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Impact of Iron Deficiency Anemia Treatment on Type 2 Diabetic Complications

Amira S Ahmed[1,2*], Rehab M Elgharabawy[1,3], Amal H Al-Najjar[4], Monerh H Al-Abdullatif[5], Mona A Al-Abdullatif[5] and Turki A Al-Mogbel[6]

[1]Department of Pharmacology and Toxicology, Faculty of Pharmacy, Qassim University, Saudi Arabia

[2]Hormones Department, National Research Centre, Egypt

[3]Pharmacology and Toxicology Department, Faculty of Pharmacy, Tanta University, Egypt

[4]Drug and Poison Information Specialist, Pharmacy Services, Security Forces Hospital, Riyadh, Saudi Arabia

[5]Faculty of Pharmacy, Qassim University, Saudi Arabia

[6]Buraydah Diabetes Center, King Fahad Specialist Hospital, Saudi Arabia

[*]**Corresponding author:** Amira S Ahmed, Department of Pharmacology and Toxicology, Faculty of Pharmacy, Qassim University, Saudi Arabia, Tel: +966163800050; E-mail: dr.amira2007@yahoo.com

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Abstract

Objectives: To evaluate the relationship between the iron deficiency anemia (IDA) and type 2 diabetes, to estimate the effect of the IDA on the level of glucose and glycated hemoglobin (Hb1Ac), and to assess the ameliorating effect of IDA treatment on progression of diabetes and its complications.

Subjects and methods: This study included 125 male Saudi adult participants divided into five groups; control subjects (group I), patients with type 2 diabetes (group II), patients with IDA (group III), patients with type 2 diabetes and untreated IDA (group IV), and patients with type 2 diabetes and treated IDA with iron supplementation (group V). Fasting blood glucose (FBG), HbA1c, CBC, ferritin, iron, and total iron binding capacity (TIBC) were assayed.

Results: The HbA1c and FBG levels were significantly higher in groups III and IV compared to group I. The results revealed a significant decline in HbA1c and FBG levels in group V compared to group IV. Negative significant correlations were observed between iron and ferritin with HbA1c and FBG. The incidence of diabetic complications was significantly associated with IDA (X2: 81.48, $p<0.001$). Ferritin was the most reliable predictor of type 2 diabetes in patients with IDA. The best cut off value for ferritin was 31.56 ng/ml.

Conclusion: Low iron level has a crucial effect on glycemic status by increasing the level of FBG and HbA1c, IDA is strongly correlated with type 2 diabetes, and the iron supplementation for diabetic patients with IDA ameliorates the progression of diabetes and its complications.

Keywords: Type 2 diabetes; Iron deficiency; Anemia; Diabetic complications; Glycated hemoglobin; Ferritin; Iron; Total iron binding capacity

Introduction

Diabetes is a common worldwide health problem and the leading cause of high percentage of mortality and morbidity because it affects more than one system in the body. The tremendous growth of the incidence of type 2 diabetes is very high in both adults and young [1]. Diabetes can elevate lipids levels (especially triglyceride and cholesterol), increasing the risk of heart disease. Thus, diabetes can be classified as the most common diseases which contributing in premature death [2,3]. There are multiple causes for type 2 diabetes including; tissues insulin resistance, impaired insulin secretion, deficiency or resistance to incretin hormones and excess glucagon secretion [4]. Generally, diabetes is related with serious long-term complications including the large and small vascular complications, and fetal acute complications like acute diabetic ketoacidosis [5]. Patient education, self and social supports as well as optimal glycemic control can lower the risk of diabetes complications and prevent acute conditions [6].

Glycated hemoglobin (HbA1c) is widely used as an important indicator of chronic glycemic control. However, the level of HbA1c is not influenced by glucose levels in the blood alone, there are multiple conditions that also can increase the HbA1c level regardless of glycemic status like, iron deficiency and hemolytic anemia, alcohol consumption, chronic blood loss, gestation and uremia [7,8].

One of the global major health issues is iron deficiency anemia (IDA). Recent studies have shown that more than two billion of world population have anemia, most of these cases are IDA [9]. There are many pathological and habitual causes of IDA such as worm infection and unhealthy low red meats containing diet, respectively [10].

Up to 30% of diabetic patients present with coexisting anemia [11]. The exact relationship between IDA and its effect on HbA1c needs further explanations. The purpose of the current study is to evaluate the relationship between IDA and type 2 diabetes, to

estimate the effect of the IDA on the level of glucose and the Hb1Ac level, and to assess the ameliorating effect of IDA treatment on progression of diabetes and its complications.

Subjects and Methods

Subjects

This study is a prospective cross-sectional study. Patients were recruited from Buraydah Diabetes Center of King Fahad Specialist Hospital, Qassim and Security Forces Hospital, Riyadh, Saudi Arabia. The study included 125 male Saudi adult participants divided into five groups of 25 subjects each: control subjects (group I), patients with type 2 diabetes (group II), patients with IDA (group III), patients with type 2 diabetes and untreated IDA (group IV), and patients with type 2 diabetes and IDA treated with iron supplementation (group V).

Demographic Characteristics (age and gender), diabetic duration and complications were monitored. Patients with hemolytic anemia, haemoglobinopathies, renal disease, hepatic disease, chronic alcohol consumption, history of acute blood loss, and malabsorption syndrome were excluded. Informed consent was collected from all participants after full explanation of the study. The study was approved by the Qassim Region Research Ethics committee (QREC) and ethical committee of Security Forces Hospital, Riyadh, KSA.

Sample collection

For all subjects, 10 ml blood samples were obtained in the morning after an overnight fast (for a minimum of 8 h) from the antecubital vein. The blood samples were divided into three parts; the first part was collected onto EDTA containing tubes for complete blood count (CBC) and HbA1c assay, the second part was collected onto NaF containing tubes for plasma fasting glucose assay, and the last part was collected in tube without anticoagulant for serum separation for determination of iron, ferritin and total iron binding capacity (TIBC) levels.

Biochemical analyses

The CBC was obtained by using Sysmex hematology analyzer (Japan). The whole blood HbA1c, by using turbidimetric inhibition immunoassay method, and serum iron were assessed using kits provided by the Siemens Healthcare (Germany) according to manufacture instructions. Fasting blood glucose (FBG) was assessed by enzymatic colorimetric method using a kit provided by the Siemens Healthcare (Germany) according to Kunst et al. [12] method. Serum ferritin was determined using a kit provided by Roche Diagnostics Elecsys and Cobas (Schweiz) according to Blackmore et al. [13] method. Serum TIBC was determined by using a kit provided by Siemens Healthcare (Germany) according to Yamanishi et al. [14] method.

Statistical analyses

Statistical Package for the Social Sciences (SPSS; V. 23.0; IBM Corp., USA) was used to carry out all statistical analyses. The mean ± SD was used to express the results. ANOVA was used to carry out the comparisons between different groups followed by post-hoc Bonferroni test. Pearson chi square cross tabulation was used to test the differences in proportions of categorical variable and assess correlation between variables. Pearson correlation was used to estimate the association between different parameters. Multiple linear stepwise regression analyses were used to adjust the effect of other covariates. Receiver operating characteristic (ROC) curves were plotted in which the value for sensitivity was plotted against 1-specificity. The overall accuracy of a biochemical marker to predict type 2 diabetes in patients with iron deficiency anemia was designed as the average of the sensitivity and spasticity. The level of significance was set at $p \leq 0.05$.

Results

The results showed that age and diabetic duration were not significant factors influencing change across the studied groups. In addition, the percentage of diabetic complications in group V (33.3%) was significantly lower than that in diabetic patients with untreated IDA (60%), (X2:81.48, P<0.001) as presented in **Table 1**.

The results revealed a significant difference across all the six independent variables of CBC test amongst five groups (**Table 2**). There was a significant decrease in hemoglobin (Hgb), hematocrit (HCT), mean corpuscular volume (MCV) and mean corpuscular hemoglobin concentration (MCHC) levels in patients with IDA, and patients with type 2 diabetes and IDA without treatment compared to groups (I and II). However, there was no significant difference between control and group (V) that received treatment in Hgb, MCV and mean corpuscular hemoglobin (MCH) levels. For hematocrit, group (V) showed a significant difference with the control, diabetic and patients with type 2 diabetes and IDA without treatment groups. Results for MCV in group (V) were significantly increased compared to both III and IV groups. For MCHC, there was a significant difference in group (V) compared to all other groups. On the other hand, red blood cell distribution width (RDW) was significantly increased in groups (III and IV) compared to both I and II groups. In group (V), diabetics who received treatment for IDA showed a significant decrease in RDW compared to groups (III and IV).

Table 1 Demographic and clinical characteristics of participants.

Groups/Parameters	Group (I)	Group (II)	Group (III)	Group (IV)	Group (V)	P-Value
N	25	25	25	25	25	
Age (Years) (Mean ± S.D)	36.8 ± 4.51	39.8 ± 3.76	39.9 ± 2.42	40.3 ± 2.05	37.8 ± 5.03	0.052

Diabetic duration (Years) (Mean ± S.D)	N.A.	4.2 ± 0.86	N.A.	4.0 ± 0.93	4.4 ± 0.74	0.439
Diabetic complications N (%)	N.A.	4 (26.7%)	N.A.	9 (60%)	5 (33.3%)	<0.001

Results are expressed as mean ± SD and number (%).

N: Number of Participants

N.A.: Not applicable. P value ≤0.05 was considered significant

The results are expressed as mean ± SD.

Hgb: Hemoglobin, HCT: Hematocrit, MCV: Mean corpuscular volume, MCH: Mean corpuscular hemoglobin, MCHC: Mean Corpuscular Hemoglobin Concentration, RDW: Red Blood Cell Distribution Width.

a) Significant difference from control group (I).

b) Significant difference from type 2 diabetic group (II).

c) Significant difference from patients with iron deficiency anemia group (III).

d) Significant difference from patients with type 2 diabetes and untreated iron deficiency anemia group (IV). P value ≤ 0.05 was considered significant.

Table 2 Complete blood count of participants.

Groups/ Parameters	Group (I)	Group (II)	Group (III)	Group (IV)	Group (V)
Hgb (%)	13.3 ± 0.87	15.1 ± 1.52a	10.8 ± 1.33a,b	10.9 ± 1.53a,b	12.1 ± 1.79b
HCT (%)	40.1 ± 2.58	44.3 ± 4.11a	34.6 ± 2.41a,b	31.1 ± 4.88a,b	36.0 ± 3.50a,b,d
MCV (ft)	85.1 ± 3.55	84.7 ± 3.28	71.7 ± 8.85a,b	73.6 ± 6.49a,b	80.3 ± 6.49c,d
MCH (pg)	27.8 ± 1.55	28.5 ± 1.85	21.8 ± 2.89b	23.6 ± 3.10a,b	25.6 ± 4.37c
MCHC (%)	33.2 ± 0.73	33.9 ± 0.90	29.8 ± 0.30 a,b	28.4 ± 1.09a,b,c	31.7 ± 1.91a,b,c,d
RDW (%)	13.2 ± 0.91	12.8 ± 0.84	17.9 ± 3.27 a,b	18.2 ± 2.19a,b	14.6 ± 1.16c,d

The FBG and HbA1c levels were significantly increased in all studied groups compared to control group. In group (V), there was a significant decline in FBG and HbA1c levels compared to groups (II, III, and IV). The IDA marker (iron and ferritin) showed a significant decline in groups (III and IV) compared to control group. However, these markers were significantly increased in group (V) compared to group (III and IV) for iron and control group for ferritin. On the other hand, TIBC was significantly elevated in IDA group and group (IV) compared to control and diabetic groups. The TIBC was significantly decreased in group (V) compared to groups (II, III and IV) (**Table 3**).

Table 3 Diabetic and iron deficiency anemia biomarkers of participants.

Groups/Parameters	Group (I)	Group (II)	Group (III)	Group (IV)	Group (V)
HbA1c (%)	5.6 ± 0.25	8.8 ± 0.74^{a}	8.1 ± 1.35^{a}	10.1 ± 0.79^{a,b,c}	6.6 ± 1.17^{a,b,c,d}
FBG (mmol/L)	4.8 ± 0.44	12.0 ± 2.01^{a}	9.1 ± 2.21^{a,b}	12.2 ± 0.48^{a,c}	6.4 ± 1.04^{a,b,c,d}
Iron (µmol/L)	16.1 ± 1.64	15.8 ± 1.16	4.7 ± 0.83^{a,b}	3.6 ± 0.67^{a,b}	15.5 ± 2.51 ^{c,d}
Ferritin (ng/ml)	68.8 ± 14.19	14.2 ± 1.16^{a}	7.9 ± 0.74^{a}	10.6 ± 1.21^{a}	14.6 ± 3.13^{a}
TIBC (µmol/L)	55.1 ± 9.95	45.4 ± 4.76^{a}	73.5 ± 5.15^{a,b}	73.4 ± 1.73^{a,b}	55.6 ± 3.83 ^{b,c,d}

The results are expressed as the mean ± SD.

HbA1c: Glycated hemoglobin, FBG: Fasting blood glucose, TIBC: Total iron binding capacity.

a: significant difference from control group (I).

b: significant difference from type 2 diabetic group (II).

c: significant difference from patients with iron deficiency anemia group (III).

d: significant difference from patients with type 2 diabetes and untreated iron deficiency anemia group (IV). P value ≤ 0.05 was considered significant.

The results revealed positive significant correlations of FBG with HbA1c, and of iron with ferritin. However, iron and ferritin were negatively significantly correlated with TIBC. There were negative significant correlations between diabetic biomarkers (HbA1c and FBG) and IDA biomarkers (iron and ferritin). Only HbA1c was positively significantly correlated with TIBC (**Table 4**).

Data in **Table 5** shows the multiple linear stepwise regression analyses using HbA1c, FBG, iron, ferritin and TIBG as dependent variables and age, CBC, diabetic and IDA biomarkers as independent variables. Only RDW (β=-0.28, P<0.05), FBG (β=0.63, P<0.001) and iron (β=-0.34, P<0.05) remained associated with HbA1c. Age (β=0.15, P<0.05), MCH (β=0.25, P<0.05), RDW (β=0.26, P<0.05), HbA1c (β=0.54, P<0.001), ferritin (β=-0.18, P<0.05) and TIBC (β=- 0.26, P<0.05) remained associated with FBG. Additionally, HbA1c (β=-0.19, P<0.05) and TIBC (β=-0.46, P<0.001) remained associated with iron. Only FBG (β=-0.37, P<0.05) remained associated with ferritin. Finally, MCH (β=0.25, P<0.05), MCHC (β=-0.44, P<0.001), FBG (β=-0.25, P<0.05) and iron (β=-0.66, P<0.001) remained associated with TIBC.

As illustrated in **Figure 1**, ROC analyses of the biochemical markers (iron, ferritin, and TIBC); the area under the curve (AUC) were 0.816, 1, and 0.354, respectively, and thus ferritin is the most reliable predictor of type 2 diabetes in patients with IDA. The best cutoff value for ferritin was 31.56 ng/ml.

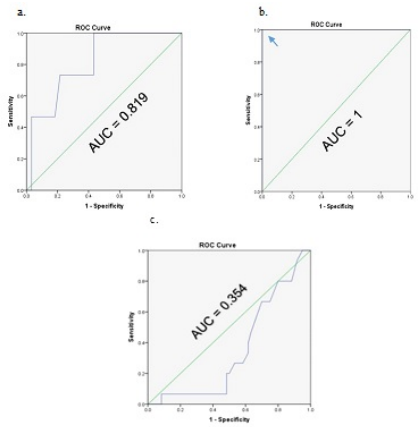

Figure 1 Receiver operating characteristic (ROC) curve for (a) iron, (b) ferritin, (c) TIBC. AUC: area under the curve. The arrow refers to a best cut off point.

Table 4 Correlations between diabetic and iron deficiency anemia biomarkers in groups (II, III, IV and V).

	HbA1c (%)	FBG (mmol/L)	Iron (µmol/L)	Ferritin (ng/ml)	TIBC (µmol/L)
HbA1c (%)	1	0.777**	-0.583**	-0.600**	0.315**
FBG (mmol/L)	0.777**	1	-0.444**	-0.604**	0.137
Iron (µmol/L)	-0.583**	-0.444**	1	0.483**	-0.798**
Ferritin (ng/ml)	-0.600**	-0.604**	0.483**	1	-0.282**

Results are expressed as correlation coefficients (r).

HbA1c: Glycated hemoglobin; FBG: Fasting blood glucose; TIBC: Total iron binding capacity.

*P-Value ≤0.05

**P-Value ≤0.01

Table 5 Multiple linear regression between different investigated parameters.

	Age (years)	Hgb (%)	HCT (%)	MCV (ft)	MCH (pg)	MCHC (%)	RDW (%)	Hb1Ac (%)	FBG (mmol/L)	Iron (µmol/L)	Ferritin (ng/ml)	TIBC (µmol/L)
Hb1Ac (%)	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	-0.28*	-----	0.63***	-0.34*	N.S.	N.S.
FBG (mmol/L)	0.15*	N.S.	N.S.	N.S.	0.25*	N.S.	0.26*	0.54***	-----	N.S.	-0.18*	-0.26*
Iron (µmol/L)	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	-0.19*	N.S.	-----	N.S.	-0.46***
Ferritin (ng/ml)	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	-0.37*	N.S.	-----	N.S.
TIBC (µmol/L)	N.S.	N.S.	N.S.	N.S.	0.25*	-0.44***	N.S.	N.S.	-0.25*	-0.66***	N.S.	-----

Results are expressed as standardized coefficients (β).

*p value ≤ 0.05.

***p value ≤ 0.001.

N.S.: Non significant correlation.

Discussion

Four hundred and fifteen million have diabetes and half of them are not yet confirmed. Ninety percent of them are exposed to bad outcomes, both those related to the large vessels or small vessels, which in turn reinforced the mental and impairment of function leading to worse healthcare budget. Regardless the social awareness about diabetic complications, and other disorders related to its occurrence, the percent of patients who suffer from diabetes increase year after year [15]. The diabetes is diagnosed using FBG that achieved by fasting 8-12 hrs and HbA1c. These are the important tests to prove the presence of diabetes. The HbA1c based on the following factors: 1. The age of RBCs 2. When HbA1c is formed in the RBCs 3. when they are released from the bone marrow [16].

Recently, clinicians prefer using HbA1c because it is characterized by a small difference between individuals and performed without fasting. The HbA1c is usually used to determine the extent to which long-term glucose levels are controlled in diabetics [17]. The level of HbA1c not only reflects the occurrence of diabetes, but also it relates to the existence of many other diseases such as hemorrhage and lack of the iron [18]. Anemia is the illness of these days and linked to the decline of iron. Women are more susceptible to IDA than male. The causes of IDA are the lack of iron in food, shortage of retention and others [19].

The present study compared the laboratory analysis of diabetics who had a shortage of iron and did not take any treatment with those who received iron supplements, most of the patients in the treated group were with lower HbA1c, ameliorating diabetic complications. This is confirmed by increasing HbA1c level in patients with IDA compared to control group and significant negative correlation between HbA1c and iron. The current results are consistent with previous studies which stated that patients with mild iron deficiency have a lower HbA1c level than those with severe deficiency [20,21].

In Turkey, Coban et al. [22] conducted a study which included 50 contributors and compared HbA1c level in both those who suffered from IDA and non-anemic. The level of HbA1c in the case of IDA was 7.4% and 5.2% in those healthier, and the level decreased when the IDA treated from 7.4% to 6.2%. The relationship between the HbA1c and the lifespan of RBCs is a positive one. The level of HbA1c increased when the lifespan of RBCs increased like in IDA and in some diseases related to Hgb. In IDA, the reduction in the iron led to raise the level of Hgb glycation which cause elevation of HbA1c [23,24]. On the other hand, when HbA1c increases, it prevents the cell turnover and production, contributing to IDA occurrence [25]. Additionally, in a study published in 2016, the results showed that the level of HbA1c increased significantly with the presence of iron deficiency, and as the level of Hgb falls as the HbA1c level becomes high.

The results of our study come in agreement with a previous one that published during 2016, which included 122 patients, where the level of HbA1c was varied between those with IDA and those non-anemic, it was high in those who suffered from iron deficiency and this increase was dependent on anemia degree. Also, the relationship of the increase of HbA1c with the level of anemia was studied and the results were consistent with our study [26].

The present work reveled the significant increase in FBG level in patients with IDA compared to control group and that level was significantly decreased in group (V) compared to group (IV). This result was confirmed by significant negative correlation between FBG and iron. Decreasing in iron increases lipogenesis and causes hyperglycemia [27]. Reduction in iron and heme synthesis lead to disruption in glucose metabolism and insulin functions. It also affects structural muscles, fatty tissues, and other tissues, leading to insulin resistance. The change in iron balance contributes to the onset of diabetes [28,29]. These results come in agreement with another research conducted on young Indians which studied the factors affecting the proportion of HbA1c, whether diabetes or others, they measured the level of HbA1c in the anemic (IDA) and the number of people with diabetes and pre-diabetes is very large, so they thought the HbA1c ratio would be high when iron deficiency and lack of ferritin as well, so using of iron capsules would reduce the likelihood of high sugar [20].

The cause of diabetic complications related to vessels, whether small or large, is the increase in the level of sugar contributing the damage of the internal tissue, increase in the clotting level, and platelet activity [15]. In 2017, it was examined the effect of intravenous iron administration on iron-deficient patients with diabetes. This research reinforced the importance of using both HbA1c and fasting glucose to maintain healthy glucose reading beside using iron to reduce complication incidence [25]. This is consistent with the results our study which revealed that the incidence of diabetic complications was significantly decreased by 1.8 fold in diabetic patients with IDA treated with iron supplements (33.3%) compared to the type 2 diabetes and IDA who didn't receive iron supplements (60%).

Conclusion

Low iron level has a crucial effect on glycemic status by increasing the level of FBG and HbA1c, IDA is strongly correlated with type 2 diabetes, and the iron supplementation for diabetic patients with IDA ameliorates the progression of diabetes and its complications.

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