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Original Article

Correlation Between Neutrophil Lymphocyte Ratio (NLR) Values And Igm Anti-*Salmonella* Results In Suspected Typhoid Fever Patients

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ABSTRACT

In Indonesia, typhoid fever remains a significant health problem and often presents with non-specific clinical symptoms, making laboratory investigations essential to support diagnosis. Common laboratory parameters include the Neutrophil Lymphocyte Ratio (NLR) as an indicator of inflammatory response and IgM anti-*Salmonella typhi* as a marker of humoral immune response. This study aimed to determine the correlation between NLR values and IgM anti-*Salmonella* test results in suspected typhoid fever patients as a diagnostic support. This study was an analytical observational study with a cross-sectional approach. A total of 30 suspected typhoid fever patients were selected using purposive sampling at dr. Sayidiman Magetan regional hospital. NLR was measured using a Sysmex XN-1000 hematology analyzer, while IgM anti-*Salmonella* was examined using a rapid immunochromatographic test. The result showed that most patients were in the mild infection category (60%), with 40% testing positive and 60% negative for IgM anti-*Salmonella*. Spearman rank correlation analysis revealed a significance value (*p*) of 0.000 and a correlation coefficient (r) of 0.849, indicating a very strong positive correlation. This means that the relationship is directly proportional, where higher NLR values tend to be associated with positive IgM anti-*Salmonella* results. In conclusion, there is a statistically significant positive correlation between NLR values and IgM anti-*Salmonella*, suggesting that both parameters can be used as complementary laboratory tests in the early diagnosis of typhoid fever.

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INTRODUCTION

Typhoid fever is a systemic infectious disease caused by *Salmonella typhi* and remains a public health challenge in many countries, particularly in developing regions. Transmission of this disease occurs via the fecal-oral route through the consumption of

contaminated food or beverages, facilitated by poor sanitation and hygiene conditions, as well as by asymptomatic carriers who sustain the chain of transmission in the community[1,2,3]. The clinical symptoms of typhoid fever including prolonged fever, gastrointestinal disturbances, and other systemic symptoms often resemble those of other infectious diseases, making clinical diagnosis difficult[4].

In *Salmonella typhi* infection, the activation of the immune response causes changes in hematological parameters as part of the body's defense mechanism against infection. This inflammatory response is characterized by an increase in neutrophils and a decrease in lymphocytes, resulting in an elevated Neutrophil-to-Lymphocyte Ratio (NLR)[5]. As a parameter derived from a complete blood count, the NLR is considered practical, easy to calculate, and has the potential to serve as a biomarker of inflammation in various bacterial infections, including typhoid fever [6].

In addition to triggering an inflammatory response, *Salmonella typhi* infection also activates a humoral immune response that produces immunoglobulin M (IgM) antibodies the first antibodies to form during the acute phase of infection[7]. The rapid test for *IgM* anti-*Salmonella* is widely used as a supportive test because it can detect infection quickly, easily, and relatively simply[8]. However, IgM test results can be influenced by the timing of antibody formation; therefore, their interpretation must be supported by other laboratory parameters that can characterize the inflammatory response during the course of the infection[9].

Nevertheless, hematological parameters such as *the* Neutrophil Lymphocyte Ratio (NLR) and immunological tests for IgM anti-*Salmonella typhi* are generally still used separately to aid in the diagnosis of typhoid fever. In fact, an elevated NLR value as an indicator of an inflammatory response and the formation of IgM anti-*Salmonella typhi* as a marker of the humoral immune response both represent the body's response to *Salmonella typhi* infection, suggesting a potential correlation between the two. However, research on the relationship between Neutrophil Lymphocyte Ratio (NLR) values and *anti-Salmonella Typhi* IgM test results in patients with suspected typhoid fever remains limited.
Based on the above, this study aims to analyze the correlation between the Neutrophil Lymphocyte Ratio (NLR) and *IgM* anti-*Salmonella* test results in patients with suspected typhoid fever at Dr. Sayidiman Magetan Regional General Hospital.

METHODS

This was an analytical observational study with a *cross-sectional* design aimed at determining the relationship between the Neutrophil Lymphocyte Ratio (NLR) and *IgM* anti-*Salmonella* test results in patients with suspected typhoid fever. The study was conducted from

09 to 31 March 2026 at the Central Clinical Pathology Laboratory of Dr. Sayidiman Magetan Regional General Hospital.

The study population consisted of all patients with suspected typhoid fever aged ≥10 years, including both outpatients and inpatients, who underwent a complete blood count and *IgM* anti-*Salmonella* testing at Dr. Sayidiman General Hospital in Magetan. The age limit of ≥10 years was determined based on a preliminary survey conducted at the research site, which showed that patients with suspected typhoid fever undergoing complete blood count and *IgM* anti-*Salmonella* testing were predominantly aged 10 years or older. Therefore, this age range was selected to reflect the characteristics of the patient population available at the research site. The study sample was selected using *purposive sampling* based on predetermined inclusion and exclusion criteria. Inclusion criteria were patients with a clinician's diagnosis of suspected typhoid fever and those who underwent rapid *IgM* anti-*Salmonella* testing. Exclusion criteria included patients with comorbidities such as chronic kidney disease, diabetes mellitus, or HIV/AIDS, as well as patients with symptoms inconsistent with suspected typhoid fever.

The research materials used consisted of whole blood and serum samples obtained from patients with suspected typhoid fever. The equipment used included 3 mL syringes, K3EDTA Vacutainer tubes, plain Vacutainer tubes, tourniquets, alcohol swabs, dry cotton swabs, a centrifuge, a Sysmex XN-1000 Hematology Analyzer, micropipettes, an *IgM* anti-*Salmonella* rapid test kit, and a timer.

The research procedure began with venous blood collection using two types of Vacutainer tubes. A total of 3 mL of blood was collected in a K3EDTA tube for hematological testing, and 3 mL in a plain tube for serological testing. The sample in the K3EDTA tube was homogenized and analyzed on the Sysmex XN-1000 Hematology Analyzer to determine neutrophil and lymphocyte counts. The sample in the plain tube was centrifuged at 3000 rpm for 10 minutes to obtain serum. The serum was then used for *IgM* anti-*Salmonella* testing using a rapid test method, with 35 µL of serum and 3 drops of buffer applied to the test device. Results were read after 15–20 minutes of incubation.

Measurements were taken of the Neutrophil Lymphocyte Ratio (NLR) and the results of the IgM anti-*Salmonella* test. The NLR value was obtained by dividing the neutrophil count by the lymphocyte count based on the complete blood count results. The Neutrophil Lymphocyte Ratio (NLR) values were categorized into three levels of infection severity: mild infection (NLR < 3.00), moderate infection (NLR 3.00–5.00), and severe infection (NLR > 5.00)[10].

The IgM anti-*Salmonella* test result was interpreted based on the appearance of colored lines in the control (C) and test (T) areas. A positive result was indicated by the appearance of two red colored lines in both the control (C) and test (T) areas. A negative result was indicated by the

appearance of one colored line in the control (C) only. The result was considered invalid if no two lines appeared in the control (C) and test (T) areas, or if only one line appeared in the test (T) area without a line in the control (C) area.

We analyzed the relationship between NLR values and IgM anti-Salmonella test results using the Spearman rank correlation test in the Statistical Package for the Social Sciences (SPSS). The strength of the correlation was interpreted based on the correlation coefficient (r), with values of 0.00–0.199 classified as very weak, 0.20–0.399 as weak, 0.40–0.599 as moderate, 0.60–0.799 as strong, and 0.80–1.000 as very strong[11]. A positive correlation indicated that higher NLR values were associated with a higher likelihood of positive IgM anti-*Salmonella* results, whereas a negative correlation indicated an inverse relationship. A significance level of $\alpha = 0.01$ was used, and a p-value < 0.01 was considered statistically significant.

RESULTS

A total of 30 patients with suspected typhoid fever who met the inclusion and exclusion criteria were included in this study. Respondent characteristics included age and sex. In addition, Neutrophil Lymphocyte Ratio (NLR) and *IgM* anti-*Salmonella* tests were performed to determine the relationship between the two study variables.

Table 1. Characteristics of the Study Participants

Characteristics	Category	Number (n)	Percentage (%)
Age	10 – 18 years old	2	6,7
	19 – 59 years old	21	70
	> 59 years old	7	23,3
	Total	**30**	**100**
Gender	Male	13	43.3
	Female	17	56,7
	Total	**30**	**100**

Table 1 shows the characteristics of patients suspected of having typhoid fever based on age and gender. The age distribution was dominated by adults (19–59 years old), comprising 21 individuals, followed by the elderly group (>59 years old) with 7 individuals and the adolescent group (10–18 years old) with 2 individuals. Based on gender, female patients were predominant, totaling 17 individuals, while male patients totaled 13 individuals. The results of the Neutrophil Lymphocyte Ratio (NLR) test are presented in Table 2.

Table 2. Neutrophil Lymphocyte Ratio (NLR) Test Results for Patients with Suspected Typhoid Fever at Dr. Sayidiman Magetan Regional General Hospital

Result	Level Severity	Number	Percentage (%)
Mild Infection	< 3,00	18	60
Moderete Infection	3,00 - 5,00	3	10
Severe Infection	> 5,00	9	30
Total		**30**	**100**

The results in Table 2 show that the majority of patients with suspected typhoid fever were in the mild infection category, totaling 18 patients (60%); 9 patients (30%) were in the severe infection category; and 3 patients (10%) were in the moderate infection category. The results of the *IgM* anti-*Salmonella* testing conducted on patients with suspected typhoid fever at the *central* laboratory of Dr. Sayidiman Magetan Regional General Hospital are shown in Table 3.

Table 3. Results of IgM anti-Salmonella Testing in Patients with Suspected Typhoid Fever at Dr. Sayidiman Magetan

Results	Number	Percentage (%)
Positive	12	40
Negative	18	60
Total	**30**	**100**

Table 3 shows that 18 people (60%) tested negative for *IgM* anti-*Salmonella*, while 12 people (40%) tested positive.

Table 4. Results of the Spearman Rank Correlation Test

Variable	Significance Value (*p-Values*)	Correlation Coefficient (r)
NLR	0.000	0.849
IgM anti-Salmonella	0.000	0.849

The results of the Spearman rank correlation test indicate a significant relationship between the NLR and the *IgM* anti-*Salmonella* test results ($p = 0.000$; $p < 0.01$). The correlation coefficient of 0.849 indicates a positive correlation with a very strong relationship, suggesting that an increase in the NLR value tends to be followed by an increased likelihood of a positive *IgM* anti-*Salmonella* result.

DISCUSSION

Based on the study results, the majority of respondents were in the adult age group (19–59 years), who have high mobility and are therefore at greater risk of exposure to food or beverages contaminated with *Salmonella typhi*[1]. There were more female respondents than

male respondents. However, this difference likely reflects the characteristics of the sample, as the incidence of typhoid fever is more influenced by environmental factors, behavior, and sanitation than by gender[12]. The variation in NLR values among patients with suspected typhoid fever may reflect differences in the systemic inflammatory response during infection. Lower NLR values may indicate that the systemic inflammatory response was not yet pronounced, whereas higher NLR values may reflect greater activation of the inflammatory response as the infection progresses[13,14].

In *Salmonella Typhi* infection, lipopolysaccharide (LPS) from the bacterial cell wall is a key stimulus of the innate immune response, recognized by Toll-like receptor 4 (TLR-4) on immune cells. Activation of this receptor stimulates the release of pro-inflammatory cytokines, including tumor necrosis factor-alpha (TNF-α), interleukin-1 (IL-1), and interleukin-6 (IL-6), which contribute to the activation of the inflammatory response[15]. Neutrophils are subsequently activated as part of the innate immune response and eliminate bacteria through phagocytosis. An increase in neutrophil numbers, accompanied by a decrease in lymphocytes due to redistribution to infected tissues and systemic inflammatory stress, can result in an increased NLR[6]. Therefore, higher NLR values may reflect greater activation of the systemic inflammatory response.

The NLR value in this study represents the ratio of neutrophil to lymphocyte counts obtained from peripheral blood. Therefore, an increase in NLR reflects changes in circulating leukocyte profiles associated with the inflammatory response[10]. Although neutrophils play an important role in the innate immune response, the NLR measurement itself does not directly measure the recruitment of neutrophils into infected tissues. Rather, changes in circulating neutrophil and lymphocyte numbers may reflect the systemic inflammatory response occurring during infection. This may explain the variation in NLR values observed among the patients in this study.

Testing for *IgM* anti-*Salmonella* revealed that 12 participants (40%) tested positive, while 18 participants (60%) tested negative. The predominance of negative results in this study was likely due to testing being conducted during the early phase of infection, when IgM antibodies had not yet been optimally produced or were still within *the window period*[7]. In *Salmonella typhi* infection, IgM antibodies generally become detectable around days 3 to 5 after the onset of clinical symptoms as part of the humoral immune response[16,17]. Therefore, patients who have experienced an inflammatory process may not necessarily show a positive IgM result immediately if antibody production has not yet reached the detection limit of the test. Conversely, a positive IgM result indicates that an adaptive immune response has occurred and the body has produced specific antibodies against the *Salmonella typhi* antigen[18].

The formation of IgM antibodies is part of the specific humoral immune response to *Salmonella typhi* infection. Bacteria that have been phagocytosed by macrophages undergo antigen processing, and the resulting antigen is presented through major histocompatibility complex class II (MHC II) molecules. The antigen-MHC II complex is then recognized by CD4+ T helper cells, which play an important role in initiating the adaptive immune response. Activation of T helper cells stimulates the proliferation and differentiation of B lymphocytes into plasma cells. These plasma cells subsequently produce antigen-specific antibodies, particularly immunoglobulin M (IgM) during the early humoral immune response[19]. Thus, the appearance of IgM anti-*Salmonella* indicates that a specific humoral immune response has developed against *Salmonella typhi* antigens.

The timing of this humoral response is important when interpreting the IgM results. Because antibody production develops after the initiation of the immune response, patients examined at an early stage of infection may already have an inflammatory response detectable through hematological parameters, while the concentration of specific IgM antibodies may still be insufficient to produce a positive rapid-test result[7]. Conversely, a positive IgM result indicates that the adaptive immune response has progressed sufficiently for specific antibodies to become detectable[18]. Therefore, the negative IgM results observed in some patients in this study may not necessarily indicate the absence of an inflammatory response, particularly when testing is performed during the early phase of infection.

The Spearman's rank correlation test revealed a significant association between NLR values and *IgM anti-Salmonella* results ($p = 0.000$), with a correlation coefficient of 0.849, indicating a positive relationship with a very strong correlation[11]. These findings suggest that the higher the NLR value, the greater the likelihood of a positive *IgM* anti-*Salmonella* result. This relationship suggests a link between the inflammatory and humoral immune responses during typhoid fever. An increase in the NLR reflects further activation of the inflammatory response, whereas the appearance of IgM antibodies indicates that the adaptive immune system has been activated to fight the infection[20 21].

The results of this study are consistent with the findings of Zerlinda et al. (2023), who reported an association between NLR levels and IgM/IgG antibody expression in patients with typhoid fever[22]. These consistent results indicate that NLR and IgM are interrelated parameters in characterizing the development of the immune response to *Salmonella typhi* infection. NLR reflects changes associated with the inflammatory response, while IgM anti-*Salmonella* reflects the development of a specific humoral immune response.

However, this study has a limitation, namely the lack of more specific confirmatory diagnostic methods such as blood culture, so the accuracy of the diagnosis still depends on

the method used[23]. In addition, the timing of sample collection relative to the onset of infection was not precisely controlled, which may have influenced the IgM results, as the development of the humoral immune response can vary with the stage of infection. The cross-sectional design also limits the ability to directly observe changes in NLR and IgM over time. Therefore, the observed correlation should be interpreted as an association between the two parameters rather than evidence of a sequential or causal relationship.

Nevertheless, despite these limitations, the NLR has the potential to serve as an easy, rapid, and cost-effective supportive parameter to aid in the interpretation of *IgM* anti-*Salmonella* test results, particularly in cases where antibodies have not yet been detected due to *the window period*. This is consistent with Herlinawati's (2022) view that a negative IgM result does not always indicate the absence of typhoid infection, as rapid tests generally have high sensitivity but low specificity[24]. Therefore, the combined interpretation of NLR and IgM anti-*Salmonella* results may provide complementary information regarding the inflammatory and humoral immune responses in patients with suspected typhoid fever.

CONCLUSION

Neutrophil Lymphocyte Ratio (NLR) values in patients with suspected typhoid fever indicate varying levels of inflammation ranging from mild, moderate, to severe in response to bacterial infection. The results of the *IgM* anti-*Salmonella rapid test* showed both positive and negative results, reflecting differences in the phase of the immune response in each patient. Spearman's rank correlation analysis revealed a significant positive correlation between NLR values and *IgM* anti-*Salmonella* test results, with a very strong correlation strength. These findings suggest that an increase in the NLR value tends to be accompanied by a higher likelihood of a positive *IgM* anti-*Salmonella* result. Thus, the NLR value has the potential to be used as a supporting parameter alongside *IgM* anti-*Salmonella* testing to aid in the diagnosis of patients with suspected typhoid fever.

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AUTHOR CONTRIBUTIONS

All authors contributed to the conceptualization and design of the study, data collection, data analysis and interpretation, and the preparation and revision of the manuscript.

CONFLICT OF INTEREST

The authors declare no conflicts of interest regarding the publication of this article.

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