The Turkish Journal of Pediatrics 1997; 39: 465-471

ABSORPTION OF IRON FROM GRAPE – MOLASSES AND FERROUS SULFATE: A COMPARATIVE STUDY IN NORMAL SUBJECTS AND SUBJECTS WITH IRON DEFICIENCY ANEMIA*

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SUMMARY: Aslan Y, Erduran E, Mocan H, Gedik Y, Ökten A, Soylu H, Değer O. (Departments of Pediatrics and Biochemistry, Karadeniz Technical University Faculty of Medicine, Trabzon, Turkey). Absorption of iron from grape-molasses and ferrous sulfate: a comparative study in normal subjects and subjects with iron deficiency anemia. Turk J Pediatr 1997; 39: 465-471.

We compared the absorption of iron from grape molasses (GM) and ferrous sulfate (FS) using the post-absorptive serum iron increase method (non-radioactive). The study involved 56 subjects, aged 6-36 months. Group I consisted of 30 subjects with iron deficiency anemia (IDA) and group II, 26 non-anemic subjects. Each group was subdivided randomly into two equal subgroups to be given either GM or FS. The absorption of the iron from GM was monitored in 15 infants with IDA and in 13 non-anemic infants, and the absorption of iron from FS was tested in the rest of the subjects in each group.

In those infants in each group given GM or FS, there was no significant difference in before-test values for serum iron (SI) and total iron binding capacity (TIBC) ($p > 0.05$). In the group with IDA, the mean after-test SI value in FS-given infants was higher and the mean TIBC value lower than those of GM-given infants ($p < 0.05$). However, in the non-anemic group, there was no significant difference in the mean after-test SI and TIBC values in either GM- or FS-given infants ($p > 0.05$).

The mean increase of serum iron in GM-given infants with IDA was 27.0 ± 13.4 µg/dl and in FS-given infants, 60.6 ± 17.0 µg/dl ($p < 0.05$). In the non-anemic group, the mean increase of serum iron of GM-given infants was 23.0 ± 4.3 µg/dl, and that of FS-given infants, 23.8 ± 10.0 µg/dl ($p > 0.05$).

We determined that in non-anemic subjects, the absorption of iron from GM was comparable to that from FS. Furthermore, we believe that grape molasses is an effective iron source in preventing iron deficiency anemia in infancy. *Key words: iron absorption, grape molasses, ferrous sulfate.*

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Iron deficiency remains the most common cause of anemia despite improvements in diet and the widespread use of well-absorbed forms of iron-fortified foods[1]. Infants who, from early infancy, are fed cow milk[2] or formula that is not iron-fortified[3] remain especially at risk for the development of iron deficiency.

The most important factors contributing to the development of iron deficiency in infants are rapid growth and an insufficient amount or absorption of dietary iron[1,4]. Growth and mental and motor development during infancy is highly dependent on the bioavailability of dietary iron. Therefore, prevention of nutritional iron deficiency is required[5,6].

A balanced weanling diet is the key to preventing iron deficiency anemia in children[7]. A rapidly growing weanling becomes susceptible to iron deficiency when neonatal iron stores have been consumed after the first few months following birth[8]. Experience has also shown that fortification of infant foods with iron is a more reliable and effective measure than providing iron medication[9].

In this study, we compare the absorption of iron in grape molasses (GM), which is traditional infant food in Turkey, to that in ferrous sulfate (FS), and form an opinion about whether or not grape molasses is an alternative to iron-containing drugs in the prevention of iron deficiency anemia (IDA) in infancy.

Material and Methods

The study was performed on 56 subjects, aged 6-36 months, after informed consent was obtained from their parents. Group I consisted of 30 subjects with IDA (hemoglobin < 11 g/dl, transferrin saturation < 10%, ferritin < 10 ng/ml). Group II consisted of 26 non-anemic subjects (hemoglobin > 12 g/dl, transferrin saturation > 15%, ferritin > 10 ng/ml). Each group was subdivided randomly into two equal subgroups. In half of the subjects in each group (i.e. 15 in group I, 13 in group II), the absorption of iron in GM was determined, and in the other half of each group, the absorption of iron in FS was tested. Subjects known to have malabsorption syndromes, acute or chronic infections, or malignancies and those taking non-steroidal anti-inflammatory drugs or having chronic blood losses were excluded.

The iron content of GM used in the study was 9.8 mg per 100 g, which was measured by the spectrophotometrical ferrozine method (Pointe Scientific Inc) with 1/25 dilution.

The post absorptive serum iron increase method (non-radioactive) was used determine the absorption of iron from GM and FS. To avoid the well-known iron absorption inhibiting effect of the many iron complexing compounds in foods, tea, milk etc., the subjects did not take any food or beverage for six hours before and three hours after oral application. Only tap water was allowed during this period. The serum iron studies started at 8 a.m. After drawing an initial venous blood sample, GM or FS, containing 1 mg/kg iron, was given orally. The second venous blood samples were drawn after three hours. All blood samples were analyzed for serum iron (SI) and total iron binding capacity (TIBC) by routine colorimetric methods, and the increase of serum iron was calculated. Transferrin-Fe-saturation was calculated using total iron and TIBC. The serum ferritin levels were only analyzed in initial blood samples in each group. Hemoglobin, SI, TIBC, serum ferritin, and transferrin-Fe-saturation were used as the diagnostic criteria for the separation into IDA and non-anemic groups.

Statistical analyses were performed using the Wilcoxon matched-pairs signed ranks, Mann-Whitney U, and chi-square tests and Kruskal Wallis variance analysis[10].

Results

There were no statistically significant differences between the two groups in the sex and mean age of the subjects (Table I), nor among the subgroups of each group in the mean hemoglobin values ($p > 0.05$). The mean transferrin-Fe-saturation and serum ferritin level were 25.5 ± 7.1% and 48.4 ± 21.3 ng/ml, respectively, in nonanemic subjects, and 6.9 ± 1.7% and 5.2 ± 2.6 ng/ml respectively, in anemic subjects (the values are mean ± SD). Hematologic results of the subjects with IDA (before and after test) are shown in Table II, and those of non-anemic subjects in Table III. There were no significant differences in before-test SI and TIBC values in either GM- or FS-given subjects in each group ($p > 0.05$). In the group with IDA, the mean after-test SI value in FS-given infants was higher, and the mean TIBC value lower, than those of GM-given infants ($p < 0.05$). However, in the non-anemic group, there were no significant differences between the mean before- and after-test values of SI and TIBC in the GM- and FS- given subjects ($p > 0.05$).

Table I: Sex and Age of the Subjects

		Sex (Male/Female)	Age (X ± SD) (Months)
Subjects with IDA	Grape molasses (n = 15)	6/9	20.5 ± 8.9 (6-36)*
	Ferrous sulfate (n = 15)	6/9	21.4 ± 9.7 (6-36)*
Non-anemic Subjects	Grape molasses (n = 13)	6/7	20.7 ± 9.4 (6-36)*
	Ferrous sulfate (n = 13)	6/7	22.2 ± 9.4 (6-36)*

* Ranges.

Table II: Hematological Data of the Subjects with Iron Deficiency Anemia (the Values are Mean ± SD and Parenthesis Represents Minimum-Maximum Values)

		Before Test	After Test	p
Serum iron (mg/dl)	Grape molasses	32.2 ± 7.0 (18-45)	59.3 ± 18.0 (30-81)	< 0.05
	Ferrous sulfate	30.0 ± 7.6 (19-42)	90.0 ± 19.9 (56.5-120)	< 0.05
p		> 0.05	< 0.05	
TIBC (mg/dl)	Grape molasses	425 ± 59 (363-572)	361 ± 73 (245-485)	< 0.05
	Ferrous sulfate	436 ± 69 (270-557)	295 ± 58 (216 ± 352)	< 0.05
p		> 0.05	< 0.05	

Table III: Hematological Data of the Non-Anemic Subjects (the values are mean ± SD and parenthesis represents minimum-maximum)

		Before Test	After Test	p
Serum iron (mg/dl)	Grape molasses	72.2 ± 21.0 (50-119)	95.0 ± 17.5 (76-136)	< 0.05
	Ferrous sulfate	70.0 ± 17.2 (52-104)	96.0 ± 19.6 (65-120)	< 0.05
p		> 0.05	> 0.05	
TIBC (mg/dl)	Grape molasses	327 ± 32 (250-357)	287 ± 30 (224-318)	< 0.05
	Ferrous sulfate	319 ± 34 (247-370)	280 ± 34 (220 ± 320)	< 0.05
p		> 0.05	> 0.05	

The mean increase of serum iron in GM-given infants with IDA was 27.0 ± 13.4 (3-47) µg/dl and in FS-given infants with IDA, 60.6 ± 17.0 (35-91) µg/dl ($p < 0.05$). On the other hand, in the non-anemic group, the mean increase of serum iron of the GM-given subjects was 23.0 ± 4.3 (17.30) µg/dl and that of the FS-given subjects, 23.8 ± 10.0 (9-40) µg/dl ($p > 0.05$).

Discussion

Despite major advances in our knowledge of iron nutrition in infancy, iron deficiency remains a significant problem, especially in developing countries[11, 12]. Nutritional iron deficiency appears after the age of six months and is undoubtedly linked to inappropriate feeding practices[12, 13].

The Committee on Nutrition of The American Academy of Pediatrics has recommended that iron supplementation (to a maximum of 15 mg per day from one or more sources) sohuld start no later than four months of age in term infants and no later than two months of age in pre-term infants[1,4]. At present, iron-containing drops and foods fortified with iron, such as infant cereals, commercial milk formulas and enriched flour and other cereal products, have been widely used as iron sources in infants[1,4].

The bioavailability of iron in a diet is more important than the amount[1,14]. There is no doubt that absorbable forms of iron in foods will improve iron balance and prevent iron deficiency[1,14]. Recently it has been reported that micronutrient-fortified rusk[15], adapted formula with bovine lactoferrin[16], modified cow milk[17] and cereal fruit products[18] are effective in preventing iron deficiency anemia in infancy.

Grape molasses (which contains 10 mg of iron, 70.6 g of carbohydrate, 0.6 g of protein, 400 mg of calcium, and 293 kcal of energy per 100 g) is boiled grape juice[19,20]. In Turkey, GM is a traditional and inexpensive food, which is widely used in all socoeconomic groups and has been suggested by physicians as a source of iron. To our knowledge, there has been no previous study reported investigating the bioavailability or absorption of iron in grape molasses. Fomon et al.[18] determined erythrocytic incorporation of the stable isotope, Fe^{58}, after feeding the following foods (extrinsically labelled with Fe^{58}) which he claimed seemed nutritionally important as iron sources: cereal-vegetable products, grape juice fortified with ferrous sulfate, and a vitamin-, mineral-, and protein-enriched rice cereal fortified with ferrous fumarate.

In the present study, the absorption of iron in GM and FS was compared using a non-radioactive method (post-absorptive serum iron increase method). In the group with IDA, the mean increase of serum iron which was measured three hours after the oral administration of GM was very low compared to the increase measured after the administration of FS ($p < 05$). However, there was no statistically significant difference between the mean increase of serum iron from GM or FS in the non-anemic group ($p > 0.05$). We were not able to determine why the mean increase of serum iron in GM-given infants was lower than that of FS-given infants in the group with IDA. However, this may be related to the amount of GM.

The post-absorptive serum iron increase method has been used since 1934 to detect the bioavailability of the therapeutic oral iron preparations[21]. This procedure cannot be used for quantitative estimation of iron absorption in individual subjects, but it is useful for comparison of several iron preparations in a sufficient number of subjects (at least 10)[21]. In this study, this method was used for comparison of the iron absorptions of two iron sources in 56 subjects. Bioavailability is not a simple synonym of iron absorption but describes both the fractional iron absorption as well as recreation of a suitable iron reserve[22]. Furthermore, Özsoylu and Özbek[23] emphasized that iron absorption does not always correspond to its utilization for heme synthesis, which is a better criterion of iron bioavailability.

Finally, we believe that grape molasses, which is inexpensive and readily available, is a reliable and effective iron source in preventing iron deficiency anemia in infancy, but it is not effective in the treatment of IDA. Thus, grape molasses may be recommended for infants after six months of age as an artifical iron source to prevent the development of iron deficiency anemia, but only if the infants have been breastfed within the first four to six months of their lives. If an infant has been fed with artificial foods during the first four to six months, grape molasses may be recommended earlier than six months of age. However, new clinical studies about the long-term effect of grape molasses on iron balance are needed.

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