Glycemic control of diabetic patients with iron deficiency anemia, HbA1c or Fructosamine, which marker should we use?

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ABSTRACT

Glycemic control is very important for the management of the risk of long-term complications in diabetic patients. The HbA1c is the most commonly used marker but its rate is affected by numerous physiological and pathological variations as seen during renal failure, hemolyse, hemoglobinopathies and also iron deficiency anemia during which levels are falsely higher.In this case, particular interest is given to fructosamine which is independent of the fluctuations incurred by hemoglobin.Our present study aims to determine the impact of iron deficiency anemia on glycated hemoglobin determination and the place of fructosamine in this situation.

We conducted a case-control study at the central laboratory of medical analysis. We realize different biochemical parameters in cases and controls such as: glycaemia, HbA1C, Fructosamine, iron, ferritin, hemoglobin and others, the statistical study was done with excel and SPSS Software

Our study focused on a series of 100 type 2 diabetic patients, 50 of whom were anemic (cases) with a sex ratio of 0.21 and 50 non anemic (controls)with a sex ratio of 0.72.

The mean HbA1c in cases (no iron treated)is 8.83% (SD=2.6) and 7.77 %(SD=1) in controls. The average fructosamine in cases (no iron treated) is 395.50 umol/dl (SD=118.03) and 449.11 umol/dl (SD=146,43)in controls. The student test demonstrated significant difference of HbA1c levels between cases and controls independently of glycaemia nevertheless no difference in fructosamine rates.

In anemic diabetics we found no significant correlation between glycated hemoglobin and Glycaemia while fructosamine was well correlated, also no correlation between HbA1c and fructosamine while this two parameters were correlated in patients without anemia.

Our results confirm that fructosamine values evolve in parallel with those of glycated hemoglobin, but are not subject to variations in hemoglobin hence the usefulness of this parameter in these situations

KEYS WORDS:

Diabetes, Iron deficiency anemia, HbA1c, Fructosamine, Glycemic control

PURPOSE

Diabetes mellitus is a public health problem,according to the WHO report the number of diabetic persons has passed from 108 million in 1980 to 422 million in 2014. The worldwide prevalence of diabetes in adults has passed from 4,7% in 1980 to 8,5 % in 2014[1].

Glycemic monitoring is essential for the management of type 2 diabetes mellitus, and various glycemic markers are available in clinical practice.

Hemoglobin A1C (HbA1c) which is formed by glycation of the NH -terminal valine residue of the β-chain of hemoglobin is the most widely used parameter. It reflects average glucose levels over 2–3 months and provides information on the risk of long-term complications in patients with diabetes. Recently (2010) the American Diabetes Association (ADA) confirmed the HbA1c as a diagnostic marker of diabetes if it is ≥ 6.5% and between 5,7% and 6,5% individuals are at high risk for developing diabetes[2].

However, HbA1c is limited in its ability to reflect short-term glycemic changes, and it cannot reflect postprandial hyperglycemia and fasting hyperglycemia separately.

In addition, HbA1c levels can be influenced by a variety of factors: analytical (standardization problems), physiological including age, ethnicity, pregnancy, red cells life span , smoking, and pathological factors like conditions that alter red cells turnover (hemoglobinopathies, hemolyse, and anemia).

40% of North Africans suffer from anemia; In Algeria, it affects 20% of the population, the prevalence of anemia is estimated about 10% to 30% in patients with diabetes.

Iron deficiency is one of the most prevalent forms of malnutrition. Globally, 50% of anemia is attributed to iron deficiency.

Anemia may be associated with more rapid erythrocyte turnover, which decreases the HbA1c level, or with slower turnover or changes in the 3dimensional configuration of Hb, which elevates the HbA1c level.

Iron and vitamin B12 deficiency, renal failure, and bone marrow suppression in alcoholism inhibit erythropoiesis and increase the mean survival duration of erythrocyte, leading to increase HbA1c. However, hemolytic anemia, chronic liver disease increase reticulocytes and decrease the mean age of erythrocyte, which can decrease HbA1c level[3].

Therefore alternative markers could be useful, such as fructosamine measuring the amount of total glycosylated proteins in serum which relates to average levels of glucose during the preceding 1 to 3 weeks[4]. It is a simple, robust and inexpensive test. It is not affected by anemia or variant hemoglobin but the level is influenced by the concentrations of serum protein, bilirubin, uric acid, and low molecular weight substances coexisting in the blood[5].

Previous studies have shown that iron deficiency elevates HbA1c levels independent of glycaemia[6][7][8][9]showed that the HbA1c levels in subjects with iron deficiency anemia (IDA) were higher than those of subjects with normal iron levels. However, these studies were performed mostly in subjects without diabetes, so they could not conclude whether the presence of IDA affected the HbA1c levels in diabetic patients.

Our study aims to determine the impact of IDA on glycated hemoglobin levels, as well as the interest of the determination of fructosamine during this situation.

MATERIAL AND METHODS

TYPE AND SCOPE OF STUDY:

This is a case-control study, conducted at the central laboratory of medical analyzes, UHC FRANTZ FANON Blida between January and April 2019.

The glycated hemoglobin assay was performed at UHC BAB EL OUED Algiers

STUDY POPULATION:

Our study included 100 type 2 diabetic patients (50 controls and 50 cases)

The subjects with low hemoglobin levels (<13 g% in males and <11 g% in female), predominantly microcytic indices (MCV <76 fL), and hypochromic indices (MCH <27 pg/cell) were considered to have iron deficiency anemia (cases), which was confirmed by low serum ferritin levels (<25 and <20 ng/ml in males and females, respectively).

All subjects with: Type 1 diabetes; kidney failure; dysproteinemia; inflammatory anemia; icterus were excluded

FACT SHEET:

Contains the following data:

Last name and first name; Demographic information (gender, age);Weight height; Age of diabetes; Diet;Risk factors (tobacco); Current treatment; physical exercise,other pathologies , body mass index (BMI), systolic blood pressure.

LABORATORY METHODS:

Complete blood count parameters: Hemoglobin, MCV, MCH, and MCHC estimation was carried out by hematology analyzer (Sysmex, Kobe, Japan); HbA1cwas measured usingimmonoturbidimetric method (Cobas 6000 analyzer) and Fructosamine by a spectrophotometric colorimetric method

Other biochemical parameters estimated using Selectra ProM analysers

ELITECH DIAGNOSTIC:

Plasma glucose was estimated by glucose oxidase/peroxidase method ; Concentrations of cholesterol, high-density lipoprotein–cholesterol (HDL-C) , non-HDL-C and triglycerides ; C-reactive protein (CRP) ; Urea and creatinine ; Proteins, albumin ; bilirubin and Ferritin Statistical analysis was carried out by the SPSS software (Statistical Package for the Social Sciences) version 25.Student and Wilcoxon tests were performed respectively, based on the results of the Normality test of quantitative variables, Chi-square test and fisher exact test for qualitative variables, and Pearson's coefficient of regression.

A value of p <0.05 was selected as statistically significant

RESULTS

TABLE 1 POPULATION QUALITATIVE CHARACTERISTICS IN CASES AND CONTROLS

CHARACTERISTICS	CATEGORY	N	N	P (KHI2)
Gender	Men	9 SR=0.21	21 SR=0.72	
	Women	47	29	**0.007**
Overweight	No	14	41	
	Yes	36	9	0.23
Diet	Yes	32	23	
	No	18	27	0.7
Smoking	Yes	2	2	
	No	33	41	
	Passive	15	7	0.196

No differences between cases and controls in term of qualitative characteristics except the sex ratio.

TABLE 2 POPULATION QUANTITATIVE CHARACTERISTICS IN CASES AND CONTROLS

CHARACTERISTICS (UNITS)		CASES	CONTROLS	P
		Mean (SD)	Mean (SD)	Student
Old	(years)	59.59 (11.95)	62.2(9.29)	0.07
BMI		28.2 (4.72)	27.42 (3.12)	0.33
Age of diabetes	(years)	8.37(6.33)	6.87(5.13)	0.19
Cholesterol	(g/l)	1.61(0.47)	1.85(0.51)	0.50
Triglycerides	(g/l)	1.28(0.57)	1.65(0.68)	0.004
Urea	(g/l)	0.39(0.17)	0.36(0.15)	0.06
Creatinine	(mg/l)	9.63 (3.71)	9.14(2.74)	0.44
Glycaemia	(g/l)	1.32(0.51)	1.44(0.45)	0.197
HbA1c	**(%)**	**8.05 (2.11)**	**7.77(1.64)**	**0.457**
Fructosamine	**(µmol/dl)**	**432.82 (126.12)**	**449.11 (146.43)**	**0.553**
Hemoglobin	(g/dl)	10.8 (0.81)	13.66(1.04)	0.0001
Red cells	(x106element)	3.99 (0.7)	4.87 (0.80)	0.00034
VGM	(fl)	76.00 (4.59)	80.68(3.56)	0.00069
Iron	(µg/dl)	45.64 (21.07)	94.03(31.06)	0.0004
Ferritin	(ng/ml)	27.34(27.97)	104.35 (65.60)	0.00036

Characteristics of cases and controls were identical without observable differences except anemia. Difference between fructosamine rate in cases and controls was no significant and also HbA1c but Knowing that iron treatment affect HbA1c values so we excluded iron treated patients and we do again the comparisonof HbA1c and fructosamine between controls and no iron treated cases

TABLE 3 COMPARISON MEAN HBA1C, GLYCAEMIA AND FRUCTOSAMINE BETWEEN CASES AND CONTROLS AFTER JUSTIFYING IRON TREATMENT EFFECT

	Cases non-iron treated n=32 Mean (SD)	Controls n=50 Mean (SD)	P
HbA1c %	8.83 (2,16)	7,77 (1.64)	**0.013**
Glycaemia g/l	1.37 (0.55)	1.44 (0.45)	0.53
Fructosamine µmol/dl	395.50 (118.03)	449.11 (146.43)	0.08

Mean of HbA1c was statistically higher in cases than controls independently of glycaemia, while no difference noticed in fructosamine rate, which means that IDA influence HbA1c values.

TABLE 4 PEARSON CORRELATION BETWEEN HBA1C AND HEMATOLOGICAL PARAMETERS

	CASES		CONTROLS	
	r	p	r	p
Hemoglobin	0.031	0.756	0.082	**0.411**
Ferritin	-0.077	0.431	0.161	**0.102**
MGV	**-0.430**	**0.002**	-0.232	**0.102**

Presence of significant negative correlation between HbA1c levels and mean globular volume (MGV)

TABLE 5 PEARSON CORRELATION BETWEEN GLYCAEMIA AND HBA1C AND GLYCAEMIA AND FRUCTOSAMINE

	GLYCAEMIA			
	CASES		CONTROLS	
	r	p	r	p
Hba1c	**0.233**	**0.107**	**0.237*** **	0.024
Fructosamine	**0.146*** **	**0.033**	**0.255*** **	0.01

HbA1c is significantly correlated with glycaemia in controls but not in cases, while fructosamine is significantly correlated with glycaemia both in cases and controls

TABLE 6 PEARSON CORRELATION BETWEEN HBA1C AND FRUCTOSAMINE IN CASES AND CONTROLS

	HBA1C			
	CASES		CONTROLS	
	r	p	r	p
Fructosamine	0.226	0.115	**0.529*** **	**0.000**

Fructosamine and Hba1c are correlated in controls but not in cases

DISCUSSION

Our study did not show any significant correlation between hemoglobin and HbA1c both in cases and controls in agreement with results obtained by Alap et al[10].However erythrocyte size had a strong negative correlation with HbA1c, compatible with the hypothesis that iron deficiency increases Hb glycation.

Ferritin is a storage form of iron. Hence, in this study, its correlation with HbA1c was assessed, but no significant. In a study by Raj and Rajan[11], ferritin showed positive correlation with HbA1c in diabetic individuals; Sharifi and al[12], Alap and al[10] did not find any significant correlation. We could not explain the lack of correlation of serum ferritin levels with HbA1c in this study may be it was due to the simple size.

At the first time, we found that the presence of IDA slightly increased the HbA1c level in cases nevertheless it was statistically insignificant. But Brooks et al.[5], Gram-Hansen et al.[13] and Coban et al.[14] showed effects of iron therapy on glycated hemoglobin and found a significant reduction in HbA1c levels after iron therapy. So after justifying this factor by excluding iron treated cases, the mean of HbA1c was statistically higher in cases independently of glycaemia comparing with controls.Our results were similar to those obtained by Alap et all in india[9], Hashimoto and al.[15]et other studies.

According to the explanation provided by Sluiter hemoglobin glycation is an irreversible process. Hence, HbA1 levels in erythrocyte will be increased with cell age. In iron deficiency, red cell production decreases, consequently an increased average age of circulating red cells ultimately leads to elevated HbA1 levels[16]

It has been suggested too that the quaternary structure of the Hemoglobin molecule may be altered and that glycation of the βglobin chains occurs more readily in patients with IDA.

El-Agouza et al. proposed that a decrease in the Hemoglobin concentration might lead to an increase in the glycated fraction at a constant glucose level because HbA1c is measured as a percentage of total HbA[17]

Saudek et al.considered measurements of HbA1c to be invalid in the presence of anemia [18]

In our study Fructosamine was correlated with Fasting serum glycaemia in cases contrary to the HbA1c; This two markers were strongly correlated in controls `in step of all studies done in this sense` but not in cases which means that really HbA1c values are affected by IDA

Therefore, IDA not only increases HbA1C levels in non-diabetic individuals but also it can interfere with its ability to determine glycemic status of diabetic individuals

Nevertheless fructosamine is not affected by IDA and may be a useful substitute to HbA1c for monitoring glycemic control.

CONCLUSION

In persons with conditions that may interfere with the interpretation of the HbA1c test fructosamine is proposed like an alternative.

We draw clinician`s attention to any conditions that could affect the red cells turnover and before altering the treatment regimen for diabetes, iron deficiency anemia should be considered.

For sure additional studies with larger number of participants with IDA would be helpful in examining the impact of IDA on measurements of HbA1c.

KEYS WORDS: Diabetes, Iron deficiency anemia, HbA1c, Fructosamine, Glycemic control

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