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ORIGINAL ARTICLE

Influence of ABO Blood Group on Fibrinogen Levels and Platelet Count in Apparently Healthy Nigerian Subjects

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Abstract

Background: A close association has been shown to exist between ABO blood type and the risk of some diseases with non-O (A, B, or AB) individuals having an increased differential disposition to thrombotic disorders.

Objective: The present study evaluated the fibrinogen levels and platelet count in subjects of different ABO blood groups with the view of ascertaining if disparity exists in the levels of fibrinogen and platelet count amongst the different ABO blood groups.

Methods: A total of hundred (100) apparently healthy young adult male and female participants comprised of 25 subjects of blood group A, B, AB and O respectively were recruited for this study. Five milliliters of blood was collected and used for the estimation of fibrinogen levels, platelet count and ABO blood grouping using standard laboratory methods. Statistical package for social science (SPSS) version 20 was employed in the analysis of the data.

Results: Blood group O individuals has a significantly lower platelet count compared to those of blood group A (157.56 ± 23.21 Vs 188.92 ± 46.94; P = 0.041). However, the mean plasma fibrinogen level did not differ significantly among the different ABO blood groups (P > 0.05) respectively. Furthermore, there were no significant gender differences in the mean plasma fibrinogen level and platelet count of participants of various ABO blood groups (P > 0.05). In blood group A individuals, there was a significant positive correlation between Fibrinogen level and platelet count (r = 0.385; p = 0.048). However, there were no significant correlations observed between these parameters in the other ABO blood groups (P > 0.05).

Conclusion: Blood group O individuals may be more predisposed to thrombocytopenia than blood group A individuals.

Keywords

Platelet count, Fibrinogen, ABO blood group, Thrombocytopenia

Introduction

In 1901, Landsteiner identified ABO blood groups as the first recognized human blood group system. The clinical significance of ABO blood type extends beyond transfusion medicine and solid organ/hematopoietic transplantation. The ABO blood group is determined by the presence of A and B antigens on the surface of the red blood cells (RBCs). In addition to RBCs, these antigens are widely expressed on the membranes of a wide variety of cells, including platelets, vascular endothelium and epithelium, as well as in saliva and body fluids [1]. The ABH blood group antigens consist of terminal carbohydrate molecules which are synthesized by the sequential action of the ABO glycosyltransferases [2]. The frequency of the ABO blood groups varies among different populations in the world with the highest frequency of the A phenotype found mainly in Northern and Central Europe, B phenotype in Central Asia while Blood group O is the most frequent phenotype globally, with parts of Africa and Australia showing highest frequencies [3].

Numerous reports have suggested important associations between ABO blood groups and various diseases, for example, gastric cancer, periodontal diseases and cardio metabolic diseases [1,2,4]. A close relationship exists between ABO blood type and the risk of venous thromboembolism [5,6]. In both arterial and venous thromboembolism, non-O (A, B, or AB) individuals show significantly increased risk [7,8], whereas group O individuals have more pronounced inherited bleeding tendency [9].

The ABO blood group is known to influence haemostasis because it is a major determinant of the von

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Willebrand factor (vWF) and FVIII plasma levels [10]. Liu, et al. [11] and Wang, et al. [12] found that plasma vWF levels and FVIII activity were significantly increased in individuals with non-O type blood compared with those with type O blood groups. Also a study by Okeke, et al. [13], showed that individuals with blood group O showed a significantly higher APTT value compared to blood groups A, B, and AB while blood group A individuals showed a significantly higher PT value compared to blood groups O, B, and AB.

Platelet count and fibrinogen are parameters that play major roles in haemostasis. Platelet is the central mediator of primary haemostasis that involves platelet adhesion, release of granules and aggregation resulting in the formation of a platelet plug. They are small, irregularly-shaped cells, 2-4 µm in diameter, which are derived from fragmentation of precursor megakaryocytes. The average lifespan of a platelet is between 8 and 12 days. Platelets play a fundamental role in hemostasis and are a natural source of growth factors [14]. According to Zhong, et al. [15] ABH antigens are expressed on both platelet membrane lipids and platelet glycoproteins, and these may influence platelet interactions with fibrinogen, thereby modulating platelet aggregation and platelet-driven thrombosis. Fibrinogen is an essential component of the blood coagulation system, being the precursor of fibrin. It is one of the coagulation factors and plays a vital role in secondary haemostasis. It is converted to fibrin by the action of thrombin leading to the formation of fibrin clot which is stabilized by factor XIII. It is a soluble glycoprotein found in the plasma, with a molecular weight of 340 kDa [16]. Fibrinogen plays a vital role in a number of physiopathological processes in the body, including inflammation, atherogenesis and thrombogenesis. Decreased levels of fibrinogen are associated with an increased risk of bleeding [17]. Increased fibrinogen levels are postulated to enhance thrombus formation by altering the kinetics of the coagulation cascade, resulting in increased fibrin formation [18].

Since both Fibrinogen and platelet are known to play a role in haemostasis and a number of previous studies has identified a link between ABO blood group and haemostatic differences in individuals this study was therefore designed to assess the influence of differences in ABO blood group on the levels of Fibrinogen and platelet count.

Material and Methods

Study site

This study was carried out at College of Health Sciences, Nnamdi Azikiwe University, Anambra State, Nigeria. The College is a multi-faculty college comprising the Faculties of Health Sciences and Technology, Basic Medical Sciences and Medicine with each faculty having many academic departments. The college has a population of over 2000 students. The laboratory testing was done in Hematology Laboratory of Nnamdi Azikiwe University Teaching Hospital, Nnewi, Anambra State.

Study design

This is a cross sectional study designed to evaluate the levels of fibrinogen and platelet count in subjects of different ABO blood groups.

Study population

The population comprised apparently healthy male and female undergraduate students of College of Health Sciences, Nnamdi Azikiwe University.

Sample size calculation and sampling technique

Sample size was calculated using G*Power software (version 3.0.10) Universitat Dusseldorf Germany. Power analysis for one-way analysis of variance with four (blood) groups was conducted in G*Power to determine a sufficient sample size using an alpha of 0.05, a power of 0.90, and a large effect size (f = 0.40). Based on these, the calculated total sample size of 96 has 90% power to detect a difference of 0.40 at a significance level of 0.05. A total of 100 subjects comprising 25 subjects belonging to each of the four blood groups (namely blood groups A, B, AB and O) were recruited for this study. They were undergraduate students aged 18 to 28 years (with mean age of 23.6 ± 4.4 years) drawn from various faculties and departments in the College of Health Sciences, Nnamdi Azikiwe University. The individuals were recruited using a convenience sampling technique in which individuals that meets the inclusion criteria were recruited consecutively until the sample size was attained.

Inclusion criteria

Apparently healthy adult subjects of both genders.

Exclusion criteria

Those excluded from this research work included; individuals with known bleeding disorders, those who were sick or have been transfused with blood in the previous 3 months, smokers and individuals who refused to give their consent.

Ethical consideration

The ethical approval for this research was obtained from Ethics Committee of Faculty of Health Sciences and Technology, Nnamdi Azikiwe University in accordance with the Helsinki declaration by the World Medical Association (WMA) on the ethical principles for medical research involving human subjects.

Informed consent

Informed consent was sought and obtained from the subjects before sample collection.

Sample collection and analysis

Five milliliters (5 ml) of venous blood was collected

Table 1: Gender distribution of the subjects.

	Male	Female	Total
Blood group	Frequency (%)	Frequency (%)	Frequency (%)
O	20 (80.0)	5 (20.0)	25 (100.0)
A	12 (48.0)	13 (52.0)	25 (100.0)
B	11 (44.0)	14 (56.0)	25 (100.0)
AB	15 (60.0)	10 (40.0)	25 (100.0)
Total	58 (58.0)	42 (42.0)	100 (100.0)

Table 2: Comparison of platelet count and fibrinogen levels among the different ABO blood groups.

Blood Group	Platelet (x10^9)	Fibrinogen
O (n = 25)	157.56 ± 23.21	331.92 ± 53.76
A (n = 25)	188.92 ± 46.94	313.00 ± 70.92
B (n = 25)	164.84 ± 32.26	329.40 ± 63.98
AB (n = 25)	178.60 ± 54.90	288.92 ± 46.53
F(P) Value	2.89 (0.040*)	2.76 (0.046)
O vs. A p-value	0.041*	0.676
O vs. B p-value	0.924	0.999
O vs. AB p-value	0.278	0.058
A vs. B p-value	0.172	0.765
A vs. AB p-value	0.813	0.484
B vs. AB p-value	0.641	0.083

*Statistically significant at $p < 0.05$

by standard venepuncture from the individuals aseptically, 3 ml was dispensed into EDTA container for ABO blood grouping and platelet count while 2 ml was dispensed into 0.22 ml of 3.2% trisodium citrate sample container. The blood was mixed and spun and platelet poor plasma separated for use in fibrinogen assay.

Method for ABO blood grouping

The ABO blood grouping was done by the tile method using commercially prepared antisera (namely anti-A, anti-B and anti-AB). A drop of anti-A, -B, and -AB was placed on a white pitted tile, and a drop of blood was placed on each of the antisera. It was mixed well and rocked gently and observed for agglutination. A control was set up by adding standard cells to each antiserum in separate square.

Procedure for platelet count

This was done by the manual method using 1% Ammonium oxalate as diluent and Improved Neaubauer counting chamber for cell count. A 1 in 20 dilution of blood was made by adding 0.02 ml of blood to 0.38 ml of 1% Ammonium oxalate in a clean tube and mixed properly. The cover slip was tightly fixed to the Improved Neaubauer counting chamber, the chamber was charged with the suspension using a clean Pasteur pipette, and placed in a petri dish containing a piece of moist filter paper and covered for 20 minutes to allow the cell settle then using 40x objective and eyepiece, the cell present was counted in the 5 of 0.04 mm area that is 4 outer square of 0.04 mm^2 and the central one. The platelet count was calculated as appropriate.

Measurement of fibrinogen level

The test (platelet poor) plasma was diluted 1:10 in buffer and incubated at 37 °C, phospholipids and thrombin were added followed by Calcium (all pre-warmed to 37 °C). On the addition of the Calcium, timing begun. The time taken for the clot to form was compared to a calibration curve and the fibrinogen concentration determined.

Statistical analysis

Statistical package for social science (SPSS) version 20 was used for the data analysis. Results were expressed as mean ± standard deviation and parameters was compared between different groups using independent t-test while One-way analysis of variance (ANOVA) was used for comparison of parameters among groups. Correlation was done using pearson correlation and level of significance was set at $P < 0.05$.

Results

About 58% of the participants were males and 42% were females. Majority of the blood group O (80%) and blood group AB (60%) subjects were males while majority of the blood group B (56%) and A (52%) were females respectively (Table 1).

There was a significant decrease in platelet count in blood group O participant when compared with the value obtained in participants of A blood group (157.56 ± 23.21 Vs 188.92 ± 46.94; $p = 0.041$). However, post-hoc comparison between other blood groups showed no significant difference platelet count ($p > 0.05$). Also, there was no significant difference in the mean fibrinogen levels of participants belonging to different blood groups ($p > 0.05$) (Table 2).

There was no significant difference in the mean levels of platelet count and fibrinogen obtained in the male participants when compared with values obtained in female participants respectively ($p > 0.05$) (Table 3).

The mean levels of parameters (platelet and fibrinogen) studied did not differ significantly when compared between various blood groups in male and female participants studied ($p > 0.05$) (Table 4).

Table 3: Comparison of platelet count and fibrinogen level between male and female subjects.

Gender (n)	Platelet (x10^9/l)	Fibrinogen
Male (n = 58)	170.88 ± 41.68	313.84 ± 64.97
Female (n = 42)	174.69 ± 43.75	318.52 ± 55.98
t-value	0.442	0.376
P-value	0.66	0.708

Table 5: Correlation of fibrinogen level and platelet count among the blood groups.

Blood Group	Correlation	r-value	P-value
O (n = 25)	Fibrinogen vs. Platelet	-0.174	0.406
A (n = 25)	Fibrinogen vs. Platelet	0.385	0.048*
B (n = 25)	Fibrinogen vs. Platelet	0.229	0.271
AB (n = 25)	Fibrinogen vs. Platelet	-0.029	0.892

*Statistically significant at $p < 0.05$

Table 4: Comparison of parameters between male and female subjects among the different blood groups.

Blood Group	Gender	Platelet (x10^9/l)	Fibrinogen
Blood group O	Male (n = 20)	159.45 ± 22.56	330.40 ± 60.11
	Female (n = 5)	150.00 ± 26.93	338.00 ± 10.95
	t-value	0.808	0.277
	P-value	0.427	0.784
Blood group A	Male (n = 12)	176.50 ± 42.24	301.25 ± 86.26
	Female (n = 13)	200.38 ± 49.75	323.84 ± 54.51
	t-value	1.288	0.79
	P-value	0.21	0.438
Blood group B	Male (n = 11)	167.18 ± 35.01	340.90 ± 52.57
	Female (n = 14)	163.00 ± 31.15	320.35 ± 72.33
	t-value	0.316	0.791
	P-value	0.755	0.437
Blood group AB	Male (n = 15)	184.33 ± 60.71	282.00 ± 47.66
	Female (n = 10)	170.00 ± 46.55	299.30 ± 45.16
	t-value	0.631	0.907
	P-value	0.534	0.374

There was a weak but significant positive correlation between fibrinogen level and platelet count in blood group A participants ($r = 0.385$; $p = 0.048$) (Table 5).

Discussion

The present study evaluated the fibrinogen levels and platelet count in subjects of different ABO blood groups with the view of ascertaining if disparity exists in the levels of platelets and fibrinogen amongst the different ABO blood groups as this may provide possible explanation for previous findings especially in ABO differential disposition to thrombotic disorders.

The mean platelet count was found to be significantly lower in individuals of blood group O than in individuals of A blood group. Though in comparison to the reference range in literature (150 to 400 × 10^9/l) the levels were normal in this study, this may be suggestive of a likelihood of thrombocytopenia in individuals of O blood group than A blood group. Several studies have shown increased risk of thrombosis in individuals belonging to the non-O (A, B and AB) blood type [19]. The finding of this study however is in contrast with the report of Ajayi, et al. [20] who observed that blood group AB had significantly higher values of Platelet counts when compared with other blood groups. Aside the significant difference obtained between blood group O and A, there were no significant differences in the mean platelet count when compared between individuals of the other blood groups. This is in consonance with the findings of a previous study that reported no significant differences in the mean platelet count between the different ABO blood groups [21].

Our finding showed no statistically significant difference in the mean level of plasma fibrinogen when compared among the participants of different ABO blood groups. This is in consonance with the report of previous studies [12,22]. However, contrary to the result obtained in this study in which there was a non-significant lower fibrinogen level in those with blood group AB compared to blood group O, Jimenez, et al. [23] documented a higher plasma fibrinogen level in the patients with blood group AB than in those with blood groups A, B and O.

Furthermore, the present study showed no significant differences in the mean level of both fibrinogen and platelet count in male and female participants of different ABO blood groups studied. This confirms that gender differences do not alter the levels of platelet count and fibrinogen level in individuals of different ABO blood group. This finding is corroborated by the results of similar studies [12,15,21].

Our study showed that in individuals of A blood group, plasma fibrinogen level correlates positively with Platelet count. This implies that in these individuals, when there is an increase in fibrinogen level an increase in platelet count should be expected and vice versa. This supports an earlier study by Aliberti, et al. [24] which found that plasma fibrinogen level was significantly correlated with the Platelet count. However, in our study, there were no significant correlations observed between platelet count and plasma fibrinogen level individuals of the other ABO blood groups (AB, B and O).

Conclusion

The present study revealed a significantly lower platelet count in O blood group participants than in A blood group individuals, this suggests the likelihood of higher risk of thrombocytopenia in blood group O individuals compared to blood group A individuals. Also in individuals of A blood group, fibrinogen level correlates positively with platelet count.

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Conflicts of Interest

There are no conflicts of interest.

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