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Original Research

Correlations of Serum Zinc with NLR, Lymphocyte Count and CRP in Tripoli, Libya: A Retrospective Analysis

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ABSTRACT:

Zinc is an essential micronutrient vital for immune function. Its deficiency can lead to immune dysregulation, but data from North African populations are scarce. This research aimed to investigate the correlation between serum zinc concentrations and primary immuno-inflammatory markers, including C-reactive protein (CRP), absolute lymphocyte count, and the Neutrophil-to-Lymphocyte Ratio (NLR), in an adult population in Tripoli, Libya. A retrospective cross-sectional analysis was performed utilizing data from 112 adult participants at Tripoli Medical Center. Serum zinc levels, complete blood count with differential, and C-reactive protein levels were documented. The NLR is determined according to the ratio of the absolute neutrophil count to the absolute lymphocyte count. The relationships among the variables were examined using Pearson's correlation analysis, while group differences according to zinc status were evaluated through an independent samples t-test. Participants had an average age of 32.3±13.8 years. The analysis indicated a substantial negative association between serum zinc concentration and NLR ($r = -0.485$, $p < 0.001$). Conversely, serum zinc concentration demonstrated a considerable affirmative association with the

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absolute lymphocyte count (r = 0.398, p = 0.002). When stratified by zinc status, the zinc-deficient group (< 70 µg/dL) exhibited a significantly higher mean NLR (3.9 vs. 2.4, p < 0.001) and a decreased mean lymphocyte count (1.6 vs. 2.3 x10³/µL, p = 0.004) compared to the zinc-sufficient group. Lower serum zinc levels are strongly associated with an elevated NLR and reduced lymphocyte counts, indicating a pro-inflammatory and immune-compromised state. These results highlight how crucial it is to maintain sufficient zinc levels for immunological homeostasis. Screening for zinc deficiency may be clinically relevant in the administration of inflammatory disorders in the Libyan population.

KEYWORDS: Zn deficiency, NLR, CRP, Hematology, Inflammatory, Libya.

INTRODUCTION

Zinc is a very important trace element that is involved in many biological processes, especially in keeping the immune system healthy as we get older. Zinc is essential for the immune system to stay healthy and work properly. It has an impact on antioxidant defense systems, cell division, and DNA synthesis, all of which are important for the immune system to work properly. Zinc is required for the proper functioning of important immune cells, like natural killer (NK) cells, lymphocytes, neutrophils, and macrophages. These cells work together to keep the immune system strong and ready to fight off infections (Wessels et al., 2021). Unlike specific nutrients such as fat-soluble vitamins or iron, which the body can preserve for prolonged durations, zinc does not have a specialized storage mechanism (Ravisankar et al., 2015). Consequently, modifications in zinc absorption, retention, or excretion can promptly result in zinc deficiency. Accordingly, the body requires a consistent diet to sustain sufficient amounts for essential physiological processes (Calder, 2025). Zinc deficiency, in particular, causes a variety of immunological abnormalities, such as lymphopenia, dysregulation and impairment of both adaptive and innate immunity, and rais vulnerability to infectious diseases, because the immune system's growth and function depend on zinc (Wong et al., 2015). This is particularly alarming for vulnerable populations, who may encounter difficulties in achieving sufficient zinc intake due to factors such as diminished appetite, alterations in taste, dietary limitations, and compromised dental health (Rudzińska et al., 2023; Leach et al., 2025; Schulz and Rink, 2025).

Recent research indicates that zinc supplementation may serve as an effective strategy to address immune dysregulation. Zinc modulates several essential immune pathways, enhancing neutrophil chemotaxis (Kuźmicka et al., 2021), increasing the NK cells' cytotoxic activity (Amling et al., 2025), helping macrophages carry out their phagocytic activity (Monteith and Skaar, 2021), and maintaining B and T cell responsiveness (Rolles et al., 2021), which are fundamental elements of adaptive immunity. Zinc's antioxidant properties play a significant role in its protective function against oxidative stress (Kaltsas, 2023). Zinc influences essential gene expression pathways, such as nuclear factor kappa-light-chain-enhancer of activated B-cells (NF-κB), and peroxisome proliferator-activated receptor (PPAR) signaling, which are essential for controlling inflammatory reactions and enhancing cellular toughness. Zinc may be able

to reduce oxidative stress and systemic inflammation by modifying these pathways, thereby promoting the health and functionality of immune cells within both adaptive and innate immune responses (Jarosz et al., 2017; Li et al., 2025).

Zinc deficiency induces lymphopenia by disrupting lymphocyte proliferation, differentiation, and T-cell homeostasis, resulting in heightened apoptosis and diminished adaptive immune function. This deficiency disrupts neutrophil function, increasing neutrophil counts and leading to a higher neutrophil-to-lymphocyte ratio (NLR) (Perestiuk et al., 2025). An increased NLR indicates systemic inflammation and disease severity, exhibiting a negative correlation with serum zinc levels in chronic kidney disease (Kim et al., 2018). At the same time, a lack of zinc weakens the inhibition of NF-κB, which increases the production of C-reactive protein (CRP) driven by IL-6 and shows significant negative correlations with CRP levels through Spearman's analysis (Perestiuk et al., 2025).

It is commonly known that zinc and immunological function are related; however, there is a paucity of data from North African populations, particularly in Libya. Given the unique dietary habits and the potential for micronutrient deficiencies in the area, investigating these correlations is of substantial public health importance. This study investigates the connection between serum zinc levels and critical immune and inflammatory indicators, namely CRP, lymphocyte count, and the Neutrophil-to-Lymphocyte Ratio (NLR), within a sample of the adult population in Tripoli, Libya.

MATERIALS AND METHODS

Research Methodology

A cross-sectional study was performed, employing retrospective data from individuals who visited outpatient clinics and laboratories at Tripoli Medical Center in Tripoli, Libya, for routine health check-ups between 2023 and 2024.

Sampling

The study comprised data from adult males and females (aged 18 years and older) for whom results for serum zinc, complete blood count (CBC) with differential, and/or C-reactive protein (CRP) were accessible. To reduce confounding variables, individuals with identified hematological malignancies, acute severe infections, or those undergoing immunosuppressive therapy were excluded. The investigation complied with the ethical principles outlined in the Declaration of Helsinki (as revised in 2000) and obtained approval from the institutional review board.

Data Collection and Laboratory Analysis

Test records were used to collect demographic information (age and gender) as well as test results. Trained phlebotomists collected venous blood samples from all subjects, the samples were permitted to clot and then centrifuged at 4000 rpm for 10 minutes to isolate the serum. The laboratory testing included the following:

- **Serum Zinc** Measurement: Serum zinc concentrations were measured utilizing atomic absorption spectrophotometry. A colorimetric zinc (Zn) assay reagent (Elabscience E-BC-K137-M, USA) was utilized for the analysis. Results were reported in micrograms per deciliter (μg/dL).
- **Hematological Analysis**: A complete blood count (CBC) with a 5-part differential was performed utilizing an automated hematological analyzer (e.g., Sysmex XN-series).
- **Absolute counts** of total white blood cells (WBC), neutrophils, and lymphocytes were measured (expressed as $x10^3/\mu L$).
- **Inflammatory marker measurements**: Serum C-reactive protein (CRP) levels

were measured using a high-sensitivity immunoturbidimetric technique. Results were expressed in milligrams per liter (mg/L).

- **Variable Calculation**: Each subject with available differential counts had their Neutrophil-to-Lymphocyte Ratio (NLR) computed by calculating the ratio of the absolute neutrophil count to the absolute lymphocyte count.

Statistical Analysis

SPSS software (Version 26.0) was employed for all statistical analyses. The attributes of the study population were summarized utilizing descriptive statistics (mean ± standard deviation for continuous variables), and the corresponding frequencies and percentages for categorical variables). Upon confirming that the data followed a normal distribution, The relationship between serum zinc levels and other continuous variables (CRP, lymphocyte count, NLR) was assessed using Pearson's correlation coefficient (r). Spearman's rank correlation was utilized for data that did not conform to a normal distribution. The definition of statistical significance is a p-value < 0.05.

RESULTS

Demographic and Baseline Laboratory Characteristics of the Research Cohort

The conclusive assessment comprised 112 participants in total. Table 1 shows the study population's demographic and baseline lab results. The average age of the people who took part was 32.2± 13.82 years old, with a minimum age of 18 years. The study population was mostly female, with 76.8% of the participants being female and 23.2% being male.

The group's average serum zinc level was 98.5 ± 70.1 µg/dL. As for, the average number of lymphocytes was 2.1 ± 0.9 x10³/µL, while the average Neutrophil-to-Lymphocyte Ratio (NLR) was 2.8 ± 1.5. The average CRP for the group of participants with data (n=12) was 6.4 ± 5.1 mg/L.

Table 1: Demographic and Laboratory laboratory characteristics of the research cohort at Baseline.

Characteristic		Mean ± SD
Age (years)		32.3±13.8
Sex	**Male**	26.0 (23.2%)
	Female	86.0 (76.8%)
	Total	112.0
Laboratory Parameters		
Serum Zinc (µg/dL)		98.5 ± 70.1
WBC Count (x10³/µL)		7.6 ± 2.5
Absolute Neutrophil Count (x10³/µL)		4.5 ± 1.8
Absolute Lymphocyte Count (x10³/µL)		2.1 ± 0.8
Neutrophil-to-Lymphocyte Ratio (NLR)		2.8 ± 1.5
C-Reactive Protein (CRP, mg/L) (n=14)		6.4 ± 5.1

SD: Standard Deviation; WBC: White Blood Cell

Correlation between Serum Zinc and Immuno-Inflammatory Markers

The correlation between serum zinc concentration and NLR, absolute lymphocyte count and C-reactive protein (CRP) level were analyzed using Pearson's correlational method-based evaluation and illustrated in Table 2. The most significant finding was the strong negative correlation between serum zinc levels and the Neutrophil-to-Lymphocyte Ratio ($r = -0.485$; $p < 0.001$). This indicates that as serum zinc declines NLR typically increases, indicating that the body is tilting towards a pro-inflammatory status.

On the other hand, serum zinc levels were substantially and favorably correlated with the absolute lymphocyte count (r = 0.398, p = 0.002), suggesting that low zinc was associated with lower numbers of circulating lymphocytes.

On the contrary, a significantly negative correlation existed between serum zinc and CRP levels in the subset of patients with available CRP data (r = -0.512, p = 0.087) as indicated in Fig 1, although this finding was not statistically significant due to the small sample size.

Table 2: Correlation between Serum Zinc with NLR, Absolute Lymphocyte Count and CPR.

Statistics	NRL	absolute lymphocyte	CPR
R value	-0.485	0.398	-0.512
P value	< 0.001	0.002	0.087

Comparison of Inflammatory Markers based on Zinc Status

Zinc deficiency was identified in 30 participants (26.8%), while optimal zinc levels were found in 82 participants (73.2%). Table 3 shows that the zinc-deficient group (n=30) had a significantly lower mean absolute lymphocyte count (1.6 ± 0.7 × 10^3/µL) relative to the zinc-sufficient group (2.3 ± 0.8 × 10^3/µL; p = 0.004) and a markedly higher mean NLR (3.9 ± 1.6 vs. 2.4 ± 1.1; p < 0.001).

Table 3: Comparison of Immuno-Inflammatory Markers by Zinc Status.

Parameters	Zinc Deficient (< 70 µg/dL) (n=30)	Zinc Sufficient (≥ 70 µg/dL) (n=82)	*p* value
Absolute Lymphocyte	1.6±0.7	2.3±0.8	0.004
NLR	3.9±1.6	2.4±1.1	< 0.001

Values are expressed as Mean ± SD. P-values were determined using an independent samples t-test

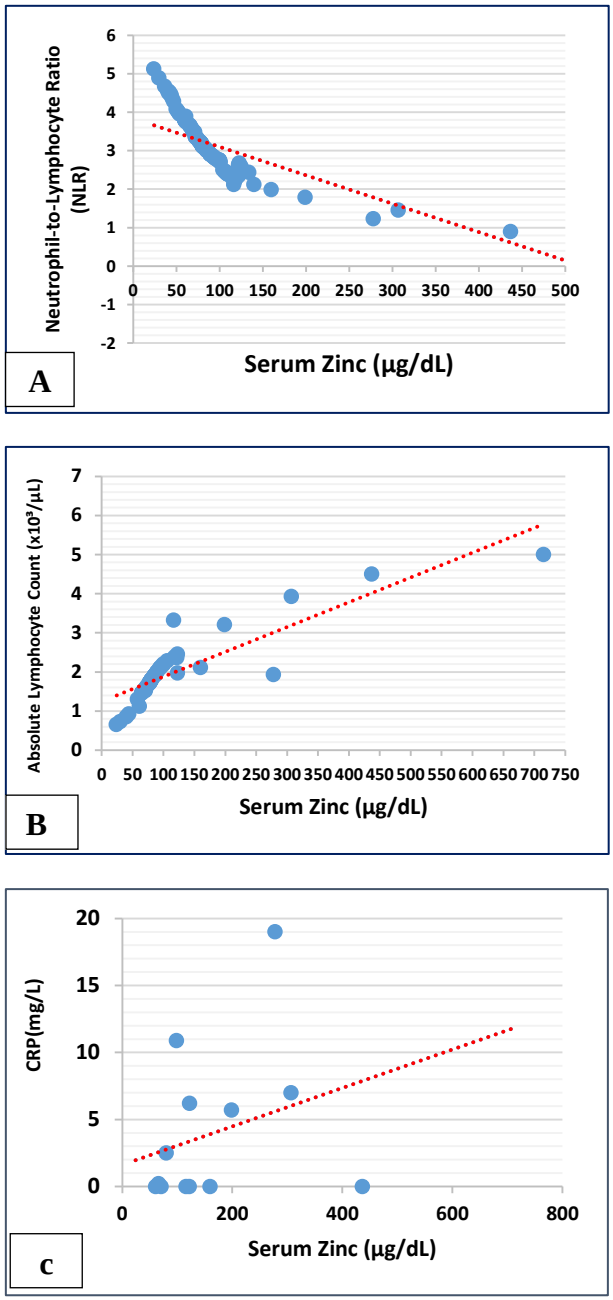

Fig (1): Scatter plot with a fitted regression line depicting the correlation among zinc levels with NLR, absolute lymphocyte count, and CRP in patients. Every single navy blue dot represents a unique piece of patient data, illustrating that patient's assessed NLR, absolute lymphocyte count, and CRP levels, with related zinc level. The red line represents the line of greatest fit (regression line), which encapsulates the general trend or association among NLR, absolute lymphocyte count, and CRP, with zinc levels.

DISCUSSION

This cross-sectional study, which included adult from Medical Tripoli Center patients in the city of Tripoli, Libya, gives local evidence to support a key role of zinc in immune regulation and inflammation. The prominent observation in our study, is the strong negative correlation between serum zinc level vs NLR and conversely positive association of zinc with absolute lymphocyte doucsnt implying immune balancing immunomodulatory role of this trace element in mediating immuno-inflammatory response. The NLR, now widely accepted as a strong indicator of systemic inflammation and physiologic stress, represents dual immunological disruption—neutrophilia and relative lymphopenia—that is driven by zinc deficiency, favoring neutrophil predominance, lymphoid suppression, and pro-inflammatory innate functions that are not effectively counter-regulated by adaptive immunity.

The findings highlight the essential immunomodulatory activity of zinc, affirming that a negative trend in anti-inflammatory markers such as CRP, albeit modest statistical significance, corresponds with the established biological relevance of this mineral. These results validate our hypothesis that lower zinc concentrations reflect a state of immune dysregulation characterized by systemic inflammation and impaired adaptive immunity.

Zinc serves as a "gatekeeper" of immune function (Wessels et al., 2021). Zinc modulates innate and adaptive immunity by obstructing NF-κB signaling, diminishing neutrophil activation, and enhancing lymphocyte proliferation and functionality (Bonaventura et al., 2015). According to (Gammoh and Rink, 2017), zinc obstructs the dissociation of NF-κB from its bound inhibitory protein, thereby obstructing the nuclear translocation of NF-κB, so mitigating future inflammation. Deficiency disrupts the equilibrium of T helper cells, resulting in an increase in NLR due to Th2 skewing and M2 macrophage impairment, while concurrently lowering cytokines that reduce inflammation, like IL-4 (Gammoh and Rink, 2017; Kido et al., 2019). This elucidates the pro-inflammatory shift ($r = -0.485$ for NLR, $p < 0.001$). Our findings are consistent with a growing body of global literature identifying zinc deficiency as a primary driver of elevated NLR. Recent studies, including those on hospitalized patients with COVID-19, have mirrored this inverse relationship, suggesting that zinc status is a reliable predictor of the systemic immune-inflammatory state (Zhou et al., 2022; Perestiuk et al., 2025). The NLR is widely recognized as a robust marker of physiological stress; the observed elevation in our zinc-deficient cohort reflects a dual immunological imbalance: neutrophilia, driven by uncurbed innate activation, and relative lymphopenia, resulting from impaired lymphocyte maturation.

Despite the negative association between serum zinc and C-reactive protein (CRP) not achieving statistical significance ($p = 0.087$), the observed trend ($r = -0.512$) is consistent with the recognised anti-inflammatory capabilities of zinc. The absence of significance is probably a Type II error due to the restricted sample size (n=14) for CRP analysis. Nevertheless, meta-analyses have confirmed that zinc supplementation effectively reduces circulating CRP levels by inhibiting pro-inflammatory cytokine signaling, particularly IL-6 (Ceylan et al., 2021). As noted by (Kim et al., 2018), patients with zinc deficiency demonstrated elevated levels of C-reactive protein ($p < 0.001$) and increased corticosteroid usage ($p = 0.04$). An investigation by (Razeghi Jahromi et al., 2021) revealed that there was an association between increased zinc levels and falling serum CRP levels.

This research appears to be among the initial inquiries into these specific immunological interactions in a Libyan environment. The findings are especially pertinent to the region, where eating habits may

predispose people to micronutrient deficiencies. The immunological dysregulation profile identified in zinc-deficient individuals could have serious public health consequences, including greater susceptibility to infections and a higher burden of chronic inflammatory disorders.

CONCLUSION

This study demonstrates a notable association between diminished serum zinc concentrations and indicators of systemic inflammation (NLR) and adaptive immune suppression (lymphopenia) in people residing in Tripoli, Libya. These findings underscore the critical importance of zinc in sustaining immunological equilibrium. We advocate for the incorporation of regular zinc screening in clinical practice, especially for people predisposed to inflammatory diseases. Future prospective and longitudinal studies are essential to investigate the causal link and possible advantages of targeted zinc supplementation in mitigating chronic inflammation and improving immunological resilience in the Libyan population.

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علاقة الزنك بنسبة العدلات NLR مع عدد الخلايا اللمفاوية وبروتين سي التفاعليCRP في المصل: تحليل استعادي طرابلس ، ليبيا

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الملخص

يعتبر الزنك عنصرًا غذائيًا دقيقًا حيويًا لوظائف الجهاز المناعي، وقد يؤدي نقصه إلى اختلال التنظيم المناعي، غير أن البيانات المتعلقة بسكان شمال أفريقيا في هذا الشأن لا تزال ضعيفة لذلك هدف البحث إلى دراسة العلاقة بين تركيزات الزنك في المصل والمؤشرات المناعية-الالتهابية الأساسية، بما في ذلك بروتين سي التفاعلي، والعدد المطلق للخلايا الليمفاوية، ونسبة العدلات إلى الخلايا الليمفاوية، لدى عيّنة من البالغين في مدينة طرابلس، ليبيا. حيثأ ُجري تحليل استعادي مقطعي باستخدام بيانات 112 مشاركًا بالغًا من مركز طرابلس الطبي، حيث تم توثيق مستويات الزنك في المصل، وبروتين سي التفاعلي، وتعداد الدم الكامل مع التعداد التفريقي. وتُحسب نسبة العدلات إلى الخلايا الليمفاوية وفقًا لنسبة العدد المطلق للعدلات إلى العدد المطلق للخلايا الليمفاوية واستُخدم تحليل ارتباط بيرسون لدراسة العلاقات بين المتغيرات، في حين قُيِّمت الفروق بين المجموعات وفقًا لحالة الزنك باستخدام اختبار (t)

بارتفاع نسبة العدلات إلى الخلايا الليمفاوية وانخفاض عدد الخلايا الليمفاوية، مما يشير إلى حالة مؤيدة للالتهاب ومصحوبة بضعف مناعي. وتُبرز هذه النتائج مدى أهمية الحفاظ على مستويات كافية من الزنك للحفاظ على التوازن المناعي وقد يكون فحص نقص الزنك ذا أهمية سريرية في إدارة الاضطرابات الالتهابية لدى السكان الليبيين.

الكلمات المفتاحية: نقص الزنك؛ نسبة العدلات إلى الخلايا الليمفاوية؛ بروتين سي التفاعلي، أمراض الدم؛ الالتهاب؛ ليبيا.

للعينات المستقلة. بلغ متوسط عمر المشاركين 32.3 ± 13.8 عامًا. وأظهر التحليل وجود ارتباط سلبي قوي بين تركيز الزنك في المصل ونسبة العدلات إلى الخلايا الليمفاوية ($r = -0.485$، $p < 0.001$). في المقابل، أظهر تركيز الزنك في المصل ارتباطًا إيجابيًا ملحوظًا مع العدد المطلق للخلايا الليمفاوية ($r = 0.398$، $p = 0.002$). وعند تصنيف المشاركين وفقًا لحالة الزنك، أظهرت مجموعة نقص الزنك (أقل من 70 ميكروغرام/ديسيلتر) متوسطًا أعلى بشكل معنوي لنسبة العدلات إلى الخلايا الليمفاوية (3.9 مقابل 2.4، $p < 0.001$)، وانخفاضًا في متوسط عدد الخلايا الليمفاوية (1.6 مقابل 2.3 × 10^3/ميكرولتر، $p = 0.004$) مقارنة بمجموعة الزنك الكافي و ترتبط مستويات الزنك المنخفضة في المصل ارتباطًا قويًا