








## Research Article

# Correlation of Serum Ferritin Levels and Liver Function Tests with ABO Blood Grouping

Oloruntoba Okojie Festus <sup>1\*</sup>, Basil Nnaemeka Obodo <sup>1,2</sup>, Ojugo G.O.<sup>1</sup>, Osaihiomwan Aigbe<sup>3</sup>, Sylvia Enorese Irogue<sup>1</sup>, Kennedy Oberhiri Obohwemu <sup>4</sup>, Efosa John Osarenren <sup>4</sup>, Gift Aimanehi Okhinoghor<sup>1</sup>, Ogbemudia Endurance Edebiri <sup>6</sup>, Okosun Hassan Blackie <sup>7</sup>, Imiefoh Basil Ekenomen<sup>8</sup> and Andrew Oseghale Eigbedion <sup>9,10</sup>

<sup>1</sup>Department of Chemical Pathology, Faculty of Medical Laboratory Science, Ambrose Alli University, Ekpoma, Edo State, Nigeria.

<sup>2</sup>Department of Biological Sciences, Purdue University, West Lafayette, United States of America.

<sup>3</sup>Department of Medical Biochemistry, Faculty of Basic Medical Sciences, Ambrose Alli University, Ekpoma, Edo State, Nigeria.

<sup>4</sup>Senior Lecturer, Leeds Trinity University, SSS Partnership, Birmingham Campus, United Kingdom; and PENKUP Research Institute, Birmingham, United Kingdom.

<sup>5</sup>Department of Biochemistry, University of Benin, Benin City, Edo State, Nigeria.

<sup>6</sup>Department of Human Physiology, Faculty of Basic Medical Sciences, Ambrose Alli University Ekpoma Edo state Nigeria.

<sup>7</sup>Department of Human Anatomy, Faculty of Basic Medical Sciences, Ambrose Alli University Ekpoma Edo state Nigeria.

<sup>8</sup>Department of Chemical Pathology, Irrua Specialist Teaching Hospital, Irrua, Edo State, Nigeria.

<sup>9</sup>Department of Paediatrics, Faculty of Clinical Sciences, Ambrose Alli University, Ekpoma, Edo State, Nigeria.

<sup>10</sup>Department of Paediatrics, Irrua Specialist Teaching Hospital, Irrua, Edo State, Nigeria.

\*Corresponding author: [festusokojie@hotmail.com](mailto:festusokojie@hotmail.com)

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## Abstract

This study was carried out to evaluate the association between serum ferritin, liver function parameters, and ABO blood grouping. A total of eighty (80) subjects were recruited, comprising nineteen (19) individuals with blood group A, fifteen (15) with blood group B, sixteen (16) with blood group AB, and thirty (30) with blood group O. Blood samples were collected from the antecubital vein into accurately labeled heparinized bottles for each participant and transported to the laboratory for the estimation of alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP), total protein, and albumin. Both ABO and Rhesus blood groups were determined using the tube method. The results of this study revealed that serum ALT activity differed significantly ( $p < 0.05$ ) among the various blood groups. Pairwise comparison showed that ALT levels were significantly different ( $p < 0.05$ ) in blood groups B and AB when compared with groups A and O. However, the serum activities of AST and ALP did not differ significantly ( $p > 0.05$ ) among the blood groups, although AST was higher in blood group AB and ALP was higher in blood group O. Similarly, pairwise comparisons for these enzymes were also not significantly different ( $p > 0.05$ ). Serum total protein and albumin levels were not significantly different ( $p > 0.05$ ) across the blood groups; total protein was higher in blood group AB, while albumin was higher in blood group B. Serum ferritin levels were likewise not significantly different ( $p > 0.05$ ) among the blood groups, although higher values were observed in blood group AB compared with the others. This study, therefore, suggests that, although most liver function and ferritin parameters did not show statistically significant differences across ABO blood groups, there are observable trends indicating that these parameters may vary among individuals with different ABO phenotypes.

## 1. Introduction

The ABO blood group system is the most important and widely used blood group system in human transfusion medicine. The naturally occurring anti-A and anti-B antibodies, predominantly of the IgM class, are produced early in life through environmental sensitization to exogenous antigens such as food components, bacteria, and viruses. ABO blood group antigens are not unique to humans; they have also been identified in certain other mammals, including rodents and non-human primates such as chimpanzees, bonobos, and gorillas [1].

Disease development (pathogenesis) is often multifactorial, involving both host genetic factors and environmental exposures. The ABO blood groups represent one such genetically determined system of agglutinogens (antigens) expressed on the surface of red blood cells. Over the years, several studies have reported associations between ABO blood groups and a variety of infectious and non-infectious diseases [2–4]. In most individuals, A and B antigens can also be secreted into body fluids and circulate in the bloodstream. Individuals who are non-secretors appear to be more susceptible to a range of infections, and this vulnerability may be related to the absence of soluble blood group substances that can competitively inhibit pathogen binding to cell-surface polysaccharides [2]. Furthermore, [5] reported that the development of fibrosis in hepatitis C virus (HCV) infection is influenced by gene–environment interplay, and that ABO blood group distribution is linked to thrombotic events, with non-O blood groups showing an increased risk of venous thrombosis.

Liver enzymes constitute a broad group of biomarkers that include alanine aminotransferase (ALT), aspartate aminotransferase (AST), together often referred to as transaminases or aminotransferases alkaline phosphatase (ALP),  $\gamma$ -glutamyl transferase, glutathione-S-transferase, serum cholinesterase, glutamate dehydrogenase, and 5'-nucleotidase [6]. Among these, ALT and ALP are the most frequently evaluated in clinical practice. Measurement of the serum or plasma activities of these enzymes remains a fundamental tool for assessing hepatic integrity and function in both health and disease [7]. Notably, some reports have suggested links between ABO blood groups and certain liver-related disorders [5]. Marked inter-individual differences in the serum concentrations of intestinal and hepatic enzymes may increase susceptibility to specific diseases and may even influence the nutritional value derived from diet [8]. For example, individuals with blood group O have been reported to be more resilient to the sequelae of acute viral hepatitis [9]. Conversely, persons with blood group A have been observed to have an increased risk of certain malignancies, including gastric, pancreatic, epithelial ovarian, and skin cancers [10], suggesting that host blood group antigens may modulate inflammatory or proliferative pathways. These observations raise the possibility that liver enzyme levels could vary by ABO phenotype. However, despite these indications, there remains limited information on the direct relationship between ABO blood group and variation in liver enzyme activities in apparently healthy individuals.

More recently, genome-wide association studies (GWAS) have provided additional mechanistic clues: two independent GWAS demonstrated that the systemic inflammatory milieu can be influenced by polymorphisms within the ABO locus. In particular, tumor necrosis factor- $\alpha$  (TNF- $\alpha$ ) and soluble intercellular adhesion molecule-1 (sICAM-1) were shown to be associated with single nucleotide polymorphisms (SNPs) in the ABO gene region [11, 12]. Since increased TNF- $\alpha$  expression has been implicated in liver inflammation and hepatocarcinogenesis [12], these findings support a biologically plausible link between ABO blood group, inflammatory signaling, and hepatic biochemical markers. In addition to liver enzymes, serum ferritin is a key acute-phase reactant and an established indicator of body iron stores. Because ferritin levels can be influenced by inflammation, infection, and liver function, exploring its behavior across different ABO blood groups may offer further insight into host–phenotype interactions.

Therefore, this study aims to assess the relationship between serum ferritin and liver function parameters (ALT, AST, ALP, and total protein) among apparently healthy individuals with ABO blood groups A, B, AB, and O. Given the paucity of data on this subject, our objective is to determine whether ABO blood grouping is associated with measurable variations in serum ferritin and hepatic enzyme activities, thereby contributing to the growing body of evidence on the clinical relevance of ABO phenotypes.

## 2. Materials and Methods

### 2.1. Study Design

This cross-sectional study was conducted among apparently healthy individuals of different ABO blood groups at Ambrose Alli University, Ekpoma, Edo State, Nigeria. The study evaluated serum ferritin, alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP), total protein, and albumin levels across the ABO phenotypes. Data collection was carried out over a four-month period. A complete medical history was obtained, including name, age, sex, dietary habits, and nutritional status, to confirm apparent good health and to exclude individuals with conditions that could alter liver function or serum ferritin levels.

### 2.2. Study Population

The study population comprised eighty (80) apparently healthy individuals of different ABO blood groups recruited from Ambrose Alli University, Ekpoma, Edo State, Nigeria. Participants included nineteen (19) individuals with blood group A, fifteen (15) with blood group B, sixteen (16) with blood group AB, and thirty (30) with blood group O. Demographic data such as name, age, and sex were obtained from all subjects. Only individuals who appeared clinically healthy and had no obvious signs of illness were included. Persons with a history or clinical evidence of liver disease, hematological disorders, recent infections, acute or chronic illnesses, or those on medications that could alter liver function tests or serum ferritin levels were excluded. This was done to ensure that the measured parameters reflected normal physiological variations across ABO blood groups.

### 2.3. Sample Collection

Blood samples were collected from the antecubital vein into accurately labeled heparinized tubes for each participant. ABO and Rhesus blood groups were determined using the tube method. The blood samples were centrifuged in a laboratory centrifuge within two hours of collection, and the serum was separated into clean, dry plain tubes labeled to correspond with the original sample bottles. Biochemical analyses were then carried out for serum ferritin, alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP), total protein, and albumin.

## 2.4. Sample Analysis

ABO and Rhesus blood groups were determined using the standard tube agglutination method as described by [13]. Commercial antisera (anti-A, anti-B, anti-AB, and anti-D) were used to identify the corresponding red cell antigens, and agglutination was read macroscopically and confirmed microscopically. Serum ferritin concentration was determined using atomic absorption spectrophotometry (AAS) following the direct method of [14]. Serum samples were prepared and aspirated into the instrument according to the manufacturer's operating conditions for ferritin/iron estimation. Serum alanine aminotransferase (ALT) and aspartate aminotransferase (AST) activities were measured by the colorimetric method of [15]. Absorbance was read spectrophotometrically, and enzyme activities were calculated and expressed in U/L. Serum alkaline phosphatase (ALP) activity was estimated using the method described by Rec (1972), in which the rate of p-nitrophenol formation was monitored spectrophotometrically and used to derive ALP activity. Total serum protein was determined by the biuret method [16], and serum albumin was measured using the bromocresol green (BCG) dye-binding method of [17], in both assays, the absorbance obtained was compared with the appropriate standard to determine the concentration.

## 2.5. Statistical Analysis

The mean and standard deviation (SD) of all measured parameters were computed. Group differences across the ABO blood types were assessed using one-way analysis of variance (ANOVA) with Fisher's least significant difference (LSD) post hoc test, implemented in SPSS software (version 21; IBM Corp). A p-value of less than 0.05 was considered statistically significant.

## 3. Results

The results of this study showed that serum alanine aminotransferase (ALT) activity differed significantly ( $p < 0.05$ ) among the ABO blood groups. ALT levels in blood groups B and AB were significantly higher ( $p < 0.05$ ) than those in blood groups A and O.

In contrast, serum aspartate aminotransferase (AST) and alkaline phosphatase (ALP) activities were not significantly different ( $p > 0.05$ ) across the groups, although AST was higher (not significant) in blood group AB and ALP was higher (not significant) in blood group O. Pairwise comparisons for these enzymes also showed no statistically significant differences ( $p > 0.05$ ). Serum total protein and albumin levels were likewise not significantly different ( $p > 0.05$ ) among the blood groups, although total protein was higher (not significant) in group AB and albumin was higher (not significant) in group B. Serum ferritin levels did not differ significantly ( $p > 0.05$ ) across the ABO groups, but values were higher (not significant) in blood group AB compared with the others Table 1.

**Table 1:** Serum ferritin levels and liver function in ABO blood groups

Parameters	Blood Group A (n=19)	Blood Group B (n=15)	Blood Group AB (n=16)	Blood Group O (n=30)	F-value	p-value
AST (U/L)	8.47±1.71	8.59±1.44	8.62±1.56	8.39±2.03	0.077	0.972
ALT (U/L)	6.99± 1.47	8.51±1.68*	7.04±1.74*	6.46±1.52	5.550	0.002
ALP (U/L)	149.84±37.00	144.40±26.94	140.36±30.92	152.00±32.76	0.520	0.670
TP (g/dl)	7.05±1.02	7.25±0.56	7.30±0.72	7.12±0.58	0.443	0.723
ALB (g/dl)	3.83±0.33	3.95±0.16	3.93±0.26	3.85±0.22	1.062	0.370
Ferritin (ug/dl)	126.93±22.34	114.50±18.38	137.53±10.64	122.09±23.24	0.586	0.630

**KEYS:** AST= Aspartate amino transferase, ALT=Alanine amino transferase, ALP= Alkaline phosphatase, TP= total protein, ALB= Albumin, n=Sample size,  $p < 0.05$ = Significant;  $p > 0.05$ = Not Significant.

When results were stratified by sex, serum ferritin and liver function parameters showed some variation across blood groups; however, only ALT activity in male subjects was significantly higher ( $p < 0.05$ ). AST, ALP, total protein, albumin, and ferritin levels were not significantly different among the blood groups with respect to gender Table 2.

**Table 2:** Serum ferritin levels and liver function in ABO blood groups with respect to Sex

Parameters		Blood Group A	Blood Group B	Blood Group AB	Blood Group O	F-value	p-value
AST (U/L)	Male	8.37±1.48	9.35±1.20	8.59±1.61	8.21±2.16	0.807	0.497
	Female	8.66±1.61	7.71±1.21	8.66±1.61	8.78±1.77	0.595	0.624
ALT (U/L)	Male	7.08± 1.30	8.81±1.88	6.92±1.70*	6.42±1.48	4.589	0.007
	Female	6.84±1.84	8.16±1.48	7.20±1.92	6.54±1.71	1.218	0.323
ALP (U/L)	Male	142.67±39.63	137.38±24.14	148.64±23.19	150.95±30.31	0.444	0.722
	Female	162.14±30.81	152.43±29.54	129.71±37.90	154.33±39.58	1.114	0.361
TP (g/dl)	Male	6.98±1.00	7.35±0.60	7.16±0.78	7.16±0.53	0.437	0.728
	Female	7.17±1.13	7.14±0.53	7.49±0.64	7.04±0.73	0.440	0.727
ALB (g/dl)	Male	3.83±0.24	3.96±0.19	3.86±0.27	3.90±0.17	0.742	0.530
	Female	3.83±0.48	3.94±0.13	4.03±0.22	3.76±0.29	1.204	0.328
Ferritin (ug/dl)	Male	120.98±24.41	116.19±13.26	132.62±8.50	122.33±23.16	0.973	0.414
	Female	121.53±15.24	116.64±9.02	132.37±8.94	124.62±16.85	1.188	0.354

## 4. Discussion

The results of this study showed that there was a significant association between serum ALT activity and ABO blood group, with ALT being significantly higher in some groups. This contrasts with the findings of [18], who reported no significant difference in ALT activity among individuals with different ABO blood groups. Similarly, [19] reported no significant relationship in ALT levels among healthy Saudi blood donors and further emphasized that serum liver enzymes may vary by sex, age, and ethnicity, which could partly explain the discrepancy between their study and the present one.

In this study, AST activity was observed to be higher in individuals with blood group AB, although this difference was not statistically significant. This pattern does not fully align with the report of [18], who also noted variations in AST but did not establish a consistent ABO-dependent trend. In addition, serum total protein and serum albumin were higher in blood group B compared with other groups, suggesting possible ABO-related physiological variability in protein synthesis or transport.

This study also demonstrated higher ALP activity in subjects with blood group O, a finding that differs from [18], who reported a significant relationship in ALP activity when comparing other blood groups. One possible explanation for the higher ALP activity observed in our blood group O subjects is differential cytokine-binding capacity among ABO phenotypes, which can modulate inflammatory pathways and enzyme expression. Previous studies [11, 12, 20, 21] have shown that ABO blood group is associated with differences in the binding of cytokines and adhesion molecules such as EGF, TNF- $\alpha$ , sICAM-1, E-selectin, and P-selectin, and such interactions may indirectly stimulate hepatic or intestinal ALP release.

This observation is partly consistent with the report of [5], who, in studying the severity of fibrosis in chronic hepatitis C infection, found that non-O blood groups (A, B, AB) were more frequently associated with thrombotic events, which can, in turn, enhance hepatic enzyme activity. The thrombotic tendency of non-O blood groups is believed to be mediated, at least in part, through higher circulating levels of factor VIII and von Willebrand factor [22, 23]. Qiang et al. [24] further suggested that increased cytokine-binding capacity in non-O groups may contribute to fibrosis progression, which supports the biological plausibility of ABO-related differences in liver-related parameters.

On the other hand, the present finding agrees with earlier reports by [8, 25], who observed higher ALP activity in individuals with blood groups O and B compared with group A. This has been attributed to a greater contribution of intestinal alkaline phosphatase in blood group O (and to a lesser extent B), whereas group A individuals produce relatively lower amounts of intestinal ALP. This physiological difference has even been used to support dietary recommendations for blood group A individuals, who may benefit from lower-fat diets due to their lower ALP activity and possible higher susceptibility to cardiovascular and neoplastic conditions.

This study also observed that serum ferritin levels were higher in individuals with blood group AB, with group A also showing relatively elevated values. This pattern is in concordance with the findings of [26], who reported that group B individuals had lower ferritin levels compared with groups A and O. Iron is an essential trace element involved in haemoglobin synthesis, cellular respiration, and several enzymatic processes; therefore, variation in ferritin across ABO groups may reflect subtle differences in iron handling, storage, or low-grade inflammation. Yamamoto et al. [27] suggested that differences in the cellular antigenic composition of A and B blood group antigens may enhance or inhibit certain biochemical processes, which may partly explain these observations.

The present study also showed that mean ferritin levels were lowest in group B and highest in group AB, but all values still fell within the normal reference range when compared with standard texts [28]. This is important because it suggests physiological, not pathological, variation. Although there is paucity of data directly linking serum ferritin to ABO blood types, several studies have documented associations between ABO groups and disease susceptibility, for example, peptic ulcer disease in group O and bronchopneumonia in group A. Some authors have also reported that elevated serum iron, ferritin, and TIBC may predispose individuals to infection because free iron can support bacterial growth [29]. Thus, ABO-related variation in ferritin, even within the normal range, may still have biological relevance.

Finally, the distribution of ABO blood groups in this study is consistent with previous reports from Southwestern Nigeria [30], supporting the representativeness of the sample and lending credibility to the observed biochemical trends.

## 5. Conclusion

In conclusion, these findings highlight the need to define more precisely the reference ranges for ALT, AST, ALP, total protein, albumin, and serum ferritin in young adults, particularly when interpreting results across different ABO blood groups. Establishing such population- and phenotype-specific reference values would improve the accuracy of diagnosis and clinical follow-up of liver-related conditions. Further, larger, multi-center studies are recommended to clarify the biological basis of the non-significant but observable variations in serum ferritin and liver enzyme activities observed in this study. Finally, routine liver function and serum ferritin screening in apparently healthy individuals of different ABO blood groups may be useful for early detection and monitoring of subclinical hepatic or inflammatory changes.

### Article Information

**Conflict of Interest:** The authors declare no conflicts of interest. The authors alone are responsible for the content and the writing of the paper.

**Ethical Permission:** Ethical approval was obtained from the University Ethics Committee, and informed consent was also sought from the subjects before the collection of blood samples.

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