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Bioavailability of Iron in Chicken Nuggets with the Addition of Chicken Liver and Moringa Leaves

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ABSTRACT

Anemia is a global health problem caused by iron deficiency, especially among vulnerable groups such as adolescents. One of the strategies to prevent anemia is through the consumption of foods rich in iron and other supporting nutrients. Chicken nuggets are a popular processed meat product with the potential to develop into a functional food. This can be achieved by fortifying them with iron-rich ingredients, such as chicken liver and moringa leaves. This research aims to determine the bioavailability of iron in chicken nugget products containing chicken liver and moringa leaves. The production of chicken nuggets with the addition of chicken liver and moringa leaves was divided into two treatments: Control (100% chicken meat) and P (75% chicken meat: 20% chicken liver: 5% moringa leaves). The P formula was selected based on previously conducted sensory tests. Nutritional content test using the proximate test. Iron bioavailability analysis using pepsin and pancreatin enzymes with 4 repetitions. Results were analyzed using an Independent-Samples T-Test. There were significant differences for energy content (p = 0.014), carbohydrate percentage (p = 0.031), and protein percentage (p = 0.001), and no significant differences for fat percentage (p = 0.207). There was no significant difference for iron content (p = 0.184), but there was a significant difference for iron absorbed (p = 0.013) and percentage of iron bioavailability (p = 0.034). The addition of chicken liver and moringa leaves was found to significantly increase the bioavailability of iron in the product.

Key Messages:

- Chicken liver and moringa leaves contain iron, which is good for preventing anemia, especially in vulnerable groups such as adolescent girls.
- The addition of chicken liver and moringa leaves can increase the bioavailability of iron in chicken nuggets.

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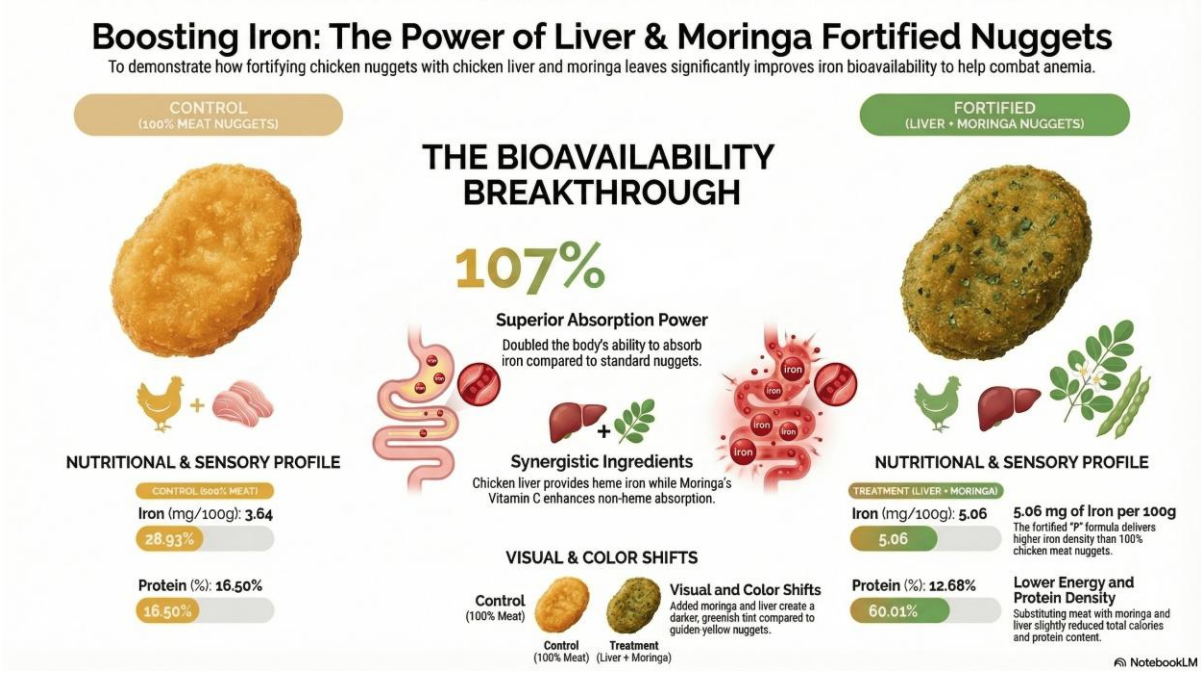
GRAPHICAL ABSTRACT



INTRODUCTION

Iron deficiency anemia remains one of the major nutritional problems in the world, especially in developing countries. The World Health Organization (WHO) estimates that more than 30% of the global population suffers from anemia, most of which is caused by iron deficiency (1)(2). Improving the bioavailability of iron in food is an important strategy in anemia prevention and control efforts.

Iron bioavailability refers to the proportion of iron that is absorbed and utilized by the body after consumption. Iron in food comes in two forms: heme and non-heme. Heme iron, which is mainly found in animal products such as chicken liver, has a higher absorption rate than non-heme iron derived from plant sources(3)(2). However, iron consumption from animal sources may not meet the body's needs, especially in vulnerable groups such as children and women of childbearing age (1).

Animal sources are the main ingredient in anemia prevention efforts. One example is chicken liver, which is known to have a high iron content of about 15.8 mg per 100 grams (4). Consumption of iron-rich animal foods, such as chicken liver, which is widely used in the food industry, can help prevent anemia due to iron deficiency. This material also has the potential to be used as a component in iron-fortified food products for people with iron deficiency anemia (5).Chicken liver is a high source of heme iron and is traditionally known to have good bioavailability.

Moringa oleifera leaves are known to be rich in micronutrients including iron, vitamin C, and other antioxidant compounds that act as enhancers of non-heme iron absorption (3)(6). The presence of vitamin C, for example, can reduce iron from the Fe^{3+} form to Fe^{2+} which is more easily absorbed in the small intestine. Therefore, the combination of chicken liver and Moringa leaves could be a synergistic strategy to improve iron bioavailability in food products.

Processed products such as chicken nuggets are very popular among various groups, including children. The development of chicken nuggets formulated with additional chicken liver and moringa leaves not only has the potential to increase total iron content, but also to improve its bioavailability. However, it is important to evaluate the extent to which the iron in these products can actually be absorbed by the body, given the presence of inhibitory factors such as phytates or tannins in plant foods that can reduce iron bioavailability(7).

Various studies have shown that fermentation methods, cooking, and food formulation can affect iron availability (3)(8) Therefore, further studies are needed on the bioavailability of iron in processed products such as chicken nuggets enriched with iron-rich ingredients and enhancers. This study aims to

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assess the potential for increasing iron bioavailability in chicken nuggets added with chicken liver and moringa leaves, with the hope of contributing to the preparation of functional food strategies for the prevention of iron deficiency anemia. The purpose of this study was to determine the bioavailability of iron in chicken nugget products with the addition of chicken liver and moringa leaves.

METHODS

Product Nugget

Nuggets were prepared using a scale, cutting board, basin, mitochiba STAR Chopper CH 250, plate, spoon, baking sheet, knife, steaming pot, freezer/refrigerator, spatula, pan, drainer, and stove.wok The main ingredients are chicken meat, chicken liver, and moringa leaves, with the addition of panir flour, eggs, wheat flour, garlic, sugar, ground nutmeg, ground coriander, salt, pepper, and oil. The process involves grinding the chicken meat and liver, mixing with the blanched moringa leaves and other seasonings, molding the dough (3 cm thick), and steaming for 30 minutes at 100°C. After cooling for 10 minutes, the nuggets were cut. The formulation for these nuggets, including the percentage of chicken replacement with chicken liver and the addition of moringa, was based on a previous study by Wibisono (9). The 'P' formulation (P1) was selected based on a previous sensory analysis.

Table 1. Preparation of chicken nuggets with the addition of chicken liver and moringa leaves

Ingredients	Control (g/%)	P (g/%)
Chicken Meat	250 (44,9)	187.5 (38,26)
Chicken Liver	0 (0)	50 (10,20)
Moringa Leaves	0 (0)	12.5 (2,55)
Flour	100 (20,4)	100 (20,4)
Egg	60 (12,2)	60 (12,2)
Fresh bread	50 (10,2)	50 (10,2)
Seasoning	30 (6,12)	30 (6,12)

Source: Wibisono (9)
Control: Chicken Nugget (Chicken Meat 100%)
P: Chicken Nugget with Chicken liver and moringa nugget formula (Percentage = Chicken Meat 75%: Chicken Liver 20%: Moringa Leaves 5%)

Proximate Examination

Proximate analysis, including energy, fat, protein, and carbohydrate content, was conducted in this study. Fat content was determined using the Soxhlet method in accordance with SNI standard No. 01-2891-1992. Measurement of protein content was carried out using the micro Kjeldahl method based on AOAC guideline No. 960.52-1961 (AOAC, 2005)(10), while carbohydrate analysis was carried out using a different method. Energy analysis in food products was not measured directly, but was calculated based on the results of proximate analysis (protein, fat, and carbohydrate content).

Examination of Iron Bioavailability in Nugget

The materials used were HCl, KH_2PO_4, Calcium, and pancreatin for buffer as well as distilled water and Pepsin. The equipment used includes a water bath (Memmert), volumetric pipettes (Pyrex), dialysis tubes (Spectra/PorP). The nugget samples to be analyzed will be thoroughly homogenized. The required nugget is 5 g. The iron bioavailability of the nugget products will be assessed using the in vitro dialysis method with 4 repetitions, which simulates the gastrointestinal digestion and absorption process. This method is based on the principles described by Miller (11) and adapted as applied in the interlaboratory study by Bueno (12) .

The homogenized nugget samples were incubated in an acidic buffer solution (pH adjusted close to gastric pH, i.e. pH 2) containing pepsin at 37°C. The incubation time is set to simulate the transit time in the stomach, which is 2 hours. After the gastric digestion phase, the pH of the mixture is raised (pH 7) to simulate the small intestine environment and pancreatin is added. Phosphat Buffer Saline (PBS) dialysis solution is used to collect dialyzable iron. Dialysis membranes will be placed in the digestion mixture.

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Incubation will continue at 37°C with constant agitation for 3 hours. After the bowel incubation period, the dialysis solution will be collected. The iron concentration in the dialysis solution will be measured using atomic absorption spectrometry (AAS) with a Bio-Rad iMark Microplate Absorbance Reader. The formula for calculating iron biovailability is as follows:

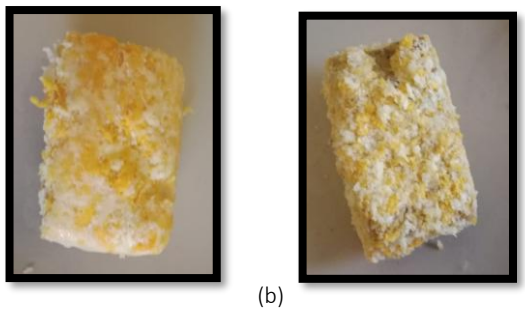
$$\textit{Iron Bioavailability (\%)} = \frac{\text{Iron Concentration in Dialysate}}{\text{Total Iron in the Initial Sample}} \times 100\%$$

Statistical Analysis

Statistical tests using the Independent t test to determine the difference between iron content in the product, iron after adding digestive enzymes (pepsin, pancreatin), and iron bioavailability between control nugget products (chicken meat) and nuggets added with chicken liver and moringa leaves. Data normality was assessed using the Shapiro-Wilk test because of the small number of repetitions. Analysis using IBM SPSS 22 software.

RESULTS

Chicken nuggets given additional chicken liver and moringa leaves have a darker color than the control. The following is a picture of chicken nuggets with chicken liver and moringa leaves.



(a) (b)

Figure 1. Chicken Nugget Products: (a) Chicken Nugget Product, (b) Chicken Nugget with the Addition of Chicken Liver and Moringa Leaf.

The macronutrient content of chicken meat and chicken liver differs. In the second formula (P), we reduced the amount of chicken meat and increased the amounts of chicken liver and moringa leaves. Based on the proximate analysis of both products, there is a significant difference in energy and protein content. The bioavailability test of iron in both products also shows a significant difference. The results of the proximate test and iron bioavailability can be seen in the following table:

Table 2. Nutritional Content and Bioavailability of Iron in Nugget Products

Nutritional Content	Control	Treatment (P)	Independent T-test
Energy	186,64 ± 0,04	156,27 ± 0,95	p = 0,014
% Carbohydrate	22.46 ± 0,59	18,40 ± 0,29	p =0,031
% Protein	16,50 ± 0,42	12,68 ± 0,16	p =0,001
% Fat	3,40 ± 0,070	3,53 ± 0,70	p =0,207
Iron (mg/100g)	3,64 ± 0,79	5,06 ± 0,91	p =0,184
Digestive Iron (mg/100g)	1,08 ± 0,55	3,012 ± 0,97	p =0,013
% Bioavailability of Iron	28,93 ± 4,33	60,01 ± 17,42	0,034

DISCUSSION

The addition of moringa leaves changed the color of the nuggets from golden yellow to greenish. This color change is in line with previous research by Prayitno and Rahim which showed that chlorophyll in moringa leaves affects the color of the product (13). In addition, another study by Nurhayati also showed that the addition of vegetables to chicken nuggets can have a significant impact on color (14). Consumers

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tend to dislike chicken nuggets with vegetables such as spinach because of their color, in contrast to nuggets with carrots or no vegetables added. The addition of only 5% (2.55 g) of moringa leaves did not cause much sensory disturbance in the respondents. Another factor that affects the color of our products is the addition of chicken liver. Since the mixing process of chicken liver is still done manually, the color also affects the final appearance of the chicken nuggets (15).

The macronutrient content of chicken meat and chicken liver has some differences. Data from the Indonesian Food Ingredients Composition Table (TKPI) in 2017 (16), explains that the energy content of chicken meat is higher than chicken liver. The results of proximate analysis conducted in the form of chicken nugget products, there is a significant difference in energy between the two groups with the control group having a higher energy content. Seong et al. (17) reported that the energy content of chicken meat will vary according to the part of the chicken itself. The percentage value of protein for each 100 grams of the sample shows that the control group has a higher protein value compared to the protein in nuggets added with chicken liver and moringa leaves and provides a significant difference. Replacing the basic ingredient of chicken meat with moringa leaves has an effect on reducing the total protein in nuggets. TKPI explains that the protein content in chicken is higher than moringa leaves (18) (16).

The percentage content of fat in the control group with treatment (P) did not show a significant difference. Seong explained that the fat content in chicken products ranges from 0.81% to 4.53%, while the fat value in both groups was 3.4% and 3.53% so there was no significant difference in the two products. There was a significant difference in the percentage of carbohydrates in the two groups. In the TKPI, it is explained that carbohydrates from chicken liver are very low, which is around 0.5 grams so that this product is not included in the carbohydrate source to meet the body's energy needs (17).

Iron content in the control group and treatment group did not differ significantly, although the treatment group had a higher iron content than the control group ($p = 0.184$). The iron content that can be absorbed after passing through several digestive enzymes will decrease. There was a significant difference in iron value between the control group and the treatment group (P), thus giving a significantly different % iron bioavailability value as well, namely for the control group 28.93% and the treatment group 60.01%. The addition of chicken liver and moringa leaves is considered to play a role in the absorption of iron in nugget products.

Chicken liver has a high iron content compared to other animal sources (19). Chicken liver also has a higher absorption capacity compared to chicken meat. Chicken liver has a bioaccessibility or bioavailability of iron around 56% while chicken meat is only 23% (20). Another study explained that oral administration of chicken liver hydrolysate in anemic rat models was shown to restore decreased hemoglobin levels, which showed a strong anti-anemia effect due to good iron bioavailability (19). The addition of chicken liver to nuggets not only increases the iron content but also its absorption. Other studies also explain that with high iron content and good bioavailability, chicken liver is often considered as a fortification ingredient in food products to prevent iron deficiency anemia (21) (22). One example is the addition of chicken liver meal in tempeh products can increase iron and protein content, which has the potential to improve iron deficiency anemia. Therefore, the addition of chicken liver has the ability to increase iron absorption from other food sources.

Moringa leaves can also affect the absorption of iron in the body. Khoja found that Moringa leaves have the highest bioaccessibility and bioavailability of iron compared to other plant sources, making Moringa leaves a potential source of iron (23). Vitamin C content in moringa leaves can affect the absorption of non-heme iron (24). Research by Merwe explained that the utilization of moringa leaves can also be done by fortification into food products (25). This is an effort to overcome iron deficiency anemia for prone or vulnerable areas such as disaster areas. The addition of moringa in this study provides the potential for adding fiber to the product. The addition of moringa leaves shows that chicken nugget formulations can increase nutritional value (26) . One strategy to make healthy nugget products is to add fiber which is beneficial for digestion and overall health (27). The combination of chicken nuggets with the addition of chicken liver and moringa leaves suggests that this product can increase the bioavailability of iron in nuggets. Future research needs to be developed by looking at the optimal temperature and time in the nugget cooking process, so that the iron in it is also optimally available.

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CONCLUSION

This study shows that the addition of chicken liver and moringa leaves to chicken nugget products can significantly increase iron bioavailability. The results of the analysis showed that the nugget formulation with the addition of 20% chicken liver and 10% moringa leaves increased the absorbable iron content and the percentage of bioavailability compared to the control nugget made from 100% chicken meat. Despite the decrease in energy, protein, and carbohydrate content due to ingredient substitution, the product still has potential as a functional food to prevent anemia. Further studies are needed on this nugget product regarding the iron status of vulnerable groups.

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CONFLICTS OF INTEREST

The authors declare no conflict of interest.

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