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Research Article

ESTIMATION OF HEMOGLOBIN A_1C USING THE COMPLETE BLOOD COUNT MEASURES IN THE DIAGNOSIS OF DIABETES

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ABSTRACT

Objective: Diabetes is a metabolic disorder occurring either due to the inadequate secretion of insulin or ineffective utilization of insulin by the body. The study was aimed to identify the variations of the complete blood count (CBC) parameters among the diabetic and normal individuals and to derive an empirical formula to estimate hemoglobin A_1c (HbA_1c) of an individual using CBC parameters.

Methods: A total of 83 subjects (mean age: 52.8±9.0 years) involved in the study, among which 39 (mean age: 49.1±8.8 years) were normal and 44 (mean age: 56±7.8 years) were diabetic. The blood was drawn from the participants and was subjected to CBC analysis using automated hematology analyzer. The stepwise linear regression model was used to determine the empirical formula to estimate HbA_1c using the CBC parameters. The Student's t-test was performed to identify the group differences.

Results: A negative correlation was observed for Hb (r=−0.35**, p<0.001) and packed cell volume (PCV) (r=−0.23**, p<0.05) against HbA_1c. The CBC parameters Hb, erythrocyte sedimentation rate, PCV, red blood cells count, mean corpuscular volume, and mean corpuscular Hb exhibited a statistically significant difference at the level (p<0.05) between the normal and diabetic groups. The empirically derived formula yielded sensitivity, specificity, positive predictive value, negative predictive value, and accuracy measures of 91%, 49%, 67%, 83%, and 71%, respectively, in diagnosing diabetes based on the estimated HbA_1c.

Conclusion: The empirical formula derived to estimate HbA_1c could be useful in the prediction of diabetes with an appreciable accuracy.

Keywords: Diabetes, Complete blood count, Estimated hemoglobin A1c, Hematology analyzer, Stepwise multivariate linear regression.

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INTRODUCTION

Diabetes is a chronic health disorder that occurs if the pancreas fails to secrete sufficient insulin or the secreted insulin cannot be effectively used by the body. Insulin is a hormone which is responsible for regulating the levels of glucose in the blood. The glucose molecules are responsible for providing adequate energy for cell survival; else if the glucose molecules are not assimilated by the cell, their concentration rises to dangerous levels in the blood. Long-term complications of diabetes can be controlled by continuing medical care and patient self-management education [1]. Morbidity and mortality are the main concern for those with diabetes, and the risk of complications increases with the degree and the duration of the disease. The elevated blood glucose levels can result in serious, life-threatening complications to the human body such as heart failure, stroke, nervous disorder, renal failure, eye blindness, impotency, and bodily infections that can even result in partial or complete amputations. The survey taken by the Center for Disease and Prevention in US says that diabetes is the sixth-leading cause of death [2]. Diabetes is categorized into three main types, namely, Type I, Type II, and gestational diabetes. Type I is those who need everlasting insulin injections for survival and develops either in their childhood or at teenage years. Type II is the one which is widespread among the majority of population and occurs due to obesity, physical inactivity, and inappropriate and inadequate diet. Gestational diabetes happens only during pregnancy and is momentary [3,4]. Worldwide statistics reveal that India and China are with more number of diabetic patients than any other nations in the world [5,6]. It has been estimated that about 62 million Indians are diabetic, which corresponds to about 7.1% of the adult population. The mean age of the onset of diabetes among the Indian population was 42.5 years, and the disease resulted in about 1 million deaths every year [6,7]. Studies by the Indian Heart Association revealed that about 109 million individuals would be diabetic by 2035. The high prevalence of the disease among the India's increasing middle-class population is mainly due to genetic vulnerability and adoption of a high-calorie, low-physical activity lifestyle [8-10].

Blood is a bodily fluid that provides necessary nutrients and oxygen to the cells and excretes the metabolic wastes away from them. Blood is composed of the major constituents, namely, erythrocytes or red blood cells (RBCs), leukocytes or white blood cells (WBCs), and thrombocytes (platelets). The main role of RBC is for transporting oxygen from the lungs to tissues and carbon dioxide from tissues to the lungs. WBC is responsible for providing the necessary immune system to the human body, and platelets are accountable for coagulation of blood [11]. A complete blood count (CBC) is a blood test performed to assess the overall health of an individual and to diagnose various disorders that affect the composition of blood. A CBC test gives the measure of RBC count, packed cell volume (PCV), RBC indices, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), MCH concentration (MCHC), WBC types, and platelet count. Abnormal levels in the cell count and composition in a CBC test may indicate an underlying medical condition that recommends for further assessment [12]. Hemoglobin (Hb) is a protein that contains an iron molecule that makes the blood to appear red in color. It is responsible for the transport of oxygen from the lungs to the other parts of the body. In the case of diabetes, excess glucose in the blood attaches to Hb molecule and elevates the level of HbA_1c [4]. The measure of HbA_1c is clinically regarded as an index of diabetes diagnosis [13]. Elevated levels of HbA_1c can impair endothelium-mediated vasoactive responses that can result in cardiovascular diseases among diabetic individuals [14]. RBC is nucleated cells composed of Hb, which is biconcave disc shaped with a diameter ranging from 6 to 9 µm and thickness in the range of 1.5-2.5 µm. The RBC appears as distributed cells with a dense and darker outer ring with a paler center, which occupies one-third of the diameter. The varying concentrations of Hb in the blood result in the variations in shape, texture, size, and color [15].

Earlier studies on RBC have revealed that the RBC undergoes morphological variations on the incidence of various diseases such as diabetes mellitus, malaria, sickle cell anemia, and hypercholesterolemia [16-18]. There are numerous intracellular mechanisms that are responsible for modifications in RBC membrane lipids and proteins which cause variations in the shape of RBC and its deformability [19-23]. Earlier studies have shown that biochemical and hematological measures exhibited significant differences in the normal with the diabetic subjects [24]. The changes in the blood composition are most common for any disease, and the CBC measures could be used to assess the extent of the disease and the treatment efficacy [25].

Hence, the aim of the study was to identify the variations of the CBC parameters among diabetes and normal individuals and to derive an empirical formula to estimate HbA_1c of individual using CBC parameters.

METHODS

Study subjects

The study was carried out at SRM Hospital and Research Centre in Kattankulathur, SRM University, with the consent of the Institutional Ethical Committee. The participants for the study were those who visited the hospital for their routine diabetes checkup during October 2015 to November 2015. An informed written voluntary consent was obtained from all participants who willingly took part in the study. A total of 83 subjects (mean age: 52.8±9.0 years) involved in the study, among which 42 (mean age: 52.1±9.5 years) were men and 41 (mean age: 53.5±8.5 years) were women.

The exclusion criteria included subjects with known cardiovascular disorders, thyroid, arthritis, and unknown ethnicity based on information furnished by the subjects in the questionnaire.

Biochemical test

The subjects considered for the study were subjected to a complete history taking and clinical examination. Blood samples (5 ml) were collected from every subject using white pore needles and were recommended for the following laboratory tests: Fasting blood glucose (FBG), post-prandial blood sugar (PPBS), HbA_1c, and estimated average glucose (eAG).The FBG was obtained using the glucose oxidase-peroxidase (GOD-POD) technique, and the reference values were set at 70-110 mg/dl. The PPBS was also obtained using the GOD-POD technique, and the reference values were set at 80-140 mg/dl. The HbA_1c measures were obtained using the high-performance liquid chromatography (HPLC) method, and the reference values were set at 4.5-6%. The eAG was estimated from HbA_1c, and the reference values were set at <126 mg/dl [26-29].

CBC parameters

The CBC analysis was performed using an automated analyzer to estimate the RBC parameters, namely, Hb (g/dl), erythrocyte sedimentation rate (ESR) (mm/hr), PCV (%), RBC count (10^{12} cells/L), MCV (FL), MCH (pg/cell), MCHC (g/dl), and platelet count (10^9/L). The various CBC measures are defined as follows:

- Hb: It is the measure of oxygen-carrying protein in the blood
- ESR: It defines the rate of RBC sedimentation in 1 hr
- PCV: It is the percentage of RBC in the circulating blood
- Platelet count: It is the measure of platelets in a defined volume of blood
- RBC count: It is the measure of the actual number of RBC per volume of blood
- MCV: It is the mean volume of the RBC in the given sample of blood
- MCH: It is mean weight of Hb per RBC in the given sample of blood
- MCHC: It is the measure of the average concentration of Hb in the given volume of packed RBC.

Statistical analyses

The data collected from the study were analyzed using the SPSS software package version 17.0 (SPSS Inc., Chicago, IL, USA). The ANOVA test was performed for comparison and to calculate the average values for all parameters considered in the study. The comparison between the normal and the diabetic groups for all the parameters included in the study was carried out by the Student's t-test. The influence of the CBC measures on the levels of HbA_1c was established by carrying out stepwise multivariate linear regression.

Performance assessment

It is essential to assess the performance of a diagnostic test based on sensitivity, specificity, and accuracy measures. Hence, a confusion matrix was formed based on the following considerations.

- True positive (TP): The person who has diabetes is also predicted as diabetic
- False positive (FP): The subject who is healthy but is predicted as diabetic
- True negative (TN): The subject who is healthy is also predicted as healthy
- False negative (FN): The subject who is diabetic but is predicted as healthy
- Sensitivity*:* It corresponds to accuracy measure for the true prediction of diabetes and is defined by the following equation:

$$\text{Sensitivity} = \frac{TP}{TP+FN}\% \quad (1)$$

- Specificity: It corresponds to accuracy measure of diagnosis of healthy subjects and is defined by the following equation:

$$\text{Specificity} = \frac{TN}{FP+TN}\% \quad (2)$$

- Overall accuracy (Efficiency): It is a measure of true findings of the diagnostic test and is defined by the following equation:

$$\text{Accuracy} = \frac{TP+FN}{TP+FP+FN+TN}\% \quad (3)$$

RESULTS

The blood samples collected from the study population were subjected to biochemical assays to quantify PPBS, FBS, HbA_1c, eAG, and the CBC parameters. The stepwise linear multivariate regression was used to empirically derive the formula to estimate HbA_1c using the CBC measures. Among the various CBC measures, it was found that only Hb plays a predominant role in the estimation of HbA_1c and is shown in the following equation:

$$\text{Estimated } HbA_1c=(-0.508\times Hb)+13.906 \quad (4)$$

The Pearson correlation test results of the chronological, CBC parameters, and the derived HbA_1c against the biochemically measured HbA_1c are presented in Table 1. It was observed that there exists a negative correlation for Hb (r=−0.35**, p<0.001) and PCV (r=−0.23**, p<0.05) against HbA_1c, which clearly indicates that elevated blood glucose levels results in reduced Hb.

The Student's t-test was performed to identify the differences that existed between the normal and diabetic groups based on the HbA_1c determined using the biochemical method and the derived empirical formula are presented in Tables 2 and 3, respectively.

Table 2 reveals that there exists a statistically significant difference at the level (p<0.05) for FBS, PPBS, HbA_1c, eAG, HB, ESR, PCV, RBC count, MCV, and MCH between the normal and diabetic groups among the entire population as well as for both the genders. Similar observations were also evident when such comparison was made between the

Table 1: Pearson correlation coefficient (r) of chronological, CBC parameters, and estimated HbA_1c against biochemically measured HbA1c among the total studied population

Characteristics	Total population (Mean±SD)	HbA_1c (%) (r)
Chronological		
Age (years)	52.8±9.0	0.2
Hb (g/dl)	13.4±1.3	−0.35**
ESR (mm/hr)	17.1±11.0	0.35**
PCV (%)	39.1±3.9	−0.23*
CBC parameters		
Platelet count (10^9/L)	236,397.6±73,891.8	−0.03
RBC count (10^{12} cells/L)	4.7±0.5	−0.18
MCV (fl)	83.3±3.8	−0.15
MCH (pg/cell)	28.5±3.8	−0.22
MCHC (g/dl)	34±1.6	0.05
Derived measure		
Estimated HbA_1c (%)	7.1±0.7	0.36**

**Correlation is significant at the 0.01 level (two-tailed), *Correlation is significant at the 0.05 level (two-tailed). SD: Standard deviation, Hb: Hemoglobin, ESR: Erythrocyte sedimentation rate, PCV: Packed cell volume, RBC: Red blood cell, MCV: Mean corpuscular volume, MCH: Mean corpuscular hemoglobin, MCHC: Mean corpuscular hemoglobin concentration, CBC: Complete blood count, HbA_1c: Hemoglobin A_1c

normal and diabetic groups based on the empirically derived HbA_1c (Table 3).

Table 4 presents the percentage changes of the biochemical and CBC measures among the normal and diabetic groups in the studied population. The increase or decreases of the various measures are common for the studied population irrespective of gender. However, the percentage level of changes was not uniform among the genders. It has been found that the biochemical measures of blood such as FBS, PPBS, HbA_1c, eAG, and estimated HbA_1c using the derived empirical formula increased by 55%, 90%, 46%, 63%, and 10%, respectively, among the entire studied population. Similarly, the CBC measures Hb, PCV, platelet count, RBC count, MCV, and MCH decreased by 8.5%, 7.1%, 1.3%, 4.2%, 3.1%, and 5.8%, respectively, among the diabetic subjects. The synthesis of the RBC is greatly affected by the onset of diabetes. Thereby resulting in decreased production of RBC and most likely leads to anemia. This results in the reduced measures of the various CBC measures associated with the RBC. Whereas, ESR and MCHC increased by 63% and 2%, respectively, among the diabetic subjects. The performance characteristics of the empirically derived formula to estimate the HbA_1c are presented in Table 5. It has been found that the sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV), and accuracy measures in the prediction of diabetes based on the estimated HbA_1c are 91%, 49%, 67%, 83%, and 71%, respectively.

DISCUSSION

Diabetes is a disease, which is regarded as a global concern occurring due to elevated blood glucose levels. The condition arises mainly due to either inadequate secretion of insulin by the pancreas or improper consumption of glucose by the human body. Earlier studies have reported that the RBC in such hyperglycemic subjects experiences numerous variations, which could alter their flow properties mainly due to their deformation and aggregation [30].

The biochemical tests which are commonly used for routine assessment of diabetes are FBG, PPBS, HbA_1c, and eAG. However, HbA_1c is the recommended diagnostic test for the clinical assessment of diabetes using a threshold (HbA_1c ≥6.5% [7.7 mmol/L]) recommended by the American Diabetes Association and the European Association for the study of diabetes [13,31]. Moreover, HbA_1c is also preferred than FBG because it requires overnight fasting, and whereas HbA_1c test can also be carried out without any restrictions. This benefits for the subjects to

Table 2: The t-test between the normal and diabetic subjects based on the HbA_1c measured using HPLC method

Characteristic features	Entire population			Male population			Female population		
	Normal (n=39)	Diabetic (n=44)	p value	Normal (n=22)	Diabetic (n=20)	p value	Normal (n=17)	Diabetic (n=24)	p value
Chronological									
Age (years)	49.1±8.8	56±7.8[a]	<0.05	46.6±7.8	58.2±7.2[b]	<0.05	52.5±9.2	54.2±8	NS
Biochemical measures									
FBS (mg/dl)	95.5±17.9	148.5±66.4[a]	<0.05	96.6±22.6	160.3±70.5[b]	<0.05	94±9.3	138.6±40.2[c]	<0.05
PPBS (mg/dl)	125.8±31.3	239.3±92.6[a]	<0.05	130±39.4	261.4±96[b]	<0.05	120.4±15.4	220.9±87.3[c]	<0.05
HbA_1c (%)	5.7±0.4	8.3±1.9[a]	<0.05	5.7±0.4	8.9±2.18[b]	<0.05	5.8±0.3	7.9±1.4[c]	<0.05
eAG (mg/dl)	118.2±10.6	192.9±53.2[a]	<0.05	117.9±11.9	208.2±62.8[b]	<0.05	118.6±8.9	180.2±40.9[c]	<0.05
CBC parameters									
Hb (g/dl)	14±1.08	12.8±1.2[a]	<0.05	14.6±0.9	13.1±1.5[b]	<0.05	13.1±0.6	12.6±0.9[c]	<0.05
ESR (mm/hr)	12.8±8.4	20.9±11.7[a]	<0.05	7.9±5.7	18.9±15.1[b]	<0.05	19.1±7	22.7±7.9	NS
PCV (%)	40.6±4.12	37.7±3.2a	<0.05	42.5±4.5	38.8±3.6[b]	<0.05	38.3±1.9	36.8±2.4[c]	<0.05
Platelet count (10^9/L)	238,076.9±71,052.4	234,909.1±77,109.4	NS	237,500±47,820	232,700±59,972	NS	238,823±94,802	236,750±90,206	NS
RBC count (10^{12} cells/L)	4.8±0.5	4.6±0.4[a]	<0.05	5±0.6	4.7±0.5[b]	<0.05	4.62±0.2	4.47±0.3	NS
MCV (fl)	84.7±4.4	82.1±2.7[a]	<0.05	85.8±5	82.7±3.2[b]	<0.05	83.2±2.9	81.5±2.22[c]	<0.05
MCH (pg/cell)	29.4±2.8	27.7±2.3[a]	<0.05	29.9±3.5	28.1±1.9[b]	<0.05	28.7±1.2	27.4±2.6[c]	<0.05
MCHC (g/dl)	33.6±1.6	34.3±1.5[a]	<0.05	33.4±2	34.5±1.7[b]	NS	33.9±1.1	34.2±1.3	NS
Derived measure									
Estimated HbA_1c (%)	6.7±0.6	7.4±0.6[a]	<0.05	6.38±0.4	7.2±0.8[b]	<0.05	7.2±0.4	7.5±0.47[c]	<0.05

[a]Statistically significant difference at the level (p<0.05) between the normal and diabetic subjects for the entire study population, [b]statistically significant difference at the level (p<0.05) between the normal and diabetic subjects for the male population, [c]statistically significant difference at the level (p<0.05) between the normal and diabetic subjects for the female population. NS: Not significant, FBG: Fasting blood glucose, PPBS: Post-prandial blood sugar, HbA_1c: Hemoglobin A_1c, eAG: Estimated average glucose, Hb: Hemoglobin, ESR: Erythrocyte sedimentation rate, PCV: Packed cell volume, RBC: Red blood cell, MCV: Mean corpuscular volume, MCH: Mean corpuscular hemoglobin, MCHC: Mean corpuscular hemoglobin concentration, CBC: Complete blood count, HPLC: High-performance liquid chromatography

Table 3: The t-test between the normal and diabetic subjects based the estimated HbA_1c

Characteristic features	Entire study population			Entire male population			Entire female population	
	Normal (n=23)	Diabetic (n=60)	p value	Normal (n=21)	Diabetic (n=21)	p value	Normal (n=3)	Diabetic (n=38)
Chronological								
Age (years)	47.17±7.6	54.9±8.6[a]	<0.05	46.9±7.6	57.3±8.3[b]	<0.05	50±7	53.74±8.6
Biochemical measures								
FBS (mg/dl)	103.9±32.8	131.1±53.8[a]	<0.05	105.5±33.9	148.4±72.6[b]	<0.05	94.67±13.3	122.11±38.9
PPBS (mg/dl)	151.7±69.3	199.1±94.7[a]	<0.05	156.2±70.9	228.9±107.9[b]	<0.05	111±15.5	184.61±84.5
HbA_1c (%)	6.1±0.9	7.5±2.01[a]	<0.05	6.2±0.98	8.3±2.5[b]	<0.05	5.87±0.6	7.11±1.6
eAG (mg/dl)	128.3±27.2	169.1±57.8[a]	<0.05	129.7±27.9	192.1±73.0[b]	<0.05	121.7±17.3	157.22±44.5
CBC parameters								
Hb (g/dl)	14.9±0.8	12.8±0.9[a]	<0.05	15±0.8	12.8±1.6[b]	<0.05	14.23±0.3	12.69±0.7
ESR (mm/hr)	7.9±5.1	20.6±10.6[a]	<0.05	7.05±3.6	19.2±14.9[b]	<0.05	15.33±8.5	21.63±7.5
PCV (%)	42.7±4.6	37.7±2.5[a]	<0.05	43.1±4.6	38.3±2.9[b]	<0.05	38.67±3.1	37.29±2.3
Platelet count (10^9/L)	239869±67267	235066±76776	NS	250857±50685	219571±52414	NS	157000±112583.3	243973.68±87705.7
RBC count (10^{12} cells/L)	5.01±0.6	4.6±0.3[a]	<0.05	5.1±0.6	4.61±0.4[b]	< 0.05	4.63±0.3	4.52±0.3
MCV (fl)	85.4±4.9	82.5±3[a]	<0.05	85.5±5.1	83.1±3.6	NS	83.67±2.1	82.13±2.7
MCH (pg/cell)	30±3.3	27.9±2.1[a]	<0.05	30.1±3.45	27.95±1.9[b]	<0.05	29.67±1.5	27.76±2.2
MCHC (g/dl)	33.8±2.2	34.1±1.3	NS	33.8±2.3	34.1±1.4	NS	35.33±1.5	34±1.1
Derived measure								
Estimated HbA1c (%)	6.2±0.3	7.4±0.4[a]	<0.05	6.2±0.3	7.4±0.6b	<0.05	6.4±0.05	7.5±0.4

[a]Statistically significant difference at the level (p<0.05) between the normal and diabetic subjects for the entire study population, [b]statistically significant difference at the level (p<0.05) between the normal and diabetic subjects for the male population. NS: Not significant, FBG: Fasting blood glucose, PPBS: Postprandial blood sugar, HbA_1c: Hemoglobin A_1c, eAG: Estimated average glucose, Hb: Haemoglobin, ESR: Erythrocyte sedimentation rate, PCV: Packed cell volume, RBC: Red blood cell, MCV: Mean corpuscular volume, MCH: Mean corpuscular hemoglobin, MCHC: Mean corpuscular hemoglobin concentration, CBC: Complete blood count

Table 4: Percentage variations of the biochemical measures and CBC parameters among the diabetic subjects on comparison with normal subjects

Characteristic features	Entire population (n=83)	Male (n=42)	Female (n=41)
Biochemical measures			
FBS (mg/dl)	55↑	66↑	47↑
PPBS (mg/dl)	89.9↑	101↑	83↑
HbA_1c (%)	45.6↑	56↑	36↑
eAG (mg/dl)	62.7↑	76.6↑	52↑
CBC parameters			
Hb (g/dl)	8.5↓	10.3↓	3.8↓
ESR (mm/hr)	63.9↑	139.2↑	18.8↑
PCV (%)	7.1↓	8.7↓	3.9↓
Platelet count (10^9/L)	1.3↓	2↓	0.9↓
RBC count (10^{12} cells/L)	4.2↓	6↓	3.2↓
MCV (fl)	3.1↓	3.6↓	2↓
MCH (pg/cell)	5.8↓	6↓	4.5↓
MCHC (g/dl)	2.1↑	3.3↑	0.9↑
Derived measure			
Estimated HbA_1c (%)	10.4↑	12.8↑	4.2↑

↑ - Percentage increase, ↓ - Percentage decrease. FBG: Fasting blood glucose, PPBS: Post-prandial blood sugar, HbA_1c: Hemoglobin A_1c, eAG: Estimated average glucose, Hb: Hemoglobin, ESR: Erythrocyte sedimentation rate, PCV: Packed cell volume, RBC: Red blood cell, MCV: Mean corpuscular volume, MCH: Mean corpuscular hemoglobin, MCHC: Mean corpuscular hemoglobin concentration, CBC: Complete blood count

undergo a HbA_1c test as it avoids the necessity to sacrifice their working hours. The main drawback of the HbA_1c test is that it is costlier due to the increased cost of the assay, reagents, and devices used for performing the test. Earlier studies have revealed that the diabetes influences in the reduced synthesis of RBC cells. CBC test is performed to analyze the composition of the various constituents in the blood. It has been found that diabetes can result in changes of RBC morphology [16-18]. This creates a requirement to explore ways to identify the extent of diabetes among individuals who have been recommended for CBC examinations for diagnosis of various diseases. In an earlier study by Khanam *et al*., it was found that the subjects with chronic renal failure with anemia possess lower levels of Hb, PCV, and RBC count than the normal subjects also the levels of the blood measures decreased greatly with the severity of the disease [32]. When a similar approach was adopted in this study to find the similarities and differences among the CBC measures among the normal and diabetic subjects. The results of this study revealed that the levels of Hb, PCV, platelet count, RBC count, MCV, and MCH are high in the normal subjects than the diabetic subjects for the entire population, and also a statistically significant difference was observed between the two groups at the level ($p<0.05$), except for platelet count. Whereas, ESR and MCHC had lower levels in the normal groups than the diabetic groups with a statistically significant difference at the level ($p<0.05$). When similar approach was performed by applying it to only male and female subjects, a similar trend was observed for both the genders as observed for the entire population. Therefore, the results of the study reveal that CBC parameters demonstrate statistically significant differences exist between the normal and diabetic subjects. The results of our study concur with the similar study on Nigerian population, wherein PCV is lesser among diabetic subjects than the normal subjects [33]. Similar studies were also carried out on Nigerian population to test the effectiveness of malaria treatment using the CBC parameters as the extent of the infection influences in the CBC measures [25].

In the present study, it was found that there exists a significant correlation for Hb ($r=-0.35^{**}$, $p<0.001$), ESR ($r=0.35^{**}$, $p<0.001$), PCV ($r=-0.23^{*}$, $p<0.05$), and estimated HbA_1c ($r=0.36^{**}$, $p<0.001$) against HbA_1c. Similarly, statistically significant correlation was observed for HbA_1c against the morphological features of RBC indicating that changes in RBC morphology are inevitable among diabetic cells [4].

Table 5: Performance characteristics of the estimated HbA_1c in identifying subjects with diabetes

Performance characteristics	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)	Accuracy (%)
Estimated HbA_1c	91 (CI: 78.3-97.5)	49 (CI: 32.4-65.2)	67 (CI: 59.2-73.4)	83 (CI: 63.9-92.7)	71

CI: Confidence interval ; PPV: Positive predictive value, NPV: Negative predictive value, HbA_1c: Hemoglobin A_1c

The RBC of a diabetic cell undergoes variations in its shape and form due to changes in blood glucose levels. Therefore, the features that correspond to these variations of RBC could be used to diagnosis diabetes. We adopted a method to estimate HbA_{1c} using the CBC parameters, and the performance measures were found to be with sensitivity, specificity, PPV, NPV, and accuracy measures of 91%, 49%, 67%, 83%, and 71%, respectively, in diagnosing diabetes based on the empirically derived HbA_1c. The major limitations of the study are that it involves a relatively small group of the study population and its confinement to a single ethnic group, which needs to be validated over a larger sample size and different ethnicities.

CONCLUSION

We found that there exists a connection between the various CBC measures that corresponds to RBC features of a normal and diabetic cell. The empirical formula derived to estimate HbA_1c using CBC parameters could be useful in the prediction of diabetes with an appreciable sensitivity and a limited specificity.

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