



Research Article

Serum Selenium Status in Neutropenic and Non-Neutropenic Nigerian Adults: A Case-Control Study

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ABSTRACT

Selenium-dependent glutathione peroxidase protects neutrophils, the body's first line of cellular defence, from the oxidative damage generated during phagocytosis, and selenium deficiency has been linked to impaired neutrophil survival and function. This study determined serum selenium status among neutropenic adults attending the General Outpatients' Department of Kwara State University Teaching Hospital, Nigeria, and examined its relationship with absolute neutrophil count (ANC). In this hospital-based, unmatched case-control study, 140 adults aged 18–70 years were recruited and classified as neutropenic cases ($n = 92$) or non-neutropenic controls ($n = 48$). Serum selenium was measured by flame atomic absorption spectrophotometry, and ANC was obtained with a five-part automated haematology analyser (Sysmex XN-1000). Group means were compared using Student's t-test, and the selenium–ANC relationship was assessed by Pearson Chi-square and linear regression. The ANC was significantly lower in the neutropenic group than in controls (1.53 ± 0.28 vs $3.01 \pm 0.63 \times 10^9/L$; $p < 0.001$), but mean serum selenium did not differ significantly between groups (70.59 ± 13.92 vs 83.40 ± 16.46 ng/mL; $p = 0.648$). No significant relationship was found between selenium level and ANC among neutropenic subjects ($p = 0.873$). Serum selenium status was not significantly associated with neutropenia or ANC in this population, suggesting limited value for routine selenium screening in neutropenic Nigerian adults; larger, longitudinal, and molecular-level studies are warranted to clarify this relationship.

Keywords: Diabetic Kidney Disease; Estimated Glomerular Filtration Rate; Neutrophil Gelatinase-Associated Lipocalin; Type 2 Diabetes Mellitus; Urinary Albumin-to-Creatinine Ratio

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INTRODUCTION

Selenium (Se) is an essential trace element with a central role in antioxidant defence, immune regulation, and cellular metabolism (Sadler *et al.*, 2024). Beyond its long-recognised antioxidant function, selenium is now understood to modulate both innate and adaptive immunity through incorporation into more than twenty-five selenoproteins (Filippini *et al.*, 2023). It has an atomic

number of 34 and an atomic weight of 78.96, with oxidation states of +6, +5, +4, +3, +2, +1 and –1; it is classified as a semi-metal with intermediate properties between a metal and a non-metal, and is stable and does not oxidise at ordinary temperatures (Burk *et al.*, 2015).

Selenium occurs naturally in soil, water, rock and food (Schrauzer, 2000), and enters the environment through the burning of hydrocarbons, volcanic

eruptions, and the mining and milling of minerals (Rayman, 2012). Excessive selenium intake may cause brittle nails, garlic breath, nausea and vomiting (Cummins & Kimura, 1971), whereas selenium deficiency is associated with an increased risk of cancer and ischaemic heart disease (Schrauzer, 2000). Its deficiency has also been shown to impair neutrophils' phagocytic function and their capacity to kill ingested organisms; this defect has been attributed to reduced cytosolic glutathione peroxidase activity within neutrophils, such that the neutrophils themselves are destroyed by oxidative injury rather than the ingested pathogen, resulting in neutropenia (Arthur *et al.*, 2003).

Selenium is incorporated into several selenoproteins and selenoenzymes with essential biological functions, including dehydrogenases, glutathione peroxidases (GPXs), deiodinases, selenoprotein-P and selenoprotein-W (Sadler *et al.*, 2024). These enzymes catalyse reactions that protect neutrophils against the oxidative stress and free-radical damage generated during the respiratory burst used to kill ingested organisms, and selenium-dependent pathways are increasingly recognised as direct regulators of neutrophil reactive-oxygen-species signalling, phagocytosis and extracellular-trap formation (Lee & Prabhu, 2025). Selenium additionally supports thyroid hormone metabolism and contributes to the regulation of immune-related transcriptional signalling (Filippini *et al.*, 2023).

Neutropenia refers to a reduction in the absolute number of circulating neutrophils (segmented cells and bands) to below $1.80 \times 10^9/L$ (Thobakgale & Ndung'u, 2014). Normal neutrophil counts must be stratified for age and ethnicity: in some ethnic groups, particularly persons of African heritage and some Middle Eastern populations, the lower limit for normal is $0.8\text{--}1.0 \times 10^9/L$ (Denic *et al.*, 2016). Benign ethnic neutropenia, characterised by lower baseline neutrophil counts without a corresponding increase in infection risk, has been reported in 25–50% of individuals of African and Middle Eastern descent (Oloyede *et al.*, 2021), a pattern also documented among Nigerian cohorts (Wonodi *et al.*, 2017). Neutropenia is therefore a relatively frequent finding that is often well-tolerated and resolves spontaneously (Hsieh *et al.*, 2007). Approximately 1% of the general population lives with mild-to-severe neutropenia, and this proportion rises to as much as 8% in populations of African origin (Donadieu *et al.*, 2013).

Selenium status may be assessed directly in whole blood, plasma, serum or tissue (e.g. liver, kidney), or

in urine, hair and toenails, or indirectly through erythrocyte glutathione peroxidase activity (Fairweather-Tait *et al.*, 2010). Analytical approaches include colorimetry, fluorimetry, atomic absorption spectrometry, inductively coupled plasma mass spectrometry, neutron activation analysis and gas chromatography, among others (Neve *et al.*, 1992); however, flame atomic absorption spectrophotometry was adopted in the present study. Despite the biological plausibility linking selenium deficiency to neutrophil loss, population-specific data comparing serum selenium status between neutropenic and non-neutropenic Nigerians remain scarce, and no prior case-control study appears to have examined this relationship in Kwara State, north-central Nigeria. This study therefore aimed to compare serum selenium levels between neutropenic cases and non-neutropenic controls attending Kwara State University Teaching Hospital, Nigeria, and to assess the relationship between selenium level and absolute neutrophil count.

MATERIALS AND METHODS

Study Area and Population

This study was conducted among adults aged 18–70 years attending the General Outpatients' Department (GOPD) of Kwara State University Teaching Hospital, Ilorin, Nigeria, following approval by the institutional research ethics committee.

Study Design

This was a hospital-based, unmatched case-control study. Neutropenic subjects (cases) and non-neutropenic subjects (controls) were recruited from the same source population and clinic, and blood samples from both groups were analysed for full blood count and serum selenium level, with results compared statistically between cases and controls.

Sample Size Determination

The minimum sample size was calculated using the formula for comparing two independent proportions in an unmatched case-control study (Charan & Biswas, 2013):

$$n = (Z\alpha/2 + Z\beta)^2 [p_1(1 - p_1) + p_2(1 - p_2)] / (p_1 - p_2)^2$$

where n is the minimum number of subjects required per group, $Z\alpha/2$ is the standard normal deviate for a two-tailed α of 0.05 (1.96), $Z\beta$ is the standard normal deviate for 80% power (0.84), p_2 is the anticipated proportion of selenium deficiency among non-neutropenic controls, and p_1 is the anticipated proportion of selenium deficiency among neutropenic cases, derived from a minimum detectable odds ratio (OR) of 3.0 between the two groups.

The control-group prevalence of selenium deficiency ($p_2 = 17.2\%$) was obtained from a comparable Nigerian case-control study of serum selenium status (Eze *et al.*, 2020). Assuming a minimum detectable Odds ratio of 3.0, the corresponding anticipated prevalence of selenium deficiency among cases was:

$$p_1 = (OR \times p_2) / (1 - p_2 + OR \times p_2) = (3.0 \times 0.172) / (1 - 0.172 + 3.0 \times 0.172) = 0.384 \text{ (38.4\%)}$$

Substituting these values, with equal allocation of cases to controls:

$$n = (1.96 + 0.84)^2 [0.384(1 - 0.384) + 0.172(1 - 0.172)] / (0.384 - 0.172)^2$$

$$n = 7.84 \times [0.237 + 0.142] / 0.0449$$

$$n = 7.84 \times 0.379 / 0.0449 \approx 66.2$$

A minimum of 67 subjects was therefore required per group (134 in total). A total of 140 participants, comprised of 92 neutropenic cases and 48 non-neutropenic controls were eventually recruited, exceeding the calculated minimum overall, although the achieved case-to-control allocation was unequal (approximately 2:1) rather than the 1:1 ratio assumed in the calculation. This reflects the relative clinical availability of neutropenic and non-neutropenic patients presenting at the GOPD during the recruitment period, and is acknowledged as a factor that may modestly reduce statistical efficiency relative to a balanced design.

Inclusion and Exclusion Criteria

Male and female subjects aged 18–70 years who consented to participate were included in the study. Subjects at the extremes of age, pregnant women, and individuals with HIV/AIDS, diabetes mellitus, or a history of smoking or alcohol use were excluded, as these conditions may compromise immune status and thereby affect selenium level (Newburger & Boxer, 2011).

Ethical Consideration and Informed Consent

Ethical clearance for the study was obtained from the Kwara State University Teaching Hospital Institutional Review Committee (KWASUTHIRC), Nigeria (Approval number KWASUTH/ADM/134/VOL.II/381). Written informed consent was obtained from all participants after the purpose and benefits of the study had been explained to them. Participation was entirely voluntary, and all data collected were treated with strict confidentiality.

Data Collection

A semi-structured questionnaire was used to obtain the socio-demographic characteristics of participants.

Collection of Samples and Analysis

Blood samples were obtained from an accessible peripheral vein of each subject under aseptic conditions. A total of 5 mL of blood was collected: 3

mL was dispensed into an EDTA anticoagulant bottle for full blood count analysis, while the remaining 2 mL was dispensed into a plain bottle, allowed to clot, and centrifuged at 3,000 rpm for 5 minutes using a bench-top centrifuge (Life Assistance Scientific, UK, model 80-2). Serum was immediately separated into cryovials for selenium assay, appropriately labelled, and stored at -20°C prior to analysis.

Full Blood Count Estimation

Complete blood count was performed using a five-part Sysmex XN-1000 automated haematology analyser.

Serum Selenium Level

Serum selenium level was analysed using flame atomic absorption spectrophotometry (PG 990 model).

Data Analysis

Data generated from this study were analysed using the Statistical Package for Social Sciences (SPSS) version 21.0 (IBM SPSS Statistics, Chicago, IL, USA). Continuous variables such as absolute neutrophil count and serum selenium level were expressed as means and standard deviations. Student's t-test was used to compare means and assess statistical differences between cases and controls, while the relationship between continuous variables was determined using Pearson Chi-square and linear regression analyses. Statistical significance was set at $p \leq 0.05$.

RESULTS

Characteristics of the Study Participants

A total of 140 participants meeting the eligibility criteria were enrolled and classified into two groups: neutropenic cases ($n = 92$) and non-neutropenic controls ($n = 48$). The mean age (mean $\pm$ standard deviation) was 43.32 ± 13.96 and 38.46 ± 14.82 years for the case and control groups, respectively, with no significant difference between the two groups ($p = 0.335$). Among the cases, 45 (48.9%) were male and 47 (51.1%) were female; among the controls, 25 (52.1%) were male and 23 (47.9%) were female, with no significant difference in sex distribution between groups ($p = 0.346$).

Comparison of Selenium Level between Cases and Controls

Absolute neutrophil count (ANC) was significantly higher in the control group than in the case group ($p < 0.001$). However, no statistically significant difference was observed in serum selenium level between cases and controls ($p = 0.648$).

Relationship between Selenium Level and Absolute Neutrophil Count in Cases

Pearson Chi-square analysis ($p = 0.847$) and linear regression modelling ($p = 0.873$) both showed no

statistically significant relationship between serum selenium level and absolute neutrophil count among the neutropenic cases.

Table 1: Characteristics of the Study Groups

Characteristic	Cases	Controls	p-value
Age (years)	43.32 ± 13.96	38.46 ± 14.82	0.335
Sex (Male/Female)	45/47	25/23	0.346

Values are expressed as Mean ± SD.

Table 2: Comparison of Selenium Assay and ANC between Cases and Controls

Parameter	Cases (Mean ± SD)	Controls (Mean ± SD)	p-value
ANC ($\times 10^9/L$)	1.53 ± 0.28	3.01 ± 0.63	<0.001*
Selenium (ng/mL)	70.59 ± 13.92	83.40 ± 16.46	0.648

*Statistically significant difference between groups ($p < 0.05$). Values are expressed as Mean ± SD; p-values were determined by Student's t-test

Table 3: Relationship between Selenium Level and Absolute Neutrophil Count in Cases

Statistical Test	Selenium (ng/mL)	ANC ($\times 10^9/L$)	p-value
Pearson Chi-square	70.59 ± 13.92	1.53 ± 0.28	0.847
Linear regression	70.59 ± 13.92	1.53 ± 0.28	0.873

Values are expressed as Mean ± SD; $p < 0.05$ was considered statistically significant

DISCUSSION

This case-control study compared adult Nigerians with neutropenia (aged 18–70 years) against an apparently healthy, age- and sex-comparable control group, all resident in Ilorin, north-central Nigeria. A structured questionnaire and interview method, adapted from a World Health Organization consent template, was used to gather participant information after informed consent had been obtained (World Health Organization, 2004).

A total of 92 neutropenic cases and 48 non-neutropenic controls were recruited for this study. Serum selenium was determined by flame atomic absorption spectrophotometry, and values within 60–150 ng/mL were considered within the normal range for this assay. Neutropenia in this cohort was, consistent with prior reports, a relatively frequent and generally well-tolerated finding (Hsieh *et al.*, 2007); normal ANC must be stratified for age and ethnicity, since the prevalence of neutropenia can reach 8% in populations of African descent compared with approximately 1% globally (Donadieu *et al.*, 2013).

The demographic characteristics of the case and control groups were comparable for age and sex, with no statistically significant difference in age between the two groups ($p = 0.335$). This is consistent with earlier reports that neutrophil concentration is influenced by age and ethnicity, and that the degree

of neutropenia varies across ethnic groups (Denic *et al.*, 2016).

The mean serum selenium level of the neutropenic cases was lower than that of the healthy controls (70.59 ± 13.92 vs 83.40 ± 16.46 ng/mL), although this difference was not statistically significant ($p = 0.648$). This numerically lower selenium level may reflect selenium's supportive role in both innate immune cells, such as macrophages and neutrophils, and adaptive immune cells, such as T- and B-lymphocytes (Arthur *et al.*, 2003). A comparable pattern has been reported in patients with cancer, whose hair selenium concentrations were lower than those of healthy controls (Ozgen *et al.*, 2007). Population-specific reference data for serum selenium remain scarce in Kwara State; the most closely comparable Nigerian data, drawn from an elderly cohort in Owerri, similarly suggest that locally derived reference intervals may differ from those established in Western populations (Okwara & Wakwe, 2023), reinforcing the need for larger, region-specific normative case-control studies.

No statistically significant relationship was observed between ANC and selenium level ($p = 0.648$; Table 2). This finding is broadly consistent with a previous study that examined the relationship between selenium level and complete blood count parameters, which reported significant positive correlations between selenium level and both haemoglobin concentration ($r = 0.43$, $p = 0.001$) and

platelet count ($r = 0.76$, $p = 0.01$), with no correlation between selenium level and white blood cell count (El-Mazary *et al.*, 2015).

Evaluation of the relationship between serum selenium level and ANC using both Pearson chi-square and linear regression models likewise showed no statistically significant relationship ($p = 0.847$ and $p = 0.873$, respectively) in the present study. This is in agreement with a study conducted in Korea, in which selenium levels of critically ill children were correlated with white blood cell count, platelet count, and haemoglobin concentration; only platelet count showed a significant negative correlation with selenium level ($r = -0.378$, $p = 0.003$), while no significant relationship was found between selenium level and either total white blood cell count ($r = -0.094$, $p = 0.472$) or haemoglobin concentration ($r = -0.124$, $p = 0.342$) (Kim *et al.*, 2013).

CONCLUSION

This case-control study found no statistically significant relationship between serum selenium level and absolute neutrophil count among neutropenic Nigerian adults attending Kwara State University Teaching Hospital, nor a statistically significant difference in serum selenium between cases and controls. Although a numerically lower selenium level was observed among neutropenic subjects, the difference relative to controls was not statistically significant. These findings suggest limited justification for routine serum selenium assessment as a standalone marker of neutropenia risk in this population, and indicate a need for larger, adequately powered, longitudinal, and mechanistic (e.g. selenoprotein and glutathione-peroxidase-activity) case-control studies to clarify whether selenium status contributes causally to neutrophil turnover in Nigerian adults.

Consent for publication

All authors have reviewed and approved the final manuscript for submission and publication.

Data availability

The datasets generated and/or analysed during the current study are available from the corresponding author on reasonable request.

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Conflicts of interest

The authors declare no conflict of interest. The sponsors had no role in the design, execution, interpretation, or writing of the study.

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