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THE CORRELATION OF INTAKE PHYTATE AND TANNIN ON SERUM TRANSFERRIN RECEPTOR AND HEMOGLOBIN IN STUNTED OVERWEIGHT ADOLESCENTS

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ABSTRACT

Stunted overweight teenagers are at risk of having iron deficiency. Iron deficiency is caused by various factors including the high food absorption inhibitors of iron such as phytate and tannins. Phytate and tannin contain polyphenol compounds which have a strong ability to bind iron so that it inhibits iron absorption in the intestine. This study aims to analyze the correlation between phytate, tannin intake and serum transferrin receptor (sTfR) and hemoglobin in stunted overweight adolescents. The research method was a cross-sectional study of 64 stunted overweight adolescents selected by consecutive sampling in four high schools/vocational high schools in Banyumanik District, Semarang City. Phytate and tannin intake data using SQ-FFQ method. The serum transferrin receptor examination uses the ELISA method and the hemoglobin level uses the Cyanomethemoglobin method. The results of the study, most of the respondents had high phytate and tannin intake of 96.9% and 89.1%. Respondents with low serum transferrin receptor were 7.8% and low hemoglobin levels were 7.8%. There was no correlation between phytate intake with serum transferrin receptor or hemoglobin (p_1 = 0.937 r_1 = -0.010, p_2 = 0.192 r_2 = 0.165). Tannins were significantly correlated with serum transferrin receptor and hemoglobin (p_1 = 0.005 r_1 = 0.344, p_2 = 0.002 r_2 = -0.374). Based on multivariate analysis, tannin is a determinant of hemoglobin (R^2 = 0.257). Conclusion is that tannin is positively correlated with serum transferrin receptor and hemoglobin in stunted overweight adolescents. Excessive tannin intake can cause deficiency in stunted overweight adolescents.

Keywords: Stunted overweight; sTfR; Hemoglobin; Fitat; Tannin

INTRODUCTION

Iron deficiency and stunted overweight are two nutritional problems that still occur in Indonesia. Iron deficiency and stunted overweight are known to be interconnected and often occur simultaneously **(Balitbang, 2013)**. Stunted overweigt is the nutritional state of a person who has a short body and is overweight. Malnutrition in early life marked by stunted, the risk of overweight and obesity in adolescence. The mechanism of obesity in stunted children due to low energy intake during growth causes high cortisol levels and low IGF-1. These hormonal changes are related to body fat storage while the low IGF-1 hormone allows interference with lipolysis in breaking down fat, therefore that, long-term adaptation in stunted children causes impaired fat oxidation **(Stanojevic, Kain and Uauy, 2007)**.

The prevalence of short and very short adolescents aged 16 – 18 years in Semarang City reached 18.3% and 3.7%. The prevalence of obese and obese adolescents is 7.6% and 2.7%. Semarang City is one of 16 districts/cities with obesity prevalence above the provincial prevalence of 5.4% obese and 1.7% obese **(Santoso et al., 2013)**. Research in several countries proves that stunted children have a risk of overweight and obesity as teenagers. In a Brazilian study, the prevalence of stunted adolescents was 11%, and 30% of them were obese **(Hoffman et al., 2000)**. Research in South Africa, amounting to 14.8% of secondary school students stunted and there was a tendency to overweight **(Mukuddem-Petersen and Kruger, 2014)**. Research in Indonesia in Bangsri Subdistrict, Jepara Regency, and stunted incidence in young women was 23.3% and 11.1% classified as obese **(Saraswati and Sulchan, 2016)**.

The increasing prevalence of stunted overweight and obesity adolescents is caused by changes in lifestyle and eating patterns to sedentary lifestyle and consumption of energy-dense foods. Energy-dense food consumption is a shift in traditional and high consumption patterns of eating vegetables and fruits and is shifting to high-energy and low micronutrient food consumption habits **(Feeley et al., 2012)**. Habits of physical activity in adolescents have decreased by an average of 7% per year **(Dumith et al., 2011)**.

Stunted overweight and obesity are body conditions where there is excess fat accumulation with an unproportionally increased characteristic of adipose tissue **(Kruger, Pretorius and Schutte, 2010)**. High fat accumulation will cause inflammation which is at risk of iron deficiency **(Aigner, Feldman and Datz, 2014)**. Iron deficiency occurs when an imbalance between erythropoiesis and the amount of body iron stores **(Urrechaga, Borque and Escanero, 2013)**. Iron status in the body is influenced by factors that inhibit iron absorption. Absorption of iron is inhibited by phytate and tannins. Fats and tannins can bind mineral elements in the form of zinc, iron, calcium, protein to form water-insoluble complexes making iron, protein difficult to be absorbed so that it can cause iron deficiency **(Hurrel and Engli, 2010)**.

Iron deficiency in adolescents will have a negative impact on health, namely the occurrence of growth and development disorders, fatigue, increased body susceptibility to infections, decreased physical ability and endurance as well as academic ability **(Hassan et al., 2016)**. This study aims to analyze the correlation of intake phytate, tannins against serum transferrin receptors and hemoglobin in stunted overweight adolescents.

Scientific hypothesis

The hypothesis of this study is that phytate and tannin intake correlates with serum transferrin receptor and hemoglobin in stunted overweight adolescents.

MATERIAL AND METHODOLOGY

Statement of Ethics

This study was approved by the Health Research Ethics Commission (KEPK) of the Faculty of Medicine, Diponegoro University as stipulated in Ethical Clreance No. 67/EC/FK UNDIP/III/2019 dated March 11, 2019. Written agreement was obtained from all participants and participating parents.

Study Design

A cross-sectional study was conducted at SMAN 9 Semarang, Hidayatullah Islamic High School, SMKN 11 Semarang, Hidayah Vocational School. Research respondents were 64 people selected by consecutive sampling according to inclusion and exclusion criteria **(Sastroasmoro and Ismael, 2014)**. Inclusion criteria are stunted overweight and obese adolescents aged 15 – 18 years and classified as stunted overweight and stunted obesity if the TB/U indicator with Z-Score < -2 SD and overweight based on BMI/U with Z-Score > 1 SD 2 SD and obesity based on BMI/U with Z-Score > 2 elementary school in the same age group **(Cashin and Oot, 2018)**. Willing to be a research respondent by filling out informed consent. Exclusion criteria were suffering from chronic illness in the last 1 month, suffering from infectious disease in the last 2 – 3 weeks, having menstruation in the last 1 week, consuming iron tablets in the last 3 months.

Data collection

Measurements of body weight using a digital Camry EB9003 scale with an accuracy of 0.1 kg, placed on a flat floor surface. Respondents took off their footwear, dressed as seminally as possible, took off their hats and took off their cell phones, watches, wallets and other objects that could affect the outcome of the weighing. Respondents are welcome to rise on the digital scale, both feet are in the middle of the scale and look straight ahead. The height is measured using Seka's microtoise with the accuracy of 0.1 cm, taped to a wall as high as at least 2 meters with a flat surface. Respondents were asked to take off their footwear and then stand upright against the wall. The heel, calf, buttocks, shoulders, head stick well to the wall and look straight ahead. The anthropometric measurements of the respondents were carried out to determine nutritional status based on TB/U < -2 SD, BMI/U overweight z-score 1 SD 2 SD and obesity z-score ≥2 SD. Nutrition status data using WHO-Antro Plus Software. Phytate intake and tannin respondents were obtained through interviews using the SQ-FFQ (Semi Quantitative-Food Frequency Questionnaire) form. Phytate and tannin intake data were processed using Nutrisoft sofware

Biochemical Analysis

Blood sampling was conducted in the morning between 8 and 10 am by experienced analysts from the Diponegoro University GAKY laboratory. Blood was taken as much as 3 cc through the antecubital vein and analysed at the GAKY Laboratories at Diponegoro University. The serum transferrin receptor level of the respondents was measured using the ELISA method. Serum transferrin receptor levels were categorized as normal (1.8 – 4.6 $mg.L^{-1}$), low (<1.8 $mg.L^{-1}$) and high (> 4.6 $mg.L^{-1}$) **(Sumarmi et al, 2016)**. Hemoglobin levels were measured using the Cyanmethomoglobin method. Categorizes hemoglobin levels which are normal in men (13.2 – 17.3 $g.dL^{-1}$), low (<13.2 $g.dL^{-1}$) and high (> 17.3 $g.dL^{-1}$) and in women categorized as normal (11.7 – 15.2 $g.dL^{-1}$), low (<11.7 $g.dL^{-1}$) and high (> 15.2 $g.dL^{-1}$) **(Association of Clinical Pathologists, 1996)**.

Statistic analysis

Statistical analysis using SPSS Version 21. Test the normality of the data using Kolmogorov-smirnov. Data are presented in the form of percentages, medians and maximum-minimum values. Spearman correlation test to see the correlation between phytate, tannin intake and serum transferrin receptor and hemoglobin with significant at *p*-value <0.05 and 95% confidence intervals. The strength of correlations was determined by *r*-value. Multivariate analysis using linear regression test **(Dahlan, 2011)**.

RESULTS AND DISCUSSION

Subject Characteristics

Table 1 shows that the number of research respondents was 64 people and the majority were women (65.6%). The age of research respondents is mostly 16 years, which is 46.9%. This result is in line with the study in Semarang City High School where most of the stunted overweight are adolescent girls by 88.46% **(Afifah, Sulchan and Nissa, 2017)**.

Table 1 Subject characteristics.

Characteristics	n	%	Average ±*SD*	Median (min – max)
Gender				
Male	22	34.4		
Female	42	65.6		
Age (Years)			15.97 ±0.73	16 (15 – 17)
15	18	28.1		
16	30	46.9		
17	16	25.0		
Nutritional Status(TB/U)			-2.14 ±0.22	-2.04 (-3.3 – (-2.01))
Very Short	1	1.6		
Short	63	98.4		
Nutritional Status (IMT/U)			2.02 ±0.71	1.86 (1.03 – 5.15)
Overweight	42	65.6		
Obesity	22	34.4		

Table 2 Fitat intake, Tannin, sTfR levels and Hemoglobin.

Characteristics	n	%	Average ± *SD*	Median (min – max)
Fitat intake			1193.7 ±334.01	1103 (674 – 2173)
High ≥718 mg	62	96.9		
Low ≤718 mg	2	3.1		
Tannin intake			0.38 ±0.25	0.29 (0 – 1.16)
High >0.29 mg	57	89.1		
Low <0.29 mg	7	10.1		
Iron Status (sTfR)			2.69 ±0.60	2.77 (1.04 – 3.53)
High (1.8 – 4.6 $mg.L^{-1}$)	59	92.2		
Low (< 1.8 $mg.L^{-1}$)	5	7.8		
High (> 4.6 $mg.L^{-1}$)	0	0		
Iron Status (Hemoglobin)			14.10 ±1.78	13.9 (9.4 – 17.8)
Male			15.88 ±1.08	15.8 (13.9 – 17.8)
Normal (13.2 – 7.3 $g.dL^{-1}$)	19	29.7		
Low(<13.2 $g.dL^{-1}$)	0	0		
High (>17.3 $g.dL^{-1}$)	3	4.7		
Female			13.17 ±1.30	13.25 (9.4 – 15.6)
Normal (11.7-15.2 $g.dL^{-1}$)	35	54.7		
Low (<11.7 $g.dL^{-1}$)	5	7.8		
High (>15.2 $g.dL^{-1}$)	2	3.1		

The accelerated linear growth in adolescent girls takes place at the age of 9.5 – 14.5 years and slows down at the age 16 years and stopped at the age of 19 years, while men started at around 14.4 years and stopped at 21 years. Age 14 years is the average maximum age for women experiencing first menstruation, where menstrual conditions are associated with changes in the hormones estrogen and progesterone which increase lipoprotein lipase activity and fat stores in the body. Adolescent girls have more fat around 22 – 26% than men around 18 – 23% **(Brown, 2011; Habánová et al., 2010)**. This makes women easier to overweight than men, especially in stunted conditions **(Kruger, Margetts and Vorster, 2004)**.

Table 2 shows that the majority of respondents had a high phytate intake of 96.9% with an average of 1193.7 ±334.01 mg. The high intake of phytate respondents came from daily foods such as rice, tempeh and tofu. Most of the respondents' tannin intake in the high category was 89.1%. The high intake of respondent tannins came from tea consumption. Respondents with low serum transferrin receptor were 7.8% and low hemoglobin levels were 7.8%.

Correlation of Intake of Fitat, Tannin with sTfR and Hemoglobin

Bivariate analysis was performed to see the correlation between independent variables including phytate and tannin intake on serum transferrin receptor and hemoglobin dependent variables. Bivariate analysis can be seen in Table 3 below.

Table 3 Correlation of intake of Fitat, Tannin to sTfR and Hemoglobin.

Variabel	sTfR		Hemoglobin	
	p	*r*	*p*	*r*
Phytate	0.937	-0.010	0.192	0.165
Tannin	0.005*	0.344	0.002*	-0.374

Note: * Significant (p <0.05).

Multivariate Analysis

Table 4 multivariate analysis shows that tannin is a weak determinant of hemoglobin. The R^2 value of the results of the multiple linear regression test was 25.7% which means tannin was able to affect hemoglobin by 25.7% while 74.3% was influenced by other variables not examined in this study.

Table 4 Double Linear Regression Test for Hemoglobin.

Variabel	Hemoglobin	
	B	***P***
Phytate	-0.000099	0.869
Tannin	-3.674	0.000*

Note: *Adjusted R Square = 0.257.

Phytate acid is the main form of phosphorus storage in cereals and legumes **(Kumar et al., 2010)**. The average daily intake of phytate a vegetarian is 2000 – 2600 mg per day, while non-vegetarians daily intake is 150 – 1400 mg phytate per day **(Reddy and Shridhar, 2002)**.

The high intake of phytate subjects came from everyday foods such as white rice, tempeh and tofu. In 100 grams of white rice there are 126 mg phytate, in 25 grams tempeh there are 99 mg phytate and in 25 grams tofu is 94 mg phytate. Fitat is an iron absorption inhibitor, its effect is influenced by the dose. 2 mg intake of phytate inhibits iron absorption by 18%, and 25 mg of phytate inhibits iron absorption by 64%, and 250 mg of phytate inhibits iron absorption by 82% **(Hallberg, Brune and Rossander, 1989)**.

Phytate intake did not correlate with serum transferrin receptor (p = 0.937) or hemoglobin (p = 0.192). This is due to the phytate intake of respondents such as rice having gone through various processes starting with the process of grinding, washing repeatedly so that the phytate content can be lost. Other sources of phytate intake such as tempeh and tofu have gone through a fermentation process so as to reduce phytate content and increase iron absorption. Research of, feeding high phytate does not affect the status of ferritin iron and serum transferrin receptor **(Armah et al.,2015)**.

Fitat has a stable tendency towards heat, the inhibitory effect of phytate can be reduced by boiling the boiled water is discarded **(Sotelo, Gonzales-Osnaya and, Sanchesz-Chinchilla, 2010)**.The process of grinding, heating, fermentation can also degrade phytate content. The fermentation process in grains can increase the bioaccessibility of iron **(Hurrel and Engli, 2010)**. Fermentation can induce phytic hydrolysis, besides the fermentation process also produces organic acids that have the potential to increase iron absorption **(Hotz and Gibson, 2007)**. Meat consumption and ascorbic acid can also overcome the inhibitory effect on phytate **(Hurrel and Engli, 2010)**.

Most respondents had tannin intake in the high category of 89.1% with a mean tannin intake of 0.29 (0 – 1.16 mg). The results of the Spearman correlation test analysis showed that tannin was significantly correlated with serum transferrin receptor (p = 0.005), positive correlation was weak (r = 0.344). This shows that the higher the tannin intake the higher serum transferrin receptor levels. Whereas tannin with hemoglobin showed a significant correlation (p = 0.002), negative correlation was weak (r = -0.374). This shows that the higher the tannin intake, the lower the hemoglobin level. This result is in line with research in high school adolescents in Makassar City which shows that there is a significant correlation between tannin intake and hemoglobin p = 0.013, **(Indriasari and Jafar, 2015)**.

Research in India, respondents who were given a full meal consumed together with 1 cup of tea decreased iron absorption by 59% (p = 0.001) in the anemia group and 49% (p = 0.01) in the control group **(Thankachan et al, 2008)**. Absorption of non-heme iron in food consumed together with water is 10 – 13% but if the same food is consumed with 200 ml of tea will reduce Fe absorption by 2 – 3% **(Nelson and Poulter, 2004)**.

The results of multiple linear regression analysis, tannin is a weak determinant factor for hemoglobin. The R^2 value of the multiple linear regression test results is 25.7% meaning tannins are able to influence hemoglobin by 25.7% while 74.3% is influenced by other variables not examined in this study.

Tea contains tannins that can bind minerals (including iron) and in some teas (especially black tea) polyphenol compounds which act as antioxidants have been oxidized, so they can bind minerals such as Fe, Zn, and Ca so that iron absorption is reduced **(Thankachan et al., 2008)**, If the body's iron needs are not met through food intake, the iron reserves in the body will decrease. If this negative balance lasts a long time then the availability of iron in the body will be compensated so that erythropoeisis occurs with iron deficiency. This will cause an initial increase in serum transferrin receptor concentration progressively and if it continues there will be a decrease in hemoglobin levels **(Zimmermann and Hurrell, 2007)**.

CONCLUSION

Tannins are positively correlated with serum transferrin receptor and negative correlation with hemoglobin. Excessive tannin intake can cause iron deficiency in stunted overweight adolescents.

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