Rapid Recovery Time of Hemoglobin Level in Female Regular Blood Donors with Ferrous Fumarate and High Dose of Ascorbic Acid Supplement

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Background: *Iron deficiency anemia has occurred more frequently in female than male regular blood donors. Iron supplement is necessary for maintaining the hemoglobin level. A combination with ascorbic acid increases the absorption of iron.*
Objective: *Compare the effect of ferrous fumarate 200 mg/day and the combination of ferrous fumarate 200 mg/day with ascorbic acid 100mg /day and 500mg /day on hemoglobin level and red blood cell indices in female regular blood donors.*
Material and Method: *Female regular blood donor volunteers were randomly divided into three equal groups to supplement with each regimen for one month. Hemoglobin (Hb), MCV, MCH, and MCHC before and after blood donation at 0.5, 1, 2, and 3 months, were compared.*
Results: *Ninety-six volunteers were included and equally divided into three groups of 32 volunteers each. There were no significant differences between age, BMI, ABO blood group, Rh, Hb, MCV, MCH, and MCHC before blood donation between each group ($p>0.05$). The duration of recovery times of Hb to before blood donation levels in group I, II, and III being 2, 3, and 1 month, respectively were statistically significant difference ($p<0.001$). The duration of recovery times of MCV, MCH, and MCHC to before blood donation levels in both group II and III were similar (0.5 months in every value), which was more rapid than in group I (>3, 3, and 1 month, respectively) with statistically significant difference ($p<0.001$). All three groups tolerated well. No participant withdrew from the present study because of side effects.*
Conclusion: *The present study shows that a combination of ferrous fumarate 200 mg and ascorbic acid 500 mg per day accelerates timing of hemoglobin and red blood cell indices in recovery to the level of before blood donation in female regular blood donors.*

Keywords: *Hemoglobin, Blood donor, Ferrous fumarate, Ascorbic acid, Iron supplement*

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Regular blood donors will have decreased iron storage and have 20 to 30% iron deficiency. There were more prevalence of anemia[1,2], iron deficiency[3,4], and risk of iron deficiency in female than in male donors. In Canada, female regular blood donors had iron deficiency (25.9%), decreased iron storage (36.8%), and normal iron storage (37.3%) after blood donation[5]. The iron storage was 200 to 400 mg in female[6] whereas, it was 1,000 mg in male[7]. After blood donation, iron is mobilized from the iron storage for erythropoiesis to replace the red blood cell loss in donation. Each time of donation will have 200 to 250 mg iron loss[8].

In iron deficiency, the erythropoiesis will decrease gradually until the hemoglobin level reach the level of anemia in several months. In normal female, after blood donation, the iron absorption will increase from dietary to 0.4 mg/day[9]. That requires 1.5 years to replace the iron from blood loss. Therefore, the chance of repeat donation will be decreased[10]. In Thai dietary, there is a lower iron amount and bioavailability than western dietary[11]. With heme iron such as in meat, blood with high bioavailability will have 20 to 30% absorption rate. However, with non-heme iron in inorganic form or in plants such as rice, nut, potato, vegetable and milk, the bioavailability depends on the factors that increases absorption such as meat and ascorbic acid or that inhibits absorption such as carbonate, oxalate, phosphate, tannate, phytate, and high fiber vegetables. The iron absorption pathway in the small intestine will be different. Heme iron is absorped directly by heme iron transporter (HCPI) under control of other factors such as iron deficiency or hypoxia[12]. Non-heme iron in dietary will be changed to ferric form in the intestine. Then,

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it has to change to ferrous form by ferric reductase enzyme such as duodenal cytochrome b (DCYTB) at brush border or by ascorbic acid in that meal[13].

The iron deficiency anemia will be improved by iron supplement[14]. Therefore, the iron supplement will be necessary in blood donors. The recommendation of iron supplement with carbonyl iron 100 mg/day for 56 days is used in regular blood donors in USA. This program can replace 85% of the iron loss from blood donation. The iron supplement in high dosage such as 600 mg/day divided in three intakes daily for one week will increase side effects and is unacceptable. Iron supplement with one tablet once daily for one month can help most of the donors that had been rejected to success repeat blood donation again[15]. Thammasat blood transfusion service unit gives the blood donor a package of 30 tablets of ferrous fumarate 200 mg for using one tablet daily. This can help the donor to repeat donation in every three months. Because the regular blood donors have a lower prevalence of infectious diseases than new blood donor two or three times and also lower unit cost in regular blood donor, so it is important to maintain the regular blood donation program. Ascorbic acid combined with iron supplement will increase the iron absorption and iron utilization[16]. High dose ascorbic acid such as ascorbic:iron = 2:1 to 4:1 molar ratio increases the iron absorption[17]. The dosage of ascorbic acid up to 200 mg/day will increase the iron absorption[18]. The supplement of ferrous fumarate combined with ascorbic acid for nine months will increase the hemoglobin to normal level (>10.5 g/dl)[19]. The intake of high dosage of ascorbic acid such as 2 g/day orally or 1 gm/day intravenously for one year is safe[20,21].

Therefore, the present study aimed to compare the effect of ferrous fumarate alone and a combination with ascorbic acid low dose and high dose on hemoglobin in female regular blood donors in order to maintain their donation ability.

Material and Method

The present study was approved by the ethical committee of Thammasat University. The volunteer female regular blood donors, those that donated four times, every three months, in consecutive years and were still active for at least one year, who were not pregnant and came to donate blood at the Thammasat blood transfusion service unit were recruited for study between April and October 2010. The standard procedure used to screen donors by the National Blood Centre, Thai Red Cross Society[22] was followed. All donors were determined to be healthy based on their histories and hemoglobin concentration as evaluated by HemoCue® (Angelheim, Sweden) was 12.5 g/dl or above for screening and by Automate cell counter (LH780, PCL Holding, USA) for baseline study levels. Informed consents were obtained from all volunteers. The volunteers were randomly divided equally into three groups by computer to take a package of 30 tablets (doses) of ferrous fumarate 200 mg in group I, combination of a package of 30 tablets (doses) of ferrous fumarate 200 mg plus a package of 30 tablets (doses) of ascorbic acid 100 mg in group II, and combination of a package of 30 tablets (doses) of ferrous fumarate 200 mg plus a package of 30 tablets (doses) of ascorbic acid 500 mg in group III. Each volunteer had taken one dose or one combination dose once a day between meals for 30 days, after blood donation. Approximately 450 ml of venous blood was collected in each volunteer. The scheduled of 0.5, 1, 2, and 3 months visits after donation for taking 3 ml of venous blood to use in testing of red blood cells indices (Hb, MCV, MCH, and MCHC) by Automate cell counter. All volunteers were informed of the importance of regular drug supplement taking and they were questioned about their compliance, any side effects including constipation, nausea, vomiting, abdominal discomfort, abdominal cramp, diarrhea, and melena at each visit.

Statistical analysis

The sample size of 32 for each group was calculated based on Hb concentration value before blood donation and used the mean difference effect size of 0.52 (mean difference divided by standard deviation) with confident at 95% and power of test at 95%. Mean ± standard error are used for reporting age, BMI, Hb, MCV, MCH, and MCHC while frequency and percentage are done for blood groups and types. ANCOVA with repeated measure was used to compare the means difference among the groups and Chi-square test was used for frequency data. All the test significances performing based on alpha level at 0.05.

Results

Ninety-six volunteer female regular blood donors were included and randomly divided equally into three groups. The mean age was 34.78±1.92, 31.94±1.70, and 33.59±1.65 years in group I, II, and III respectively, with no significant difference (p = 0.52). The distribution percentage of A, B, O, and AB blood groups was 18.75, 21.88, 56.25, and 3.12% vs. 18.75,

Table 1. Age, body mass index and the distribution of blood group of female regular blood donors in all 3 groups

Group	Age (year) (mean±SE)	BMI* (mean±SE)	Blood group, n (%)				
			A	B	O	AB	Rh(+)
Group I ferrous fumarate (200 mg/day)	34.78±1.92	21.23±0.29	6 (18.75%)	7 (21.88%)	18 (56.25%)	1 (3.12%)	32 (100%)
Group II ferrous fumarate (200 mg/day) + ascorbic acid (100 mg/day)	31.94±1.70	21.04±0.30	6 (18.75%)	13 (40.63%)	12 (37.50%)	1 (3.12%)	32 (100%)
Group III ferrous fumarate (200 mg/day) + ascorbic acid (500 mg/day)	33.59±1.65	20.75±0.29	3 (9.38%)	10 (31.25%)	18 (56.25%)	1 (3.12%)	32 (100%)
p-value	0.52	0.53	0.49	0.27	0.23	na**	na**

* Body mass index (BMI) = weight (kg)/height^2 (m^2)
** na = staitistical test is not applicable

40.63, 37.50, and 3.12% vs. 9.38, 31.25, 56.25, and 3.12% in group I vs. II vs. III respectively, with no significant difference (p>0.05). The distribution percentage of A, B, O, and AB blood groups were no significant difference between each group (p>0.005). All of them were Rh positive. The mean body mass index was 21.23±0.29, 21.04±0.30, and 20.75±0.29 kg/m^2 in group I, II, and III respectively. There was no significant difference (p = 0.53) (Table 1).

The mean values of Hb, MCV, MCH, and MCHC before blood donation were 12.82±0.10 g/dl, 88.82±1.29 fl, 29.24±0.42 pg, and 32.66±0.09 g/dl vs. 13.09±0.12 g/dl, 88.63±1.06 fl, 29.13±0.42 pg, and 32.78±0.09 g/dl vs. 12.93±0.10 g/dl, 88.11±0.85 fl, 28.80±0.35 pg, and 32.81±0.10 g/dl in group I vs. II vs. III, respectively, with no significant difference (p>0.05). During 0.5 months after blood donation the mean value of Hb level was significantly decreased in all three groups (p<0.001) (Fig. 1). The recovery time of mean value of Hb to before blood donation level was two months (12.72±0.10 g/dl) vs. three months (13.01±0.10 g/dl) vs. one month (12.68±0.10 g/dl) in group I vs. II vs. III, respectively with statistically significant difference (p<0.001) (Fig. 1). During 0.5 months after blood donation the mean values of MCV, MCH, and MCHC in group I were significantly decreased (p<0.001) (Fig. 1-4). The recovery times of mean values of MCV, MCH, and MCHC to before blood donation level in both groups II and III were similar, being 0.5 months (88.61±0.14 fl and 88.48±0.14 fl), 0.5 months (29.12±0.07 pg and 29.11±0.07 pg), and 0.5 months (32.80±0.08 g/dl and 32.81±0.08 g/dl), which more rapid than in group I being >3 months [The value of 88.21±0.16 fl at 3 months was still below the value of 88.82±1.29 fl at the time before blood donation with statistically significant difference (p<0.001).], 3 months (29.14± 0.21 pg) and 1 month (32.52±0.08 g/dl), respectively with statistically significant difference (p<0.001) (Fig. 1-4).

Significance of mean difference were found on every point (p<0.001) during following-up (Fig. 1-4).

Side effects of ferrous fumarate or ascorbic acid supplements such as constipation, abdominal discomfort, abdominal cramp, nausea, diarrhea, and melena in the volunteers in each group were not serious and no significant difference (p>0.05) (Table 2).

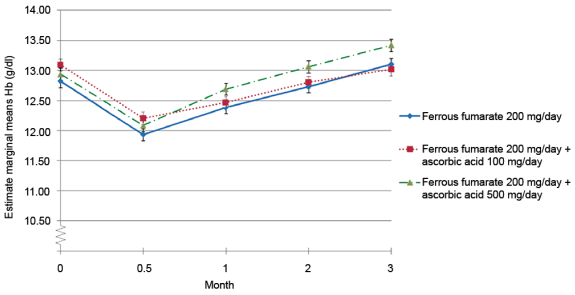

Fig. 1 The Hb level before and after blood donation of the female regular blood donors in all 3 groups.

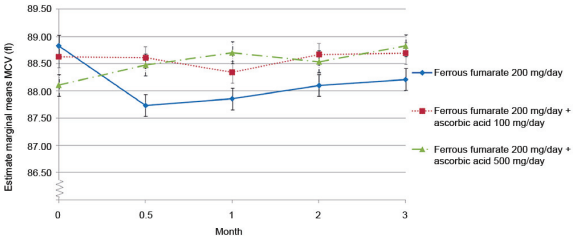

Fig. 2 The MCV value before and after blood donation of the female regular blood donors in all 3 groups.

Table 2. Side effects of ferrous fumarate or ascorbic acid supplements in the volunteers female regular blood donors in all 3 groups

Group	Side effect (n)					
	Constipation	Abdominal discomfort	Abdominal cramp	Nausea	Diarrhea	Melena
Group I ferrous fumarate (200 mg/day)	19	15	2	2	1	32
Group II ferrous fumarate (200 mg/day) + ascorbic acid (100 mg/day)	17	17	1	1	0	32
Group III ferrous fumarate (200 mg/day) + ascorbic acid (500 mg/ day)	16	14	1	1	0	32
p-value	0.75	0.75	na*	na*	na*	na*

* na = statistical test is not applicable

Discussion

In the present study during 0.5 months after blood donation, the Hb level was still significantly decreased in all three groups. The present study showed that in the ferrous fumarate 200 mg per day plus ascorbic acid 500 mg per day supplement group, the Hb level increased most rapidly within one month after blood donation to reach the level of before blood donation. This was followed by the group of supplement with ferrous fumarate 200 mg per day alone, the recovery time of Hb level was two month,

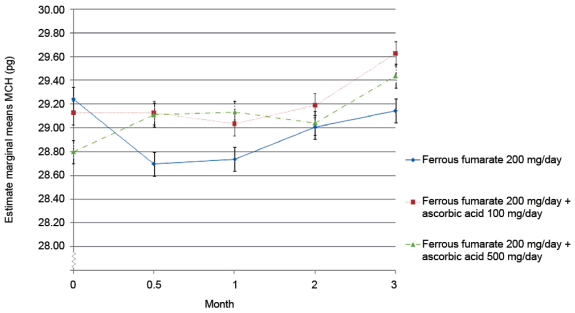

Fig. 3 The MCH value before and after blood donation of the female regular blood donors in all 3 groups.

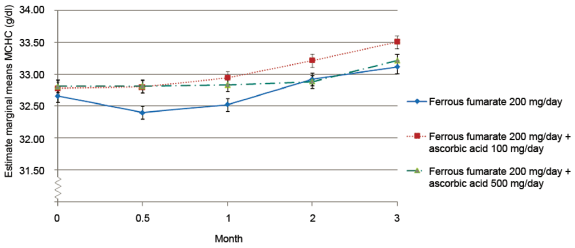

Fig. 4 The MCHC value before and after blood donation of the female regular blood donors in all 3 groups.

and the group of ferrous fumarate 200 mg per day plus ascorbic acid 100 mg per day the recovery time was the slowest (3 months). The present study supported the previous evidences of ascorbic acid increases the iron absorption, which result in rapidly increasing hemoglobin level[17,18]. The present study also showed that the other red blood cell indices in both ascorbic acid supplement groups had similar and simultaneous rapid recovery times (MCV, MCH and MCHC in 0.5 months). These values still significantly decreased in this period in the group of supplement with ferrous fumarate alone. The supplement with low dose ascorbic acid 100 mg per day plus ferrous fumarate 200 mg per day seemed to be not effective in increasing hemoglobin level when compared to ferrous fumarate 200 mg per day alone. The recovery time of Hb level was longer but the recovery times of other red blood cell indices(MCV, MCH and MCHC) were shorter, respectively. This result is similar to the previous study showing that the dosage of ascorbic acid supplement should be 200 mg per day in increasing the iron absorption[18]. Additionally, the recent study found that the dosage of ascorbic acid supplement with iron that would be able to increase iron absorption should be required up to 2:1 to 4:1 molar ratio[17].

The present study showed that there was no difference in the side effects of ferrous fumarate 200 mg per day supplement alone and either plus ascorbic acid 100 mg per day or 500 mg per day for one month. No participant withdrew from the present study because of side effects. This could also be supported by the other report that it was safe to take ascorbic acid 2 grams per day orally for one year[20].

The results of the present study will be useful in regular blood donor retention program especially

female donors who had decreased hemoglobin level and increased risk of iron deficiency anemia. The supplement of ascorbic acid 500 mg per day with ferrous fumarate 200 mg per day for one month may be more convenient to the regular blood donor and shorter time than taking ferrous fumarate 200 mg per day for 56 days as the recommendation dosage in USA. Additionally, it is also more effective than the supplement with ferrous fumarate 200 mg per day alone for one month. This regimen is safe, has no serious side effect, and no withdrawal result of side effect. This regimen will be applicable to autologous blood donation program because of rapid recovery of hemoglobin level and other red blood cell indices within one month that is more suitable to the timing of the elective surgical treatment than conventional program. This protocol is also more friendly to the donor compared to the increasing dosage of ferrous fumarate alone protocol with having more side effects[10].

The disadvantage of this protocol is that the ascorbic acid is more expensive and should be kept in dark storage.

The present study did not study the serum ferritin level that would be more accurate in evaluation the recovery of iron storage in blood donors; it will be worthwhile for further study.

Further study should pay attention to the cost effectiveness of the appropriate dosage of ascorbic acid supplement with ferrous fumarate using in an expanded regular blood donor retention program.

Conclusion

The recovery time of the value of Hb to the level of before blood donation in the ferrous fumarate 200 mg per day plus ascorbic acid 500 mg per day group is the most rapid within one month. It is followed by within two months in the ferrous fumarate 200 mg per day alone group whereas the ferrous fumarate 200 mg per day plus ascorbic acid 100 mg per day group is the slowest within three months. The recovery times of the values of other red blood cell indices (MCV, MCH, and MCHC) to the level of before blood donation in the ferrous fumarate 200 mg per day plus ascorbic acid 500 mg per day group and the ferrous fumarate 200 mg per day plus ascorbic acid 100 mg per day group are similar within 0.5, 0.5 and 0.5 months, respectively which more rapid than within >3 months, 3, and 1 months respectively in the ferrous fumarate 200 mg per day alone group.

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Potential conflicts of interest

None.

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การเสริมธาตุเหล็กร่วมกับวิตามินซีในขนาดสูง ช่วยให้ระดับฮีโมโกลบินกลับคืนสู่ระดับก่อนการบริจาคโลหิตได้เร็วในผู้หญิงที่บริจาคโลหิตเป็นประจำ

ณิชาภา เจียมจรรยา

ภูมิหลัง: *ภาวะโลหิตจางจากการขาดธาตุเหล็ก เนื่องจากการบริจาคโลหิตบ่อยครั้ง โดยเฉพาะในผู้บริจาคโลหิตเพศหญิงมีความเสี่ยงที่จะขาดธาตุเหล็กมากกว่าเพศชาย การเสริมธาตุเหล็กมีความจำเป็นสำหรับการรักษาระดับฮีโมโกลบินให้อยู่ในเกณฑ์ปกติ และการให้วิตามินซีร่วมด้วยจะเพิ่มการดูดซึมธาตุเหล็ก*

วัตถุประสงค์: *การศึกษาครั้งนี้เพื่อเปรียบเทียบผลของการให้ธาตุเหล็ก (เฟอรัส ฟูมาเรต 200 มิลลิกรัม) วันละ 1 เม็ด ทุกวันเป็นเวลา 1 เดือน และการให้ธาตุเหล็ก (เฟอรัส ฟูมาเรต 200 มิลลิกรัม) ร่วมกับวิตามินซี 100 มิลลิกรัม หรือ 500 มิลลิกรัม อย่างละ 1 เม็ด ทุกวันเป็นเวลา 1 เดือน แก่ผู้บริจาคโลหิตเพศหญิงที่บริจาคโลหิตเป็นประจำ เพื่อช่วยเพิ่มระดับฮีโมโกลบินและค่าดัชนีของเม็ดเลือดแดง*

วัสดุและวิธีการ: *แบ่งอาสาสมัครที่จะบริจาคโลหิตเป็น 3 กลุ่ม โดยการสุ่มด้วยคอมพิวเตอร์ ในการรับประทานยาอย่างใดอย่างหนึ่งหลังจากการบริจาคโลหิตแล้วทำการเจาะเลือดจำนวน 4 ครั้ง คือ 0.5 เดือน, 1 เดือน, 2 เดือน และ 3 เดือนหลังให้ยา เพื่อวัดค่าฮีโมโกลบิน (Hb), ปริมาตรของเม็ดเลือดแดงโดยเฉลี่ย (MCV), ปริมาณเฉลี่ยฮีโมโกลบินในเม็ดเลือดแดง (MCH) และความเข้มข้นเฉลี่ยของฮีโมโกลบินในเม็ดเลือดแดง (MCHC)*

ผลการศึกษา: *อาสาสมัครจำนวน 96 ราย แบ่งออกเป็น 3 กลุ่ม กลุ่มละ 32 คน มีค่า Hb, MCV, MCH และ MCHC ก่อนเจาะเลือดดังนี้ กลุ่มที่ 1 รับประทานธาตุเหล็กเพียงอย่างเดียวมีค่า 12.82±0.10 g/dl, 88.82±1.29 fL, 29.24±0.42 pg และ 32.66±0.09 g/dl ตามลำดับ กลุ่มที่ 2 รับประทานธาตุเหล็กร่วมกับวิตามินซี 100 มิลลิกรัม มีค่า 13.09±0.12 g/dl, 88.63±1.06 fL, 29.13±0.42 pg และ 32.78±0.09 g/dl ตามลำดับ กลุ่มที่ 3 รับประทานธาตุเหล็กร่วมกับวิตามินซี 500 มิลลิกรัม มีค่า 12.93±0.10 g/dl, 88.11±0.85 fL, 28.80±0.35 pg และ 32.81±0.10 g/dl ตามลำดับ ซึ่งทั้ง 3 กลุ่มไม่มีความแตกต่างกัน จากผลการศึกษาพบว่าค่า Hb จะเพิ่มขึ้นจนเทียบเท่ากับระดับก่อนบริจาคโลหิตในระยะเวลาดังนี้ กลุ่มที่ 1 ในระยะเวลา 2 เดือน (12.72±0.10 g/dl) กลุ่มที่ 2 ในระยะเวลา 3 เดือน (13.01±0.10 g/dl) กลุ่มที่ 3 ในระยะเวลา 1 เดือน (12.68±0.10 g/dl) ซึ่งมีความแตกต่างกันอย่างมีนัยสำคัญทางสถิติ ($p<0.001$) ค่า MCV, MCH และ MCHC จะเพิ่มขึ้นจนเทียบเท่ากับระดับก่อนบริจาคโลหิตในระยะเวลาเท่ากัน ในกลุ่มที่ 2 และ 3 คือ 0.5 เดือน, 0.5 เดือน และ 0.5 เดือน ตามลำดับ ซึ่งเร็วกว่ากลุ่มที่ 1 ที่มีระยะเวลามากกว่า 3 เดือน, 3 เดือน และ 1 เดือน ตามลำดับ อย่างมีนัยสำคัญทางสถิติ ($p<0.001$) โดยพบว่าอาสาสมัครทั้ง 3 กลุ่ม สามารถรับประทานธาตุเหล็กหรือวิตามินซี และทนต่ออาการข้างเคียงได้ดี*

สรุป: *กลุ่มที่รับประทานธาตุเหล็กร่วมกับวิตามินซี 500 มิลลิกรัม จะมีค่า Hb, MCV, MCH และ MCHC กลับมาเท่ากับระดับก่อนบริจาคโลหิตในระยะเวลาเร็วที่สุด*