# Incidence and Association of Glycated Hemoglobin Levels with Iron Deficiency Anemia in Patients with or without Diabetes – A Study in a Semi Urban Area

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## Abstract

**Introduction:** HbA1c and other hemoglobins constitute the HbA1 fraction of the adult HbA[3]. HbA1c is also affected by pregnancy[5,6], uremia[7], hemolytic anemia[8], hemoglobinopathies[9], acute and chronic blodd loss[10,11], Vitamin B12, folate deficiencies. Iron deficiency anemia is also shown to have a considerable effect on HbA1c levels[12]. Studies have shown that reduced iron levels are correlated with increased levels of HbA1c leading to false high levels of HbA1c in individuals.

**Materials and Methods:** 1000 patients each with and without diabetes were included into the study. Hemoglobin levels, mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MHCH), mean corpuscular volume (MCV), hematocrit, platelet count, total blood picture, differential leucocyte count was done for all samples. Samples from patients with IDA were tested for all the above parameters at base line, after 1 week, 1 month and 2 months following iron treatment. Type of anemia was categorized and mild moderate and severe based on hemoglobin levels.

**Results:** Preponderance of females were seen in the anemic cases over males. 11.1% of non-diabetic and 16.3% of the diabetic patietns were anemic. The prevalence of low hemoglobin and low iron content was seen in 2.8% of the cases among non-diabetics while low Hb and normal iron levels were seen among 4.6% of the cases. Normal Hb with iron deficiency were seen among 3.7% of the cases while among the diabetics it was 2.1%, 7.3% and 6.9% respectively. Mean hemoglobin levels of all the severe anemic patients at base line was 5.9 ± 1.2, 9.1 ± 0.9 after 1 month and 10.6 ± 1.5 after 2 months of treatment.

**Conclusion:** There is a significant association of HbA1c levels and Iron Deficiency Anemia according to our study, though more studies need to be conducted to assess a proper clinical diagnosis

**Keywords**: Iron Deficiency anemia, HbA1c, glycated hemoglobin, diabetic patients, non-diabetic patients

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## Introduction

HbA1c is formed by the glycation of the terminal valine of the β-chain of hemoglobin. It is used as an indicator of patients glycemic status over the past 3 months[2,3]. HbA1c and other hemoglobins constitute the HbA1 fraction of the adult HbA[3]. A1c is the most common fraction found in the HbA1 fractions. HbA1c should be present below 7% in all diabetic patients according to the American Diabetes Association, so as to prevent the development of micro vascular complications[4]. Apart from being affected by the blood glucose levels, HbA1c is also affected by pregnancy[5,6], uremia[7], hemolytic anemia[8], hemoglobinopathies[9], acute and chronic blood loss[10,11], Vitamin B12, folate deficiencies. Iron deficiency anemia is also shown to have a considerable effect on HbA1c levels[12].

Iron deficiency is one of the most prevalent type of malnutrition. Ferritin is the form in which iron is stored, and testing amount of ferritin reflects the iron status. Globally, 50% of anemia is attributed to iron deficiency[13]. Studies have shown that reduced iron levels are correlated with increased levels of HbA1c leading to false high levels of HbA1c in non-diabetic individuals[14]. Though one of the most common cause of nutritional deficiency, there have been many reports of inconsistency in HbA1c levels and the clinical implications[12,14].

We have therefore conducted this study to investigate the effects of Iron deficiency anemia on HbA1c levels in diabetic and non-diabetic patients.

## Material and Methods

This retrospective study was conducted in the department of Biochemistry at Acsr Government Medical College over a period of two years.1000 diabetic patients and 1000 non diabetic patients aged between 18 – 70 years with or without anemia who underwent hemoglobin concentration and HbA1c level tasting were taken into consideration. All female patients of child bearing age, who had amenorrhea were screened for pregnancy and were excluded from the study if found positive. Patients were screened for disorders like hemolytic anemias, hemoglobinopathies, and uremia with high creatinine and urea levels and were also excluded if found positive.

Written informed consent was taken from all patients. Detailed history was taken and physical examination was done for all patients. Hemoglobin levels, mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MHCH), mean corpuscular volume (MCV), hematocrit, platelet count, total blood picture, differential leucocyte count was done for all samples. Samples from patients with IDA were tested for all the above parameters at base line, after 1 week, 1 month and 2 months following iron treatment.

The anemia type was defined by peripheral smear examination. The type of anemia was divided into mild, moderate and severe based on the hemoglobin levels. Mild anemia was categorized as hemoglobin levels of 12 – 12.9 g/dL in males and 11-11.9 g/dL in females; moderate anemia with 9-11.9 g/dL in males and 8-10.9 in females ; severe was <9g/dl in males and <8 g/dL in females[1].

Patients with microcytic (MCV <80fL) or hypochromic (MCH < 26 pg/cell) indices were considered as iron deficient anemia (IDA). IDA was confirmed by serum ferritin levels < 29 ng/ml in males and <10 ng/ml in females. The tests for these levels was redone after 1 month and 2 month.

## Results

Out of total 2000 patients, 64% were females and 36% were males in the diabetic group and 68% females and 32% males in the non-diabetic group (Fig: 1). In the control group, 60% were females and 40% were males. The mean age for the patient group was 32.7± 1.8 while that of control group was 33.2±1.4.

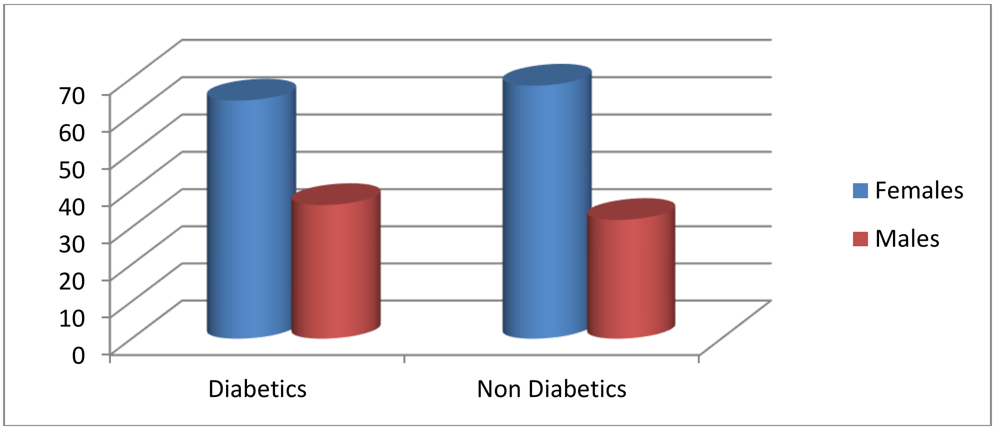


**Fig. 1: Sex wise distribution of anemia in patients with and without Diabetes mellitus**

Among the non-diabetic patients, 111(11.1%) of them were anemic, with 95(85.6%) of them severely anemic, 15(13.5%) moderately anemic and only 1(1%) had mild anemia. This was probably due to the fact that persons with mild anemia were not normally sent for HbA1c testing. 163(16.3%) of the diabetic patients, who underwent HbA1c testing were anemic. Of these, 117(71.8%) were having severe anemia, 44(27%) were with moderate anemia. (Fig: 2). There were no cases of mild anemia among the diabetic patients.

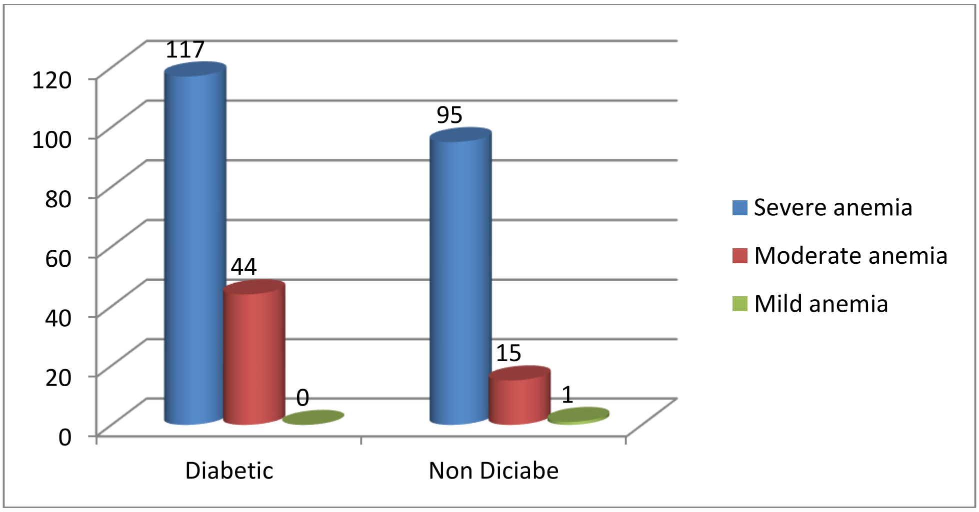


**Fig. 2: Type of anemia among diabetic and non-diabetic patients**

The most common symptom observed was weakness and pallor in 100% of the anemic cases and malaise in 96%. 46% of the patients had no energy or interest in work and dyspnea was seen in 42% of the cases. Splenomegaly was seen in 12% of the cases and systolic murmur in 50% of the patients (fig: 3).

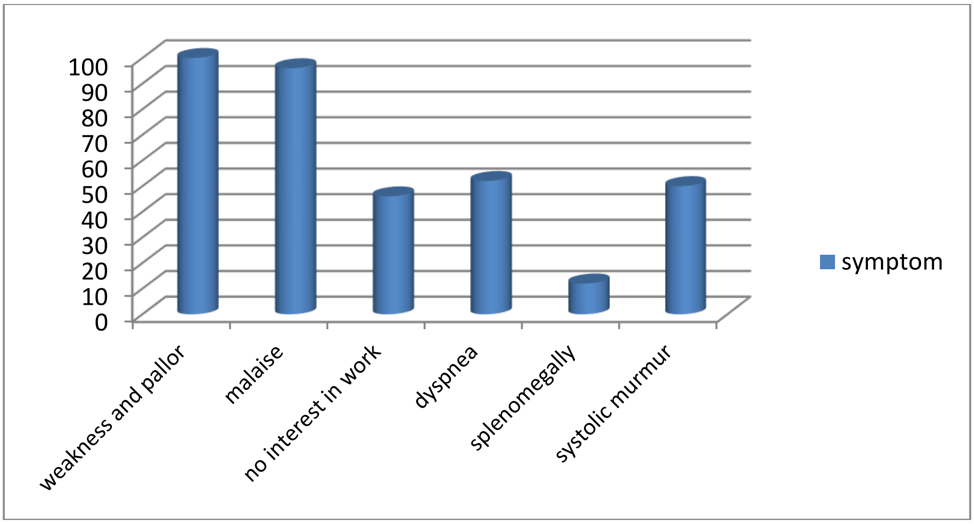


**Fig. 3: Symptoms seen among the anemic patients**

The prevalence of low hemoglobin and low iron content was seen in 2.8% of the cases among non-diabetics while low Hb and normal iron levels were seen among 4.6% of the cases. Normal Hb with iron deficiency were seen among 3.7% of the cases while among the diabetics it was 2.1%, 7.3% and 6.9% respectively. (Table: 1)

| | **Diabetic N=1000** | | **Non Diabetic N=1000** | |
|---|---|---|---|---|
| | **Females (%) (n = 642)** | **Males (%) (n = 358)** | **Females (%) (n = 679)** | **Males (%) (n = 321)** |
| Low Hb, low iron | 16 (2.5%) | 5 (1.4%) | 21 (3.1%) | 7 (2.2%) |
| Low Hb, Nornal iron | 54 (8.4%) | 19 (5.3%) | 34 (5.0%) | 12 (3.7%) |
| Normal Hb, low iron | 47 (7.3%) | 22 (6.1%) | 26 (3.8%) | 11 (3.4%) |
| Normal Hb, Normal iron | 525 (81.8%) | 312 (87.2%) | 598 (88.1%) | 271 (90.7%) |

Females were predominantly affected with anemia, both iron deficiency and non-iron deficiency type than males. Mean hemoglobin levels of all the severe anemic patients at base line was 5.9 ± 1.2, 9.1 ± 0.9 after 1 month and 10.6 ± 1.5 after 2 months of treatment. The mean hemoglobin level of the normal patients (Normal Hb and normal iron content) it was 12.6 ± 3.8. It was observed that though treatment with iron supplements were given to the patients for 3 months, the iron content was still lower than that of normal non anemic patients

**Table 2: Ferritin, hemoglobin and HbA1c levels in IDA patients before and after treatment**

| | **Patients at baseline** | **Patients at 1 month** | **Patients at 2 months** | **Normal patients** |
|---|---|---|---|---|
| Serum Ferritin level (ng/ml) | 17.7 ± 5.8 | 145.6 ± 67.1 | 256 ± 65.2 | 235.0 ± 35.5 |
| Hemoglobin level (g/dL) | 5.9 ± 1.2 | 9.1 ± 0.9 | 10.6 ± 1.5 | 12.6 ± 3.8 |
| HbA1c levels (%) | 6.3 ± 0.3 | 5.4 ± 0.7 | 5.1 ± 0.4 | 5.6 ± 0.5 |

Among the diabetics whose fasting blood sugar levels was <100mg/dL, the mean Hb levels was found to be 10.1± 1.9 in males and 9.4 ±2.3 in females. Mean HbA1c value was found to be 6.14 ±1.1% among the moderate anemia cases while it was 5.5 ± 1.3 among the severe anemia patients.

## Discussion

Iron deficiency anemia is one of the most common types of anemia. HbA1c is a glycated hemoglobin that is used to determine the glycemic status of a patient for the past 3 months. But there are a few conditions which also affect the levels of HbA1c like pregnancy, uremia, hemolytic anemias, hemoglobinopathies, acute and chronic blood loss[8-11].

Our study shows that Iron deficiency anemia is more common among women than men. This was observed in a similar study by Nitin Sinha et al who reported females to be more affected with IDA than males.

Our results show that there is a positive correlation between hemoglobin and HbA1c concentrations. HbA1c levels tend to be higher in cases of iron deficiency. On treatment with iron supplements, the HbA1c levels decrease. This was observed first by Brooks et al who reported that HbA1c levels in 35 non diabetic patients with IDA were significantly higher in IDA patients before treatment with iron supplements and decreases after treatment[14]. They estimated that the reason could be due to the fact that in iron deficiency, the quaternary structure of the hemoglobin molecule was altered, and that glycation of the globin chain occurred more readily in the relative absence of iron [14].

Van Heyningen et al reported that there was no difference in HbA1c concentration and IDA before and after treatment in non-diabetic patients. They reported that the difference is due laboratory testing[15]. This was corroborated by a study by Hansen et al, who demonstrated that there was no difference in the HbA1c concentrations between IDA and healthy individuals[16].

Corroborating our study, studies by Cogan et al, and el Agouza et al showed that the HbA1c leels were higher in patietns with IDA and decreased significantly on treatment with iron supplements. According to them, elevated HbA1c levels in iron deficiency anemia could be explained by the assumption that if serum glucose remains constant, a decrease in the hemoglobin concentration might lead to an increase in the glycated fraction[17,18].

In contrast, in a study by Sinha et el, there was a significant rise in the HbA1c levels 2 month after treatment with iron supplements in IDA patients. There have been a few studies on diabetic patients which has proven that controlled plasma glucose levels for 3 months correlates very well with controlled HbA1c. Hence, patients with controlled plasma glucose levels are expected to have A1C below 6.5 %[19].

Ferritin, the storage form of iron reflects its true status of iron in the plasma[20]. We have assessed the levels of ferritin during the treatment of severely anemia patients. We observed that there was a considerable increase of ferritin levels on treatment in 1 and 2 months which was associated with marked rise in the hemoglobin levels. This results were corroborated by Nitin Sinha et al while in yet another study by Christy et al[20], no significance was found.

Raj et al[22] investigated 86 patients with type 2 diabetes mellitus and demonstrated that serum ferritin positively correlated with HbA1c and was increased with increasing duration of disease, indicating that poor glycaemic control can contribute to elevated ferritin levels independent of iron status.

## Conclusion

There is a significant association of HbA1c levels and Iron Deficiency Anemia according to our study. But there are few studies done in this area. This only reiterates the need of further investigations to be performed with more number of participants in order to estimate the significance of this association so as to address the situation in clinical practice.

**Conflict of Interest: None**

**Source of Support: Nil**

## References:

1. Rastogi T, Mathers C, editors. Global burden of iron deficiency anaemia in the year 2000. [Updated on Apr 2007]. http://www.who.int/entity/healthinfo/statistics/bod_irondeficiencyanaemia.pdf
2. International Expert Committee. International Expert Committee report on the role of the A1c assay in the diagnosis of diabetes. Diabetes Care2009;32:1327–1334.
3. Telen MJ, Kaufman RE. The mature erythrocyte. In: Greer JP, Forester J, et al., editors. Wintrobe's clinical hematology. 11th ed. Lippincot: Williams and Wilkins; 2004. p. 230.
4. American Diabetes Association. Position statement: Standards of medical care in diabetes-2007. Diabetes Care. 2007;30(S1):S9.
5. Lind T, Cheyne GA. Effect of normal pregnancy upon the glycosylated haemoglobins. Br J Obstet Gynaecol. 1979; 86:210–213.
6. Phelps RL, Honig GR, Green D, Metzger B, Frederiksen MC, Frienkel N. Biphasic changes in hemoglobin A1c concentrations during normal human pregnancy. Am J Obstet Gynecol. 1983;147:651–653.
7. Flückiger R, Harmon W, Meier W, Loo S, Gabbay KH. Hemoglobin carbamylation in uremia. N Engl J Med. 1981;304:823–827.
8. Horton BF, Huisman TH. Studies on the heterogeneity of hemoglobin. VII. Minor hemoglobin components in haematological diseases. Br J Haematol. 1965;11:296–304.
9. Eberentz-Lhomme C, Ducrocq R, Intrator S, Elion J, Nunez E, Assan R. Haemoglobinopathies: a pitfall in the assessment of glycosylated haemoglobin by ion-exchange chromatography. Diabetologia.1984;27:596-598. [PubMed]
10. Bernstein RE. Glycosylated hemoglobins: hematologic considerations determine which assay for glycohemoglobin is advisable. Clin Chem. 1980;26:174–175. [PubMed]
11. Starkman HS, Wacks M, Soeldner JS, Kim A. Effect of acute blood loss on glycosylated hemoglobin

determinations in normal subjects. Diabetes Care. 1983; 6:291–294.
12. Nitin Sinha, Mishra TK, Tejinder Singh, Naresh Gupta; Effect of Iron Deficiency Anemia on Hemoglobin A1c Levels Ann Lab Med. 2012 Jan;32(1):17–22.
13. John A. Iron Deficiency and Other Hypoproliferative Anemias. In: Longo D, Fauci A, Kasper D, Hauser S, Jameson J, Loscalzo J, editors, editors. Principles of Internal Medicine by Harrisons. 17th ed. United States of America: McGraw-Hill; 2008. pp. 628–35.
14. Brooks AP, Metcalfe J, Day JL, Edwards MS. Iron deficiency and glycosylated haemoglobin A1. Lancet. 1980 Jul;316(8186).
15. Van Heyningen C, Dalton RG. Glycosylated haemoglobin in iron-deficiency anaemia. Lancet.1985;1:874.
16. Gram-Hansen P, Eriksen J, Mourits-Andersen T, Olesen L. Glycosylated haemoglobin (HbA1c) in iron- and vitamin B12 deficiency. J Intern Med. 1990;227:133–136.
17. El-Agouza I, Abu Shohla A, Sirdah M. The effect of iron deficiency anaemia on the levels of haemoglobin subtypes: possible consequences for clinical diagnosis. Clin Lab Haematol. 2002;24:285–289.
18. 21. Coban E, Ozdogan M, Timuragaoglu A. Effect of iron deficiency anemia on the levels of hemoglobin A1c in nondiabetic patients. Acta Haematol. 2004;112:126–128.
19. Standards of Medical Care in Diabetes—2011. Diabetes Care. 2011;34(1):S13.
20. John A. Iron Deficiency and Other Hypoproliferative Anemias. In: Longo D, Fauci A, Kasper D, Hauser S, Jameson J, Loscalzo J, editors , editors. Principles of Internal Medicine by Harrisons. 17th ed. United States of America: McGraw-Hill; 2008. pp. 628–35.
21. Alap L Christy, Poornima A Manjrekar, Ruby P Babu, Anupama Hegde, Rukmini MS. Influence of Iron Deficiency Anemia on Hemoglobin A1C Levels in Diabetic Individuals with Controlled Plasma Glucose Levels Iran Biomed J. 2014 Apr;18(2):88–93.
22. Raj S, Rajan GV (2013) Correlation between elevated serum ferritin and HbA1c in type 2 diabetes mellitus. Int J Res Med Sci 1:12–15.