Original article

Development of tempe flour-enriched snacks to prevent anemia in adolescent girls

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Abstract. *Background:* Anemia is still a nutritional problem among adolescent girls around the world. Snack consumption habits among adolescent girls are increasing, even though snacks contain much energy but are low in protein and iron. Tempe, a nutritious and inexpensive indigenous food of Indonesia, steadily gaining global recognition, is rich in protein and iron. This study aimed to develop tempe flour-enriched snacks to prevent anemia among adolescent girls. *Method:* This research and development (R&D) study consists of four stages. Firstly, to determine a snack carrier through a survey of snack preferences among 356 female adolescent students of Junior High School in Tasikmalalaya City. Secondly, the product formulation uses three different percentages of tempe flour, i.e., 40% (S1), 30% (S2), and 20% (S3) in the 100g dough. Thirdly, sensory tests were carried out on 80 young women. Finally, the determination of the nutritional content (energy, protein, carbohydrate, fat, iron, and beta carotene) of the selected formula results from a sensory test. *Results:* The preference test results showed that snack made of cassava flour, salted and not fried, was chosen as "carrier" based on the most preferred by participants (90.4%). The results showed no difference in all sensory tests (taste, smell, color, texture, and overall) between the three formulas. Formula S2, with the addition of 30% tempe flour, was determined to be the chosen formula because it had the highest average of all sensory scores compared to the others. Analysis of the nutritional content of the selected recipe (S2) per 100g showed an energy content of 239.89 kcal, protein 12.6 g, carbohydrate 27.1 g, fat 9.01 g, iron 1.46 mg, and beta carotene 251.46 µg. *Conclusions and suggestions:* Adding tempe flour to cassava flour based-snack can increase protein and iron content. This makes it suitable as a food-based approach (FBA) to prevent or ameliorate anemia in adolescent girls. Clinical trials are needed to prove that Tempe flour-enriched snacks can prevent anemia in adolescent girls.

Key words: snacks, tempe, anemia, adolescents, girls.

Introduction

Adolescent girls serve as an avenue to overcome the problem of undernutrition because taking adequate care of them provides cross-generational benefits in adulthood, pregnancy, and newborns (outcomes of pregnancy) (1,2). Nutritional investment in adolescents is highly needed to measure up with the increase in the number of adolescents worldwide (3). Anemia is a nutritional deficiency in the adolescent female, which is still a public health problem in developing countries (4). Among adolescents, anemia affects not only current health status but also has adverse effects on the future. During pregnancy and childbirth, an adolescent girl who suffers from anemia is at risk of giving birth to a child with low birth weight (LBW), prematurity, and an increased risk of neonatal death (5).

Adolescent girls are prone to anemia due to increased iron requirements during rapid growth (6) and iron loss due to menstruation (7). Another cause is the low consumption of iron-rich foods with good bioavailability (8) due to the increasing habit of consuming snacks in female adolescents from time to time (9). Compared to main meals, snacks contain less protein and micronutrients such as iron (10).

The food-based approach (FBA) strategy, through increasing the consumption of foods rich in protein and iron, is an excellent approach to overcome anemi (11,12). Factors related to eating habits, food preferences, and availability are needed for the FBA strategy's sustainability (13). A combination of FBA strategies can be done by increasing the consumption of iron-rich foods, adding absorption-boosting components, and applying food processing techniques to reduce absorption barriers such as heating, germination, or fermentation (11).

Tempe or soybean cake is a fermented soybean food that is popularly known and has been part of the eating culture of the Indonesian people since the 16th century. Tempe consumption is increasing rapidly, not only in Indonesia but also in the United States and Europe. Although Tempe is difficult to export, it is consumed and produced in many countries (14). At this time, tempe has been produced globally as a highly nutritious plant food (15). Tempe made into nuggets can be used as inexpensive and nutritious food to improve protein and energy intake (16). In addition, tempe is also rich in iron, and phytic acid is degraded due to the fermentation process, thereby increasing iron bioavailability (16,17).

Increased absorption of iron also occurs due to the consumption of animal foods. Animal foods, besides containing heme iron easily absorbed by the body, its presence is also thought to stimulate iron absorption (18). Animal foods can increase the absorption of non-heme iron in plant foods and are often referred to as the Meat Fish Poultry (MFP) factor (18,19). Tilapia fish is a group of MFP containing animal protein that can help increase iron absorption (18). In addition, fish flesh is also a food rich in protein and iron (20). Beta-carotene is also a nutrient that can increase iron absorption (21,22). Pumpkin (Cucurbita moschata) is a potential source of natural provitamin A which contains 1569 μg beta-carotene per 100 grams (23). Therefore, in addition to tilapia fish, pumpkin is also a potential food to stimulate iron absorption. Another advantage is that Tilapia fish (24) and pumpkin (25) are foods that are commonly consumed by people worldwide and have low prices.

In Indonesia, 23.4% of snack foods sold in schools are traditional snacks (26). One of the raw materials commonly used to make traditional snacks is cassava flour (27). However, cassava flour does not have to be fortified with iron like wheat flour (28). Therefore, food-to-food fortification of snacks made from tempe combined with tilapia and pumpkin has the potential to increase iron in snacks made from cassava flour. Modification of this snack can be used as a means of preventing anemia in adolescent girls. Anemia in adolescent girls is still categorized as a moderate public health problem in the West Java province, which is the province with the largest population in Indonesia. The prevalence of anemia in adolescent girls in the West Java province is 32.4% (29). This study aims to develop snacks enriched with tempe flour to prevent anemia in adolescent girls.

Method

Research design

This is a research and development (R&D) study of tempe flour-enriched snack food products for adolescent girls, consisting of four stages. The first stage is the determination of "carrier," the second stage is product formulation, the third stage is the sensory test, and the fourth stage is the analysis of nutrients in the selected product from the sensory test.

Stage 1. Determination of the "carrier."

Determination of snacks that will be selected as a carrier is done by collecting data on the snack preferences in adolescents. The snacks that the participants most liked will be selected as carriers. The data collection of snack preferences used a survey method with 356 female junior high school students as participants. Participants came from six Junior High Schools in

Tasikmalaya City, West Java, Indonesia. Participants' age were as follows: 54(9.9%) 12-years, 257(47.1%) 13-years, and 235(43.0%) 14-years.

The questionnaire that used snacks as the preference survey instrument contains a list of types of snacks. The list emanated from observing snacks sold at the survey location (six junior high schools) with two possible answers: likes and dislikes. The inclusion criteria were snack and made of flour. Snacks made of flour are then differentiated based on taste (sweet and salty) and processing methods (fried and non-fried). Furthermore, snack foods made of flour are divided into four groups; 1). Sweet and fried; 2). Salty and fried; 3). Sweet and non-fried, and 4) salty and non-fried.

Data collection is carried out in class using paper-based. Before filling out the questionnaire, the subjects were explained that the answers were based on the preferences felt each. In collecting data, researchers were assisted by three research assistants.

Stage 2. Products formulation

Cassava flour, the main basic ingredient in snack foods, is enriched with tempe and other additives, i.e. Tilapia fish and pumpkin, as food for food fortification in snacks. All raw materials were purchased from the market located in the study area. The tempe is then converted into flour by cutting into small pieces, steaming for 10 minutes, drying for 5 hours at 60° C, then mashing by blending for 3 minutes and sifting. Tilapia fish weighing 200-250g per piece is cleaned, washed, filleted, and then mashed. Pumpkin is peeled and seeded, steamed, and then mashed. All the ingredients were added to the cassava flour as a basic ingredient for snack food products, and we also added seasonings (salt, pepper, and garlic). All the ingredients are stirred until thoroughly mixed, then hot water is added. The dough is then made into balls weighing 15 g per piece and steamed for approximately 15 minutes. The same person makes snacks at the same time to maintain product homogeneity. Products are served in sealed, labeled containers and warm. The manufacture of snacks is done using the three formulations based on the composition of the materials. The addition of Tilapia and pumpkin were as much as 14% and 16%, respectively, for the entire formula. While the ratio between tempe flour: and cassava flour is different, i.e. 40%:30% for snack formula 1 (S1), 30g%:40% for snack formula 2 (S2), and 20%:50% for snack formula 3 (S3).

Stage 3. Sensory test

The sensory test aims to determine the selected product from the three formulations made in the second stage. There were 80 young women aged between 12-18 years as a non-trained panelist on the sensory test. Before participating in the sensory test, all panelists who agreed to participate in the study and their parents signed informed consent. The inclusion criteria for panelists were being in good health and being not allergic to soybeans and fish as self-reported. A sensory test was carried out in the room at 10-11 am. All panelists were given three formulas that were labeled as a combination of three random numbers, and each time they switched from one formula to another, they were asked to drink mineral water to neutralize the taste. Panelists were asked to rate the sensory of the three formulas using three preference scales: dislike, neutral, and like (30). The parameter of the sensory test includes taste, aroma, color, texture, and overall. The scoring for each answer is as follows: if the subject answered "dislike" was given 1 point, "neutral" was given 2 points, and "like" was given 3 points. The data from the sensory test (taste, aroma, color, texture, and overall) are then calculated as the mean ±SD value. The Kruskal Wallis test was used because the data are not normally distributed based on the Kolmogorov Smirnov test. The statistical analyses were performed using the Statistical Package for Social Science (SPSS) version 26.0. The data were considered statistically significant with a p-value <0.05

Stage 4. Nutrient analysis

The product selected in the sensory test was then analyzed for its nutritional content. Analysis of protein, fat, and carbohydrate content was conducted using a proximate test, while AAS (Atomic Absorption Spectrophotometer) was used for iron. The test was carried out in the Central Agro-Industry Center (CAIC)

laboratory of Bogor, Indonesia. Energy calculation was done by multiplying factors 4, 9, and 4 for protein, fat, and carbohydrates, respectively (31). Meanwhile, the beta carotene content was determined based on the Indonesian Food Composition Table (23).

Ethical considerations

The health research ethics committee of the Faculty of Public Health, Universitas Diponegoro has approved this research with approval number: 39/EA/KEPK-FKM/2020

Results

Table 1 shows the participants' preferences for snacks made of flour. All 356 participants (100%) have filled out the questionnaire. The results showed that the flour-based snack selected as the "carrier" was salty and non-fried. This snack was liked by 322 participants (90.4%) and became the most preferred by participants.

The process of making snack made of flour, salty and non-fried, was done by adding tempe flour, tilapia fish, and pumpkin to the cassava flour mixture as the main ingredients (food to food fortification) (Figure 1). Previously, tempe was first made into flour, tilapia fish were filleted and mashed, and pumpkin was steamed and mashed. All ingredients (cassava flour, tempe, tilapia fish, and pumpkin) were added with water, formed into balls, and steamed until cooked. Snack food product was made in three formulations with different percentages of tempe flour.

Table 2 shows the sensory test results of the three formulations of snacks made of cassava flour, salty, and non-fried (steamed), enriched with tempe, tilapia fish, and pumpkin (S1, S2, and S3). The results of the Kruskal Wallis test showed that there was no difference in participant preferences among the three formulas (S1, S2, and S3) for all sensory indicators, i.e., taste, aroma, color, texture, and overall with p=0.465; 0.085; 0.951; 0.114; and 0.493, respectively. These results indicate that the participants' preferences for the three formulas were the same. However, formula 2 (S2) had the highest average preference score for taste, aroma, color, texture, and overall compared to formula 1 (S1) and 3 (S3). And finally, formula 2 (S2) was chosen for further analysis of nutrient content.

Table 3 shows the results of the analysis of the nutritional content of the selected snack food products in the sensory test (S2). Snacks weighing 100g with a composition of 40% cassava flour, 30% tempe flour, 16% pumpkin, and 14% tilapia fish contain 239.89 kcal of energy, 12.6g of protein, 27.1 of carbohydrate, 9.01g of fat, 1.46g of iron, and 251.46μg of beta carotene.

Table 1. The preference for the snack made of flour

No	Snack	Preferences			
		like		dislike	
		n	%	n	%
1.	Flour, sweet, fried	279	78.4	77	21.6
2.	Flour, salted, fried	304	85.4	52	14.6
3.	Flour, sweet, non-fried	311	87.4	45	12.6
4.	Flour, salted, non-fried	322	90.4	34	9.6

Discussion

Adolescence is an important period in life where food choices have begun to be determined individually. The eating habits of adolescents, such as skipping meals, eating irregularly, low consumption of fruits and vegetables, and frequent consumption of energy-dense snacks, often lead to long-term adverse health effects (32). Daily snacking is very common among children and adolescents, increasing more than 20% over the last 30 years (33). Teenagers choose snacks based on taste rather than nutritional content and prefer salty flavors (34). More than 23% of adolescents in Mexico consume salty snacks daily (35). Also, research in Serbia showed that adolescents consume salty snacks several times per week (36). This study also found that snacks made of flour with a salty taste were the most preferred by adolescent.

Snack foods contain high energy but are low in protein and micronutrients such as iron (10). Tempe made of soybeans are used as food fortification in snacks to improve its nutritional content (31,32). Food products produced from soybean can have good nutritional quality and sensory acceptance (37).

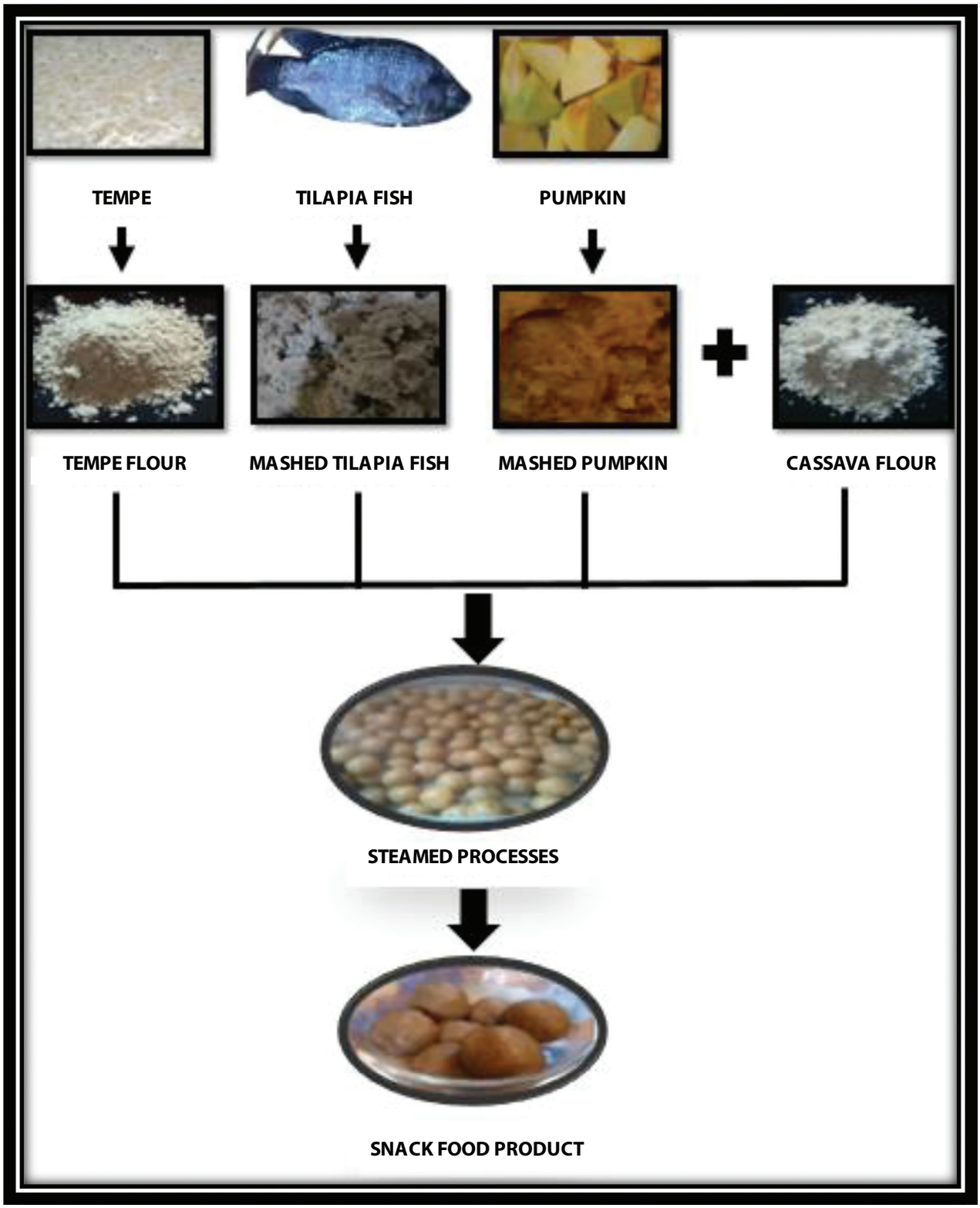

Figure 1. The process of making a snack food product

Table 2. Sensory test results for tempe flour-enriched snacks made of cassava flour, salty and non-fried

Sensory Test	Snack formula 1 (S1) Mean±SD	Snack formula 2 (S2) Mean±SD	Snack formula 3 (S3) Mean±SD	*p*-value
Taste	2.71±0.48	2.75±0.44	2.66±0.48	0.465
Aroma	2.35±0.58	2.55±0.55	2.45±0.63	0.085
Color	2.53±0.50	2.56±0.49	2.55±0.50	0.951
Texture	2.24±0.68	2.44±0.71	2.38±0.68	0.114
Overall	2.45±0.53	2.55±0.50	2.51±0.53	0.493

Table 3. Comparison of nutritional content between the selected formula (S2) and the original snack

Nutrients	Composition per 100 g	
	The selected formula (S2)	Original snack
Energy (kcal)	239.89	363
Protein (g)	12.6	1.1
Carbohydrates (g)	27.1	88.2
Fat (g)	9.01	0.5
iron (mg)	1.46	1.0
Beta carotene (µg)	251.46	0

A preference test using five main parameters is used in snacks enriched with tempe, including taste, aroma, color, texture, and overall assessment (38). For consumers, nowadays, the primary consideration for choosing and eating a food commodity is the delicacy or quality of the food product, while parameters such as nutritional content are the second consideration (39).

Taste is an important factor for consumers to accept or reject a food product. Panelists can accept the taste of snacks enriched with tempe, with the highest preference score for snacks and the addition of 30g of tempe flour. The flavor of snacks is influenced by the addition of tempe and spices. The addition of fish also affects the flavor because fish has a good taste (40). Likewise, the sweet taste of pumpkin and the salty taste of snack products will produce a savory taste (41).

The aroma of food is strongly influenced by the raw materials used. The more tempe added, the stronger the smell will be. The formation of a distinctive aroma in tempe is due to the degradation of the components in tempe during the fermentation process (42). Aroma is detected when volatile compounds enter the nasal passages and are received by the olfactory system and transmitted to the brain (43). The heating process during steaming can result in volatile evaporation (44), so the distinctive aroma inherent in tempe-enriched snacks tends to disappear. The increasing use of tempe in steamed snacks shows a tendency to increase the value of participants' preferences for the aroma of the snacks produced.

Color is a perception in the brain that results from the detection of light after interacting with an object which is influenced by the physical and chemical composition of the object, the source of light that illuminates the object, and the sensitivity of a person's eyes. Change in any of the factors can change the color of the object (39). Color is the fastest and easiest parameter to give an impression on a product. The color difference in snacks enriched with tempe is influenced by the color of the tempe. Tempe flour has a brownish white color, so more addition of tempe flour will tend to make the snack food products darker in color and less desirable. The average sensory test score for the lowest color was 2.53 (S1), with the most addition of tempe flour (40%).

Tempe flour-enriched snacks have a chewy texture because the basic ingredient is cassava flour. Most of the content of cassava flour is starch (45) and the steaming process can retain starch (46). Starch contains amylopectin, which binds water, thereby increasing elasticity (47). The addition of tempe flour will reduce the amount of cassava flour, invariably reducing food elasticity. Snacks with the addition of 40%tempe flour have the lowest texture preference score compared to the addition of 30% and 20%. The overall appearance of a product is essential to shape consumer perceptions regarding the quality of a food product before consumption (48).

The study showed that the preference score for all tempe flour-enriched snacks was the addition of 30% of tempe flour.

Snacks enriched with tempe flour as much as 30% contain 12.6g of protein so they can contribute 19.4% to protein adequacy in adolescents (49). Insufficient protein intake from food consumed daily (main menu and snacks) has the likelihood of doubling the incidence of anemia (50). The iron content in snacks enriched with tempe flour is 1.46 mg, contributing almost 10% of the daily iron requirement. The iron content in tempeflour-enriched snacks was higher than in sprouted soybeans-based drinks (1.32mg). The results showed that the provision of sprouted soybeans-based drinks with an iron content of 1.32mg and protein content of 6.26g was proven to increase Hb levels in adolescent girls (51).

Conclusion

Snacks with the addition of 30g tempe flour are the most preferred formula overall. Adding tempe, Tilapia fish, and pumpkin can improve the nutritional quality of snack foods, especially protein and iron, so they have the potential as a food-based approach (FBA) strategy to prevent anemia in an adolescent female. Clinical research in the field is needed to test the efficacy of tempe-enriched snacks in preventing anemia in a female adolescents.

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Conflict of Interest: Each author declares that he or she has no commercial associations (e.g. consultancies, stock ownership, equity interest, patent/licensing arrangement etc.) that might pose a conflict of interest in connection with the submitted article.

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