STUDY OF PROGNOSTIC ROLE OF NEUTROPHIL AND LYMPHOCYTE RATIO IN ACUTE ISCHEMIC STROKE

Kishan P Halkati[1], Nitin Jadhav[2], Makarand Mane[3]

[1] Senior Resident, Department of General Medicine, Krishna Institute of Medical Sciences, Karad - 415110, Maharashtra, India

[2] Associate Professor, Department of General Medicine, Krishna Institute of Medical Sciences, Karad - 415110, Maharashtra, India

[3] Associate Professor, Department of General Medicine, Krishna Institute of Medical Sciences, Karad - 415110, Maharashtra, India

Address of Correspondence:
Dr Kishan P Halkati
Senior Resident,
Department of General Medicine,
Krishna Institute of Medical Sciences,
Karad - 415110, Maharashtra, India
Phone No.: +919844227915
Email ID: halkatikishan@gmail.com

Conflict of interest: none
Acknowledgements: None

ABSTRACT

Background: Management of acute ischemic stroke (AIS) can be more apt if timely diagnosis and proper assessment of prognosis are performed. The study aimed to evaluate the risk factors of acute ischemic stroke and assess prognostic role of the neutrophil to lymphocyte ratio (NLR) in acute ischemic stroke patients.

Materials and methods: This was a single-centre, hospital-based, observational, descriptive study which included patients with AIS who were admitted in the this hospital over a period of 18 months. Clinical history of the patient such as severity of stroke during admission and the outcome of their health was collected in the database. The NLR was calculated from the blood sample collected at the time of admission. Further, the NLR was evaluated in relation to each risk factor and the significance of association with them was evaluated.

Results: Total 100 patients were included in the study, of them, 65% were male and the mean age of study population was 56.4 (SD: 17.3) years. On analysing the blood samples of 100 patients for NLR data, 23 patients were admitted in hospital had normal NLR of range 1-3, 36 patients were admitted in hospital had NLR of range 3 to 6, 22 patients had NLR ranging from 6-9 NLR, 16 patients had NLR ranging from 9-18, and 3 patients had NLR ranged above 18. The cut off value of NLR in the present study was found to be ≥11.31. The area under curve was 0.999. The NLR was found to be statistically associated ($p < 0.05$) with NIHSS, MRS, duration of hospital stay, and three months outcomes.

Conclusion: In the present study, it was observed that NLR has been significantly associated with poor three months outcomes. This is a readily available and relatively cheap marker that may be a useful tool in routine clinical practice. This marker can be very beneficial in resource-poor settings for predicting clinical outcomes in patients with acute ischemic stroke, as it is inexpensive as well as easily obtainable.

Keywords: Acute ischemic stroke, Neutrophil to lymphocyte ratio, outcomes, prognosis, risk factor

INTRODUCTION

Stroke has transpired as the second major cause of disability and death globally. On the higher side, it stands as the top-most burden in low- and middle-income countries. [1] Acute strokes are broadly classified as either ischemic or hemorrhagic. Of which, ischemic stroke represents 80% of cases. [2] Acute ischemic stroke (AIS) can be described by the abrupt loss of blood circulation towards some part of the brain, causing a subsequent loss of neurologic function. There are numerous risk factors for ischemic stroke, including both modifiable (eg, diet and comorbid conditions) and nonmodifiable risk factors (eg, age, race). [3]

Initially, the management of patients with AIS was mostly dependent on the time from the onset of stroke, severity of neurologic deficit, and outcomes on neuroimaging. [4] The severity

of neurologic deficit has been evaluated by the National Institutes of Health Stroke Scale (NIHSS), ranging from 0 to 42, indicating that the higher the value of score severe the deficits. [5] The neuroimaging constituted reports of non-contrast CT and MRI.

Recently, inflammation has demonstrated to possess a strong association with the incidence of stroke, and has shown its negative effects, both experimentally as well as clinically. [6] Among many, chemokines, cytokines, neutrophils and recruited peripheral circulating leukocytes have been the major factors in the Ischemic response. Neutrophils tend to reach the ischemic sites in the initial few hours of onset of stroke and thereby release some chemical mediators, that are linked with the tissue damage aggravation and poor neurologic improvement. [7, 8] On the other hand, it is reported that lymphocytes, the key cerebroprotective immunomodulators, tend to decrease in such situation thus causing the loss of neuroprotective function and leading to neurological deterioration. [9] The raised neutrophil to lymphocyte ratio (NLR) level with neutrophils escalation and lymphocyte reduction signifies the imbalance in interface amongst stroke-induced central inflammation and peripheral inflammation. [6] Thus, NLR can be correlated with outcomes of patients with AIS. [10] Higher NLR is linked to higher levels of inflammation. Advantage of performing NLR as a prognostic factor is that it can be evaluated through blood routine test, it is performed under low cost and the test is commonly available. [11] Many studies have been performed relating to the association between NLR and outcomes after AIS and also about the prognosis after AIS. Several studies have reported that higher NLR is a negative prognostic indicator in AIS. [6] Majorly, the patients with high NLR have been at high risk of hospital mortality and show less improvement within the 90 days of ischemic stroke treatment. [12] In this study, we aimed to evaluate the risk factors of acute ischemic stroke and assess prognostic role of NLR in patients with acute ischemic stroke.

MATERIALS AND METHODS

This single-centre, hospital-based, observational, descriptive study was conducted at Tertiary care Hospital in India. The study was conducted in patients with acute ischemic stroke admitted to the General Medicine Ward and Medical Intensive Care Unit over a period of 18 months from April-2020 to October-2022. The study was approved by institutional ethics committee before initiation. Written informed consent was obtained from the patients prior to the enrolment of subjects in the study. **Figure 1** indicates study flow chart.

In this study, patients with following categories were included: only the patients who had post-acute ischemic stroke, patients who were admitted within 72 hrs of onset of symptoms, patients above 18 years of age. Patients with following categories were excluded: patients with more than 3 days of onset of symptoms, patients with history of infection within 1 week or within 72 hours of onset of symptoms of stroke, patient with malignancies or haematological symptoms, patients who were taking immune suppressant drugs, patients had history of recent stroke (for past 6 months) or with previous stroke disability, patients with severe pulmonary disorder or chronic renal failure undergoing treatment i.e., dialysis, patients with intra-cerebral haemorrhage.

Data from the patients who are enrolled in this study included their demographic factors, co morbidities, duration of hospital stay, day-to-day life activities such as food habits and lifestyle of the patient. Blood samples were collected once the patient was admitted, to find the neutrophils and lymphocyte counts in their blood. Patients with features of acute ischemic stroke were subjected to detailed history, examination, and thorough investigations.

Clinical history of the patient such as severity of stroke during admission and the outcome of their health was collected in the database. Neurological examination and Neuro-imaging

method used to diagnose the stage of severity of AIS. Based on the NIHSS, the intensity of stroke is classified and functional outcome at the time of discharge is determined using MRS. Patients are followed up in the hospital to retrieve clinical data from the time of admission until they discharge from the hospital. Information collected from the patients are maintained securely as per the informed consent.

The defined co-morbidities include hypertension, type 2 diabetes mellitus, history of CVA, ischemic heart disease, depression, hypothyroidism, and rheumatic heart disease. During the discharge day of the patient, the number days patient got admitted and stayed in the hospital for treatment was collected in database. The patients were followed up for 3 months and any incidence of symptomatic intracranial haemorrhage or death were noted.

Statistical analysis

Continuous variables are expressed as means and standard deviations (range) and categorical variables are reported as frequencies and percentages. Descriptive analysis was performed using the Statistical Package for Social Sciences (SPSS) version 16.0 software (SPSS Inc., Chicago, IL, USA). Chi square test or ANOVA test was applied where applicable. P value ≤ 0.05 was considered statistically significant.

RESULTS

A total 100 patients with diagnosis of acute ischemic stroke were enrolled for present observational study. Of them, 65 (65%) were male and 35 (35%) were female. The mean age of the study population was 56.4 (SD: 17.3) years. The results were distributed as ≤30 years (9%), 31-45 years (16%), 46-60 years (31%), 61-75 years (35%), 76-90 years (9%). Among 100 patients, 72 patients were without any habits of drinking, smoking or tobacco chewing, 6

patients were alcoholic, 13 patients were smokers. Analysing the co-morbidities, 38 patients had only hypertension (HTN), 2 patients had only Type 2 Diabetes mellitus (T2DM), 26 patients had both HTN as well as T2DM. Ten patients had a history of previous stroke, while 90 patients did not have any history of stroke (**Table 1**).

On analysing the blood samples of 100 patients for NLR data, 23 patients were admitted in hospital had normal NLR of range 1-3, 36 patients were admitted in hospital had NLR of range 3 to 6, 22 patients had NLR ranging from 6-9 NLR, 16 patients had NLR ranging from 9-18, and 3 patients had NLR ranged above 18. The NIHSS was taken on the admission of AIS patient and was found that 17 patients were affected by minor stroke, 65 patients were affected by moderate stroke, 11 patients were found to be in the stroke scale of moderate to severe, and only 7 patients were severely affected. The modified Rankin scale was calculated for enrolled patients, 15 patients showed no significant disability, 20 patients showed slight disability, 20 patients showed moderate disability, 19 patients showed moderately severe disability, 14 patients showed severe disability and one patient passed away (**Table 2**).

Of total 100 patients, 19 patients stayed less than 4 days, 52 patients stayed in the range of 5-8 days, 22 patients stayed 9-12 days, 5 patients stayed 13-16 days and only two stayed for 18 days. Of all patients, 22 patients were treated with Tab Ecosprin (75 mg), Tab Rozat (40 mg), and 78 patients were treated with Tab Ecosprin (75 mg), Tab Rozat (40 mg), and Tab Clopitab (75 mg). At three months, ten patients experienced adverse events, and 90 patients did not demonstrate any such event. The adverse event reported at three months, in all ten patients was intracranial haemorrhage. Course of hospitalisation and outcomes have been outlined in **Table 3.**

Figure 2 depicts receiver operating curve (ROC) showing predictive value of NLR for poor outcome. The cut off value of NLR in the present study was found to be ≥11.31. The area under curve was 0.999.

The NLR was calculated in relation to each risk factor and the significance of association with them was evaluated. The NLR was found to be statistically associated ($p < 0.05$) with NIHSS, MRS, duration of hospital stay, and three months outcomes.

DISCUSSION

The main findings of the present study were: (1) elevated neutrophil lymphocyte ratio was the strong predictor of poor outcomes at 3 months in patients after acute ischemic stroke; (2) the optimal cut off value for NLR as a predictor for 3 months outcome was found to be 11.31; (3) increased NLR was significantly related to longer the duration of hospitalisation of patients; (4) NLR was significantly associated with severity and disability scales, NIHSS and modified Rankin scale. The role of NLR in prognosis of patients with acute ischemic stroke has been evaluated in various recent studies, but the cut off values and NLR's association with other risk factors have been varying greatly among studies. The variation amongst various studies can be attributed to the regional, cultural, ethnic, and sampling differences.

The present study population was predominated by male gender (65%). Similarly, in a study, Qun S, et al. had reported 55.9% male patients. [13] Whereas, in another study by Lin Y, et al., the number of males were slightly higher, i.e., 71.9%. [14] There is a male predominance found in the studies of AIS, however the proportions have been varying. The mean age of the study population was 56.4 ± 17.3 years. The distribution of age groups was: ≤30 years (9%), 31-45 years (16%), 46-60 years (31%), 61-75 years (35%), 76-90 years (9%). This was comparable to the age range and mean age of patients in various other studies. [14, 15] Ten patients had reported a previous stroke. Similarly, in a study by Zafar A, et al. 10.2% patients had reported to have past history of stroke. [16] Alcoholism, smoking, and tobacco were the lifestyle risk factors in the patients, of which smoking was the most prevalent (13%).

Literature suggests that cigarette smoking has been documented as eminent risk factor for stroke and exhibit a strong association with athero-thrombotic process related to stroke. [17]

In the present study, the most prevalent comorbidities that presented as risk factors were hypertension (66%) and diabetes (28%), followed by old cerebrovascular accidents (12%), hypothyroidism (4%), ischemic heart diseases (3%), and depression (1%); either individually or co-existence of more than one in a patient. Zafar A, et al. documented that hypertension, diabetes mellitus, and hyperlipidaemia were prevalent modifiable risk factors, followed by ischemic heart disease and history of previous stroke. [16] Soliman RH, et al. [17] reported 62.3%, 34.7%, and 5.4% patients of acute ischemic stroke with hypertension, disbetes and old CVA as a risk factor.

In the present study, mean NIHSS was 9.64 ± 6.01 which was comparable to a study by Li L, et al., [15] which was 9.2±7.8. The distribution of NIHSS in the patients of this study was similar to the study by Soliman RH, et al. [17], i.e., 19.8%, 61.7%, 11.4%, 7.2%, respectively. In the present study, NLR was found to be significantly correlated with the NIHSS ($p = 0.002$). Similarly, in a case–control study of 225 AIS patients, Cai et al. reported that higher NLR was positively associated with higher NIHSS and infarct sizes. [18] In the present study, the majority proportion of the degree of disability were slight (20%) and moderate (20%). Whereas in a study by Soliman RH, et al. [17], moderate (25.7%) and severe (17.4%) disabilities were more prevalent in the study patients.

In the present study, mean duration of hospitalisation was 7.21 ± 3.13 days. About half of the patients stayed in hospital from 5 to 8 days (52%). Duration of hospitalisation was significantly associated with NLR ($p <0.001$). It was lower than the admission days reported by Lin Y, et al. [14] [11 (8 – 15) days] and Wang L, et al. [10] [10 (8 – 13) days]. Up to three months, 10 patients reported with adverse events. The outcomes at 3 months were closely related to the NLR values. Higher NLR was associated with poor outcomes ($p <0.001$). Wan

J, et al. in their meta-analysis demonstrated that, for patients with stroke, a higher NLR was related with poor functional outcome at three months, however its link with death at 3 months was not concluded. [19] In the present study, the mean NLR was 6.42 ± 4.50. The cut off value for NLR was ≥11.31. The NLR was found to be significantly associated ($p < 0.05$) with NIHSS, modified Rankin Scale, duration of hospitalisation, and three months outcomes. Whereas it was not statistically associated with other risk factors such as comorbidities or lifestyle habits or diet. Similar to our study, a multi-centre study by Duan Z, et al reported that NLR ≥ 7.0 was independently related with poor functional outcomes at 3 months. [20] Another retrospective study by Brooks et al. showed that NLR ≥ 5.9 was a predictor of poor outcome and mortality at 3 months. [8] On contrary, in a study by Qun S, et al. the cut off value for NLR as a predictor for 3 months outcome was lower (≥2.995). [13]

There were some limitations of this study, as the sample size of the presents study was relatively small as compared to many larger studies. This study was designed to collect data from only one centre, which might cause selection bias. Moreover, several other diseases and factors that could influence inflammatory markers were not considered.

CONCLUSION

In the present study that included 100 patients with acute ischemic stroke, we observed that neutrophil lymphocyte ratio is a simple marker for predicting stroke severity on admission after acute ischemic stroke. Baseline NLR level can be a promising predictor of acute ischemic stroke prognosis, such that raised NLR can also be linked with high risk of ischemic stroke recurrence. This is a readily available and relatively cheap marker that may be a useful tool in routine clinical practice. This marker can be very beneficial in resource-poor settings for predicting clinical outcomes in patients with acute ischemic stroke, as it is inexpensive as well as easily obtainable. The National Institutes of Health Stroke Scale (NIHSS) at admission, modified Rankin scale (MRS) at admission, and duration of hospitalisation of the

patients have been closely associated with ranges of NLR. Other risk factors like gender, age, hypertension, type-2 diabetes mellitus, etc were not significantly related to NLR of patients at the time of admission. It was observed that NLR has been significantly associated with poor three months outcomes. However, further prospective studies with larger sample sizes, targeting NLR, are necessary.

REFERENCES

1. Saini V, Guada L, Yavagal DR. Global epidemiology of stroke and access to acute ischemic stroke interventions. Neurology. 2021;97(20 Supplement 2):S6-S16.
2. Ikram MA, Wieberdink RG, Koudstaal PJ. International epidemiology of intracerebral hemorrhage. Current atherosclerosis reports. 2012;14(4):300-6.
3. Boehme AK, Esenwa C, Elkind MS. Stroke risk factors, genetics, and prevention. Circulation research. 2017;120(3):472-95.
4. Powers WJ. Acute ischemic stroke. New England Journal of Medicine. 2020;383(3):252-60.
5. Lyden P, Brott T, Tilley B, Welch K, Mascha E, Levine S, et al. Improved reliability of the NIH Stroke Scale using video training. NINDS TPA Stroke Study Group. Stroke. 1994;25(11):2220-6.
6. Song S-Y, Zhao X-X, Rajah G, Hua C, Kang R-j, Han Y-p, et al. Clinical significance of baseline neutrophil-to-lymphocyte ratio in patients with ischemic stroke or hemorrhagic stroke: an updated meta-analysis. Frontiers in neurology. 2019:1032.
7. Anrather J, Iadecola C. Inflammation and stroke: an overview. Neurotherapeutics. 2016;13(4):661-70.
8. Brooks SD, Spears C, Cummings C, VanGilder RL, Stinehart KR, Gutmann L, et al. Admission neutrophil–lymphocyte ratio predicts 90 day outcome after endovascular stroke therapy. Journal of neurointerventional surgery. 2014;6(8):578-83.
9. Zhang J, Ren Q, Song Y, He M, Zeng Y, Liu Z, et al. Prognostic role of neutrophil–lymphocyte ratio in patients with acute ischemic stroke. Medicine. 2017;96(45).
10. Wang L, Song Q, Wang C, Wu S, Deng L, Li Y, et al. Neutrophil to lymphocyte ratio predicts poor outcomes after acute ischemic stroke: A cohort study and systematic review. Journal of the neurological sciences. 2019;406:116445.

11. Bi Y, Shen J, Chen S-C, Chen J-X, Xia Y-P. Prognostic value of neutrophil to lymphocyte ratio in acute ischemic stroke after reperfusion therapy. Scientific reports. 2021;11(1):1-11.
12. Campbell BC, Mitchell PJ, Kleinig TJ, Dewey HM, Churilov L, Yassi N, et al. Endovascular therapy for ischemic stroke with perfusion-imaging selection. New England Journal of Medicine. 2015;372(11):1009-18.
13. Qun S, Tang Y, Sun J, Liu Z, Wu J, Zhang J, et al. Neutrophil-to-lymphocyte ratio predicts 3-month outcome of acute ischemic stroke. Neurotoxicity Research. 2017;31(3):444-52.
14. Liu Y-L, Wu Z-Q, Yin H-P, Xia P-S, Qiu D-H, Liang M-Q, et al. High neutrophil-to-lymphocyte ratio is a predictor of poor short-term outcome in patients with mild acute ischemic stroke receiving intravenous thrombolysis. Brain and Behavior. 2020;2020.
15. Li L-H, Chen C-T, Chang Y-C, Chen Y-J, Lee I-H, How C-K. Prognostic role of neutrophil-to-lymphocyte ratio, platelet-to-lymphocyte ratio, and systemic immune inflammation index in acute ischemic stroke: a STROBE-compliant retrospective study. Medicine. 2021;100(25).
16. Zafar A, Al-Khamis FA, Al-Bakr AI, Alsulaiman AA, Msmar AH. Risk factors and subtypes of acute ischemic stroke: A study at king fahd hospital of the university. Neurosciences Journal. 2016;21(3):246-51.
17. Soliman RH, Oraby MI, Fathy M, Essam AM. Risk factors of acute ischemic stroke in patients presented to Beni-Suef University Hospital: prevalence and relation to stroke severity at presentation. The Egyptian journal of neurology, psychiatry and neurosurgery. 2018;54(1):1-9.
18. Cai W, Liu S, Hu M, Huang F, Zhu Q, Qiu W, et al. Functional dynamics of neutrophils after ischemic stroke. Translational stroke research. 2020;11(1):108-21.
19. Wan J, Wang X, Zhen Y, Chen X, Yao P, Liu W, et al. The predictive role of the neutrophil-lymphocyte ratio in the prognosis of adult patients with stroke. Chinese neurosurgical journal. 2020;6(03):161-70.
20. Duan Z, Wang H, Wang Z, Hao Y, Zi W, Yang D, et al. Neutrophil-lymphocyte ratio predicts functional and safety outcomes after endovascular treatment for acute ischemic stroke. Cerebrovascular Diseases. 2018;45(5-6):221-7.

Tables

Table 1: Baseline demographics of study patients with acute ischemic stroke

Variables	**Total Patients (n = 100)**
Age (Mean ± SD; Years)	56.4 ± 17.3
Male, n	65
Habits	
Without any habits	72
Alcoholic	6
Smoker	13
Tobacco Chewer	5
Both Alcoholic and Smoker/Tobacco Chewer	3
Smoker, Alcoholic, and Tobacco Chewer	1
Mixed diet	58
Risk factors	
Only HTN	28
Only T2DM	2
HTN with T2DM	26
HTN with other comorbidity	12
Other comorbidities	3
Presence of more than 2 comorbidities	3
Without any Comorbidities	26
History of previous stroke	10
Door-to-needle time, mins	61.5 ± 5.9

Table 2: Distribution of patients as per NLR, NIHSS, and MRS

Variable	**Total Patients (n = 100)**
NLR	
1-3 (Normal)	23
3-6 (Mild Stroke)	36
6-9 (Mild to moderate Stroke)	22
9-18 (Moderate Stroke)	16
>18 (Severe Stroke)	3
NIHSS	
Minor Stroke	17
Moderate Stroke	65
Moderate to Severe Stroke	11
Severe Stroke	7
MRS	
No symptoms	11
No significant disability	15
Slight disability	20
Moderate disability	20
Moderately Severe Disability	19
Severe disability	14
Dead	1

Table 3: Course of hospitalisation and outcomes

Variables	**Total Patients (n = 100)**
Number of hospitalisation days	
≤ 4 days	19
5-8 days	52
9-12 days	22
13-16 days	5
17-20 days	2
Treatment after admission	
Tab Ecosprin 75 mg Hs, Tab Rozat 40 mg Hs	22
Tab Ecosprin 75 mg Hs, Tab Clopitab 75 mg Hs, Tab Rozat 40 mg Hs	78
Outcomes at three months	
No	90
Yes	10

Figures

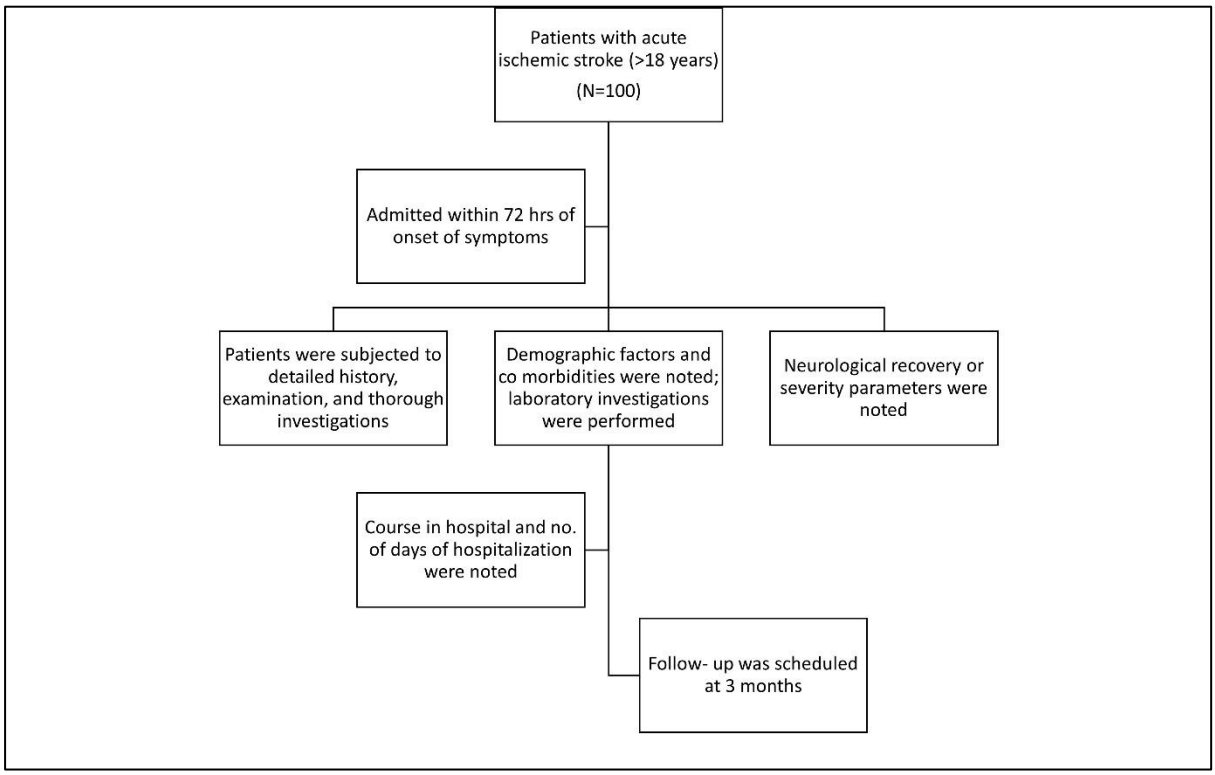

Figure 1: Study flow chart

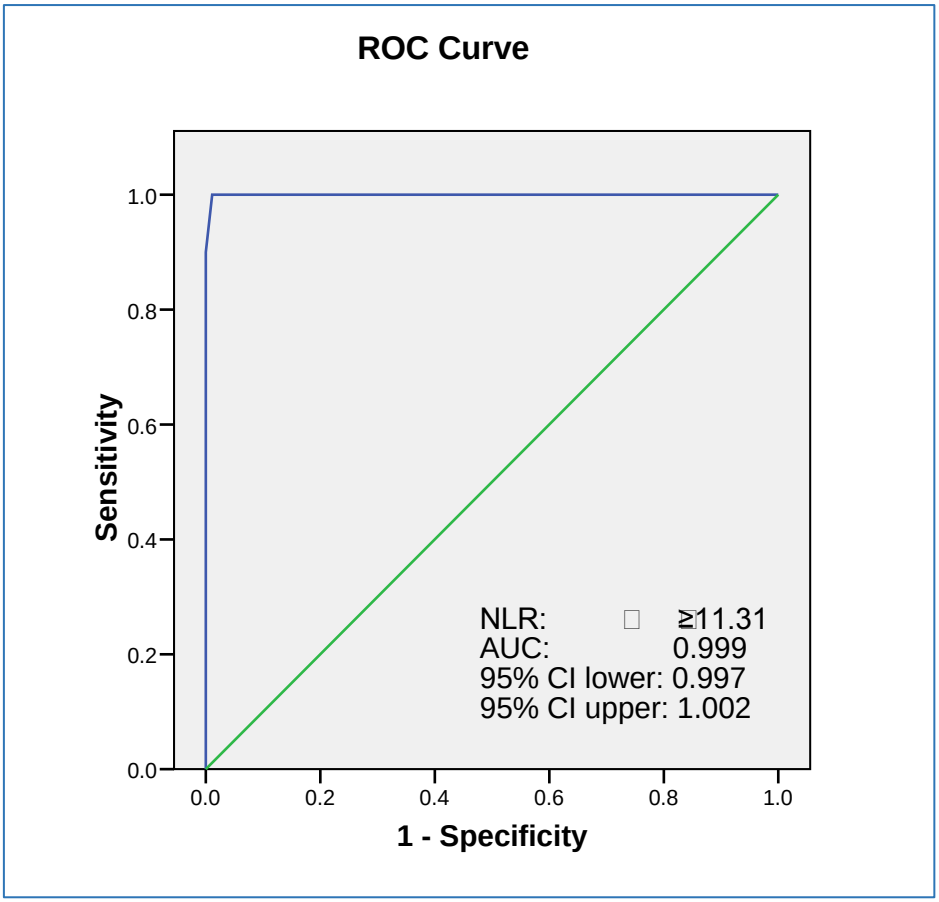

Figure 2: Receiver operating curve (ROC) showing predictive value of NLR for poor outcome