#### **Original article:**

# The Effectiveness of Giving Crystal Guava Juice (*Psidium guajava L. "Crystal"*) on Haemoglobin Levels in Female Adolescents with Anaemia

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#### Abstract

Anaemia is one of the health problems in female adolescents with haemoglobin levels in the blood circulation is below normal, which is <12 g/dl at the adolescent threshold. Crystal guava is a fruit that is rich in iron and vitamin C, which helps increase haemoglobin levels in the blood. This study aims to analyse the effectiveness of giving crystal guava juice (Psidium guajava L."Crystal") on haemoglobin levels in female adolescents with anaemia. The design of this study was a quasi-experiment with pre and post-test control group design with 36 subjects divided into 3 group. This study was tested using SPSS version 25 with a significant level of 0.05. The result showed that there was an effect of giving crystal guava juice on haemoglobin levels in female adolescents with anaemia by p=0.018 (p<0.05). Giving crystal guava juice with iron tablets is effective to increase haemoglobin levels in female adolescents with anaemia.

**Keywords:** iron, vitamin C, crystal guava juice, haemoglobin levels, female adolescents with anaemia

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## Introduction

Adolescence is a period of transition from childhood to adulthood when physical, mental and emotional growth and development proceed very rapidly. According to WHO, the age limit for adolescents is 10-19 years old.<sup>1</sup> Adolescents are a group at high risk of developing iron deficiency and anemia. Periods of puberty and development to significant additional iron requirements. Very rapid growth, psychological changes, and increased activity lead to increased nutritional requirements in adolescents. Irregular eating patterns, skipping breakfast and skipping lunch adversely affect the health of adolescents.<sup>2</sup> Anemia is a condition in which circulating hemoglobin levels are below the adolescents threshold of <12 g/dl.<sup>3</sup>6 Anemia is a health problem worldwide. According to WHO (2015), 30% or more than 2 billion people worldwide suffer from anemia. Indonesia is a country with a high incidence of anemia, over 20%.<sup>4</sup> Based on Basic Health Research data for 2007, 2013, and 2018, there was an increasing trend in the prevalence of anemia among adolescents. In 2018, he 32% of young people in Indonesia suffered from anemia.<sup>5</sup> Percentage of iron deficiency anemia among pre-school age children and women at reproductive age ranged from 25% and 37% respectively. Several studies have shown that untreated iron deficiency anemia can have a serious impact on health.<sup>6</sup> Anemia, or low hemoglobin levels, impairs cognitive

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and motor development, leading to fatigue and decreased productivity.<sup>7</sup>

Female Adolescents with anemia who become pregnant are at increased risk of premature birth, low birth weight, and maternal and neonatal mortality.8 Of the various determinants of anemia, iron deficiency is common in developing countries.9,10 Iron deficiency is estimated to be responsible for approximately 50% of anemia.<sup>2</sup> Giving iron tablets along with other micronutrients is more effective in raising hemoglobin levels because it increases the body's absorption of iron, so iron supplementation should be combined with other micronutrients such as vitamin C. Vitamin C is known to affect iron metabolism and specifically does not promote heme iron uptake or increase iron mobilization from stores. Iron is soluble. Therefore, it is efficient and effective in its iron form  $(Fe^{2+})$ . Vitamin C changes the acidic atmosphere of the stomach and helps convert iron ( $Fe^{3+}$ ) to iron ( $Fe^{2+}$ ) in the intestine.<sup>11</sup> Vitamin C deficiency may also contribute to hemolysis due to capillary hemorrhage leading to oxidative damage to red blood cells and blood loss. Vitamin C supplementation has been shown to increase hemoglobin levels.<sup>2</sup> Witjaksono (2014) stated that the effectiveness of taking iron with natural vitamin C from fruit is superior to taking iron with high-dose vitamin C tablets. One is crystal guava. Crystal guava has the highest content of vitamin C compared to citrus fruits, papaya and bananas.12 Food processing needs to change to increase demand for fruit consumption. One of his processed fruits that is very popular in the community is juice. Based on research in Chem-Mix Pratama's laboratory in Yogyakarta, the content of crystal guava juice per 200 ml is 6.9 mg iron and 423 mg vitamin C. Iron (Fe<sup>3+</sup>) is converted to iron  $(Fe^{2+})$  in the small intestine, making it easier for the body to absorb. The reductive process is even greater when the stomach pH becomes more acidic, which can increase the iron absorption process by up to 30%.13

In previous studies, most of the interventions were red guava juice, but in this study we wanted to compare the effects with crystal guava juice. A previous study found a 2.44 g/dl increase in hemoglobin levels before and after administration of blood products and guava juice. Another study conducted in adolescent females given red guava juice found a 0.6 g/dl increase in hemoglobin levels with p<0.05.<sup>14</sup>

Based on the above explanation, this study aimed

to analyze the effectiveness of giving crystal guava juice (*Psidium guajava L. "Crystal"*) on hemoglobin levels in female adolescents with anemia.

## **Materials and Methods**

The design of this research is quasi-experiment with pre and post-test control group design conducted in June-July 2022. The research subjects were female adolescents of the Nurul Qur'an Islamic Boarding School and Miftahul Huda Islamic Boarding School in Boyolali Regency.

The subject selection technique in this study used purposive sampling, the determination of the sample was carried out according to the inclusion and exclusion criteria. Inclusion criteria include female adolescents aged 12-19 years, have hemoglobin levels <12 g/dl, are willing to follow the research procedure by agreeing and signing informed consent, and can communicate, read, and write while the exclusion criteria include female adolescents who are sick such as tuberculosis (TB), helminthiasis, HIV, and malaria are based on a doctor's diagnosis and has a guava allergy. The total number of subjects is 36 subjects divided into 3 groups, 12 subjects in the K group, 12 subjects in the P1 group, and 12 subjects in the P2 group.

The K group was given iron tablets, the P1 group was given iron tablets and red guava juice, while the P2 group was given iron tablets and crystal guava juice. This study was conducted for 30 days and the treatment group was given red guava juice (P1) and crystal guava juice (P2) every 200 ml per day before eating. The recommended daily supplement dose for non-pregnant women of child bearing potential living in countries where anemia is prevalent is 60 mg of elemental iron and 400 mcg of folic acid in an iron tablet.<sup>15</sup>

Crystal guava obtained from crystal guava plantations. Examination of the nutritional content of crystal guava juice was carried out at the Chem-Mix Pratama food laboratory, Yogyakarta. The nutritional content of crystal guava juice per 200 ml is 6.9 mg of iron and 423 mg of vitamin C. Crystal guava juice is made naturally using the cold pressed juice technique. This technique works with high pressure (pressed) and produces very minimal heat so that it can maintain the nutritional content in the fruit such as iron and vitamin C compared to using a regular blender and can be stored longer.<sup>16-17</sup> Research subjects signed informed consent before screening and intervention. Anemia screening questionnaire in female adolescents to examine the early symptoms of anemia experienced and has been tested for validity in previous studies.<sup>18</sup> Body weight was measured using a digital scale with an accuracy of 0.1 kg and height measurements using a microtoice with an accuracy of 0.1 cm to determine nutritional status. Measurement of hemoglobin levels using Easy Touch GCHb at preliminary studies and during the research using cyanmethemoglobin method with a spectrophotometer. Measurement of food intake was carried out using the food record form and the calculation using Nutrisurvey 2007 to obtain data on nutrient intake.

Data analysis in this study was carried out using

the SPSS version 25 program. Analysis of the characteristics of the subject using the Chi Square test. The normality test of the data used the Shapiro Wilk test because the subjects were less than 50 people, while the Wilcoxon test and Kruskal Wallis test were conducted to determine whether or not there was an effect on the intervention given.

#### Result

The description of the data regarding the characteristics of the respondents in this study includes the gender of the subjects in this study, 100% female adolescents with anemia, there is no difference in each group. In addition, the education level of female adolescents is at junior high school. Characteristics of age and nutritional status (BMI/Age) are shown in table 1.

Characteristics	К		P1		P2		р
	n	%	n	%	n	%	
Age							
Early Adolescence (10-13 yrs)	1	8.33	1	8.33	5	41.67	0.233
Middle Adolescence (14-17 yrs)	11	91.67	11	91.67	7	58.33	
Late Adolescence (18-24 yrs)	0	0	0	0	0	0	
Amount	12	100.00	12	100.00	12	100.00	
Nutritional Status (BMI/U)							
Malnutrition	1	8.33	1	8.33	0	0	0.816
Good Nutrition	8	66.67	8	66.67	10	83.33	
Overweight	3	25	3	25	2	16.67	
Obesity	0	0	0	0	0	0	
Amount	12	100.00	12	100.00	12	100.00	

Table 1. Characteristics of age and nutritional status (BMI/Age) of subject

In table 1, it is known that the total of all research subjects was 36 subjects with the distribution of early adolescence (10-13 years) as many as 1 subject (8.33%) in K group, 1 subject (8.33%) in P1 group, and 5 subjects (41.67%) in P2 group. Middle adolescents (14-17 years) were 11 subjects (91.67%) in K group, 11 subjects (91.67%) in P1 group, 7 subjects (58.33%) in P2 group. While at the age of late adolescents (18-24 years) as much as 0% of both K, P1, and P2 groups. From the results of the Chi Square test, it was found that there was no difference between the subjects in groups K, P1, and P2 with p=0.233 (p>0.05), which means that the subject was homogeneous.

In the characteristics of nutritional status (BMI/Age), it was found that in the K group there was 1 subject with undernutrition (8.33%), 8 subjects with good nutrition (66.67%), and 3 subjects with overweight (25%). In the P1 group, the

distribution was the same as the control group, namely 1 subject with undernutrition (8.33%), 8 subjects with good nutrition (66.67%), and 3 subjects with overweight (25%). In P2 group there were 10 subjects with good nutrition (83.33%) and 2 subjects with overweight (16.67%). From the statistical test results of the Chi Square test, it was found that there was no difference between the subjects in each group with p=0.816 (p>0.05), which means the subjects were homogeneous.

 Table 2. The average results of hemoglobin levels before and after being given crystal guava juice in female adolescents with anemia

Group	n	Before	After	Δ Mean	р
		$Mean \pm SD$	$Mean \pm SD$		
К	12	10.33±1.88	10.85±1.18	0.52±1.03	0.052*)
P1	12	11.23±1.08	11.88±0.99	0.65±0.62	0.008*)
P2	12	11.12±0.74	12.31±0.75	1.19±0.51	0.002*)
р				0.018**)	

 $\Delta$  : Difference in haemoglobin levels before and after intervention

\*) : Wilcoxon test

\*\*) : Kruskal Wallis test

Table 2. shows that the mean haemoglobin level of the K pre-test group was  $10.33\pm1.88$  and the post-test haemoglobin level was  $10.85\pm1.18$  with a mean difference before and after the intervention was  $0.52\pm1.03$ . The mean haemoglobin level in the P1 pre-test group was  $11.23\pm1.08$  and the haemoglobin level in the P1 post-test group was  $11.88\pm0.99$  with a mean difference of  $0.65\pm0.62$ . The mean haemoglobin level in the P2 pre-test group was  $11.12\pm0.74$  and the haemoglobin level in the P2 post-test group was  $12.31\pm0.75$  with a mean difference of  $1.19\pm0.51$ .

The results of the statistical test of 2 groups before and after the intervention with the Wilcoxon test for each group before and after the intervention showed that K group with p=0.052 (p>0.05) means that there is no difference before and after the intervention in K group. In the P1 group before and after the intervention, the results obtained with p=0.008 (p<0.05) meaning that there was a difference before and after the intervention in the P1 group. In the P2 group before and after the intervention, the results were p=0.002 (p<0.05), meaning that there was a difference before and after the intervention in the P2 group. The results of the statistical test between the 3 groups from the average difference before and after the intervention with the Kruskal Wallis test showed that there was a difference in haemoglobin levels before and after the intervention with p=0.018 (p<0.05).

#### Discussion

Based on the results of the study found that the average haemoglobin level in the K pre-test group was 10.33 g/dl with a standard deviation of 1.88. In the P1 group, the mean pre-test haemoglobin level was 11.23 g/dl with a standard deviation of 1.08. Meanwhile, in the P2 group the mean pre-test haemoglobin level was 11.12 g/dl with a standard deviation of 0.74. All samples that have been examined have an average haemoglobin level below the normal threshold <12 g/dl, classified as mild, moderate, and severe anaemia. When these female adolescents become anaemic, many of them look pale, get sleepy easily, can't concentrate on their studies, get tired easily, and have no endurance.<sup>19</sup>

The average haemoglobin level in the K post- test group was 10.85 g/dl with a standard deviation of 1.18. The mean haemoglobin level of the P1 post-test group was 11.88 g/dl with a standard deviation of 0.99. Meanwhile, the average haemoglobin level of the P2 post-test group was 12.31 with a standard deviation of 0.75. In K group there was an increase in the average haemoglobin level. However, the haemoglobin level is still classified as mild anaemia. The P1 group also experienced an increase but also included mild anaemia. Meanwhile, in the P2 group there was also an increase in haemoglobin levels already above the normal threshold by >12 g/dl.

Underweight is an anthropometrically assessed sign of malnutrition and has been associated with anaemia in some, but not all, studies is related to various factors such as inappropriate home and community environment, inappropriate complementary feeding practices leading to inadequate intake of micronutrients and animal products, contaminated water and poor sanitation, weight gain. environment and infectious diseases. Furthermore, overweight and obesity are associated with iron deficiency, and data from several countries suggest that overweight and obese people are at increased risk of iron deficiency. It may be due to hepcidin, a peptide hormone involved in iron homeostasis and produced primarily in the liver. Hepcidin levels are elevated in overweight and obese individuals compared to lean individuals, had elevated hepcidin levels and poorer iron status than normalweight adolescents.

Giving red guava juice (P1) and crystal guava juice (P2) each can increase hemoglobin levels in female adolescents with anemia. However, giving crystal guava juice was more effective in increasing hemoglobin levels than red guava juice given to female adolescents with anemia significantly with p=0.018 (p<0.05). Anemia can be treated by consuming iron-rich foods. It can aid in the process of iron absorption. In addition, dietary components that may contribute to iron absorption, such fruits rich in vitamin C, iron is also absorbed in the small intestine, absorbed by the body. The highest iron and vitamin C content is found in crystal guava juice even when compared to red guava juice. Crystal guava juice contains 6.9 mg of iron and 423 mg of vitamin C, while red guava juice contains 6.8 mg of iron and 321.3 mg of vitamin C.

The effectiveness of crystal guava juice on hemoglobin levels in female adolescents with anemia based on the Wilcoxon test showed that there was a significant difference between hemoglobin levels before and after giving crystal guava juice. So it can be concluded that giving crystal guava juice has an effect after the intervention. When compared to the three treatment groups, namely groups K, P1 and P2, P2 group with the addition of iron tablets and crystal guava juice was the most effective in increasing hemoglobin levels compared to the other groups with an increase in hemoglobin levels of 1.19 g/dl. This study is consistent with the results of Damayanti (2020), who found differences in hemoglobin levels between groups who received iron tablets and those who consumed guava juice.<sup>20</sup> Additionally, this study is consistent with studies done on anemic pregnant women who were given vitamin C. There was a significant increase in hemoglobin levels.<sup>21</sup> This study is supported by other studies, namely, the provision of iron supplements also affects the increase in iron in pregnant women who experience anemia with  $p<0.05.^{22}$ 

Food sources rich in iron and vitamin C, such as guava juice, can help increase iron absorption and can be used as an alternative therapy for anemic patients, especially adolescent women with anemia. Iron in these food sources is absorbed with the help of vitamin C by reducing iron  $(Fe^{3+})$  to iron  $(Fe^{2+})$  in the small intestine to heme. As the pH of the stomach becomes more acidic, the iron breakdown process becomes even more pronounced.23 Each heme binds to globin made by polylinosomes. Tetramers are composed of four globins, each with a heme group, and formed into hemoglobin molecules, increasing hemoglobin levels.<sup>24</sup> Absorption of non-heme iron increases four-fold in the presence of vitamin C to approximately 30%. Most blood transferrins transport iron to the bone marrow and other parts of the body. Bone marrow uses iron to make hemoglobin. Since the bone marrow requires precursors such as iron and vitamin C for the formation of red blood cells and hemoglobin, increased levels of iron and vitamin C lead to increased red blood cell formation in response to increased hemoglobin levels.25

Prevention and control strategies should also be implemented. Improve food intake and increase food variety through increased iron bioavailability, targeted food fortification, and iron supplementation for at-risk groups such as the young. In order to prevent anemia, it is necessary to improve knowledge and education about anemia. Since early life is a critical period for intellectual and psychomotor development, anemia prevention must continue to be supported, promoted and embraced.<sup>26</sup>

#### Conclusion

Giving crystal guava juice with iron tablets is effective to increase hemoglobin levels in female adolescents with anemia. Giving crystal guava juice together with the consumption of iron tablets was proven to be the most effective in increasing hemoglobin levels in female adolescents with anemia compared to other groups with p=0.018 (p<0.05).

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# **Conflicts of interests:**

The authors declared no conflict of interest.

## **Ethical clearance**

This study were approved by the Health Research Ethics Committee of the Faculty of Medicine, Sebelas Maret University, Surakarta based on the ethics committee letter with letter number 36/ UN27.06.6.1/KEP/EC/2022.

# **Authors' contribution**

All authors were equally involved in the preparation of this study and manuscript.

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