

Original Research Article

Relationship between fasting blood sugar and some haematological parameters in diabetic patients attending Nigerian Navy Reference Hospital Calabar, Cross River State, Nigeria

Comment [PE1]: Non-diabetics were also included in the study too.

ABSTRACT

Background of study: A survey in Cross River State (CRS) gave a prevalence of Diabetes Mellitus (DM) at 6.5% in the age group 20 and above. This translates to an estimated 97,500 Diabetics within the population of 3 million.

Aim of study: This study was undertaken to evaluate fasting blood sugar and some haematological parameters in patients with diabetes mellitus visiting Nigeria Navy Reference Hospital Calabar, Cross River State, Nigeria.

Materials and Methods: A total of hundred (100) subjects (males and females) comprising of both diabetic and non-diabetic (controls) subjects who were within the ranges of 20-55 years of age were enrolled in this study.

Results: It showed that diabetes patients exhibited significant ($p < 0.05$) decrease in the following parameters compared with the control group: Lymphocyte, Eosinophil and Platelets count. The TWBC, Neutrophil, Monocyte, Basophil and FBS did not vary between the diabetes patients and the control group and only platelet established significant relationship in terms of age compared with other parameters. Based on the findings of the study, it can be concluded that there is a statistically significant increases ($P < 0.05$) in platelet in the males compared with the females. Also, diabetic patients showed no significant in PCV and Hb value than the control subject.

Conclusion: It is important that haematological profile should be included as a routine screening investigation to diagnose diabetic patient and treat accordingly. Full blood count as one of the routine laboratory tests is required in the management of diabetic patients.

Keyword: Diabetes Mellitus, fasting blood sugar, haemoglobin, white blood cell, Platelet

Introduction

Diabetes mellitus is a clinical syndrome characterized by hyperglycemia due to absolute or relative deficiency of insulin. Lack of insulin, whether absolute or relative, affects the metabolism of carbohydrate, protein, fat, water and electrolytes (Sacks, 2013).

As stated by Vcelakova, Blatny, Halbhuber, Kolar and Neuwirth, (2013) there are 3 types of diabetes namely: Type 1, Type 2 and Gestational diabetes. However, they further maintained that Type 1 diabetes (T1D) is a heterogeneous disorder associated with the destruction of pancreatic beta cells, with the resultant effect of absolute insulin deficiency. Type 2 diabetes on the other hand is characterized by resistance to insulin action and suboptimal insulin secretory response, while gestational diabetes is when a woman without diabetes, develops high blood sugar levels during pregnancy. The body uses glucose for energy. But too much glucose in the

blood is not good for pregnant women or the baby (Vcelakov *et al.*, 2013). Furthermore, Boitard, 2012) argued that the causes of diabetes range from autoimmune mediated destruction of beta cells and idiopathic destruction or failure of beta cells.

Wang, Lin, Liu, Cheung and Lai, (2014) conducted epidemiological study and noted relationship between some components of metabolic syndrome and leukocytes. They concluded that reduced haemoglobin concentrations are common findings in diabetic patients. In addition, it has been observed that peripheral blood leukocytes produce polymorphonuclear cells, including monocytes as well as lymphocytes. Some previous studies showed that peripheral white blood cell (WBC) count might be associated with type-2 diabetes, coronary artery disease (CAD), stroke, micro and macro vascular complications (Oshita, Yamane, Hanafusa, Mori, Mito, Okubo, Hara and Kohno, (2014). Increased differential cell counts, including counts of eosinophils, neutrophils, and monocytes, also indicate the future incidence of coronary artery disease (CAD) (Madjid, 2013). Fu-mei Chung, Jack, Tsai, Dao-Ming Chang, Shyi-Jang Shin and Yau-Jiunn Lee (2005), showed that the white blood cells (WBC) might play a role in the development and progression of diabetic complications. However, there is no investigation concerning the differential leukocyte count in relation to diabetic nephropathy.

Moreover, altered level of many haematological parameters such as red blood cells (RBCs), white blood cells (WBC), and the platelet function has been observed in patients with diabetes (Almamory, 2014). According to Almamory, (2014) many studies have advocated the importance of raised level of WBC and RBC count in the diagnosis of metabolic syndrome. Many epidemiologic studies have also suggested a close relationship between haematological parameters and different components of metabolic syndrome. Jabeen, (2013) added that even a number of studies have supported the association between hematologic parameters with insulin resistance. According to Ezenwaka, (2014) abnormal hematological parameters are observed in patients with chronic renal failure and among them anemia is the most common abnormality seen in patients with diabetes mellitus. Ezenwaka further maintained that chronic renal failure is associated with variety of haematological abnormalities. Anemia is the most common, consistent and severe of the various haematological abnormalities

Furthermore, anaemia is relatively common in patients with diabetes mellitus, and low haemoglobin concentration contributes to many clinical aspects of diabetes mellitus or its progression. Low haemoglobin concentration in patients with diabetes mellitus is associated with a more rapid decline in glomerular filtration rate than that of other kidney diseases (Rossing and Coulson, 2004). Therefore, total peripheral white blood cells (WBC) count, a nonspecific marker of inflammation, has also been suggested to be associated with diabetes risk (Nakanishi and Piette, 2003).

Materials and Methods

Comment [PE2]: More information on the sampling technique, sampling frame, sampling procedures (what method of randomization was used), did you use research assistants? who collected the blood samples? Could you also add the ethical approval number here?

The study area was Nigerian Navy hospital Calabar Cross River State where they had their diabetic clinic days on Thursdays 8am-1pm.

A total of hundred (100) subjects (males and females) comprising of both diabetic and non-diabetic (controls) subjects was enrolled in this study. A random sampling method was used in the selection of both diabetic and controls who were within the ranges of 20-55 years of age in the Nigerian Navy hospital Calabar. Ethical approval was gotten from Ministry of Health, Cross River State and approval to use establishment was gotten from the Commanding Officer Nigerian Navy Hospital Calabar Cross River State before commencement of the project.

The inclusion criteria were as follows: diabetic and non-diabetic subjects of both sexes and subjects who were willing to participate in the study and gave their consent, while exclusion criteria was subjects with denial of consent were not forced to join in the study, since participation was voluntary.

Two milliliters (2mls) of blood was aseptically collected from each diabetic and non-diabetic subjects using a venepuncture and dispersed into Ethylene Diamine Tetra-acetic Acid (EDTA) container in the concentration of 2mg/ml of blood. 4mls of blood was aseptically collected from each control using a venepuncture and 2mls was dispersed into an Ethylene Diamine Tetra-acetic Acid (EDTA) container for the hematological analysis and the remaining 2mls was dispersed into a fluoride oxalate container for fasting blood sugar. All samples were put in a flask containing ice blocks and transported to the laboratory within 30minutes for immediate analysis.

Full Automatic Blood Cell Counter for Full Blood Count (Full automatic blood cell counter: Model= SYSMEX KX-21N (SYSMEX CORPORATION JAPAN, S/No B4 577)

Principle: The principle states that particles pulled through an orifice concurrent with an electric current, produces a change in impedance that is proportional to the volume of the particles transversing the orifice. This pulse in impedance originates from the displacement of electrolyte caused by the particle. The coulter principle was named for its inventor Wallace H. Coulter.

Glucose Oxalate Method for Fasting Blood Sugar

Principle: In this enzymatic method glucose is converted to glucose-6-phosphate (G-6-P) by hexokinase in the presence of ATP, a phosphate donor. Glucose-6-phosphate dehydrogenase then converts the G-6-P to gluconate-6-P in the presence of NADP⁺. As the NADP⁺ is reduced to NADPH during this reaction, the resulting increase in absorbance at 340 nm (secondary wavelength = 700 nm) is measured. This is an endpoint reaction that is specific for glucose.

Comment [PE3]: How do you come about the 100 subjects to be selected for this study? What is the population that visits the diabetic clinic on its days?

Comment [PE4]: Are the control healthy subjects? Non-diabetics are not the same as healthy subjects. A definition of the control group should be given.

Comment [PE5]: How were the non-diabetics selected in this study?

Statistical Data

Data was presented in tables and graphs as \pm standard deviation (SD). The statistical package for social science (SPSS) 23 was used in the analysis of the results. Comparison was made between two and three groups using Student t-test and ANOVA respectively. Pearson correlation was also used to determine relationships. $P \leq 0.05$ were considered statistically significant.

Results

Mean of Control subjects and Diabetic subjects

Table 1 shows the result of control subject and diabetes subject for haematological parameters, there was a significant mean age difference between diabetic patients compared to non-diabetic controls [$P < 0.05$]. Hence there was no significant difference on diabetic haematological parameters on packed cell volume (PCV) and haemoglobin (Hb) examined, while other haematological parameters such as platelet and fast blood sugar were significant.

Some haematological parameters of control subject and Diabetes subject based on gender

Table 2 shows some haematological parameters of control subjects and Diabetic subjects based on gender. It shows that diabetes patients exhibited significant ($p < 0.05$) decrease in the following parameters compared with the control group: Lymphocyte, Eosinophil and Platelets count. Diabetic patients showed no significant in PCV and Hb value than the control subject. While the TWBC, Neutrophil, Monocyte, Basophil and FBS did not vary between the diabetic patients and the control group. Table 2 also shows a statistically significant increases ($P < 0.05$) in platelet in the males compared with the females (250 ± 8.29 vs 221 ± 16.24).

Shows some haematological parameters of control subject and Diabetes subject based on age

Table 3a shows significant relationship ($p < 0.05$) between the diabetes patients and the control group in age range 23-32 and compared to the age range 33-42 and 43-52. Table 3b revealed that only platelet established significant relationship in terms of age compared with other parameters.

Correlation graph of FBS verses PCV, Hb, TWBC, Platelet count of diabetes subjects

Correlation graph between FBS and PCV of control subject showed significant when $p < 0.05$ and strong correlation when $r = 0.159$. There also was a correlation between FBS and Hb estimation of control when $r = 0.136$ and significant at $p < 0.05$. It was shown that there was no significant correlation between FBS and TWBC of control subject with $p > 0.05$ and 0.072 . Correlation between FBS and Platelet count of control subject and diabetes subject showed

Comment [PE6]: What was the rationale for the choice of haematological parameters?

Comment [PE7]: Modify your sub-headings

Comment [PE8]: How did you come about these age categories?

significant while correlation between FBS and PCV of diabetes subject was not significant when $r=0.046$ and $p>0.05$. Correlation between FBS and Hb estimation of diabetes subject was not significant. It was as well shown there was significant correlation between FBS and TWBC of diabetes subject.

Table 1: Mean \pm SEM of control subject and Diabetes subject

Parameters	Control (n = 50)	Diabetes (n = 50)	P value	Remark
Age	31.3 \pm 1.17	36.9 \pm 1.14	P<0.05	S
PCV (%)	39.6 \pm 0.49	39.2 \pm 0.43	P>0.05	NS
Hb (g/L)	13.2 \pm 0.19	12.9 \pm 0.15	P>0.05	NS
TWBC (X10 ⁹)	4.4 \pm 0.15	6.2 \pm 0.50	P<0.05	S
Neutrophil (%)	58.6 \pm 1.24	55.4 \pm 1.62	P>0.05	NS
Lymphocyte (%)	37.7 \pm 1.42	39.6 \pm 1.59	P>0.05	NS
Eosinophil (%)	2.4 \pm 0.18	2.3 \pm 0.18	P>0.05	NS
Monocyte (%)	2.1 \pm 0.11	2.8 \pm 0.16	P>0.05	NS
Basophil (%)	0.08 \pm 0.04	0.00 \pm 0.00	P<0.05	S
Platelet (X10 ⁹)	268 \pm 5.72	236 \pm 9.05	P<0.05	S
FBS (mmol/L)	4.6 \pm 0.06	12.2 \pm 0.32	P<0.05	S

Table 2a: Shows some haematological parameters of control subject and Diabetes subject based on gender

Parameters	FEMALE			MALE		
	Control (n = 19)	Diabetes (n = 24)	P Value	Control (n = 31)	Diabetes (n = 26)	P value
PCV (%)	39.1±1.03	38.9±0.69	P>0.05	39.9±0.49	39.7±0.54	P>0.05
Hb (g/L)	12.9±0.39	12.7±0.26	P>0.05	13.3±0.18	13.0±0.18	P>0.05
TWBC (X10 ⁹)	4.6±0.22	6.8±0.92	P<0.05	4.3±0.19	5.8±0.47	P<0.05
Neutrophil (%)	58.7±2.19	55.8±2.67	P>0.05	58.5±1.52	55.0±1.96	P>0.05
Lymphocyte (%)	36.7±2.29	39.0±2.67	P>0.05	38.3±1.83	40.1±1.88	P>0.05
Eosinophil (%)	2.4±0.33	2.5±0.34	P>0.05	2.4±0.21	2.2±0.18	P>0.05
Monocyte (%)	2.2±0.16	2.9±0.22	P>0.05	2.0±0.15	2.7±0.22	P>0.05
Basophil (%)	0.05±0.05	0.00±0.00	P<0.05	0.10±0.05	0.00±0.00	P<0.05
Platelet (X10 ⁹)	280±11.2	221±16.2	P<0.05	260±5.89	250±8.29	P>0.05
FBS (mmol/L)	4.6±0.10	11.7±0.42	P<0.05	4.5±0.84	12.7±0.48	P<0.05

Table 2b: Shows some haematological parameters of control subject and Diabetes subject based on gender

Parameters	CONTROL			DIABETES		
	Female (n = 19)	Male (n = 31)	P Value	Female (n = 14)	Male (n = 26)	P Value
PCV (%)	39.1±1.03	39.9±0.49	P<0.05	38.8±0.69	39.7±0.54	P>0.05
Hb (g/L)	12.9±0.39	13.3±0.19	P<0.05	12.7±0.26	13.0±0.18	P>0.05
TWBC (X10 ⁹)	4.6±0.22	4.3±0.19	P>0.05	6.8±0.91	5.8±0.47	P>0.05
Neutrophil (%)	58.7±2.19	58.5±1.52	P>0.05	55.8±2.67	55.0±1.96	P>0.05
Lymphocyte (%)	36.7±2.29	38.1±1.83	P>0.05	39.0±2.67	40.1±1.88	P<0.05
Eosinophil (%)	2.4±0.33	2.4±0.21	P>0.05	2.5±0.33	2.2±0.18	P<0.05
Monocyte (%)	2.2±0.16	2.0±0.15	P>0.05	2.9±0.22	2.7±0.22	P>0.05
Basophil (%)	0.1±0.05	0.1±0.05	P>0.05	0.00±0.00	0.00±0.00	P>0.05
Platelet (X10 ⁹)	280±11.24	260±5.89	P>0.05	221±16.24	250±8.29	P<0.05
FBS (mmol/L)	4.6±0.10	4.5±0.08	P>0.05	11.7±0.42	12.7±0.48	P>0.05

Table 3a: Mean±SEM age range of control subject and Diabetes subject

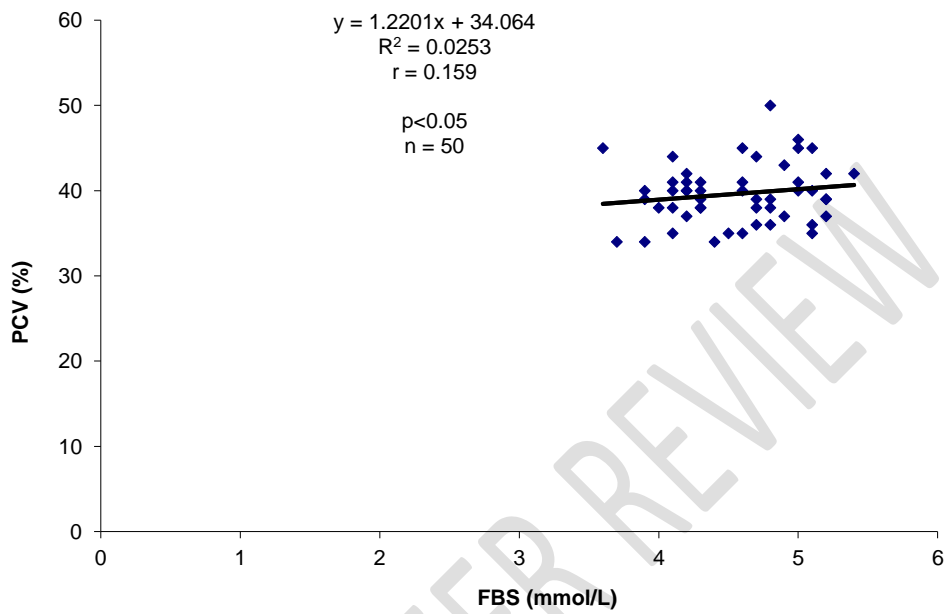
Age Range	Control	Diabetes	P Value
23 – 32 (n = 36, 17)	26.6±0.39	28.1±0.82	P<0.05
33 – 42 (n = 7, 19)	39.4±0.99	37.6±0.69	p>0.05
43 – 52 (n = 7, 14)	47.1±1.34	46.9±0.79	p>0.05

Comment [PE9]: Not sure what this means?

Table 3b: Shows some haematological parameters of control subject and Diabetes subject based on age

Parameters/Age Range	23 – 32 (n = 17)	33 – 42 (n = 19)	43 – 52 (n = 14)	P Value
PCV (%)	38.4±0.92	39.5±0.59	39.9±0.74	p>0.05
Hb (g/L)	12.5±0.33	12.9±0.19	13.1±0.27	p>0.05
TWBC (X10 ⁹)	7.2±1.29	5.2±0.32	6.5±0.74	p>0.05
Neutrophil (%)	54.4±3.62	56.2±1.59	55.6±3.29	p>0.05
Lymphocyte (%)	40.4±3.61	38.7±1.45	39.9±3.28	p>0.05
Eosinophil (%)	2.4±0.42	2.4±0.27	2.1±0.23	p>0.05
Monocyte (%)	3.2±0.27	2.7±0.29	2.4±0.17	p>0.05
Basophil (%)	0.1±0.01	0.0±0.00	0.0±0.00	p>0.05
Platelet (X10 ⁹)	197±18.73	262±10.53*	249±12.05*	P<0.05
FBS (mmol/L)	11.7±0.56	12.1±0.54	13.0±0.58	p>0.05

*significant from age range 23 - 32



Correlation graph between FBS and PCV of control subject

Fig 1: Correlation graph between FBS and PCV of control subject

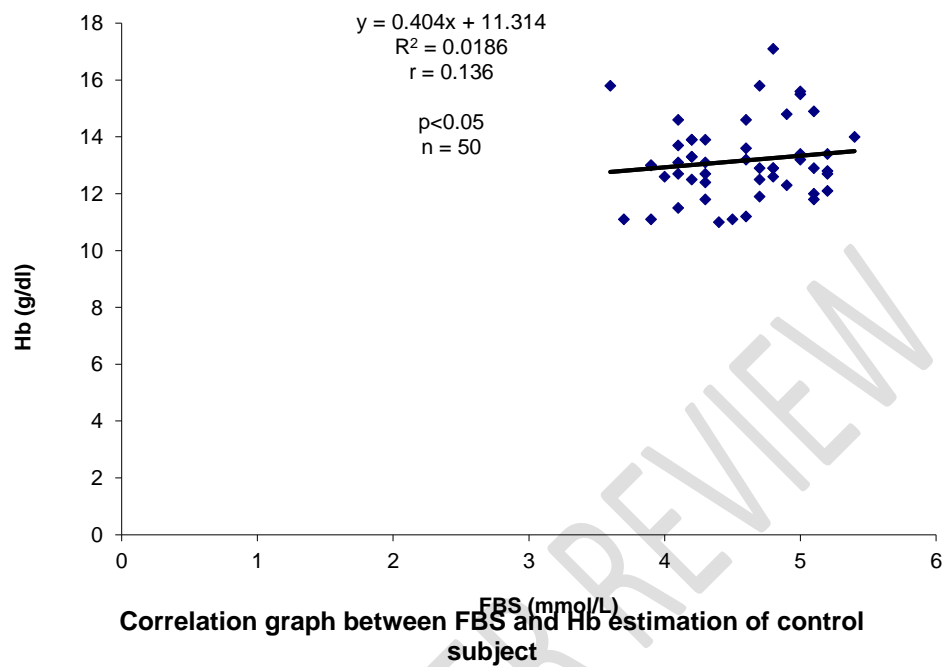
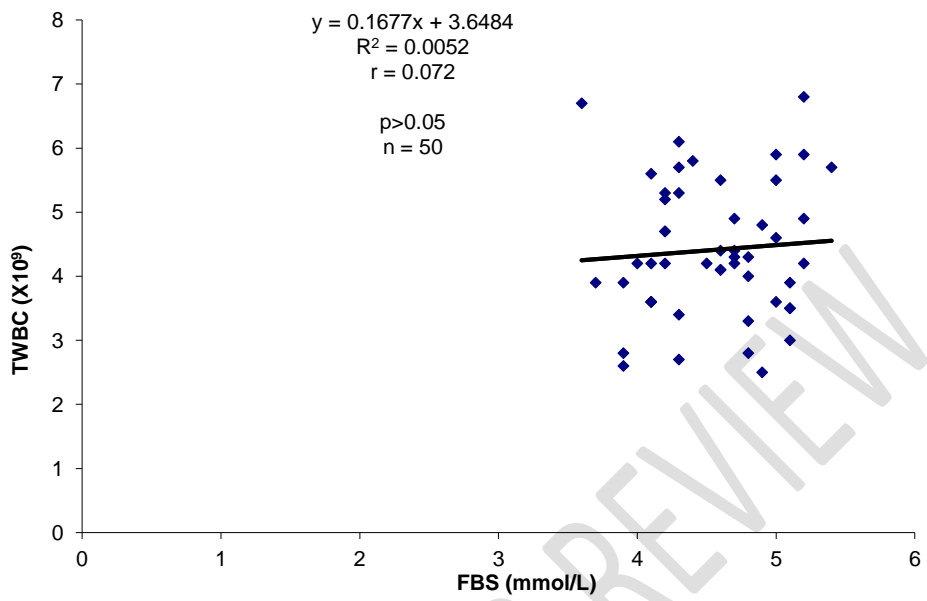


Fig 2: Correlation graph between FBS and Hb estimation of control subject



Correlation graph between FBS and TWBC of control subject

Fig 3: Correlation graph between FBS and TWBC of control subject

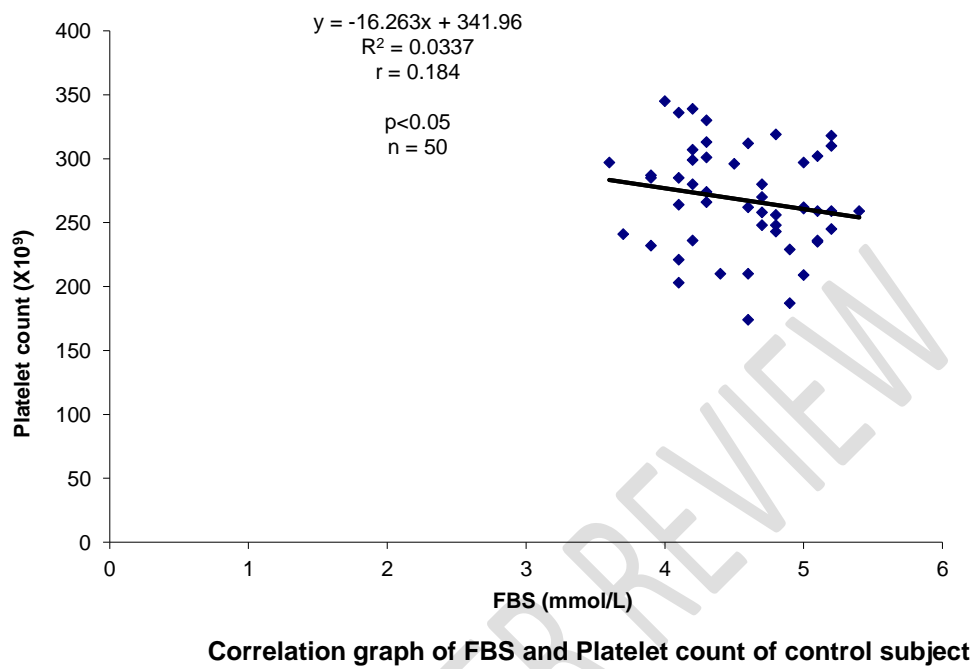
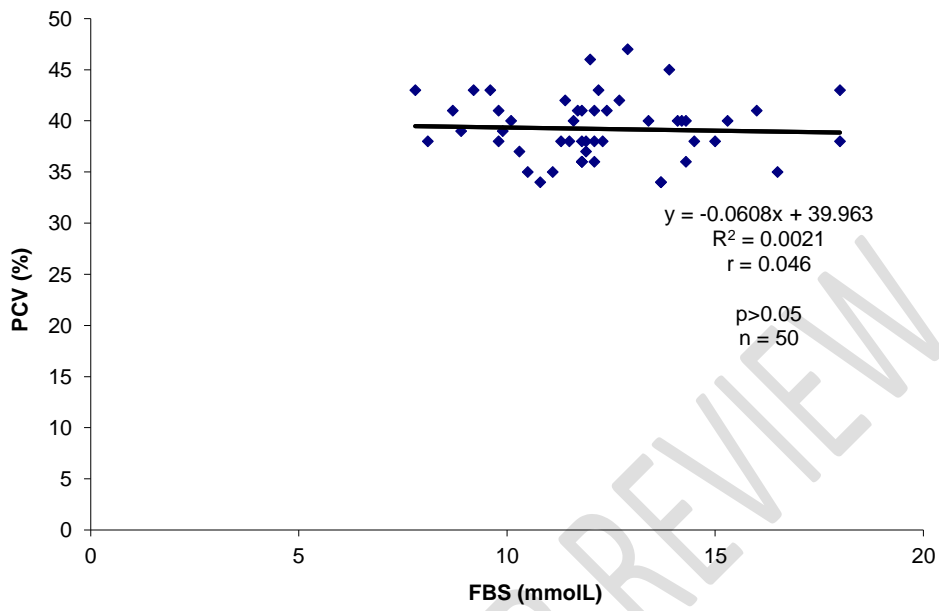


Fig 4: Correlation graph of FBS and Platelet count of control subject



Correlation graph between FBS and PCV of Diabetes Subject

Fig 5: Correlation graph between FBS and PCV of Diabetes Subject

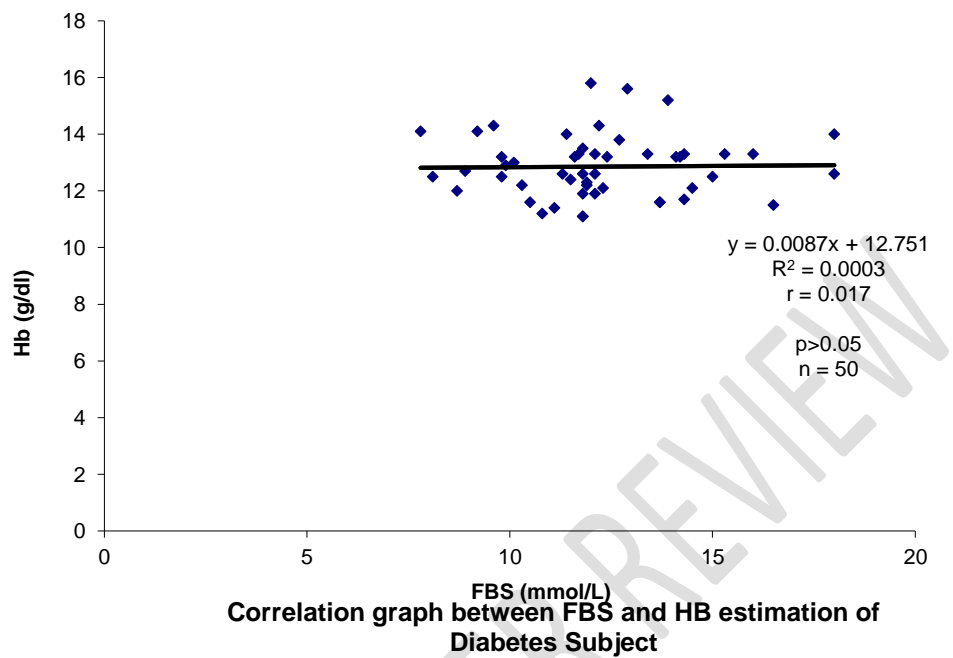


Fig 6: Correlation graph between FBS and HB estimation of Diabetes Subject

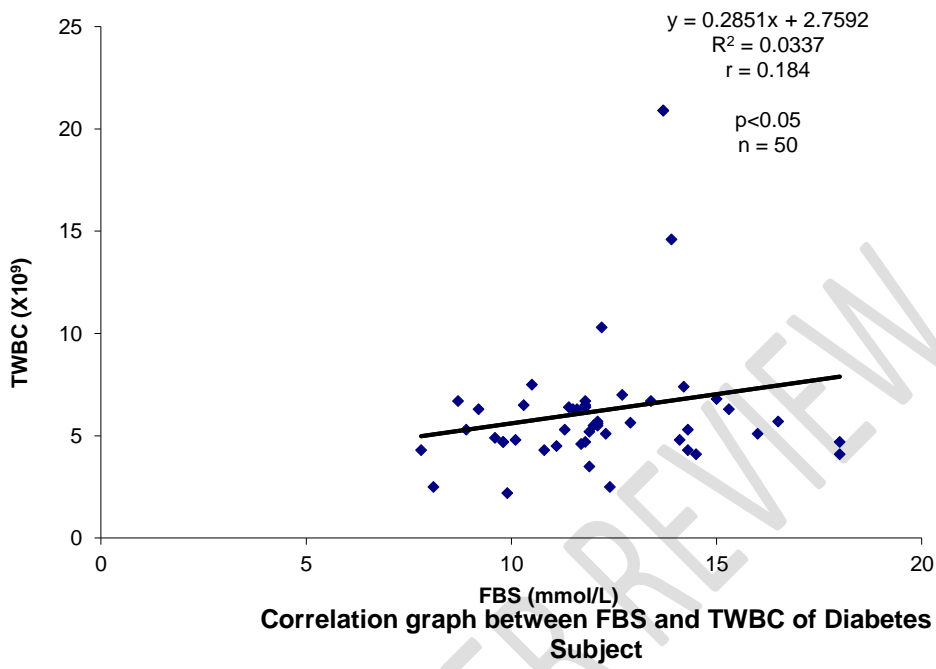


Fig 7: Correlation graph between FBS and TWBC of Diabetes Subject

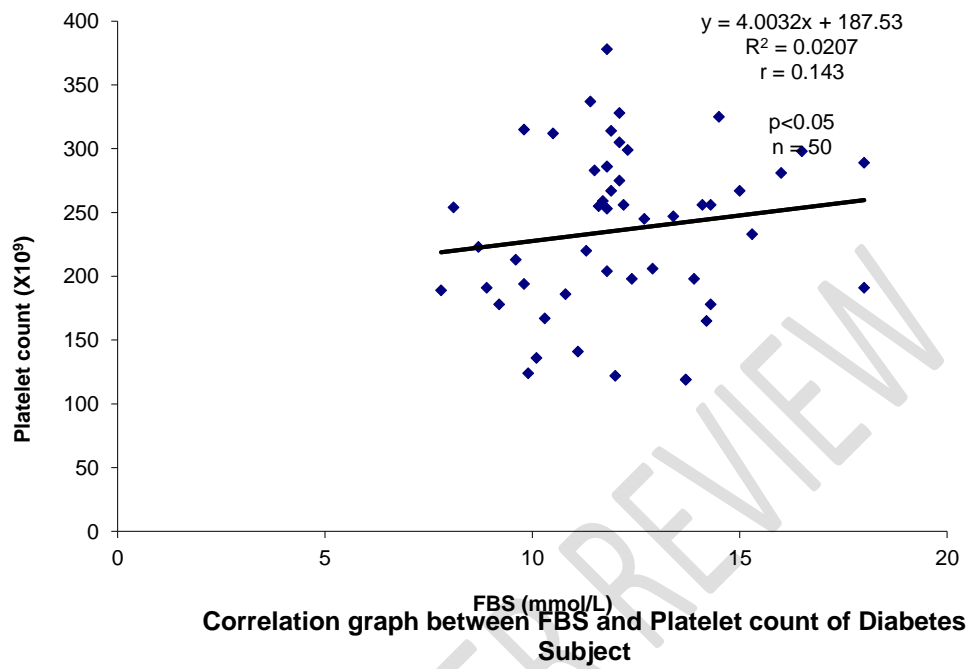


Fig 8: Correlation graph between FBS and Platelet count of Diabetes Subject

Discussion

Table 1 shows the result of control subject and diabetes subject for haematological parameters, there was a significant mean age difference between diabetic patients compared to non-diabetic controls [$P < 0.05$]. Biadgo, (2016) also performed a similar comparative study on 296 participants and reported significant difference in red blood cell distribution width between patients with diabetes and non-diabetes. Almost similar results were depicted by the present study. Biadgo, also reported that platelet indices such as mean platelet volume and platelet distribution width were significantly higher in diabetes patients, but in present study we found the contrary results (Biadgo, 2016). Hence there was no significant difference on diabetic haematological parameters on packed cell volume (PCV) and haemoglobin (Hb) examined, while other haematological parameters such as platelet and fast blood sugar were significant. This finding disagreed with Mbata, Adegoke and Nwagu, (2015) who on one study examined the effect of chronic diabetes on haematological parameters. The tests that were carried out include the determination of Total Red Cell Count, Haemoglobin estimation (Hb), Packed Cell Volume (PCV), Total White Cell Count, Differential leucocytes count and Platelet Count. It was observed in their study that there was a significant difference ($P < 0.005$) on the values of packed cell volume (PCV) of diabetic subjects both male and female (31.36 ± 2.96 and 30.25 ± 3.56). Other haematological parameters that were slightly significantly affected include Red blood cell (RBC) count of diabetic female subject.

Table 2 shows some haematological parameters of control subjects and Diabetic subjects based on gender. It shows that diabetes patients exhibited significant ($p < 0.05$) increase in the following parameters compared with the control group: Lymphocyte, Eosinophil and Platelets count. Diabetes patients showed no significant in PCV and Hb value than the control subject. While the TWBC, Neutrophil, Monocyte, Basophil and FBS did not vary between the diabetes patients and the control group. Table 2 also shows a statistically significant increases ($P < 0.05$) in platelet in the males compared with the females (250 ± 8.29 vs 221 ± 16.24). Statistical analysis showed that the mean of total white blood cells and basophil were significant ($P < 0.05$ and $p < 0.05$) respectively in the control group and diabetes patients whereas the mean cell hemoglobin ($p > 0.05$) and mean cell hemoglobin concentration ($p > 0.05$) were not significant the in the both group, but there was no significant variation between patient and control group in platelets and lymphocytes count respectively and this result corroborates Ferreiro and Angiolillo, (2010).

Table 3a shows significant relationship ($p < 0.05$) between the diabetes patients and the control group in age range 23-32 and compared to the age range 33-42 and 43-52. This finding is in with a study of Bangladeshi with adult population there was significant association between control group and diabetes patients after adjustment of data for age and sex (Biadgo, 2016). Table 3b revealed that only platelet established significant relationship in terms of age compared with other parameters. It has been established that raised platelet counts are frequently observed in diabetics with a long duration of disease while it was also shown that the presence of higher

hemoglobin level in non-diabetic subjects compare to that of diabetic subjects can be attributed as a constitutional feature. Correlation graph between FBS and PCV of control subject showed significance when $p < 0.05$ and strong correlation when $r = 0.159$. There also was a correlation between FBS and Hb estimation of control when $r = 0.136$ and significant at $p < 0.05$. It was shown that there was no significant correlation between FBS and TWBC of control subject with $p > 0.05$ and 0.072 . Correlation between FBS and Platelet count of control subject and diabetes subject showed significance while correlation between FBS and PCV of diabetes subject was not significant when $r = 0.046$ and $p > 0.05$. Correlation between FBS and Hb estimation of diabetes subject was not significant. It was as well shown there was significant correlation between FBS and TWBC of diabetes subject. This study is not in line with Karthikeyan, (2009) who after a study reported that there was significant difference in red blood cell distribution width (47.3 ± 2.6 fL vs 45.2 ± 3 fL) between diabetic patients and controls. Total white blood cells in $10^3/\mu\text{L}$ (6.59 ± 1.42 vs 5.56 ± 1.38), absolute lymphocyte count in $10^3/\mu\text{L}$ (2.60 ± 0.70 vs 2.04 ± 0.63), and absolute neutrophil count in $10^3/\mu\text{L}$ (3.57 ± 1.46 vs 3.11 ± 1.04) increased significantly in diabetic patients compared with controls, respectively. Among platelet indices, mean platelet volume (10.4 ± 1.1 fL vs 9.9 ± 1.1 fL) and platelet distribution width (14.5 ± 2.1 fL vs 13.4 ± 2.1 fL) were found to be significantly increased in the diabetic patients ($P < 0.05$). Anthropometric measurements significantly correlated with white blood cell and platelet indices (Karthikeyan, 2009).

Conclusion

Based on the findings of the study, it can be concluded that males are more affected with diabetes than females. Also, findings of this study uphold that diabetes patients showed no significance in PCV and Hb value than the control subject. There was a statistically significant increases ($P < 0.05$) in platelet in the males compared with the females (250 ± 8.29 vs 221 ± 16.24). The result of this study revealed that there was no significant correlation between FBS and TWBC of control subject with $p > 0.05$ and 0.072 while there was significant correlation between FBS and TWBC of diabetes subject.

Based on the findings in this study, the following recommendations were made:

1. Full blood count as one of the routine laboratory tests is required in the management of diabetic patients.
2. There should be development and implementation of diabetes programme
3. Hematological profile should be included as a routine screening investigation to diagnose diabetic patient and treat accordingly.
4. There should be attention given to therapeutic prevention in diabetes which can retard the progress of diabetic nephropathy and its complications.
5. Treatment of diabetes should focus on maintaining optimal hematocrit to reduce cardiovascular risk.

COMPETING INTERESTS DISCLAIMER:

Authors have declared that no competing interests exist. The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

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