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Correlation between Estimated Average Glucose and Fasting Blood Sugar Levels in Diabetic Anaemic Patients

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Abstract: Diabetes mellitus, characterized by chronic hyperglycemia, requires accurate monitoring of blood glucose levels for effective management. This study investigated the relationship between estimated Average Glucose (eAG) calculated from HbA1c levels and Fasting Blood Sugar (FBS) in diabetic patients. The research aimed to assess the correlation between these glycemic parameters and their implications for evaluating glucose control. A total of 54 diabetic patients (20 males, 34 females) were included in this cross-sectional study. Blood samples were collected to measure FBS and HbA1c levels on the same day. eAG was calculated using Nathan's regression equation as recommended by the American Diabetes Association. Statistical analyses were performed using SPSS, including Pearson's correlation coefficient to determine the relationship between eAG, FBS, and HbA1c.Results showed a significant positive correlation between eAG and FBS ($r=0.629$, $p<0.0001$). A perfect correlation was observed between eAG and HbA1c ($r=1.000$, $p<0.0001$), as expected due to the direct calculation of eAG from HbA1c. The study found that males had higher mean FBS (225±91 mg/dl) and eAG (209.2±80.4 mg/dl) levels compared to females (FBS: 177±43 mg/dl; eAG: 184.6±49.1 mg/dl), although these differences were not statistically significant. The findings suggest that while eAG and FBS are significantly correlated, they cannot be used interchangeably. The strength of the association between eAG and FBS appeared to depend on the level of glucose control, with a stronger correlation observed in poorly controlled diabetes. This study highlights the potential utility of eAG in conjunction with HbA1c for assessing long-term glycemic control, while emphasizing the continued importance of FBS measurements for day-to-day management. Further research with larger sample sizes and consideration of additional factors influencing glycemic control is warranted to validate these findings and explore their clinical implications.

Keywords: Diabetes mellitus, Glycated hemoglobin (HbA1c), Estimated Average Glucose (eAG), Fasting Blood Sugar (FBS), Glycemic control

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Introduction

Diabetes mellitus (DM) is a metabolic condition that is adapted by the rise in blood sugar levels due to the body's complications of processing carbohydrates, fats, and proteins. Globally, more than 400 million adults suffer from DM leading to 1.6 million deaths in the year 2015. Predictions

indicate that it can affect every 1 in 10 individuals by the year 2035 (Taderegew *et al*., 2020). Diabetes can be categorized into 2 types: Type 1 and Type 2 diabetes. Type 1 diabetes occurs when the body is unable to produce insulin, which is a result of autoimmune response. Type 2 diabetes, on the other hand, occurs when the body is not making enough insulin or is not absorbed by cells.

This condition arises from defects in insulin secretion by pancreatic B cells, insulin action, or both. Insulin is produced by pancreatic beta cells in response to increased blood glucose levels and attempts to lower it. It aids in the absorption of glucose by most body cells for utilization of energy, simultaneously involving the process of conversion of glucose to glycogen for storage in the liver and muscle cells. Therefore, it is mandatory to maintain a constant insulin level as a reduction in insulin secretion can cause hyperglycemia. This results in the disruption of carbohydrate, lipid, and protein metabolism, leading to the diagnosis of diabetes. Hence, untreated diagnosis induces the dysfunction of various organs, notably the eyes, kidneys, nerves, heart, and blood vessels (Farrar *et al*., 2017).

The measurement of the glycated form of haemoglobin (HbA1c) is the product of a non-enzymatic reaction between haemoglobin and glucose, reflecting HbA1c within 3 months. This test is widely used to diagnose diabetes, followed by the recommendation from the International Expert Committee, which stated the importance of HbA1c in diagnosing diabetes with a threshold of above 6.5% (Alzahrani *et al*., 2023).The lifespan of RBCs is approximately 120 days, which aids in the determination of the average blood glucose levels for the past 2 to 3 months. However, slower RBC production could lead to falsely elevated HbA1c levels. This can occur due to conditions such as hemoglobinopathies, sickle cell anaemia, and anaemia related to iron, vitamin B_{12}, or folate deficiency (Polgreen *et al*., 2003). Moreover, HbA1c is not a reliable marker for blood glucose in newborns with diabetes. This is because most haemoglobin is fetal haemoglobin (HbF), with less than 10% being haemoglobin A. As a result, HbA1c levels tend to be low even when blood glucose is high in neonatal diabetes. This happens because when the HbF alters with age, HbA1c does not accurately reflect blood glucose control in newborns (Suzuki *et al*., 2011; Suzuki *et al*., 2013). HbA1c levels do not accurately show short-term changes in blood sugar. They change gradually even when the blood sugar control improves rapidly. Therefore, HbA1c should be measured about 12 weeks after starting a new diabetes treatment to assess its effectiveness (Koga *et al*., 2011). In cases of fulminant type 1 diabetes, where blood sugar levels spike promptly, HbA1c may not be a reliable indicator as it might remain normal or only slightly elevated (Imagawa *et al*., 2000). Hence, average HbA1c levels are linked to the development and progression of diabetic complications (Nathan *et al*., 1993). Studies have found a connection between post-meal high blood sugar and cardiovascular disease (Wang *et al*., 1993). Therefore, better control of blood sugar variability is crucial to preventing cardiovascular disease in diabetes. HbA1c mainly reflects average blood sugar levels and does not distinguish between high blood sugar after meals and high fasting blood sugar (Reichard *et al*., 1993).

Several studies have tested the relationship between average blood glucose and HbA1c levels, resulting in various equations (Rohlfing *et al*., 2002; Nathan *et al*., 2007; Sacks *et al*., 2023). Similarly, this study sought to establish the relationship between estimated Average Glucose (eAG) levels calculated by Nathan's regression equation as recommended by the American Diabetes Association (ADA) and Fasting Plasma Glucose (FBS) levels.

Materials and Methods

In this study, FBS levels of 54 diabetic patients (20 males, 34 females) were observed using data from the Laboratory Information System at Sri Ramachandra Institute of Higher Education and

Research. Blood samples collected on the same day have been used to measure FBS and HbA1c. The individuals were characterised into the anaemic diabetic group, including patients with fasting blood sugar levels≥ 126 mg/dl and Hb < 13 g/dl in men and 12 g/dl in women. The estimated blood glucose levels (mg/dl) could be calculated using the formula 28.7 x HbA1c – 46.7. Glucose levels were measured using Beckman kits and the hexokinase method on Beckman AU 5800 and AU680 Automated Clinical Chemistry (Beckman Coulter Inc., Brea, CA, USA). HbA1c levels were detected using High-Performance Liquid Chromatography (HPLC).

All statistical analyses were performed using the Statistical Package for Social Sciences (SPSS, version 25.0 for Windows, Chicago, IL, USA). Data are expressed as the Means of Standard Error (SEM). A p-value of 0.05 was accepted as significant. The Spearman correlation coefficient (r) was used to test the relationship between the FBS and eAG levels.

Results and Discussion

This study was conducted at the Department of Biochemistry in Sri Ramachandra Institute of Higher Education and Research, Chennai, India. In a total sample size of 54, 20 were male and 34 were female. The mean age of males in the sample was around 60 years, while the mean age of females was approximately 55. Despite this apparent difference, the p-value of 0.0001 suggested that this difference is statistically significant. This means the age distribution between males and females was meaningfully different in this sample. The high standard deviation in both groups indicated a wide range of ages among participants. The mean FBS level for males is 225 mg/dl, notably higher than the 177 mg/dl observed in females (Table 1). The p-value (0.709) indicated that this difference was not statistically significant. This suggested that males in the sample have significantly higher FBS levels compared to females, which could indicate a higher prevalence of glucose regulation issues or diabetes among males in this group. The significant standard deviation, particularly in the male group, indicated considerable variability in FBS levels within each gender. The eAG estimated average blood glucose levels over the previous months based on HbA1c. The mean eAG is higher in males (209 mg/dl) than in females (184 mg/dl). The p-value of 0.709, though not below the standard threshold of 0.05, is relatively close, indicating that the difference might be of some interest but is not definitively statistically significant.

There exists a potential trend towards higher average glucose levels in males compared to females, but the result (Tables 1, 2) is not strong enough to conclude a significant difference. HbA1c reflects long-term glucose control, providing an average of blood sugar levels over approximately 2-3 months. The mean HbA1c percentage is higher in males (9.1%) compared to females (8.2%). Similar to eAG, the p-value is 0.732, suggesting that while there may be a trend toward higher HbA1c in males, it is not statistically significant in this sample. This could suggest that, on average, males have poorer long-term glucose control than females, though this conclusion is tentative given the borderline p-value.

Pearson's correlation coefficient between eAG and FBS was equivalent to zero. This represented that there was a significant positive relationship between these two factors. The correlation coefficient between eAG and HbA1c is r=1.000, also with a p-value of 0.0001. This perfect correlation suggests that eAG is directly derived from or closely associated with HbA1c values. The correlation coefficient between FBS and HbA1c is r=0.629** with a p-value of 0.0001, indicating a strong positive and statistically significant correlation. The diagonal line signified the ideal 1:1 relationship between eAG and FBS (Fig. 1). Figure 2 reveals a perfectly linear relationship between HbA1c and eAG. Each data point falls exactly on the regression line, further supporting the strong correlation in the correlation table. This linear relationship underscores that eAG can

Table 1: Comparison of glycaemic parameters in males and females

	Male	Female	p-value
n	20	34	
Age (y)	60.4±11.6	55.1±12.3	0.0001
FBS (mg/dl)	225±91	177±43	0.709
eAG (mg/dl)	209.2±80.4	184.6±49.1	0.732
HbA1c (%)	9.1±2.9	8.2±1.7	0.732

Table 2: Correlation between FBS and eAG

Parameters	eAG (mg/dl)		FBS (mg/dl)		HbA1c (%)	
	r value	p-value	r value	p-value	r value	p-value
eAG (mg/dl)	-	-	0.629**	0.0001	1.000**	0.0001
FBS (mg/dl)	0.629**	0.0001	-	-	0.629**	0.0001
HbA1c (%)	1.000**	0.0001	0.629**	0.0001	-	-

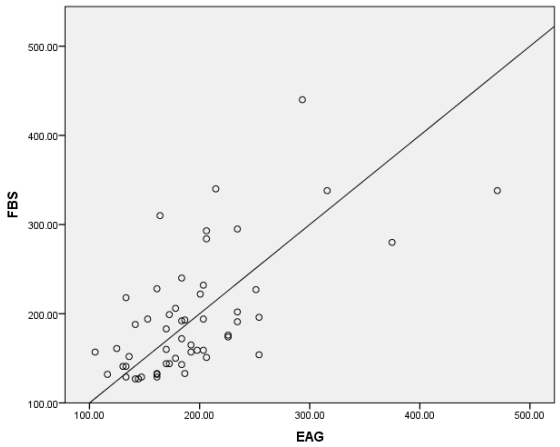

Fig. 1: Scatter-plot of FBS vs. eAG.

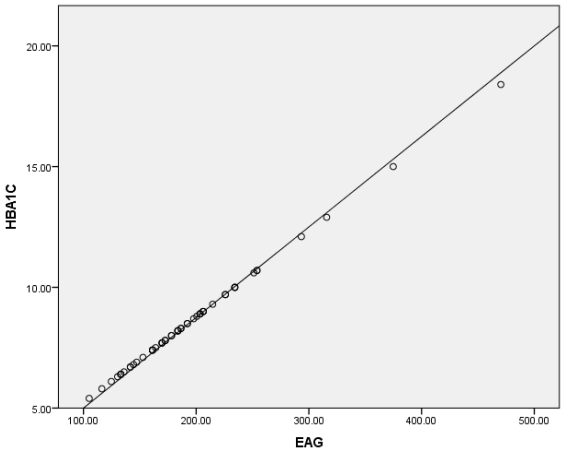

Fig. 2: Scatter-plot of HbA1c vs. eAG.

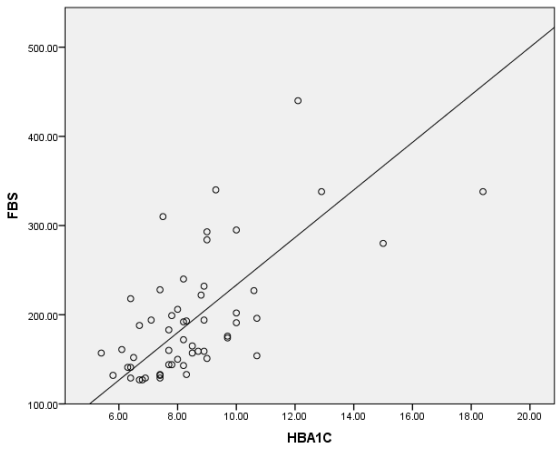

Fig. 3: Scatter-plot of HbA1c vs. FBS

be reliably predicted from HbA1c values. Moreover, in Figure 3, a positive linear trend between HbA1c and FBS, though with more variability compared to the previous plot, indicated that while there is a positive correlation, it is not as strong as the one between HbA1c and eAG. The scatter plot includes some outliers; significantly higher FBS values deviate from the regression line. This suggests that FBS is less consistent in predicting HbA1c compared to eAG.

HbA1c is one of the standard diagnostic methods for diabetes mellitus as it is a trusted biomarker with 6.5% or more levels indicating diabetes (Sherwani *et al*., 2016). It represents the average blood sugar level over eight to twelve weeks, thus being an alternative to fasting plasma glucose or oral glucose tolerance tests. Unlike other methods used for diagnosis, HbA1c testing can be done at any time without any special preparation; hence, it is more convenient (Yun *et al*., 2023). Long-term blood sugar control and evaluation for complications in diabetics are monitored by measuring HbA1c. According to the American Diabetes Association, most adults with diabetes, except pregnant ladies, should aim at achieving a target HbA1C <7%. On the other hand, targets may vary depending on the patient's disease type, duration, and overall health status. It is possible to express HbA1c results as either percentage (%) or as millimoles per mole (mmol/mol) (Abera *et al*., 2022). Standardization and stringent quality control are necessary steps to measure HbA1c levels correctly. However, certain medical conditions, such as sickle cell traits, may affect the measurement result. Patients must communicate with their doctors about their HbA1c levels to understand their health circumstances based on these results. An HbA1c of 6.5% is equivalent to 48 mmol/mol, and 7% equals 53 mmol/mol (El-Haddad *et al*., 2020). The formula 28.7 × HbA1c − 46.7= eAG (estimated average glucose) can convert HbA1c to glucose levels similar to those seen in self-monitoring.

In our study, we applied the formula to ascertain the estimated mean glucose levels (eAGs) of patients and established their association with fasting plasma glucose (FBS) levels. Notably, there was a close match between eAGs and FBSs when both were measured on the same day. The relationship between eAG and FBS showed a significant correlation and very similar averages. Specifically, FBS (Fasting Blood Sugar) in males had a mean of 225±91 mg/dl, while females had 177±43 mg/dl. The eAG (Estimated Average Glucose) in males had a mean of 209.2±80.4 mg/dl, while females had 184.6±49.1

mg/dl. Despite these similarities in mean values, the difference between eAG and FBS was statistically significant (p = 0.001). For the 20 male samples, the eAG levels were slightly lower than the FBS levels, whereas for the 34 female samples, the eAG levels were higher than the FBS levels. These results indicate that eAG and FBS cannot be used interchangeably. The apparent similarity in their mean levels across the entire group seems coincidental. This similarity disappeared when we separated the participants into subgroups based on their FBS levels. Furthermore, the significant decrease in the correlation coefficient suggested that the strength of the association between eAG and FBS depended on the level of glucose control in the patients. As glucose control worsened, the association between eAG and FBS became stronger (Akoglu *et al*., 2018).

Moreover, most clinic patients will likely be more disciplined with fasting and certain dietary restrictions concerning plasma glucose testing. This might partly explain why the eAG levels in the present study group were majorly higher than the FBS levels. At the same time, FBS demonstrates glucose levels in a single set of measurements, usually after fasting. On the other hand, eAG gives an average blood glucose level for the last three months, including the period after food intake. Hence, compared to the previous studies, the slightly high eAG recorded in this study may be due to the consideration of postprandial blood glucose level in the eAG formula (Hempe *et al*., 2010). We also discussed the view that some patients with higher FBS levels may experience stress before coming to the hospital, and their glucose levels temporarily can be elevated as a result of stress. However, these patients might have omitted their use of the anti-diabetic medications before the test, hence the higher FBS than the basal glycemia. This might explain one of the intra-FBS variation where the FBS levels are higher than the eAG levels.

Our study showed a significant difference in age between males and females (p-value= 0.0001). Fasting blood sugar levels between males and females, with males having higher levels of FBS with p-value= 0.709 and not statistically significant. The difference in HbA1c levels between males and females is also not statistically significant, with p-value= 0.732.

Our study shows a strong correlation between eAG and FBS (r=0.629**). A similar study found a stronger correlation between eAG and FBS in poorly controlled people living with diabetes, meaning that above-normal levels of FBS also translate to increased eAG which signifies poorly regulated glycemia among sufferers (Garg *et al*., 2022). Hypothesis testing for the results obtained indicates that the observed probability (p value= 0. 0001) is statistically significant, reducing the likelihood of expressing a correlation between eAG and FBS by chance to a negligible point.

Current data showed that according to the IDF (International Diabetes Federation), around the world, 285 million people suffer from diabetes. If no measures are taken, this number will rise to 438 million in the next twenty years (Satman *et al*., 2002). Thus, the given condition is relatively prevalent in Turkey, where the population is approximately 70 million people; about 9% of the population suffers from diabetes or impaired glucose tolerance; however, few of the population have even a passing awareness of the disease. From the case analysis, it can be suggested that there is a demand for enhanced self-management among diabetic patients. One of them is using estimated average daily glucose (eAG) and glycated haemoglobin figures (Miguel *et al*., 2009).

While the clinical benefits of eAG are still in question, we propose that the determination and provision of eAG values, in addition to patients' HbA1c levels, should be considered standard practice. It may assist the patient in learning how regulating the amount of glucose in the blood is essential and how proper diet and drink could minimize the demand for invasive methods of achieving glucose homeostasis.

Conclusion

According to the presented data, it is possible to state that patients with good to moderate glycaemic control still are not entirely autonomous in glycaemic control, as far as their eAG figures are concerned. Furthermore, it was equally observed that the association of FBS with eAG levels has some tendencies to be affected by glycaemic control. This means the eAG may indicate fluctuation or inconsistency in the FBS management even when the FBS level is within the target range. The higher correlation between FBS and eAG, when the patient's glucose control is poor, illustrates the need for more extensive monitoring to achieve better, steadier, and more precise blood glucose control.

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