



Journal of Home Economics

Volume 25, Number (1), 2015

<http://homeEcon.menofia.edu.eg>

**Journal of Home
Economics**

ISSN 1110-2578

Comparative Study of Chelated and Non Chelated Iron in biological indices in adult Rats

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Abstract

The main objective of this study is to compare the absorption configuration and its correlated signs of biological indices of adult male Albino rats by using of different chelated and unchelated iron forms. Thirty two white male albino rats, weighting 145-150g were fed on basal diet for adaptation after adaptation period. Rats were divided into 8 groups (4 rats each group), all groups were fed for 12 weeks on experimental diet as follows: four groups were fed on 50% Fe- deficient diet (17.5mg/kg) as ferrous sulfate, ferric citrate, glycine-chelated iron, lysine-chelated iron. Another four groups were fed on adequate Fe diet (35mg/kg) as ferrous sulfate, ferric citrate, glycine-chelated iron, lysine-chelated iron. At the end of the experimental period (12 weeks), rats were fasted over night before sacrificing, blood were collected to determine biological analysis. The obtained results revealed that groups of rats fed on iron amino acid chelate were significantly increased than ferrous sulfate on hemoglobin. The results were clearly that Red blood cells and HCT was the highest in groups feeding on Fe-diets as glycine type comparing to other groups under study. Red blood cell distribution width, it could be noticed that RDW% of group of rats fed on adequate Fe diets as glycine-chelated iron showed the lowest level and significantly decreased when compared to groups of rats fed on 50% Fe- deficient diets as ferrous sulfate, ferric citrate, glycine-chelated iron and lysine-chelated iron. Serum ferritin, the best result was recorded for the group of rats fed on adequate Fe as glycine chelated iron which it. Serum iron the best results were recorded for groups of rats fed on adequate Fe diet as ferric citrate, glycine -chelated iron. Results concluded that iron amino acid chelate had a positive impact on hematological indices and iron status of adult rats.

Key words: Iron chelated. Ferrous sulfate . Ferric citrate . hematological indices.

Introduction

Iron is the most abundant trace element in mammals. It serves as a constituent in proteins (e.g. hemoproteins: hemoglobin, myoglobin; non- heme proteins: ferritin, transferrin) and as a cofactor for many important iron dependent enzymes (e.g. cytochromes ,peroxidases and catalases). Hemoglobin makes up 80% of the entire iron body pool. Iron is present in biological systems in one of two oxidation states, and redox inter conversions of the ferrous (Fe) (II) and ferric (Fe) (III) forms are central to the biological properties of this trace element. As a constituent of hemoglobin, it is involved in oxygen and carbon dioxide transport. It plays a central role as cofactor for most of the enzymes of the Krebs cycle and functions as electron carrier (**McDowell, 2003 and Suttle, 2010**).

Iron deficiency (ID) is the most common nutritional deficiency in children. The World Health Organization (WHO) estimated that anemia affects one quarter of the world's population and is concentrated within pre-school age children and women. Iron deficiency is a particularly challenging problem for developing nations in Asia and Africa (**Stoltzfus,2003 and DeBenoist et al., 2008**).

Food fortification is advocated to tackle iron deficiency in anemic populations (**Nogueira et al., 2012**).

Iron amino acid chelates, such as iron glycinate chelates, have been developed to be used as food fortificants and therapeutic agents in the prevention and treatment of iron deficiency anemia. Ferrous bis-glycine chelate (FeBC), ferric tris-glycine chelate, ferric glycinate, and ferrous bis-glycinate hydrochloride are available commercially (**Hertrampf and Olivares, 2004**).

McDowell, (2003) reported that the fraction of iron absorbed from the amount ingested is typically low, but may range from 5 to 35% depending on circumstances and type of iron.

Iron absorption from FeBC is affected by enhancers and inhibitors of iron absorption, but to a lesser extent than ferrous sulfate. Its absorption is regulated by iron stores. FeBC is better absorption from milk, wheat, whole maize flour, and precooked corn flour than is ferrous sulfate (**Hertrampf and Olivares, 2004**).

Kamdi and Palkar,(2015) showed that ferrous asparto glycinate is an effective iron-amino acid chelate in the management of IDA in pregnant women as compared with ferrous ascorbate. Nevertheless, additional large scale prospective, randomized trials are

warranted to confirm the findings of the present efficacy trial, and also to find out the anemia eradication rate.

Finally, there is an urgent need for more rigorous efficacy trials designed to define the relative merits of amino acid chelates when compared with bioavailable iron salts such as ferrous sulfate and ferrous fumarate and to determine appropriate fortification level (**Hertrampf and Olivares, 2004**). Therefore, the objective of this study was to compare the absorption configuration and its correlated signs of biological indices of adult male Albino rats by the use of different chelated and unchelated iron forms.

Materials And Methods

Materials:

Source of Materials:

Casein as main source of protein obtained from Technogen Company for Chemicals and Drugs, Giza, Egypt. Mineral mixture and Vitamin mixture were purchased from El - Gomhoria Company for Drugs, Chemical and Medical instruments Cairo, Egypt. While, soybean oil and starch were purchased from local market of Shebin El-Kom City, Minufiya governorate, Egypt. Ferrous sulfate and ferric citrate were obtained from El-Gomhoryia Company for Chemicals and Drugs, Elameria, Cairo, Egypt. While lysine-chelated iron and glycine-chelated iron were introduced from Germany.

Animals:

Thirty two adult male Albino rats, with an average weight $145 \pm 5g$ were used in this study. Adult male albino rats, Sprague Dawley strain, were obtained from Research Institute of Ophthalmology, Giza, Egypt. Rats were housed in stainless steel wire cage under controlled condition. Diets were offered to the rats in a special non-scattering feeding cup to avoid loss of feeding and contamination. Tap water was provided to rats by mean of glass tubes projecting through wire cages from inverted bottles supported to one side of the cage..

The Composition of Basal and Tested Diets:

The Basal diet consisted of casin (14%), Soybean oil (4%), choline bitartrate (.25%), and vitamin mixture (1%), cellulose (5%), corn starch (up to 100%) and salt mixture (3.5%), according to **AIN,(1993)**.

Methods:

Experimental Designs and Animal Groups:

All rats were fed on standard diet for one week as adaptation period. Then, rats were distributed into 8 groups 4 rats per each , in which means of rats weight for all groups were nearly equal. Four groups of rats fed on adequate Fe –diet (35mg/kg diet) as different forms such as ferrous sulfate, ferric citrate, glycine-chelated iron and lysine-

chelated iron, respectively. Other four groups fed on low Fe diet (17.5mg/kg) as previous sources for 12 weeks according to the following groups:

Group 1: Rats fed on 50% Fe- deficient as ferrous sulfate.

Group 2: Rats fed on adequate Fe diet as ferrous sulfate.

Group 3: Rats fed on 50% Fe- deficient as ferric citrate.

Group4: Rats fed on adequate Fe diet as ferric citrate.

Group5: Rats fed on 50% Fe- deficient glycine- chelated iron.

Group6: Rats fed on adequate Fe diet as glycine-chelated iron.

Group7: Rats fed on 50% Fe- deficient lysine-chelated iron.

Group8: Rats fed on adequate Fe diet as lysine-chelated iron.

Blood Sampling and Organs .

From the previously mentioned groups, blood samples were collected after 12 hours fasting at the end of the experiment (12) weeks by using the retro- orbital method, by means of a micro capillary glass, blood was collected in to a dry clean centrifuge tube, and left to clot at room temperature for half an hour. The blood was centrifuged for 10 minutes at 3000 r.p.m. to separate the serum. Serum was carefully aspirated and transferred in to clean quit fit plastic tubes and kept frozen at (-20⁰ C) for biochemical analysis as described by **Schermer, (1967)**. The organs (Liver, Kidney, heart) were removed and washed in saline solution and weighted.

Biochemical analysis:

Complete Blood Count (CBC) test, serum iron and serum ferritin:

At the end of experimental period, a 5 ml blood sample were taken to determine hemoglobin (HB), hematocrit, red blood cell (RBC), serum iron, serum ferritin, were estimated according to the method described by **Dacie and Lewis, (1998)**.

Estimation the mean corpuscular volume (MCV) as calculated by dividing the hematocrit value by RBC count **Lee and Nieman,(1996)** as the following formula:

$$MCV = \frac{Hct}{RBC} \times 10$$

Statically Analysis:

The data were statistically analyzed using a completely randomized factorial design **SAS, (1985)**. A computerized costat program was used by one way ANOVA. In which a significant mean effect was detected and the means were separated with the Dunkn,s test. The results are presented as mean±SD. Differences between treatments at (p ≤ 0.05) were considered significant.

Results and Discussion:

Hemoglobin:

Data presented in table (1) showed the effect of 50% Fe-deficient diets and adequate Fe diets as ferrous sulfate, ferric citrate, glycine-chelated iron and lysine-chelated iron for 12 weeks on hemoglobin gm/dl. As expected in hemoglobin levels in groups fed on 50% Fe-deficient diets significantly decreased when compared to groups fed on adequate Fe diets in each type of Fe. The highest levels of Hb were recorded for adequate Fe diets groups as glycine chelated iron and lysine chelated iron, being 16.2 ± 1.44 and 16 ± 1.44 g/dl which didn't significantly change either between each other or compared to adequate Fe diets group as Ferric citrate which were 15.8 ± 1.34 g/dl.

This similarity in regulation of iron transferred to the plasma from iron amino acid chelate or FeSO_4 is seen in a dose-response study in which 100 anemic adolescents received either 120 mg of iron as FeSO_4 (n=27), (or 120 mg of iron (n=26), 60 mg of iron (n=21), or 30 mg of iron (n=26) as iron amino acid chelate, daily, for 28 days. Each source and quantity of iron was equally effective ($p < 0.01$) in restoring the anemic adolescents' hemoglobin levels to normal (<11 to <13.5 g/dL), implying the greater bioavailability of the iron amino acid chelate compared to ferrous sulfate (**Pineda et al., 1994**).

The obtained results are in agreement with (**layrisse et al., 2000-a**) who showed that iron bis-glycine chelate has better iron bioavailability than does ferrous sulfate, especially when the vehicles of consumption are foods that inhibit non heme-iron absorption.

Similar results were obtained by **Ashmead, (2001)** who reported a significant increase in the intestinal absorption of iron from iron amino acid chelate as compared to inorganic iron salts. While these increased the uptakes of iron from the amino acid chelate into mucosal tissue are highly significant, also demonstrated that there is a mechanism in the mucosal tissue which controls the quantity of iron from the amino acid chelate that is transferred to the plasma. For example, the higher the hemoglobin value, the less iron transferred. When considered together these studies demonstrated that iron amino acid chelate is both a safe and effective source of iron for treatment of iron deficiencies. Iron from the bis-glycine chelate competed with ferrous sulfate for the non heme-iron absorption pathway.

Many studies showed that Fe-Gly has a high bioavailability in the bodies of rats, human beings and other animals, comparing with FeSO_4 as revealed by **Ashmead, 2001 and Allen, (2002)**.

Table (1): Effect of feeding on 50% Fe deficient diets and adequate Fe diets as ferrous sulfate, ferric citrate, glycine-chelated iron and lysine-chelated iron for 12 weeks on hemoglobin(g/dl).

Groups	HB (g/dl)		
	Mean±SD		
Group (1) 50% Fe-deficient diet as ferrous sulfate	12.3	±	1.06 ^d
Group (2) Adequate Fe diet as ferrous sulfate	15.2	±	1.54 ^b
Group (3) 50% Fe-deficient diet as ferric citrate	12.7	±	1.06 ^{cd}
Group (4) Adequate Fe diet as ferric citrate	15.8	±	1.34 ^{ab}
Group (5) 50% Fe-deficient diet as glycine-chelated iron	13.1	±	1.15 ^c
Group (6) Adequate Fe diet as glycine-chelated iron	16.2	±	1.44 ^a
Group (7) 50% Fe-deficient diet as lysine-chelated iron	13.0	±	1.15 ^{cd}
Group (8) Adequate Fe diet as lysine-chelated iron	16.0	±	1.44 ^a
LSD	0.74		

Data are presented as Mean±SD – Significant at p<0.05 using one way ANOVA test. Values which have different letters in each column differ significantly. While those with have similar or partially are not significant.

Red blood cells (RBC):

Results of table (2) showed RBC of normal rats which feeding on 50% Fe- deficient diets (17.5mg/kg) and adequate Fe-diets (35 mg/kg) as ferrous sulfate, ferric citrate, glycine-chelated iron and lysine-chelated iron for 12 weeks. The RBC values were 4.10±0.36, 4.20±0.36, 4.37±0.38 and 4.30±0.36 10¹²/L for groups feeding on 50% Fe-deficient diets as ferrous sulfate, ferric citrate, glycine-chelated iron and lysine-chelated iron, respectively. While, it were 5.07±0.58, 5.27±0.40, 5.40±0.45 and 5.30±0.46 10¹²/L for groups feeding on adequate Fe-diets as ferrous sulfate, ferric citrate, glycine-chelated iron and lysine-chelated iron, respectively. It is worthy noticed that RBC values of groups which feeding on Fe-diets as glycine type significantly increased than that ferrous sulfate type, it was being the best treatments in each of 50% Fe deficient diets groups and adequate Fe diets groups. At the same time, RBC values of lysine-chelated iron and ferric citrate groups, didn't significantly differed as compared to glycine chelated iron.

The most common disorder associated with red blood cells are anemia's. Anemia is a condition in which the body does not have enough healthy red blood cells. Red blood cells provided oxygen to body tissues (Speter Klinken, 2002).

Regular RBC transfusions are the principal supportive therapy for many rare anemias involving a decrease in RBC production, an increase in cell destruction, or chronic blood loss as showed by Shander *et al.*, (2009).

Table (2): Effect of feeding on 50% Fe- deficient diets and adequate Fe diets as ferrous sulfate, ferric citrate, glycine-chelated iron and lysine-chelated iron for 12 weeks on red blood cells.

Groups	RBC ($10^{12}/L$)		
	Mean \pm SD		
Group (1) 50% Fe-deficient diet as ferrous sulfate	4.10	\pm	0.36 ^d
Group (2) Adequate Fe diet as ferrous sulfate	5.07	\pm	0.58 ^b
Group (3) 50% Fe-deficient diet as ferric citrate	4.20	\pm	0.36 ^{cd}
Group (4) Adequate Fe diet as ferric citrate	5.27	\pm	0.40 ^{ab}
Group (5) 50% Fe-deficient diet as glycine-chelated iron	4.37	\pm	0.38 ^c
Group (6) Adequate Fe diet as glycine-chelated iron	5.40	\pm	0.45 ^a
Group (7) 50% Fe-deficient diet as lysine-chelated iron	4.30	\pm	0.36 ^{cd}
Group (8) Adequate Fe diet as lysine-chelated iron	5.30	\pm	0.46 ^{ab}
LSD	0.23		

Data are presented as Mean \pm SD – Significant at $p < 0.05$ using one way ANOVA test. Values which have different letters in each column differ significantly. While those with have similar or partially are not significant

Hematocrit (HCT):

Results of table (3) showed HCT of normal rats which feeding on 50% Fe- deficient diets (17.5 mg/kg) and adequate Fe-diets (35 mg/kg) as ferrous sulfate, ferric citrate, glycine-chelated iron and lysine-chelated iron for 12 weeks. It could be showed that the mean values of HCT were 36.9 ± 4.28 , 38.1 ± 2.69 , 39.3 ± 2.60 and $39.0 \pm 2.62\%$ for 50% Fe- deficient diets groups as ferrous sulfate, ferric citrate, glycine and lysine chelated iron, there were no significant differences between them. On the other hand, there were being 45.6 ± 5.84 , 47.4 ± 4.78 , 48.6 ± 2.60 and $48 \pm 5.22\%$ for adequate Fe diets groups as ferrous sulfate, ferric citrate, glycine-chelated iron and lysine-chelated iron, respectively, and the result didn't revealed any significant differences ($p \leq 0.05$) between these groups. From the same table, it could be noticed that groups (2, 4, 6 and 8) in which rats fed on adequate Fe-diets as ferrous sulfate, ferric citrate, glycine-chelated iron and lysine-chelated iron showed a significant increased when compared with groups (1, 3, 5 and 7) in which rats fed on 50% Fe-deficient diets as ferrous sulfate, ferric citrate, glycine-chelated iron and lysine-chelated iron, respectively. The best treatments was recorded for rats fed on glycine chelated type.

Table (3): Effect of feeding on 50% Fe deficient diets and adequate-Fe diets as ferrous sulfate, ferric citrate, glycine-chelated iron and lysine-chelated iron for 12 weeks on HCT%.

Groups	HCT%		
	Mean±SD		
Group (1) 50% Fe-deficient diet as ferrous sulfate	36.9	±	4.28 ^b
Group (2) Adequate Fe diet as ferrous sulfate	45.6	±	5.84 ^a
Group (3) 50% Fe-deficient diet as ferric citrate	38.1	±	2.69 ^b
Group (4) Adequate Fe diet as ferric citrate	47.4	±	4.78 ^a
Group (5) 50% Fe-deficient diet as glycine-chelated iron	39.3	±	2.60 ^b
Group (6) Adequate Fe diet as glycine-chelated iron	48.6	±	2.60 ^a
Group (7) 50% Fe-deficient diet as lysine-chelated iron	39.0	±	2.62 ^b
Group (8) Adequate Fe diet as lysine-chelated iron	48.0	±	5.22 ^a
LSD	3.59		

Data are presented as Mean±SD – Significant at p<0.05 using one way ANOVA test. Values which have different letters in each column differ significantly. While those with have similar or partially are not significant.

Mean cell volume (MCV):

Results presented in table (4) showed mean corpuscular volume, or mean cell volume (MCV) expressed as (Mean±SD) of different rats which fed on 50% Fe-deficient dietS and adequate Fe diets as ferrous sulfate, ferric citrate, glycine-chelated iron and lysine-chelated iron for 12 weeks.

Data revealed that (Means±SD) values of MCV were 90.00±4.94, 90.7±8.12, 89.9±7.47 and 90.00±7.41fl for 50% Fe-deficient diets groups as ferrous sulfate, ferric citrate, glycine-chelated iron and lysine-chelated iron, respectively. While it were 89.9±7.79, 89.9±8.02, 90.00±8.14 and 90.06±8.09 fl for adequate Fe diets groups as ferrous sulfate, ferric citrate, glycine-chelated iron and lysine-chelated iron, respectively. The results revealed that there were non-significant differences among all tested groups.

Table (4): Effect of feeding on 50% Fe deficient diets and adequate-Fe diets as ferrous sulfate, ferric citrate, glycine-chelated iron and lysine-chelated iron for 12 weeks on mean corpuscular volume, or mean cell volume (mcv).

Groups	MCV(fl)		
	Mean±SD		
Group (1) 50% Fe-deficient diet as ferrous sulfate	90.0	±	4.94 ^a
Group (2) Adequate Fe diet as ferrous sulfate	89.9	±	7.79 ^a
Group (3) 50% Fe-deficient diet as ferric citrate	90.7	±	8.12 ^a
Group (4) Adequate Fe diet as ferric citrate	89.9	±	8.02 ^a
Group (5) 50% Fe-deficient diet as glycine-chelated iron	89.9	±	7.47 ^a
Group (6) Adequate Fe diet as glycine-chelated iron	90.0	±	8.14 ^a
Group (7) 50% Fe-deficient diet as lysine-chelated iron	90.0	±	7.411 ^a
Group (8) Adequate Fe diet as lysine-chelated iron	90.06	±	8.09 ^a
LSD	7.91		

Data are presented as Mean±SD – Significant at p<0.05 using one way ANOVA test. Values which have similar or partially are not significant.

Red blood cell distribution width (RDW):

Results presented in table (5) showed red blood cell distribution width(RDW%) as (Mean±SD) of different rats which fed on 50% Fe-deficient diet and adequate Fe diet as ferrous sulfate, ferric citrate, glycine-chelated iron and lysine-chelated iron for 12 weeks.

In each type of Fe, there were significant decreases in RDW for groups of rats fed on adequate Fe diets compared to groups of rats fed on 50% Fe deficient diets generally. This results due to Fe deficient regardless of Fe type. Also, it could be noticed that RDW% of group (6) in which rats fed on adequate Fe diets as glycine-chelated iron showed the lowest level being 14.0±1.10% and significantly decreased when compared to groups (1, 3, 5 and 7) in which rats given 50% Fe- deficient diets as ferrous sulfate, ferric citrate, glycine-chelated iron and lysine-chelated iron, being 16.0±1.70, 15.9±1.30, 15.3±1.34 and 15.5±1.41%, respectively. At the same time RDW% of this group didn't showed any significant differences when compared to other groups given adequate Fe- diets. Meanwhile, RDW for rats given 50% Fe- deficient diet as glycine-chelated iron (group5) being 15.3±1.34% and didn't showed a significant changes when compared to groups (2, 4 and 8) in which rats fed on adequate Fe diet as ferrous sulfate, ferric citrate and lysine-chelated iron which it were being 14.5±1.50, 14.3±1.22 and 14.3±1.28%, respectively. These results showed that glycine chelated iron type was the perfect.

These data are in agreement with **Henery, (1991)** who showed that there was a system of classification of anemias based on the RDW and the MCV. This scheme has been used to help discriminate between uncomplicated heterozygous thalassemia (With a normal RDW and Low MCV), and iron deficiency (high RDW and normal to Low MCV). For all the red cell measurements performed by multichannel instrument, the RDW appears to be the first to become abnormal in iron deficiency anemia due to a sensitive but non-specific indicator of iron deficiency in chronic hemodialysis patients. The RDW, Therefore, can be a helpful adjunct in the classification and management of disorders involving the erythrocyte.

Iron deficiency is usually diagnosed with laboratory tests. Low serum hemoglobin in the setting of a reduced MCV is usually the initial finding on a routine complete blood count. RDW has been proposed as a sensitive indicator for IDA. Increased RDW represents heterogeneity in the red blood cell volume distribution, equivalent to anisocytosis observed in a peripheral blood smear. A significant increase in mean RDW can be used to diagnose IDA (sensitivity 81.0%, specificity 53.4%) (**Aulakh et al., 2009**).

Table (5): Effect of feeding on 50% Fe deficient diets and adequate-Fe diets as ferrous sulfate, ferric citrate, glycine-chelated iron and lysine-chelated iron for 12 weeks on red blood cell distribution width (RDW%).

Groups	RDW%		
	Mean	±	SD
Group (1) 50% Fe-deficient diet as ferrous sulfate	16.0	±	1.70 ^a
Group (2) Adequate Fe diet as ferrous sulfate	14.5	±	1.50 ^{bcd}
Group (3) 50% Fe-deficient diet as ferric citrate	15.9	±	1.30 ^a
Group (4) Adequate Fe diet as ferric citrate	14.3	±	1.22 ^{cd}
Group (5) 50% Fe-deficient diet as glycine-chelated iron	15.3	±	1.34 ^{abc}
Group (6) Adequate Fe diet as glycine-chelated iron	14.0	±	1.1 ^d
Group (7) 50% Fe-deficient diet as lysine-chelated iron	15.5	±	1.41 ^{ab}
Group (8) Adequate Fe diet as lysine-chelated iron	14.3	±	1.28 ^{cd}
LSD	1.04		

Data are presented as Mean±SD – Significant at p<0.05 using one way ANOVA test. Values which have different letters in each column differ significantly. While those with have similar or partially are not significant.

Serum ferritin level (SF):

Data given in table (6) contained the effect of feeding on 50% Fe deficient diets and adequate Fe diets as ferrous sulfate, ferric citrate, glycine-chelated iron and lycine-chelated iron for 12 weeks on serum ferritin in normal rats. By focusing on serum ferritin results in table(6), it could be noticed that, there were no significant differences between rats fed on 50% Fe deficient diets. Also, there were no significant differences between rats given adequate Fe diets as ferrous sulfate, ferric citrate, glycine -chelated iron and lycine-chelated iron. moreover the results showed that the mean \pm SD levels of serum ferritin for groups of (2, 4, 6 and 8) in which rats fed on adequate Fe diets as ferrous sulfate, ferric citrate, glycine-chelated iron and lycine-chelated iron which were being 146.2 \pm 12.11, 142 \pm 12.28, 147 \pm 2.08 and 143 \pm 2.007 ng/ml, significantly increased compared to groups (1, 3, 5 and 7) in which rats fed on 50% Fe-deficient diets as ferrous sulfate, ferric citrate, glycine-chelated iron and lycine-chelated iron, being 130.7 \pm 11.92, 132.5 \pm 11.02, 135 \pm 11.78 and 133 \pm 11.6 ng/ml, respectively. The best value was recorded for group (6) inwhich rats fed on adequate Fe diets as glycine chelated iron, which it was 147 \pm 12.28 ng/ml.

In similar study, **Righetti *et al.*, (2012)** showed that a low iron dose of 30 mg/day for 90 days with either ferrous sulfate or iron bis-glycinate chelate significantly increased serum ferritin concentration in schoolchildren with low iron stores but no anemia and had negligible side effects. The effect of increasing iron status was sustained up to 6 months after supplementation, rendering both treatments as safe and effective. These results support the preventive effectiveness of this low-dose iron intervention to increase serum ferritin, which will help prevent iron deficiency anemia and may be relevant where iron deficiency is a public health concern and where a preventive daily low dosage of iron supplementation can be used to help the school-age population maintain an adequate nutritional iron status.

Also, **Elghetany and Banki, (2011)** and **Brittenham, (2012)** revealed that the lower in the ferritin level, even within the "normal" range, is more likely in the patient does not have enough iron.

Table (6): Effect of feeding on 50% Fe deficient diets and adequate-Fe diets as ferrous sulfate, ferric citrate, glycine-chelated iron and lysine-chelated iron for 12 weeks on serum ferritin level.

Groups	Serum ferritin (ng/ml) Mean±SD		
	Group (1) 50% Fe-deficient diet as ferrous sulfate	130.7	±
Group (2) Adequate Fe diet as ferrous sulfate	146.2	±	12.11 ^a
Group (3) 50% Fe-deficient diet as ferric citrate	132.5	±	11.02 ^b
Group (4) Adequate Fe diet as ferric citrate	142.0	±	12.28 ^a
Group (5) 50% Fe-deficient diet as glycine-chelated iron	135.0	±	11.78 ^b
Group (6) Adequate Fe diet as glycine-chelated iron	147.0	±	12.28 ^a
Group (7) 50% Fe-deficient diet as lysine-chelated iron	133.0	±	11.26 ^b
Group (8) Adequate Fe diet as lysine-chelated iron	143.0	±	11.26 ^a
LSD	4.73		

Data are presented as Mean±SD – Significant at $p < 0.05$ using one way ANOVA test. Values which have different letters in each column differ significantly. While those with have similar or partially are not significant

Serum iron(SI):

Results presented in table (7) Showed Serum iron as (Mean±SD) of different rats fed on 50% Fe- deficient diet and adequate Fe diets as ferrous sulfate, ferric citrate, glycine-chelated iron and lysine-chelated iron for 12 weeks.

Results in table (7) showed that there were significant increases in serum iron for groups of rats fed on adequate Fe- diets as ferrous sulfate, ferric citrate, glycine -chelated iron and lycine-chelated iron when compared with groups of rats fed on 50% Fe- deficient diets as ferrous sulfate, ferric citrate, glycine -chelated iron and lycine-chelated, expect of group (2) in which rats fed on adequate Fe diets as ferrous sulfate was nearly closed with all group given 50% Fe deficient diets. At the same time didn't significantly different from all groups given adequate Fe diets.

The values of serum iron were 1.47 ± 0.21 , 1.65 ± 0.20 , 1.61 ± 0.21 and 1.50 ± 0.15 ug/dl for groups fed on adequate Fe- diet as ferrous sulfate, ferric citrate, glycine -chelated iron, lycine-chelated iron. In order 1.20 ± 0.12 , 1.25 ± 0.11 , 1.30 ± 0.12 and 1.20 ± 0.12 ug/dl for groups fed on 50% Fe- deficient diet as ferrous sulfate, ferric citrate, glycine -chelated iron and lycine-chelated, respectively. The best levels recorded for groups (4 and 6), rats fed on adequate Fe diets as ferric citrate and glycine -chelated iron, being 1.65 ± 0.20 and 1.61 ± 0.21 ug/dl, respectively.

Table (7): Effect of feeding on 50% Fe deficient diets and adequate-Fe diets as ferrous sulfate, ferric citrate, glycine-chelated iron and lysine-chelated iron for 12 weeks on serum iron level.

Groups	Serum iron (ug/dl)		
	Mean±SD		
Group (1) 50% Fe-deficient diet as ferrous sulfate	1.20	±	0.12 ^b
Group (2) Adequate Fe diet as ferrous sulfate	1.47	±	0.21 ^{ab}
Group (3) 50% Fe-deficient diet as ferric citrate	1.25	±	0.11 ^b
Group (4) Adequate Fe diet as ferric citrate	1.65	±	0.20 ^a
Group (5) 50% Fe-deficient diet as glycine-chelated iron	1.30	±	0.12 ^b
Group (6) Adequate Fe diet as glycine-chelated iron	1.61	±	0.21 ^a
Group (7) 50% Fe-deficient diet as lysine-chelated iron	1.20	±	0.12 ^b
Group (8) Adequate Fe diet as lysine-chelated iron	1.50	±	0.15 ^a
LSD	0.19		

Data are presented as Mean±SD – Significant at p<0.05 using one way ANOVA test. Values which have different letters in each column differ significantly. While those with have similar or partially are not significant

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دراسة مقارنة لصور الحديد المخلي والغير مخلي على بعض القياسات الحيوية للفئران البالغة

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الملخص:

تهدف تلك الدراسة إلى مقارنة تأثير عنصر الحديد بأشكال مختلفة عندما يقدم في صور تقليدية مع الحديد المخلي والمحمل على أحماض أمينية مختلفة (جليسين، ليسين) ودراسة أثر ذلك على مستوى بعض القياسات البيولوجية المرتبطة بصورة وثيقة مع مستوى الحديد في الدم. أجريت الدراسة على 32 فأر من ذكور الألبينو تتراوح أوزانهم بين 145-150 جم، تم تغذيتهم على الوجبة الأساسية لمدة أسبوع ثم قسمت بعد ذلك إلى ثماني مجموعات تبعاً للصورة التي يقدم فيها عنصر الحديد كمدعم غذائي من خلال الوجبة الغذائية أربعة مجاميع تم تغذيتهم على وجبة منخفضة من الحديد (17.5 ملليجرام/كيلوجرام) في صورة "كبريتات الحديدوز، سترات الحديد، حديد مخلي محمل على الجليسين، حديد مخلي محمل على الليسين" على الترتيب وباقي المجاميع تم تغذيتهم وجبة كافية من الحديد (35 ملليجرام/كيلوجرام) في صورة "كبريتات الحديدوز، سترات الحديد، حديد مخلي محمل على الجليسين، حديد مخلي محمل على الليسين" على الترتيب. وفي نهاية التجربة (12 أسبوع) تم تصويم الفئران طوال الليل قبل الذبح، ثم جمع عينات الدم من كل فأر على حده وتم طرد الدم مركزاً لتقدير التحاليل البيولوجية وقد أوضحت النتائج أن الفئران التي تغذيت على وجبة قياسية بها حديد المخلي المحمل على أحماض أمينية مختلفة أظهرت ارتفاعاً معنوياً ($p \leq 0.05$) في مستوى الهيموجلوبين بالدم مقارنة بالفئران التي تغذت على وجبة قياسية تحتوي على الحديد في صورة كبريتات الحديدوز. كما سجلت أفضل النتائج لمستوى كرات الدم الحمراء والهيماتوكريت وسيرم الحديد وسيرم الفيرتين للفئران التي تغذت على وجبة قياسية بها حديد المخلي وخاصة المحمل على الجليسين كما أظهرت النتائج انخفاضاً في قيمة عرض توزيع خلايا الدم الحمراء للفئران التي تغذيت على وجبة كافية من الحديد في حديد المخلي المحمل على الجليسين وسجلت أدنى المستويات مقارنة بالفئران التي تغذت على وجبة منخفضة من الحديد في صورة كبريتات الحديدوز وسترات الحديد وحديد مخلي المحمل على الجليسين وحديد مخلي محمل على الليسين وفي نفس الوقت كان حجم الكرية الوسطى في حدود المستوى الطبيعي لذلك لخصت النتائج أن الحديد المخلي المحمل على أحماض أمينية له أثر إيجابي على مؤشرات الدم ونسبة الحديد في الفئران البالغة.

الكلمات الكشافة: حديد مخلي، كبريتات حديدوز، سترات حديد، المؤشرات الدموية