

Original Research Article

THE STUDY OF NEUTROPHIL TO HDL CHOLESTEROL RATIO IN ACUTE ISCHEMIC STROKE SUBJECTS WITH RELATION TO IN HOSPITAL OUTCOME

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ABSTRACT

Background

A stroke, or cerebrovascular accident, is defined as an abrupt onset of a neurologic deficit that is attributable to a focal vascular cause. 85% of stroke cases are due to cerebral infarction while 15% are due to intra-cerebral bleed.^[1] Stroke is the second leading cause of death worldwide, with 6.2 million dying from stroke in 2015, an increase of 830,000 since the year 2000. Stroke is likely to remain the second most common disabling condition in individuals aged 50 or older worldwide.^[2] Inflammation plays a significant role in the development, evolution, and prognosis of Ischemic Stroke. There is growing evidence indicating that inflammation contributes to the immune response in the affected site, promoting brain cells' death and exacerbating neurological dysfunction.^[3,4]

Methods

A total of 50 subjects with Acute Ischemic stroke who were admitted in hospital or visited to OPD were studied. Routine blood investigations were done along with brain imaging. Neutrophil to HDL-Cholesterol ratio (NHR) was calculated for all patients and comparison was made based on gender, age, and outcome in terms of death or discharge.

Results

Stroke incidence was almost same in males and females with slight male preponderance. Though NHR showed positive predictive value, there was no statistically significant association between NHR and in hospital outcome in subjects with Acute Ischemic stroke (AIS).

Conclusion

This study showed positive predictive value for NHR to be a hazard factor in AIS, but it is not statistically significant to show strong association between NHR and AIS with respect to in hospital outcome.

Keywords: Cerebrovascular accident, Neutrophil, HDL Cholesterol.

INTRODUCTION

Various factors influence the prognosis of ischemic stroke, with the immune-inflammatory response playing a crucial role in post-stroke processes.^[2] White blood cells (WBCs), including neutrophils, lymphocytes, and monocytes, are central to the inflammatory response. Neutrophils play a role in destabilizing atherosclerotic plaques by causing endothelial dysfunction, attracting monocytes, and promoting foam cell formation. High-density lipoprotein (HDL) possesses anti-inflammatory and antioxidant properties, regulating monocyte activation, inhibiting macrophage migration, and preventing low-density lipoprotein (LDL) oxidation.^[3] Neutrophils, being the earliest cells to reach the lesion site, engage in intricate inflammatory reactions, impacting both tissue destruction and repair in cerebral infarction. Changes in peripheral blood inflammatory cells after stroke correlate with local inflammatory responses, offering a clinical basis for prognosis assessment. Notably, studies have demonstrated the association between peripheral neutrophil count and stroke prognosis, indicating its potential as a prognostic marker. Additionally, lipids play a significant role in the inflammatory response, with high-density lipoprotein cholesterol (HDL-C) shown to inhibit various detrimental processes such as neutrophil deformation, platelet aggregation, and neutrophil trap formation, ultimately preventing further tissue damage post-thrombosis.^[2] high-density lipoprotein cholesterol (HDL-C) is integral to lipid metabolism and has been associated with stroke risk, particularly in hypertensive populations. Integrating Neutrophil and HDL-C into a single index, such as the neutrophil to high-density lipoprotein cholesterol ratio (NHR), may serve as a comprehensive biomarker for predicting AIS.^[4]

OBJECTIVES

- To study Neutrophil to HDL Cholesterol ratio in subjects of acute ischemic stroke.
- To study the Neutrophil to HDL Cholesterol ratio in subjects of Acute Ischemic Stroke with respect to in hospital outcome.

MATERIALS AND METHODS

This is a cross sectional, observational study which was conducted in Mysore Medical College and Research Institute, Mysore among patients admitted in ward or ICU and patients visiting outpatient department from the month of December 2023 to February 2024.

Inclusion Criteria

- Age between 18-85 years with Acute Ischemic Stroke.

Exclusion Criteria

- Cases with hemiparesis or any focal neurological deficit other than AIS like head injury, brain tumour, brain abscess, metabolic encephalopathy, migraine.
- Transient ischemic attack.

Procedure

- After taking the institutional ethical clearance for the study, purpose of study will be explained to the subjects and attenders. Written consent was taken from the subjects.
- Relevant history and clinical examinations were done. Blood samples for complete blood count, differential leucocyte count and fasting lipid profile was taken. Brain imaging will be done by CT or MRI.

Descriptive Analysis

Descriptive statistics including mean, standard deviation for quantitative variables, and frequency/proportions for qualitative variables was calculated.

ROC Curve Analysis

ROC curves were generated to determine the cut-off values, sensitivity, and specificity of Neutrophil count, HDL cholesterol levels, and Neutrophil/HDL ratio in predicting outcomes.

Significance Level

A p-value was not less than <0.05.

Statistical Analysis

SPSS (Statistical Package for Social Sciences) version 21. (IBM SPSS statistics [IBM corporation: NY, USA]) was used to perform the statistical analysis

- Data was entered in the excel spread sheet.
- Descriptive statistics of the explanatory and outcome variables were calculated by mean, standard deviation for quantitative variables, frequency and proportions for qualitative variables.
- Inferential statistics like.
- ROC curve was computed to find the cut-off values, sensitivity and specificity of Neutrophil, HDL and Neutrophil/HDL ratio to predict Outcome.
- The level of significance is set at 5%.

RESULTS

	N	Minimum	Maximum	Mean	S.D
Age	50	38.0	83.0	59.38	12.47

Table 1: Mean Age Distribution of the Subjects

Age Groups	Frequency	Percent
38 to 45 yrs	9	18.0
46 to 55 yrs	12	24.0
56 to 65 yrs	8	16.0
66 to 75 yrs	18	36.0
> 75 yrs	3	6.0
Total	50	100.0

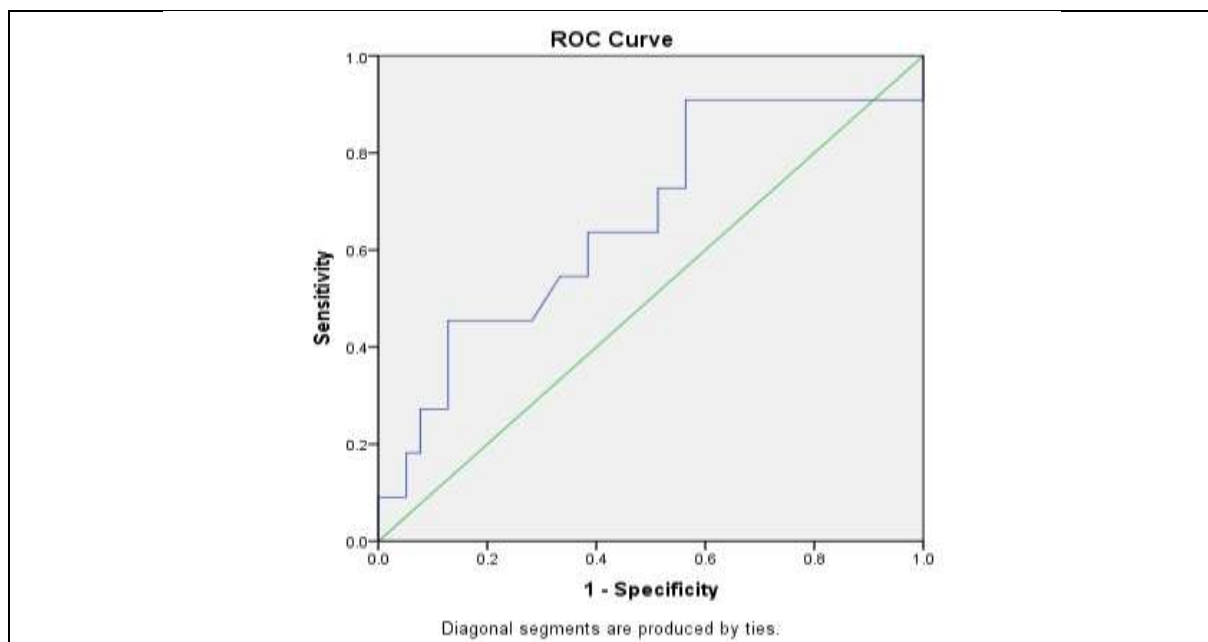
Table 2: Distribution of the Subjects Based on Age Groups

Gender	Frequency	Percent
Females	24	48.0
Males	26	52.0
Total	50	100.0

Table 3: Distribution of the Subjects Based on Gender

	N	Minimum	Maximum	Mean	S.D
Neutrophil(*109/L)	50	2.60	15.40	7.20	3.09
HDL (mmol/L)	50	.31	3.00	1.19	0.61
Neutrophil/HDL	50	1.33	26.77	7.74	5.43

Table 4: Mean Scores



Area Under the Curve

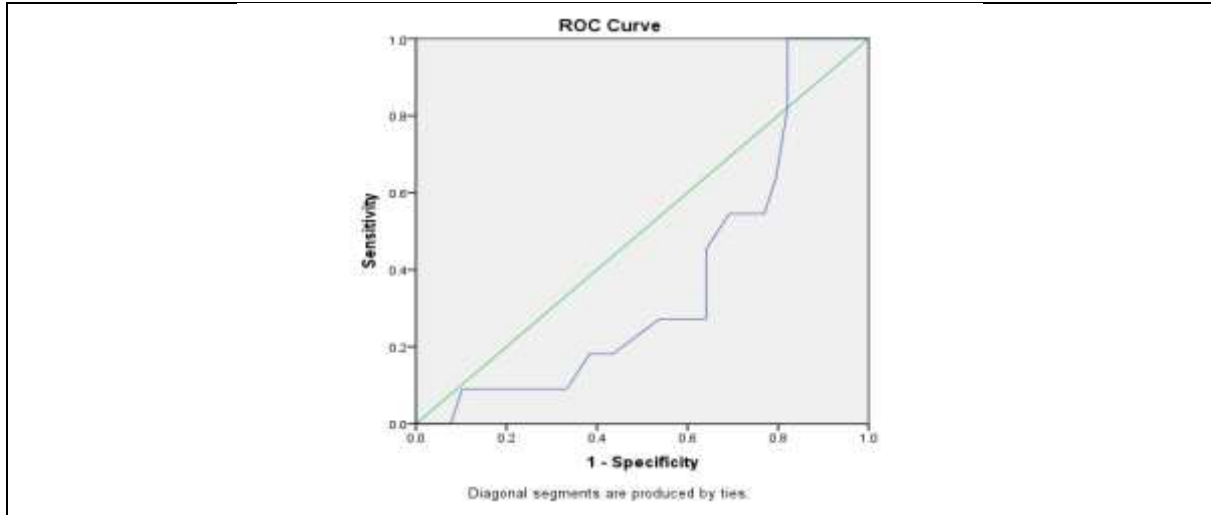
Test Result Variable(s):

Area	Std. Error	p value	Asymptotic 95% Confidence Interval	
			Lower Bound	Upper Bound
.662	.098	.104	.471	.853

Table 5: ROC Curve to Predict Outcome with Neutrophil as Risk Factor

*significant

The area under the curve is 0.662 with cut off for neutrophil being 6.52 and sensitivity of 63.6 % and specificity of 48.7%.



Area Under the Curve

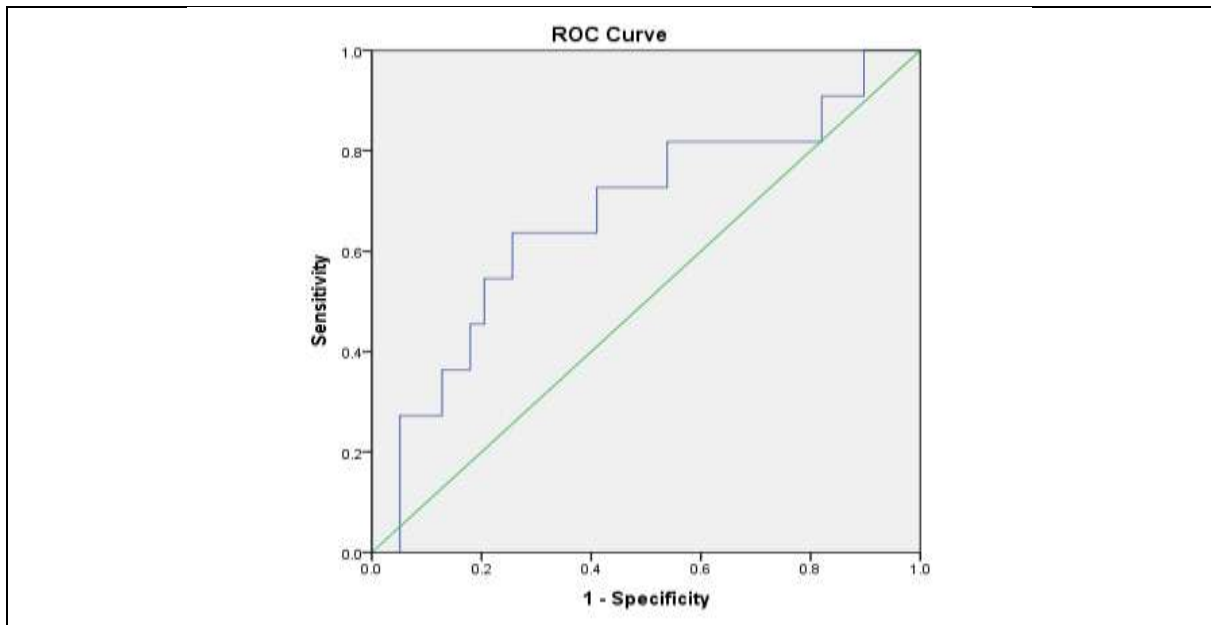
Test Result Variable(s):

Area	Std. Error	p value	Asymptotic 95% Confidence Interval	
			Lower Bound	Upper Bound
.371	.088	.194	.199	.543

Table 6: ROC Curve to Predict outcome with HDL as Risk Factor

*significant

The area under the curve is 0.371 with cut off for HDL ratio being 0.88 and sensitivity of 54.5 % and specificity of 30.8%.



Area Under the Curve

Test Result Variable(s):				
Area	Std. Error	p value	Asymptotic 95% Confidence Interval	
			Lower Bound	Upper Bound
.674	.097	.081	.483	.865
<i>Table 7: ROC Curve to Predict Outcome with Neutrophil/HDL AS Risk Factor</i>				
*significant				

The area under the curve is 0.674 with cut off for Neutrophil/HDL ratio being 6.49 and sensitivity of 72.7 % and specificity of 59%.

DISCUSSION

Inflammation emerges as a novel target for stroke risk prevention, and addressing vascular risk after stroke necessitates innovative treatment approaches. Consequently, there is a demand for new early warning indicators and markers. Presently, predictive models predominantly incorporate conventional risk factors, underscoring the necessity for the advancement of models integrating inflammation and lipid parameters.^[5]

Gkantzios A et.al, conducted a study which included MHR and NHR in their study^[6] but in our study only NHR was included so addition of other inflammatory biomarker help for better clinical application.

Lina Yu et.al, conducted a study on neutrophil to HDL-C ratio with AIS in which the ROC curve showed specificity (87.0%) and sensitivity (56.3%) of NHR which indicates that NHR has a powerful predictive value for AIS and maybe a hazard factor^[7] whereas in our study the ROC curve showed specificity(72.7%) and sensitivity(59%) of NHR which shows similar results. So this indicates that the NHR might be a powerful predictive value and hazard factor for AIS.

Li and Nakamura et al^[8,9] have recently studied about the association between inflammatory and lipid-related biomarkers and cerebrovascular diseases. Both inflammatory responses and abnormal lipid metabolism are key factors in the development of atherosclerosis. Prior research has shown that inflammation plays a crucial role in thrombosis, which can lead to thrombotic inflammatory responses affecting tissue damage severity, clinical outcomes, and functional recovery by recruiting leukocytes.^[10]

Experimental studies by Jin R and Kim E et al^[11,12] showed that following brain ischemia, pro-inflammatory cells such as monocytes, T cells, and neutrophils become activated initially, but gradually decrease over time, while anti-inflammatory monocytes increase. Interestingly, inhibiting the inflammatory response has been associated with a reduction in infarct size. Since our study is an observational study , there are more drawbacks.

A systematic assessment and meta-analysis of prospective cohort studies done by Qie R et al^[13] revealed a correlation between high-density lipoprotein cholesterol (HDL-C) levels and atherosclerotic ischemic events (ASI), indicating a reduction in the risk of ischemic stroke with higher levels of HDL-C .

A study conducted by Huang et al^[14] highlighted the predictive role of NHR in determining clinical outcomes among elderly patients diagnosed with acute myocardial

infarction. Additionally, they found a correlation between NHR and the severity of coronary artery disease.

In a study conducted by Jiang M et al^[15], showed that the Neutrophil-to-High-Density Lipoprotein Ratio (NHR) stands out as an independent predictor of cardiovascular mortality and holds significance in forecasting clinical outcomes in cardiovascular disease.

Our study didn't show any statistically significant results may be due to small sample size, short duration of follow up , not considering other comorbidities, selecting only one hospital which can cause selection bias.

CONCLUSION

Our study showed the powerful predictive NHR value for AIS with respect to our hospital outcome and its potential clinical applications as a novel inflammatory biomarker in stroke prognosis. NHR is identified as a novel serum biomarker, offers the benefits of simplicity, rapidity, and efficiency, rendering it a promising biological target for assessing the prognosis of patients with acute ischemic stroke (AIS). These Biomarkers being economic makes it promising to use in our clinical practice. P value in our study showed that it is not statistically significant due to the small sample size and many other drawbacks mentioned earlier ,hence more multicentred trails are required for more statistically significant results. It is understood that there is a scope of improving the clinical prognosis with respect to AIS.

REFERENCES

- [1] Maheshwari A, Norby FL, Inciardi RM, Wang W, Zhang MJ, Soliman EZ, et al. Left atrial mechanical dysfunction and the risk for ischemic stroke in people without prevalent atrial fibrillation or stroke: a prospective cohort study. *Ann Int Med* 2023;176(1):39-48.
- [2] Sonaglioni A, Di Cara M, Nicolosi GL, Eusebio A, Bordonali M, Santalucia P, et al. Rapid risk stratification of acute ischemic stroke patients in the emergency department: the incremental prognostic role of left atrial reservoir strain. *Journal of Stroke and Cerebrovascular Diseases* 2021;30(11):106100.
- [3] Garcia-Bonilla L, Moore JM, Racchumi G, Zhou P, Butler JM, Iadecola C, et al. Inducible nitric oxide synthase in neutrophils and endothelium contributes to ischemic brain injury in mice. *J Immunol* 2014;193:2531-7.
- [4] Fassbender K, Rossol S, Kammer T, Daffertshofer M, Wirth S, Dollman M, et al. Proinflammatory cytokines in serum of patients with acute cerebral ischemia: Kinetics of secretion and relation to the extent of brain damage and outcome of disease. *J Neurol Sci* 1994;122:135-9.
- [5] Wang N, Liu H, Tian M, Liang J, Sun W, Zhang L, et al. A Nomogram That Includes Neutrophils and High-Density Lipoprotein Cholesterol Can Predict the Prognosis of Acute Ischaemic Stroke. *Front Neurol* 2022;13:827279.
- [6] Gkantzi A, Tsiptsios D, Karapepera V, Karatzetzou S, Kiamelidis S, Vlotinou P, et al. Monocyte to HDL and Neutrophil to HDL Ratios as Potential Ischemic Stroke Prognostic Biomarkers. *Neurology International* 2023;15(1):301-17.
- [7] Yu L, Ma K, Hao J, Zhang B. Neutrophil to high-density lipoprotein cholesterol ratio, a novel risk factor associated with acute ischemic stroke. *Medicine* 2023;102(26):e34173.

- [8] Li JJ, Gu HQ, Peng YJ. The association of lipid profile and bleeding in patients with minor stroke or transient ischemic attack on antiplatelet therapy: subgroup analysis of CHANCE. *Zhonghua Nei Ke Za Zhi* 2018;57(10):723-30.
- [9] Nakamura A, Otani K, Shichita T. Lipid mediators and sterile inflammation in ischemic stroke. *Int Immunol* 2020;32(11):719-25.
- [10] Wang MQ, Sun YY, Wang Y. Platelet-to-neutrophil ratio after intravenous thrombolysis predicts unfavorable outcomes in acute ischemic stroke. *Curr Neurovasc Res* 2020;17(4):411-9.
- [11] Jin R, Yang G, Li G. Inflammatory mechanisms in ischemic stroke: role of inflammatory cells. *J Leukoc Biol* 2010;87:779–789. [CrossRef]
- [12] Kim E, Yang J, Beltran CD, Cho S. Role of spleen-derived monocytes/ macrophages in acute ischemic brain injury. *J Cereb Blood Flow Metab* 2014;34:1411–1419. [CrossRef]
- [13] Qie R, Liu L, Zhang D, et al. Dose-response association between high-density lipoprotein cholesterol and stroke: a systematic review and meta-analysis of prospective cohort studies. *Prev Chronic Dis.* 2021;18:E45.
- [14] Huang JB, Chen YS, Ji HY, et al. Neutrophil to high-density lipoprotein ratio has a superior prognostic value in elderly patients with acute myocardial infarction: a comparison study. *Lipids Health Dis.* 2020;19:59.
- [15] Jiang M, Sun J, Zou H, et al. Prognostic role of neutrophil to High-Density lipoprotein cholesterol ratio for All-Cause and cardiovascular mortality in the general population. *Front Cardiovasc Med.* 2022;9:807339.