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Original Research Article

# The Relation between Neutrophil Lymphocyte Ratio (NLR) and Grade of Severity in Dengue Infection

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**Abstract:**

**Introduction:** Dengue virus infection is one of the public health problems resulting in social and economic impacts. The prevalence of dengue virus infection increases every year with the wide endemic area. DHF is mostly found in tropical and sub-tropical climates, especially in urban areas. It is estimated that worldwide more than 50 million infections occur each year including 500,000 hospitalizations for dengue hemorrhagic fever, mainly among children with the case fatality rate exceeding 5% in some areas.

**Objectives:** To identify the possible correlation between neutrophil to lymphocyte ratio and severity of dengue patients. Methods: This hospital based cross sectional study was conducted at Sardar Patel Medical College and Associated Group of Hospitals, Bikaner, Rajasthan. Randomly enrolled total of 141 patients of Dengue fever with NS1 positive.

**Results:** In our study we found that At day-3 majority of DF cases has NLR>3 (65.38%), majority of DHF cases has NLR between >1-≤2 (44.44%) and in DSS cases 40% has NLR>3 followed by 30% has NLR between >1-≤2, at day-6 majority of DF cases has NLR between >1-≤2 (39.42%), majority of DHF cases has NLR between >1-≤2 (48.15%) and in DSS cases 50% has NLR≤1 followed by 40% has NLR between >1-≤2 and at day-9 majority of DF cases has NLR ≤1 (36.54%), majority of DHF cases has NLR between ≤1 (70.37%) and in DSS cases 50% has NLR≤1 followed by 20% has NLR between >1-≤2 and >2-≤3. Thus, NLR was low in DSS and we found statistically significant difference in NLR with grade of severity.

**Conclusion:** The study concludes that NLR may be used as prognostic marker in management of dengue fever. There is a significant relation between NLR with grade of severity in dengue infection. This means that the lower the level of NLR, the heavier the clinical degree of dengue.

**Keywords:** Dengue, Dengue hemorrhagic fever, Neutrophil-Lymphocyte Ratio (NLR), Platlet



## Introduction

Dengue virus infection is one of the public health problems resulting in social and economic impacts. The prevalence of dengue virus infection increases every year

with the wide endemic area. DHF is mostly found in tropical and sub-tropical climates, especially in urban areas. [1] Dengue virus (DENV) belongs to the family Flaviviridae, genus Flavivirus, and is transmitted to humans by Aedes mosquitoes, mainly Aedes aegypti. Based on neutralization assay data, four serotypes (DENV-1, DENV-2, DENV-3, and DENV-4) can be distinguished. DENV infection is a major cause of disease in tropical and subtropical areas. Infection with any of the DENV serotypes may be asymptomatic in the majority of cases or may result in a wide spectrum of clinical symptoms, ranging from a mild flu-like syndrome (known as dengue fever [DF] to the most severe forms of the disease, which are characterized by coagulopathy, increased vascular fragility, and permeability (dengue hemorrhagic fever [DHF]). The latter may progress to hypovolemic shock (dengue shock syndrome [DSS]). [2] The WHO criteria for the clinical diagnosis of DHF requires the presence of acute and continuous fever of 2 to 7 days, haemorrhagic manifestations associated with thrombocytopenia (100,000 cells/ c.mm or less) and haemoconcentration (haematocrit >20% from baseline of patient or population of same age). Haemorrhagic manifestations could be mucosal and or skin or even a positive tourniquet test which is the commonest. Hepatomegaly occurs at some stage of DHF and open precedes plasma leakage and hence a valuable early predictor of plasma leakage. [3] Unlike other inflammatory biomarkers e.g., ESR and CRP, the Neutrophil–lymphocyte ratio (NLR) is derived from routine complete blood count (CBC) tests. It does not need a special request. It is also a rapid, easy method, and cost-effective. Many studies have reported an increase of NLR during the inflammatory conditions in different diseases such as pancreatitis, inflammatory bowel diseases, and acute coronary syndrome. [4–9] Neutrophil lymphocyte ratio (NLR) has been proposed as a surrogate marker for endothelial dysfunction and inflammation in distinct populations and has prognostic and predictive values. [10] Dengue severity grade is also known that it is related to the inflammation occurred on dengue infection because of the exaggerating response of the immune system. [11]

From the problems mentioned above, we will be wants to know the relation between NLCR and grade of severity in dengue infection in S.P. Medical College and A.G. of P.B.M. hospital, Bikaner (Rajasthan).

**Objectives:**

1. To determine the severity of dengue patients.
2. To estimate the neutrophil to lymphocyte ratio in dengue patients.
3. To identify the possible correlation between neutrophil to lymphocyte ratio and grade of severity in dengue patients.

**Methodology:**

**Study design:** Hospital based cross-sectional study.

**Study place**: Department of Medicine, S.P. Medical College and Associated Group of P.B.M. Hospitals, Bikaner (Rajasthan).

**Sample size:** Sample size of 56 patients required at 80% study power and alpha error 5%. It is round of 60 patients for present study expecting approx.

**Sampling Method:** Simple random sampling

**Inclusion Criteria:**

- Cases were willing to participate in study.
- All patients of age above 14 yrs. & any sex, coming with symptoms of dengue fever and had positive serology (Dengue NS 1positive cases / IgM/IgG Ab positive cases)

**Exclusion Criteria:**

- Cases were not willing to participate in study

- All patients with fever who were Dengue NS1 Ag negative.
- Dengue with any chronic disease like CLD, CKD, CAD.
- Patient with history of intake of any hepatotoxic or similar drugs causing derangements of liver functions.
- Dengue patients having other known infections causing hepatitis such as acute or chronic viral hepatitis, malaria, enteric fever.
- Patients age less than 14 years were excluded from the study.

**Data Collection:**

- Detailed history was taken in each patient and information of every patients were recorded in a separate proforma (as per proforma attached).
- The complete blood count was measure by CBC analyzer

**Data Analysis:**

- All data were analyzed on EPI-info statistical software.
- Qualitative data were expressed in the form of proportion.
- Quantitative data were expressed in mean ± SD
- Qualitative data were compared by Chi square test
- Unpaired t test was used to infer the difference in means.
- For significance, following at the level of "p" value was taken
- P > 0.05 = Not significant
- P = 0.05 = Just significant
- P <0.05 = Significant
- P < 0.001 = Highly significant.

**Results & Discussion:**

In our study out of total 141 cases, majority of cases have dengue fever (73.76%) followed by 19.15% cases of dengue hemorrhagic fever and only 7.09% cases have dengue shock syndrome. **Yuditya and Sudirgo [12]** study showed that according to the dengue severity grade, 54 patients (71.1%) suffered from Dengue Hemorrhagic Fever (DHF) grade I, 14 patients (18.4%) suffered from DHF grade II, and 8 patients (10.5%) suffered from DHF grade III. In **Triana et al [13]** highest degree of dengue severity in grade I was 19 patients (54.29%). In **Bhati et al [14]** study according to the dengue severity grade, maximum patients were diagnosed as Dengue Fever with 65 patients (65%), 21 suffered from Dengue Haemorrhagic Fever (DHF) grade I, 15 patients (15%) suffered from DHF grade II, and 8 patients (8%) suffered from DHF grade III and IV. **(Figure 1)**

**Figure 1: Distribution of cases according severity of dengue**

| Grade of severity | No. | % |
|---|---|---|
| DF | 104 | 73.76 |
| DHF | 27 | 19.15 |
| DSS | 10 | 7.09 |
| **Total** | **141** | **100.00** |

All cases are NS1 positive. Out of total cases 80 cases showed IgM positivity, 61 cases showed IgG positivity and only 30 cases out of 141 showed all 3 card positive for dengue. **(Figure 2)**

**Figure 2: Distribution of the cases according to Serology**

| Serology | No. of patients |
|---|---|
| NS1positive | 141 |
| IgM positive | 80 |
| IgG positive | 61 |
| All 3 card positive | 30 |

The mean age of cases having dengue fever is 28.37 years, mean age of cases having dengue hemorrhagic fever are 27.44 years and mean age of cases having dengue shock syndrome is 32.40 years. Majority of the patients were in the adult age group, because young persons are more involve in outdoor activities. **(Figure 3)**

**Figure 3: Distribution of cases according to age.**

| Grade of severity | Age (In yrs.) | |
|---|---|---|
| | **Mean** | **SD** |
| DF | 28.37 | 10.77 |
| DHF | 27.44 | 9.73 |
| DSS | 32.40 | 17.20 |
| **P-value** | **0.4670** | |

In our study we found that at day-3 majority of DF cases has NLR>3 (65.38%), majority of DHF cases has NLR between >1-≤2 (44.44%) and in DSS cases 40% has NLR>3 followed by 30% has NLR between >1-≤2. At day-6 majority of DF cases has NLR between ≤ 1 (40.38%), majority of DHF cases has NLR between >1-≤2 (48.15%) and in DSS cases 50% has NLR≤1 followed by 40% has NLR between >1-≤2. At day-9 majority of DF cases has NLR ≤1 (36.54%), majority of DHF cases has NLR between ≤1 (70.37%) and in DSS cases 50% has NLR≤1 followed by 20% has NLR between >1-≤2 and >2-≤3. Thus, NLR was low in DSS and we found statistically significant difference in NLR with grade of severity. A study from Thailand conducted by **Pancharoen et a**l [15] reported that primary dengue infection presented with significantly lower maximal percentage of neutrophils compared with secondary dengue infection. However, **Phan et al [**16 ] reported a significant decrease in neutrophil counts, complement activity, and platelet counts in DHF/DSS patients. Also, a study in Mexico conducted by **Murillo-Llanes et al [**17] in adult dengue patients found that neutropenia, prolonged partial thromboplastin time, and elevated transaminases were observed more often in DHF patients. **Bhati et al [**18] study also shows relation between NLR and number of patients with bleeding manifestations and patients with features of shock. It shows statistically highly significant relation between occurrence of bleeding and shock with reduction in NLR in patients with dengue. (P<0.001). **(Figure 4–6)**

**Figure 4: Correlation between NLR and Grade of Severity at day-3 of fever**

| Grade of Severity | NLR | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | ≤1 | | >1-≤2 | | >2-≤3 | | >3 | |
| | N | % | N | % | N | % | N | % |
| DF | 3 | 2.88 | 16 | 15.38 | 17 | 16.35 | 68 | 65.38 |
| DHF | 1 | 3.70 | 12 | 44.44 | 3 | 11.11 | 11 | 40.74 |
| DSS | 1 | 10.00 | 3 | 30.00 | 2 | 20.00 | 4 | 40.00 |
| **Total** | **5** | | **31** | | **22** | | **83** | |
| **P-value** | **0.0396** | | | | | | | |

**Figure 5: Correlation between NLR and Grade of Severity at day-6 of fever**

| Grade of severity | NLR | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | ≤1 | | >1-≤2 | | >2-≤3 | | >3 | |
| | N | % | N | % | N | % | N | % |
| DF | 42 | 40.38 | 41 | 39.42 | 14 | 13.46 | 7 | 6.73 |
| DHF | 9 | 33.33 | 13 | 48.15 | 5 | 18.52 | 0 | 0.00 |
| DSS | 5 | 50.00 | 4 | 40.00 | 0 | 0.00 | 1 | 10.00 |
| **Total** | **56** | | **58** | | **19** | | **8** | |
| **P-value** | **0.035** | | | | | | | |

**Figure 6: Correlation between NLR and Grade of Severity at day-9 of fever**

| Grade of Severity | NLR | | | | | | | | Total |
|---|---|---|---|---|---|---|---|---|---|
| | ≤1 | | >1-≤2 | | >2-≤3 | | >3 | | |
| | N | % | N | % | N | % | N | % | |
| DF | 38 | 36.54 | 32 | 30.77 | 23 | 22.12 | 11 | 10.58 | 104 |
| DHF | 19 | 70.37 | 7 | 25.93 | 1 | 3.70 | 0 | 0.00 | 27 |
| DSS | 5 | 50.00 | 2 | 20.00 | 2 | 20.00 | 1 | 10.00 | 10 |
| **Total** | **62** | | **41** | | **26** | | **12** | | |
| **P-value** | **0.043** | | | | | | | | |

## Conclusions:

**Total** 141 serologically confirmed cases of dengue viral infection, randomly enrolled. The study concludes that NLR may be used as prognostic marker in management of dengue fever. There is a significant relation between NLR with grade of severity in dengue infection. This means that the lower the level of NLR, the heavier the clinical degree of dengue. Thus CBC is important parameter to early identify the severity of disease. CBC parameters are easily available also at primary health centres.

## Future scope:

One of the primary limitations to the generalization of the results of this study is the lack of a standard cut-off value for NLR. Further studies are warranted to be able to come up with a standard normal value of NLR. The current study was restricted to only hospitalized patients and conducted in a single hospital. If the patients those who got treated at the out-patient department level were also included, that could have caused some changes in the results because mainly the patients who fulfill the criteria for admission got admitted to the wards. Therefore, further researches are necessary to focus more on other factors that may affect dengue virus infection as well as to perform with more stringent inclusion criteria. Further researches are also essential to focus on the neutrophil/lymphocyte count ratio in children with DHF.

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