 indicates a denser tissue structure, which may be attributed to stronger myofibrillar protein binding and the formation of a more compact gel network.<sup>27</sup> This aligns with findings that show sarcoplasmic and myofibrillar protein structures play a crucial role in determining the physicochemical properties of meat-based food products.<sup>28</sup>

Chewiness measures the amount of energy required to chew a sample until it reaches a swallowable consistency. The chewiness of sample S1 ( $6.2 \pm 0.01$ ) is higher compared to S2 ( $4 \pm 0.01$ ) and S3 ( $3.2 \pm 0.3$ ). The firmer and denser texture observed in sausage S1 compared to S2 and S3 can be attributed to its higher protein to fat ratio, which significantly influences the structural and textural properties of fish sausages. Proteins play a crucial role in forming a three dimensional gel matrix during heating, where myofibrillar proteins such as actin and myosin denature and aggregate to create a compact network that traps water and fat. This network formation increases hardness, elasticity, and chewability. In S1, the lower fat content reduces the lubricating effect that usually softens the texture, resulting in greater firmness and cohesiveness. Conversely, higher fat levels in S2 and especially S3 act as fillers that interrupt the protein matrix, leading to softer, more tender textures but reduced elasticity. Fat globules weaken the gel structure by decreasing protein–protein interactions, thereby lowering hardness and chewiness. The combination of strong protein cross-linking and limited fat dispersion in S1 thus produces a more compact, elastic, and chewy texture compared to S2 and S3, which exhibit more tender and less cohesive characteristics due to their higher fat content and reduced protein network density.

Proteins in catfish sausages play a crucial role in determining the texture properties of the product. Myofibrillar proteins, which account for 55–65% of total muscle proteins, form the structural network that defines the hardness and elasticity of the product.<sup>29</sup> In contrast, the more water-soluble sarcoplasmic proteins contribute to the cohesiveness of food products, enabling the structure to be more cohesive and less prone to breaking.<sup>30</sup> The interaction between these two protein types is a key factor in creating the optimal texture for fish-based processed foods.

#### **Amino Acid Analysis of Catfish Sausages**

Amino acid analysis was conducted only on sample S2, as it demonstrated the most balanced formulation among all treatments, characterized by optimal crude protein content, desirable texture, and stable pH values. These parameters indicate that S2 possessed superior nutritional and physicochemical properties, making it the most representative sample for detailed biochemical evaluation. Considering that amino acid profiling is a complex and resource-intensive process, focusing on the best-performing formulation ensures a more efficient and scientifically relevant assessment of the protein quality in catfish sausages.

The test results showed that the amino acid content in catfish sausages is sufficient to meet daily amino acid requirements, particularly for children with stunting. A deficiency in essential amino acids can suppress protein and lipid synthesis, thereby inhibiting growth.<sup>31</sup> Amino acids play a crucial role in stunting interventions, including lysine, histidine, leucine, isoleucine, arginine, and phenylalanine.

**Lysine**, as an essential amino acid, plays a vital role in the growth and development of children. It aids in protein synthesis, particularly collagen, which is critical for bone and tissue health. Additionally, lysine supports calcium absorption in the intestines, a vital element for bone formation and remodeling.<sup>32</sup> Lysine deficiency has been shown to reduce bone mineral density and femur size, thereby affecting physical growth.

**Histidine**, as a precursor of histamine, plays a role in tissue repair and supports the immune system. Additionally, histidine contributes to the synthesis of carnosine, which acts as an antioxidant buffer in muscles and brain tissue, protecting these tissues from damage caused by free radicals and heavy metals. This function is particularly important for tissue regeneration.<sup>30</sup>

**Leucine and isoleucine**, two branched-chain amino acids (BCAAs), play critical roles in muscle protein synthesis, tissue recovery, and energy balance. Leucine, in particular, activates the mTOR (mechanistic target of rapamycin) pathway, which supports protein anabolism and accelerates tissue regeneration. These effects make it especially essential for supporting the physical growth of children, particularly those experiencing stunting.<sup>33</sup>

**Arginine**, on the other hand, not only enhances the release of growth hormones through the activation of the nitric oxide (NO) pathway but also improves blood flow and nutrient distribution to body tissues. Arginine supports the repair of damaged tissues, cell regeneration, and wound healing. Additionally, arginine plays a role in modulating immune responses, contributing to overall tissue regeneration.<sup>31</sup>

**Phenylalanine**, as a precursor of tyrosine, contributes to the synthesis of neurotransmitters such as dopamine and norepinephrine, which play crucial roles in cognitive function and the development of the



nervous system. The consumption of phenylalanine from food sources such as fish and eggs has been shown to enhance brain function and support the development of children's nervous system.

By understanding these various critical functions, nutritional needs can be formulated to support stunting interventions, as shown in Table 5. These nutritional requirements will serve as a reference for the consumption of catfish sausages, tailored to the conditions of stunting and the needs of toddlers.

**Table 5. Functions and Requirements of Amino Acids for Stunting Intervention<sup>29,33</sup>**

Amino Acid	Requirement (mg/g)	Role in Stunting Intervention
Lysine (Lysine)	48-52	Supports collagen formation, bone growth, and calcium absorption.
Histidine (Histidine)	14	Tissue repair and promotes muscle and nerve health.
Leucine (Leucine)	61-63	Muscle protein synthesis and body tissue repair.
Isoleucine (Isoleucine)	30-31	Tissue recovery and provides stable energy.
Arginine (Arginine)	30-50	Supports blood flow and growth hormone release.
Phenylalanine (Phenylalanine)	41	Produces neurotransmitters essential for cognitive and mental function.

## CONCLUSIONS

Catfish sausages (*Clarias spp.*) fortified with duck eggs have potential as a nutritional intervention food product to reduce the prevalence of stunting in toddlers. Fortification with duck eggs increases the protein and fat content of sausages, and enriches the profile of essential amino acids, including lysine, histidine and leucine, which are important for children's growth and development. Texture and pH analysis shows that this product has quite good shelf life at chiller temperatures. With appropriate formulation and nutritional content, duck egg fortified catfish sausage can be integrated as an alternative local food in efforts to deal with stunting, especially in areas with high prevalence such as Tegal City.

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