

KONSUMSI TEH DAN ANEMIA DEFISIENSI BESI PADA WANITA HAMIL DI KABUPATEN BOGOR, INDONESIA

Tea Consumption and Iron-Deficiency Anemia among Pregnant Woman in Bogor District, Indonesia

Putri Bungsu Machmud^{1*}, Ratna Djuwita Hatma², Ahmad Syafiq³

^{1,2}Department of Epidemiology, Faculty of Public Health, Universitas Indonesia
Gedung A Lantai Dasar, Kampus Baru UI Depok, Indonesia

³Department of Public Health Nutrition, Faculty of Public Health, Universitas Indonesia
Gedung F Lantai 2, Kampus Baru UI Depok, Indonesia

*e-mail: putri.bungsu10@ui.ac.id

Submitted: April 5th, 2019 revised: June 14th, 2019 , approved: June 21st, 2019

ABSTRACT

Background. Anemia in pregnancy is an essential problem due to affect to not only the mother's life but also to baby's life. An iron deficiency causes about 75 percent of anemia during pregnancy. **Objective.** This study aimed to identify risk factors for iron deficiency among pregnant women and determine the possible link between iron status and tannin levels associated with tea consumption. **Method.** The population-based cross-sectional studies were conducted from secondary data of previous thesis-research in 2013, "Pengaruh Kadar Tanin pada Teh Celup terhadap Anemia Gizi Besi (AGB) pada Ibu Hamil di UPT Puskesmas Citeureup Kabupaten Bogor Tahun 2012". The study population consisted of 94 randomly selected pregnant women. The inclusion criteria were pregnant women who participated in the previous study and have a complete antenatal care record. Demographic data were collected, including data on age, working status, gestational stage, time since last pregnancy, and parity. The information included nutritional variables, such as iron supplements, nutritional status, and iron intake. Also, data for tannin level grouped as low, medium, and high based on the frequency of daily tea consumption and tea-making habits. The linear model analysis was used to determine the influence of tea consumption on serum ferritin levels. **Results.** The results showed that time since last pregnancy (<2 years), parity (more than two children), reduced consumption of foods containing heme, and levels of tannin consumption (low, medium, or high) were predictors of iron-deficiency anemia. The results also suggested that tannin levels were inversely proportional to serum ferritin levels. **Conclusion.** Pregnant women who consumed tea with a low tannin level had the highest serum ferritin levels, whereas those who drank tea with medium and high tannin levels had the lowest serum ferritin levels.

Keywords: anemia, ferritin, pregnancy, tannin, tea

ABSTRAK

Latar belakang. Anemia pada masa kehamilan merupakan masalah yang esensial, karena tidak hanya berpengaruh pada hidup ibu tetapi juga pada hidup janin dan bayi yang dilahirkan. Sekitar 75 persen anemia selama kehamilan disebabkan oleh kekurangan zat besi. **Tujuan.** Penelitian ini bertujuan untuk mengidentifikasi faktor risiko kekurangan zat besi diantara wanita hamil dan menentukan hubungan potensial antara status zat besi dan kadar tanin yang terkait dengan konsumsi teh. **Metode.** Penelitian ini menggunakan data sekunder dari penelitian tesis dengan desain *cross-sectional* pada tahun 2013, "Pengaruh Kadar Tanin pada Teh Celup terhadap Anemia Gizi Besi (AGB) pada Ibu Hamil di UPT Puskesmas Citeureup Kabupaten Bogor Tahun 2012". Populasi penelitian terdiri dari 94 wanita hamil yang dipilih secara acak. Kriteria inklusi adalah wanita hamil yang berpartisipasi dalam penelitian sebelumnya dan memiliki catatan perawatan antenatal yang lengkap. Data demografis dikumpulkan, termasuk data usia, status

kerja, tahap kehamilan, waktu sejak kehamilan terakhir, dan paritas. Informasi variabel gizi meliputi suplemen zat besi, status gizi, dan asupan zat besi juga dikumpulkan. Selain itu, data mengenai tingkat tanin rendah, sedang dan tinggi diperoleh berdasarkan frekuensi konsumsi teh harian dan kebiasaan membuat teh. Analisis model linier digunakan untuk mengetahui pengaruh konsumsi teh terhadap kadar feritin serum. **Hasil.** Hasil penelitian menunjukkan bahwa waktu sejak kehamilan terakhir (<2 tahun), paritas (lebih dari dua anak), pengurangan konsumsi makanan yang mengandung heme, dan tingkat konsumsi tanin (rendah, sedang, atau tinggi) merupakan prediktor anemia defisiensi besi. Hasilnya juga menunjukkan bahwa kadar tanin berbanding terbalik dengan kadar feritin serum. **Kesimpulan.** Wanita hamil yang mengonsumsi teh dengan kadar tanin rendah memiliki kadar feritin serum tertinggi dan wanita hamil yang mengonsumsi teh dengan kadar tanin sedang dan tinggi memiliki kadar feritin serum rendah.

Kata kunci: anemia, feritin, kehamilan, tanin, teh

INTRODUCTION

Anemia is a major public health problem worldwide, with the highest prevalence found in children and pregnant women (46.2 and 38.2 percent, respectively), according to a study conducted in 2011.¹ Pregnant women have a higher risk of anemia as compared with that of childbearing women of similar age.²

Anemia among pregnant women is a problem in both developed and developing countries.³ About 32.4 million pregnant women worldwide suffer from anemia, with the highest number of cases (48.7 percent) reported in the South Asia region, followed by Africa, the Middle East, Europe, and the U.S. (46.3, 38.9, 25.6, and 24.9 percent, respectively) and the lowest number (24.3 percent) reported in the Western Pacific region.¹ In Indonesia, based on a National Health Survey in 2013, about 37.1 percent of pregnant women suffered from anemia.³

According to studies, an estimated 20 percent of deaths are associated with low levels of hemoglobin during pregnancy that related to bleeding, eclampsia, and infection.⁴ In pregnancy, the most common cause of anemia is a nutrient deficiency. The underlying causes of nutritional anemia include inadequate nutritional intake,

inadequate nutrient absorption, insufficient nutrient intake to fulfill increased nutritional needs during pregnancy, and insufficient intake of hematopoietic nutrients.^{4,5} About 75 percent of anemia during pregnancy is caused by iron deficiency.⁵

A previous study conducted in West Java concluded physiological changes due to pregnancy exacerbated pre-existing malnutrition and vitamin B12, folic acid, and vitamin C deficiencies, thereby contributing to anemia.⁵ High parity, short birth spacing, inadequate antenatal care or pregnancy care, and a low socioeconomic level were also reported to be risk factors for anemia among pregnant women.⁶⁻⁸

According to the literature, tea consumption may contribute to anemia by absorbing minerals as a form of iron.⁹ Tea contains various substances, such as phytate and tannin, that can inhibit iron absorption.⁹ Mineral in iron reacts with tannin to form a complex bond, which is not broken down in the gastrointestinal system. Thus, the minerals no longer function and are released from the body in the form of feces. As tannins can bind proteins, they have the ability to absorb nutrients.¹⁰ The ease of tea consumption indirectly contributes to the increased impact

of anemia on pregnant women and low birth weight or prematurity and mortality.¹¹ Although laws exist to ensure the safety of various foods and beverages, information on safe levels of consumption, including safe levels of tea consumption, is lacking among communities.¹² A lack of awareness and knowledge impede programs aimed at improving maternal and child health and reducing maternal and infant mortality and morbidity.¹⁰ This study aimed to identify risk factors for iron deficiency among pregnant women and determine the potential link between iron status and tannin levels associated with tea consumption.

METHODS

The population-based cross-sectional studies were conducted from secondary data of previous thesis-research in 2013, "*Pengaruh Kadar Tanin pada Teh Celup terhadap Anemia Gizi Besi (AGB) pada Ibu Hamil di UPT Puskesmas Citeureup Kabupaten Bogor Tahun 2012*".¹³ This study showed that pregnant women who consume high tannin in daily have risk 2.8 times have low ferritin.¹³

The study population consisted of 94 selected pregnant women 18 to 42 years old from seven health centers (Puskesmas). The inclusion criteria were pregnant women who participated in previous study and have a complete antenatal care records. Individuals with uncomplete data of research, have a comorbidities, including infections and any diseases resulting in blood anomalies, were excluded from this study.

The participants' anonymity and confidentiality were respected. The study also approved by ethical committee of Public Health Faculty, Universitas Indonesia. Demographic

data, including age, working status, gestational stage, time since last pregnancy and parity, were collected using a standard questionnaire (modified version of the Indonesian Household Health Survey 2010). The questionnaire contained items on nutritional variables (iron supplements, nutritional status, and iron intake). The nutrition instrument used in this study comprised a Food Frequency Questionnaire. The samples were collected and analyzed by trained laboratory (phlebotomies) staff in Jakarta that collaborated in previous study. The blood samples underwent centrifugation to separate serum from blood. All samples were analyzed less than 8 hours after blood collection.

A linear model was used to determine the influence of tannin consumption levels on serum ferritin levels. From the model, an equation was developed to predict the effect of difference levels of tannin consumption. The level of significance was $p \leq 0.05$.

Information was also collected on the frequency of daily tea (black tea) consumption and tea-making habits. The following tea-making habits were recorded: type of water used (i.e., boiled water vs. non-boiled [hot] water) and duration (1 min, 5 min, and 8 min) of tea bag dipping when making tea.

The tannin contents of six samples of tea were tested in the laboratory. For each sample, the tea bag was dipped in boiled water and hot water for 1 min, 5 min, and 8 min (Table 1). The tannin content was then tested using the titrimetric method (permanganate). Tannin levels were classified as low, medium, or high based on the frequency of daily tea consumption by using quartile as a cut-off point.

Table 1. Tannin Level of Tea Based on Behavior of Tea Processing

Source of hot water	Duration of dipping the tea bags	Number of Tannin (mg/mL)	Percentage of participants based on tea-making habit
Not drinking tea		0	10.6
	1 minute	0.26	22.3
Boil water	5 minute	0.29	26.6
	8 minute	0.35	6.4
	1 minute	0.21	14.9
Electric hot water	5 minute	0.25	16.0
	8 minute	0.30	3.2
Total			100

RESULTS

Out of 104 women invited to take part in the study, 94 agreed, giving a participant rate of 90.4 percent. Table 2 presents the characteristics of the study population. In the study, 38.3 percent of women had a low

serum ferritin level (defined as less than 13 µg/L). The median serum ferritin level was 18.17 µg/L. More than 50 percent of the women had two or more children, with the time since last delivery < 2 years in 44.7 percent of cases.

Table 2. Characteristic of Participants

Variable	Median (range) / Number (%)
Age ^a	29 (18 - 42)
Work status (working) ^b	7 (7.4)
Gestation (week) ^a	31 (16 - 40)
Distance of pregnancy (less than 2 years) ^b	42 (44.7)
Parity (more than 2 child) ^b	58 (61.7)
Iron supplement (not consume) ^b	10 (10.6)
Nutritional status (14) ^b	13 (13.8)
Serum Ferritin (µg/L) ^a	18.17 (3.67 - 116.3)

^a Median (range)

^b Number (%)

Table 3 presents data on daily food consumption according to iron content (heme and nonheme), inhibitors, and enhancers of iron content. The median level of tannin consumption per day based on the frequency of daily tea consumption was 0.157 mg/mL (median

frequency: 0.57 times per day). The participants consumed more foods containing nonheme iron than heme iron (114 vs. 71 g per day). They also consumed more foods containing inhibitors than foods containing enhancers of iron absorption (200 vs. 100 g day).

Table 3. Diet and Nutrition Intake of Participants per Day

Consumption	Median (range)
Heme (gram)	71.43 (6.7 – 610)
Nonheme (gram)	114.28 (0 – 642.9)
Inhibitor (gram)	200 (0 – 603.3)
Enhancer (gram)	100 (0 – 900)
Tea (frequency)	0.57 (0 – 7)
Tannin level (mg/mL)	0.157 (0 – 2.0)

According to the linear model analysis adjusted for serum ferritin levels, time since last pregnancy <2 years, parity (more than two children), reduced consumption of foods containing heme, and level of tannin consumption were predictors of iron-deficiency anemia (Table 4).

Table 4. General Linear Model on the Association between Serum Ferritin and Tannin Level

Parameter	Coef.	95% CI	
		Lower	Upper
Distance of pregnancy	-10.308	-19.626	-0.991
Parity	5.102	-4.605	14.809
Heme consumption	-0.043	-0.089	0.003
Tannin level (Low)	7.325	-5.250	19.901
Tannin level (Med)	-1.165	-13.006	10.676
Tannin level (High)	-1.835	-14.577	10.908
Constant	28.923		

Based on this model, this study developed an equation, which source from the effect size (coefficient):

$$\text{Predicted SF } (\mu\text{g/l}) = 28.923 \text{ (intercept)} - 10.308 \times \text{Time since last pregnancy (more than two-years)}^a + 5.102 \times \text{Parity (more than two children)}^b - 0.043 \times \text{Consumption of food containing heme (gram/day)} + 7.325 \times \text{Tannin low (mg/mL/day)}^c - 1.165 \times \text{Tannin medium (mg/mL/day)} - 1.835 \times \text{Tannin high (mg/mL/day)}^c$$

^a As opposed to less than two years
^b As opposed to less than two children
^c As opposed to not consumption of tea

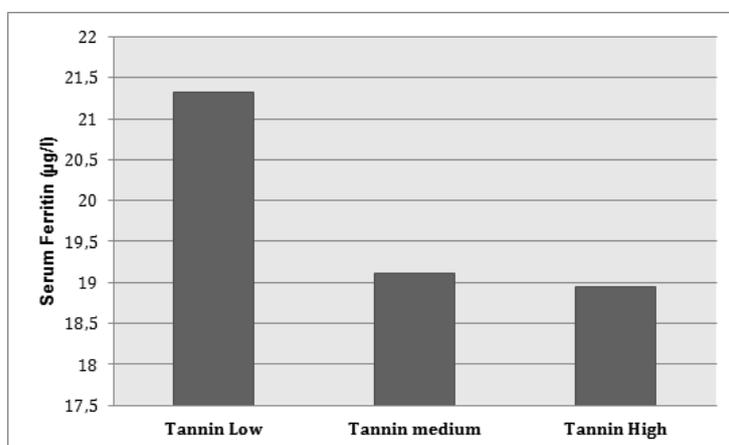


Figure 1. Predicted Serum Ferritin Related to Tannin Consumption according the Linier Model

Figure 1 shows predicted serum ferritin levels based on average tannin consumption levels and average heme consumption per day. The results suggested that the tannin level was inversely proportional to the serum ferritin level. Pregnant women who consumed tea with a low tannin level had the highest serum ferritin level as compared with pregnant women who consumed tea with medium and high tannin levels.

DISCUSSION

In this study, 94 of 104 pregnant women (participant rate 90.4%) were included in the final analysis. There are six of whom consumed tea made from loose leaves rather than tea bags and two who never drank tea were excluded from this study. The other two respondents could not take part in the interview due to health reasons.

According to the findings of the present study, time since last pregnancy, parity, protein consumption, and tea consumption (inhibit factor) predicted iron deficiency. A previous review also concluded that a short recovery between pregnancies had a strong impact on iron-deficiency anemia.⁴ A number of studies reported that parity was a strong predictor of iron-deficiency anemia among pregnant women.^{6,15,16} A study conducted in Oman reported a hazard ratio of 3.7 (95% confidence interval [CI] = 3.80-4.91) for a high-parity group as compared with that of a low-parity groups.⁶ A study results of a multivariable analysis revealing that the likelihood of anemia was increased among high-parity women as compared with a low parity group.⁷

In this study, heme consumption seemed to confer a protective effect against anemia or iron deficiency in pregnant women. A previous study found a similar result, reporting that heme iron absorption derived from animal products (meat, fish, poultry, and eggs) conferred more protection against anemia than nonheme obtained from plant products.¹⁷ The same study reported that tea consumption inhibited iron absorption only when it was consumed simultaneously with foods containing nonheme iron.¹⁷

Protein compounds can increase iron absorption in the body, which the animal-based products provide a good quality of protein material and aid iron absorption.¹⁸ In the present study, plant-based products, such as tofu and tempeh, were the most common types of food consumed by the respondents. These products have low iron contents. A previous study of the effects of dietary factors on the absorption of iron, including heme, reported that they were not necessarily associated with iron status and iron stores (as assessed by serum ferritin concentrations).¹⁰ In the study, only meat intake was consistently (positively) associated with elevated serum ferritin concentrations.

The present study suggested that tea consumption played an important role in iron deficiency, with tannin levels of tea showing a positive correlation with serum levels of ferritin. Previous research demonstrated that tea consumption decreased iron absorption in the gastrointestinal tract.¹⁷ The authors attributed this finding to polyphenols in tea forming

insoluble complexes with iron, therefore making iron unavailable for absorption.⁸ A previous study reported that drinking tea without milk inhibited the absorption of iron from solutions of Iron Chloride ($t = 2.68, p < 0.05$) and Ferrous Sulfate containing ascorbic acid ($t = 4.46, p < 0.01$).¹² A case study conducted in Italy showed that a young woman with hypermenorrhea and iron-deficiency anemia failed to respond to oral iron treatment until cessation of a long-established habit of consuming large quantities of tea.⁹ Another study reported a significant reduction in iron absorption when a test meal was accompanied by tea instead of water.¹⁹ A previous research found that there was no association between serum ferritin concentrations of various groups (men, premenopausal women, and postmenopausal women) and the type of tea (e.g., black, green, or herbal) consumed.¹⁵ The same study found no difference in serum ferritin concentrations of different tea-drinking groups with respect to tea strength, infusion time, or drinking time. In a systematic review of 35 studies on the impact of tea consumption on iron status in a U.K. study population, the authors concluded that although tea consumption limited the absorption of nonheme iron from the diet, there was insufficient evidence to draw conclusions about the effect of tea consumption on indicators of overall iron status.¹⁶ They also concluded that whether milk was added to tea had little impact on the findings. They suggested that there was no reason for healthy people with a minimal risk of iron deficiency to restrict tea consumption,

whereas those with a risk of iron deficiency should avoid tea consumption at mealtimes.

Previous studies suggested that infant feeding practices, food habits, parasitic infection, parity, early age at marriage, gestational length, and geographical location were among the most important factors associated with iron-deficiency anemia in pregnant women.²⁰ Some studies reported that maternal age, gestational age, serum ferritin concentrations, and C-reactive protein were not predictors of anemia, whereas primigravida status (OR = 2.7, 95% CI = 1.1-6.7, $p = 0.02$), low S-albumin (OR = 5.9, 95% CI = 1.4-25.2, $p = 0.01$), and low S-zinc (OR = 2.6, 95% CI = 1.0-6.6, $p = 0.03$) were predictors of anemia.¹¹ Some studies also found that gestational age, ethnicity, residence, and income were significantly associated with hemoglobin concentrations and the prevalence of anemia.^{10,11,17,24,25} They also reported that hemoglobin concentrations of pregnant women decreased with increased gestational age.^{10,21} Socioeconomic status and breastfeeding were reported to influence the onset of anemia.²¹ Women from minority groups had higher odds of anemia as compared with those from nonminority groups.^{22,23} In addition, women with high parity, long duration of breastfeeding, and high socioeconomic levels had low rates of anemia.^{22,23} Previous research suggested that the association of tea consumption with iron status did not hold true in Western populations, where most people had adequate iron stores, as determined by serum ferritin concentrations.^{21,23}

We attempted to minimize the existence of selection bias or information bias in this study. We used a structured questionnaire to prevent diagnostic bias during the interviews and also confirm the iron status through the results of laboratory tests of serum ferritin concentrations. There was a potential recall bias in this study related to nutritional data information although this study already use the Food Frequency Questionnaire (FFQ) for assess the consumption, which included questions about daily tea consumption and tea-making habits, based on 24-h recall. Measurement bias may have been introduced by assuming that the tea bag soaking times, hot water sources, and daily tea consumption frequencies remained unchanged.

CONCLUSIONS

According to the findings of the present study, four factors appeared to affect serum ferritin levels of pregnant women: time since last pregnancy (<2 years), parity (more than two children), reduced consumption of foods containing heme, and level of tannin consumption. The tannin level was inversely proportional to serum ferritin levels. Thus, pregnant women who consumed tea with a low tannin level had the highest serum ferritin levels, whereas those who consumed tea with medium and high tannin levels had the lowest serum ferritin levels.

SUGGESTION

Pregnant women should not consume tea combined with meals due to the negative effects of tannin that affected iron binding in food.

ACKNOWLEDGEMENT

The author would like to thank to Bogor District Health Office for taking research data.

REFERENCES

1. World Health Organization. *The Global Prevalence of Anaemia in 2011*. Geneva: World Health Organization;2015.
2. Api O, Breyman C, Cetiner M, Demir C, Ecdar T. Diagnosis and Treatment of Iron Deficiency Anemia during Pregnancy and the Postpartum Period: Iron Deficiency Anemia Working Group Consensus Report. *Turk J Obstet Gynecol*. 2015;12(3):173-81.
3. Badan Penelitian dan Pengembangan Kesehatan. *Riset Kesehatan Dasar (Riskesdas) 2013*. Jakarta: Badan Penelitian dan Pengembangan Kesehatan; 2013.
4. Breymann C. Iron Deficiency Anemia in Pregnancy. *Expert Review Obstet Gynecol*. 2013;8(6):587-96.
5. Santi B. Pengaruh Pemberian Suplemen Tablet Besi Folat dan Suplemen Multivitamin Mineral terhadap Kadar Hemoglobin pada Ibu Hamil Anemia di Kabupaten Kuningan Tahun 2006. Tesis. Depok: Universitas Indonesia, 2007.
6. Al-Farsi YMI. Effect High Parity on Occurance of Pre-diabetes, Low Birth Weight, and Anemia in Pregnancy in Tha Sultanate of Oman. Dissertation. Boston: Boston Universty, 2008.
7. Derso T, Abera C, Tariku A. Magnitude and Associated Factors of Anemia among Pregnant Women in Dera District: A Cross-

- sectional Study in Northwest Ethiopia. *BMC Research Notes*. 2017;10(359):1-8.
8. Samman S, Sandstorm B, Toft MB, Bukhave K, Jensen M, Sorensen SS, et al. Green Tea or Rosemary Extract Added to Foods Reduce Nonheme-Iron Absorption. *American Journal Clinical Nutrition*. 2001;73(3):607-12.
 9. Gabrielli GB, Sandre GD. Excessive Tea Consumption can Inhibit the Efficacy Oral Iron Treatment in Iron-Deficiency Anemia. *Haematologica*. 1995;80(6):518-20.
 10. Xing Y, Yan H, Dang S, Zhuoma B, Zhou X, Wang D. Hemoglobin Levels and Anemia Evaluation during Pregnancy in the Highlands of Tibet: A Hospital-based Study. *BMC Public Health*. 2009;9(336):1-7.
 11. Mohamed AA, Ali AA, Ali NI, Abusalama EH, Elbashir MI, Adam I. Zinc, Parity, Infection, and Severe Anemia among Pregnant Women in Kassala, Eastern Sudan. *Biol Trace Elem Res*. 2011;140(3):284-90.
 12. Disler PB, Lynch SR, Charlton RW, Torrance JD, Bothwell TH, Walker RB, et al. The Effect of Tea on Iron Absorption. *Gut*. 1975;16(3):193-200.
 13. Machmud PB. Pengaruh Kadar Tanin pada Teh Celup terhadap Anemia Gizi Besi (AGB) pada Ibu Hamil di UPT Puskesmas Citeureup Kabupaten Bogor Tahun 2012. Tesis. Depok: Universitas Indonesia, 2013.
 14. Mwau M, Bwana P, Kithinji L, Ogollah F, Ochieng S, Akinyi C, et al. Mother-to-Child Transmission of HIV in Kenya: A Cross-Sectional Analysis of the National Database Over Nine Years. *PLoS One*. 2017;12(8):1-15.
 15. Hyder SZ, Persson LA, Chowdhury A, Ekstrom EC. Anaemia among Non-Pregnant Women in Rural Bangladesh. *Public Health Nutrition*. 2000;4(1):79-83.
 16. Eckhardt C, Torheim L, Monterrubio E, Barquera S, Ruel M. The Overlap of Overweight and Anaemia among Women in Three Countries undergoing the Nutrition Transition. *European Journal of Clinical Nutrition*. 2008;62(2):238-46.
 17. Beck KL, Conlon CA, Kruger R, Coad J. Dietary Determinants of and Possible Solutions to Iron Deficiency for Young Women Living in Industrialized Countries: A Review. *Nutrients*. 2014;6(9):3747-76.
 18. Nguyen P, Gonzales-Casanove I, Nguyen H, Pham H, Truong T, Nguyen S, et al. Multicausal Etiology of Anemia among Women of Reproductive Age in Vietnam. *European Journal of Clinical Nutrition*. 2015;69(1):107-13.
 19. Kaltwasser JP, Werner E, Schalk K, Hansen C, Gottschalk R, Seidi C. Clinical Trial on the Effect of Regular Tea Drinking on Iron Accumulation in Genetic Haemochromatosis. *Gut*. 1998;43(5):699-704.
 20. Musaiger AO. Iron Deficiency Anaemia Among Children and Pregnant Women in the Arab Gulf Countries: The Need for Action. *Nutr Health*. 2002;16(3):161-71.
 21. Khader A, Madi H, Riccardo F, Sabatinelli G. Anaemia among Pregnant Palestinian Women in the Occupied Palestinian Territory. *Public Health Nutrition*. 2009;12(12):2416-20.
 22. Looker AC, Dalman PR, Carrol MD,

Gunter EW, Johnson CL. Prevalence of Iron Deficiency in United States. *JAMA*. 1997;277(12):973-6.

23. Pei L, Ren L, Wang D, Yan H. Assessment

of Maternal Anemia in Rural Western China between 2001 and 2005: A Two-Level Logistic Regression Approach. *BMC Public Health*. 2013;13(366):1-9.