**Scholars Journal of Applied Medical Sciences**
Abbreviated Key Title: Sch J App Med Sci
ISSN 2347-954X (Print) | ISSN 2320-6691 (Online)
Journal homepage: www.saspublishers.com

OPEN ACCESS

Biochemistry

# Serum Neuron Specific Enolase in Hyperglycemic and Normoglycemic Ischemic Stroke Patients – A Comparative Study

Dr. P. Deepalakshmi, Dr. Christal Viji Shalini C*

Assistant Professor, Department of Biochemistry, Govt. Thoothukudi Medical College, Thoothukudi, Tamil Nadu, India

**DOI:** 10.36347 /sjams.2019.v07i10.050 | **Received:** 17.09.2019 | **Accepted:** 23.09.2019 | **Published:** 30.10.2019

***Corresponding author:** Dr. Christal Viji Shalini C

**Abstract** **Original Research Article**

Cerebrovascular stroke remains largely a clinical diagnosis. The use of biomarkers in diagnosing stroke and assessing prognosis is an emerging and rapidly evolving field. Neuron Specific Enolase (NSE) is mentioned as a possible reliable marker of neuronal tissue damage. It is released from neurons after infarction. High levels of serum NSE is correlated with worse clinical outcome. This study is aimed to investigate the difference in level of serum neuron specific enolase within 72 hours of stroke in hyperglycemic and normoglycemic ischemic stroke patients with left middle cerebral artery infarct and its correlation with NIHSS score & modified Rankin scale. This cross-sectional study was conducted in tertiary health care. Newly diagnosed cases of 25 hyperglycemic (FBS > 126mg/dl) and 25 normoglycemic (FBS< 126mg/dl) ischemic stroke patients (diagnosed by CT scan, clinical signs &symptoms) within the age group of 22 to 86 years of both sexes were included in the study. Serum NSE showed a positive correlation with serum FBS, NIHSS, modified Rankin scale and it was found statistically significant (*P* value <.0.05). The increased NSE serum levels correspond to the ischemia-induced cytoplasm loss of NSE in the neurons and are detectable before irreversible neuronal damage takes place. The rise in blood glucose levels increases serum NSE level. NSE in blood is the earliest parameter for prognostic classification, earlier than any other methods such as regularly repeated neurological examinations, neuroimaging (cranial CT or magnetic resonance imaging) or electrophysiological assays so NSE can be used as a diagnostic parameter of stroke thereby to assess stroke severity.

**Keywords:** Cerebrovascular stroke, Ischemic stroke, Hyperglycemia, Neuron Specific Enolase (NSE), National institutes of health stroke scale (NIHSS).



## INTRODUCTION

Stroke or cerebral vascular accident, is the sudden death of brain cells due to inadequate blood flow. The WHO clinically defines stroke as "the rapid development of clinical signs and symptoms of focal neurological disturbance lasting more than 24 hours or leading to death with no apparent cause other than vascular origin [1]." Stroke is the second commonest cause of death worldwide after coronary heart disease (CHD). Cerebrovascular stroke remains largely a clinical diagnosis. The use of biomarkers in diagnosing stroke and assessing prognosis is an emerging and rapidly evolving field. Neuron-specific enolase is considered as specific neurobiochemical marker of brain damage after brain infarctions in humans [2].

NSE is released from neurons after infarction; hence, as the name implies, its specificity allows localization. After acute central nervous system insults, such as cerebral infarction, hypoxia, trauma, and seizure, the blood brain barrier is altered and astroglial disintegration substantially makes the NSE leak into cerebrospinal fluid and serum [3].

The neuron specific enolase, increases in blood circulation because it is not used anymore in damaged neuron. The half-life of NSE in serum has been reported to be about 48 hours and therefore blood levels of NSE would be expected to rise as long as infarct damage continued and NSE was washing out of brain tissue. We evaluated the serum NSE level rather than the CSF level, because the daily serum sampling was practical and posed no risk for older patients [4]. Stroke severity was a difficult variable to evaluate because of its relationship with stroke volume. Generally, larger infarcts are associated with greater stroke severity. Elevating NSE level is also connected with infarct volume and the extent of brain damage [5, 6].

Increased levels of serum NSE is correlated with worse clinical outcome [7]. Studies have shown that an initial high blood glucose concentration following a stroke is a predictor of poor outcome [8]. A high proportion of patients suffering an acute stress such as stroke may develop hyperglycemia even in the absence of a pre-existing diagnosis of diabetes [9]. Hyperglycemia defined as fasting blood glucose >126mg/dl [10] is correlated with elevated levels of NSE and the NIHSS score. Hyperglycemia may be directly toxic to the ischemic brain**.** Accumulation of lactate and intracellular acidosis in the ischemic brain (produced through anaerobic cerebral glucose metabolism) may contribute. Intracellular acidosis may promote and accelerate ischemic injury by enhancing lipid peroxidation and free radical formation [11], allowing accumulation of intracellular calcium [12] (a key component of the glutamate-dependent excitotoxicity seen in ischemic neurons) [13], and impairing mitochondrial function [14]. These neurotoxic effects may be particularly important in the ischemic penumbra [15]. The lipid profile also might have an important role in those ischemic strokes that are the consequence of atherosclerosis of larger arteries.

The aim of this cross-sectional study is therefore to compare serum neuron specific enolase levels in hyperglycemic and normoglycemic ischemic stroke patients with left middle cerebral artery infarct since neuron specific enolase is a marker of neuronal tissue damage and a prognostic parameter in cerebrovascular disease. Assessment of stroke severity is done by NIHSS score and modified Rankin scale in our study.

## AIMS AND OBJECTIVES

### AIMS

To find out difference in serum neuron specific enolase level in hyperglycemic and normoglycemic ischemic stroke patients.

### OBJECTIVES

- To correlate serum neuron serum neuron specific enolase with NIHSS score and modified Rankin scale
- To study the variation of lipid profile level in hyperglycemic and normoglycemic ischemic stroke patients.

## MATERIALS AND METHODS

It is a Hospital based cross-sectional study conducted in a tertiary health care. 25 hyperglycemic and 25 normoglycemic ischemic stroke patient with informed consent are included for the study after approval of institutional ethical committee.

### Inclusion Criteria

- Newly diagnosed cases of hyperglycemic and normoglycemic ischemic stroke patients within the age group of 22 to 86 years of both sex.
- hyperglycemic ischemic stroke -diagnosed by CT scan, clinical signs & symptoms& FBS > 126mg/dl
- normoglycemic ischemic stroke - diagnosed by CT scan, clinical signs &symptoms& FBS < 126mg/dl

### Exclusion Criteria

Patients with stroke of more than 72 hours, central nervous system infections, peripartum stroke, head trauma, central nervous system, tumors, seizures.

About 5 ml of blood was drawn after 12 hours of fasting using disposable syringes and needles under strict aseptic precautions, from cubital vein and collected in a test tube. Serum NSE, fasting blood glucose, Fasting lipid profile were analyzed. Serum NSE was estimated by enzyme immunoassay based on biotin double antibody sandwich technology. Blood glucose was assayed by the glucose oxidase method. Total cholesterol, HDL and TGL were estimated by CHOD-PAP method, Direct method and Enzymatic method respectively. FRIEDEWALDS formula was used to calculate LDL = TC – (HDL + TGL / 5).

## ANALYSIS AND RESULTS

Statistical analysis was performed using SPSS for windows version 17. The mean and standard deviation for quantitative variables and percentage for qualitative variables were calculated for 25 hyperglycaemic and 25 normoglycemic ischemic stroke patients. Chi square test was used to compare differences in the percentage of qualitative variables between the groups. Differences in means of quantitative variables between the two groups were compared by student t test. *P* value of less than 0.05 is considered significant. Pearson correlation coefficient was obtained to study correlation between serum NSE, fasting blood glucose and lipid profile parameters.

**Table-1: Comparison of test parameters between hyperglycaemic and normoglycemic ischemic stroke patients**

| Variables | Stroke patients | n | Mean | S.D | t | *P* |
|---|---|---|---|---|---|---|
| **FBS** | Hyperglycemic | 25 | 215.6 | 64.1 | 8.016 | <0.001 |
| | Normoglycemic | 25 | 109.0 | 17.6 | | |
| **NSE** | Hyper glycemic | 25 | 13.5 | 5.0 | 4.361 | <0.001 |
| | Normoglycemic | 25 | 8.8 | 2.2 | | |
| **Modified** | Hyperglycemic | 25 | 3.8 | .5 | 8.017 | <0.001 |
| **Rankin scale** | Normoglycemic | 25 | 2.6 | .5 | | |
| **NIHSS score** | Hyperglycemic | 25 | 13.2 | 5.2 | 5.633 | <0.001 |
| | Normoglycemic | 25 | 6.4 | 2.9 | | |
| **Total** | Hyperglycemic | 25 | 203.5 | 38.3 | 3.294 | .002 |
| **cholesterol** | Normoglycemic | 25 | 169.2 | 35.3 | | |
| **HDL** | Hyperglycemic | 25 | 37.9 | 7.1 | -.831 | .410 |
| | Normoglycemic | 25 | 40.2 | 11.8 | | |
| **TAG** | Hyperglycemic | 25 | 125.4 | 48.1 | 1.262 | .213 |
| | Normoglycemic | 25 | 109.7 | 39.6 | | |
| **LDL** | Hyperglycemic | 25 | 140.6 | 37.7 | 3.222 | .002 |
| | Normoglycemic | 25 | 107.1 | 35.7 | | |

(FBS- Fasting blood sugar, HDL-High density lipoprotein, LDL- Low density lipoprotein, NSE-Neuron Specific Enolase, NIHSS – National institutes of health stroke scale).

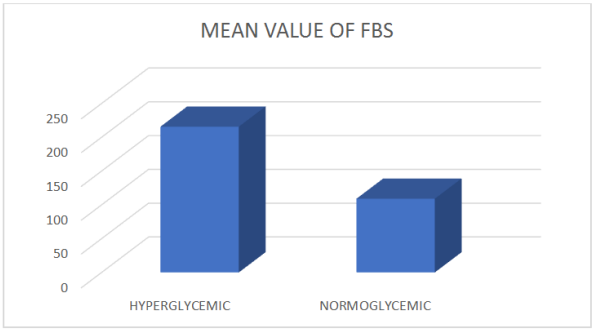


**Fig-1: Mean value of FBS in study groups**

The mean FBS values among hyperglycaemic stroke patients were 215.6 mg/dl and among normoglycemic stroke patients were 109. The difference was found to be statistically significant (*P* value < 0.05)**.**

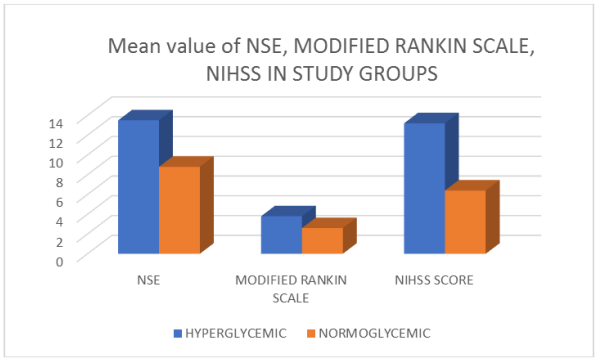


**Fig-2: Mean value of NSE, Modified Rankin Scale and NIHSS in study groups**

The mean serum NSE level was 13.5 among hyperglycemic stroke patients while it was 8.8 among normoglycemic stroke patients. This difference was found to be statistically significant (*P* value <.05). The mean NIHSS score was 13.2 among hyperglycemic stroke patients while it was 6.4 among normoglycemic stroke patients. This difference was found to be statistically significant (*P* value <.05). The mean modified Rankin scale was 3.8 among hyperglycemic stroke patients while it was 2.6 among normoglycemic stroke patients. This difference was found to be statistically significant (*P* value <.05).

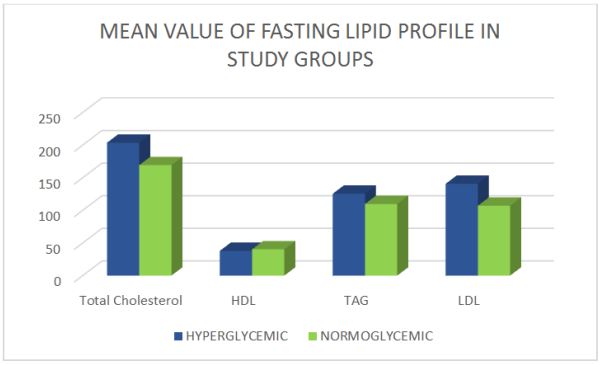


**Fig-3: Mean value of lipid profile in study group**

The mean serum cholesterol was 203.5 among hyperglycaemic stroke patients while it was 169.2 among normoglycemic stroke patients. This difference was found to be statistically significant (*P* value <.05). The mean serum Triacylglycerol was 125.4 among hyperglycaemic stroke patients while it was 109.7 among normoglycemic stroke patients. This difference was found to be statistically insignificant (*P* value >.05). The mean serum LDL was 140.6 among hyperglycaemic stroke patients while it was 107.1 among normoglycemic stroke patients. This difference was found to be statistically significant (*P* value <.05). The mean serum HDL was 37.9 among hyperglycaemic stroke patients while it was 40.2 among normoglycemic stroke patients. This difference was found to be statistically insignificant (*P* value >.05).

**Table-2: Pearson correlation between test parameters**

| Variables | Pearson Correlation (r) | *P* |
|---|---|---|
| glucose | .585** | <0.001 |
| Modified Rankin scale | .637** | <0.001 |
| NIHSS | .763** | <0.001 |
| Cholesterol | .200 | .163 |
| HDL | -.019 | .894 |
| TAG | -.036 | .806 |
| LDL | .214 | .136 |

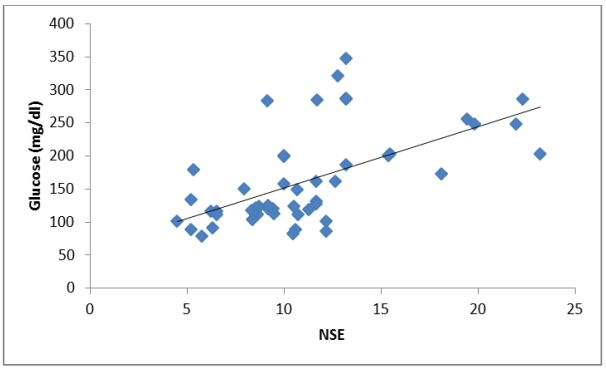


**Fig-4: Correlation between NSE and Glucose**

Serum NSE showed a positive correlation with serum FBS and it was found to be statistically significant (*P* value <.05).

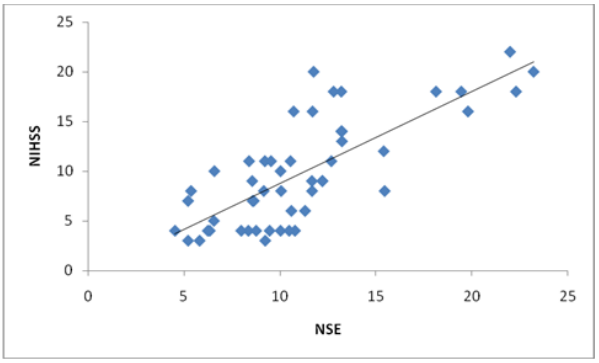


**Fig-5: Correlation between NSE and NIHSS score**

Serum NSE showed a positive correlation with NIHSS and it was found statistically significant (*P* value <.05).

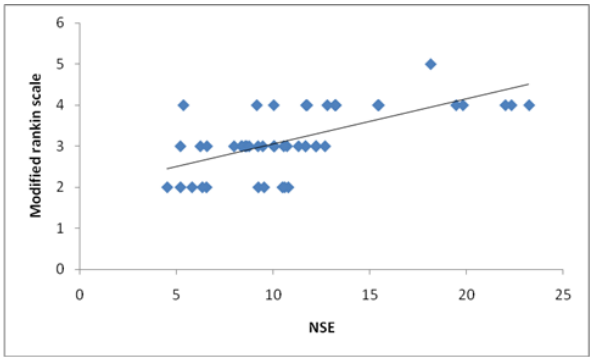


**Fig-6: Correlation between NSE and Modified Rankin Scale**

Serum NSE showed a positive correlation with modified Rankin scale and it was found statistically significant (*P* value <.05).

## DISCUSSION

A major proportion of stroke patients have stress hyperglycemia which increases the neuronal damage. The present study deals with the role of NSE as a marker of neuronal damage in ischemic stroke patients thereby comparing the severity of damage between hyperglycemic and normoglycemic ischemic stroke patients. Severity of neuronal damage was assessed by correlating NSE level with NIHSS score and modified Rankin scale. The increased NSE serum levels correspond to the ischemia-induced cytoplasm loss of NSE in the neurons and are detectable before irreversible neuronal damage takes place [16]. The mean serum NSE level among hyperglycemic ischemic stroke patients was 13.5± 5.0 and in normoglycemic ischemic stroke patients was 8.8 ± 2.2. Student t test was done to analyse the difference in mean NSE levels between these groups and the difference was found statistically significant. Pearson correlation was done to analyse the correlation between serum NSE and FBS levels. There was strong positive correlation between these two variables (r = 0.585) .This shows that as FBS levels increase, NSE levels increase.This was in accordance with the study by Pandey *et al.*, in 2011which had shown raised serum NSE level among hyperglycemic ischemic stroke patients [17].

NSE level shows strong positive correlation with NIHSS score (r= 0.763) and modified Rankin scale (0.637). These results was comparable with the study of Kiers L, Davis SM et al who have concluded hyperglycemia appears to be associated with more severe stroke, as assessed with a clinical stroke scale [18]. Our study is also comparable to studies reported by Selakovic V *et al*., [4] and Myers *et al.*, [19]. There was no significant difference in the distribution of NSE levels in accordance with age or sex among the two groups. This was in accordance with Missler *et al*., who showed that the serum level of NSE was not related with age or sex [6].

Various lipid profile parameters like serum total cholesterol, triglyceride, HDL and LDL were compared between hyperglycemic and of the normoglycemic ischemic stroke patients. There was no statistically significant relation on doing chi square test.NSE level has positive correlation with serum cholesterol (r= 0.200) and LDL level (r = 0.214). NSE level has negative correlation with TAG (r = -0.36) and HDL level (-0.19).The mechanism of lipid changes remains unclear, but it is thought to relate in part to the stress and associated catecholamine overproduction of an acute stroke.

## CONCLUSION

NSE in blood is the earliest parameter for prognostic classification, earlier than any other methods such as regularly repeated neurological examinations, neuroimaging (cranial CT or magnetic resonance imaging) or electrophysiological assays. Serum NSE levels are high in hyperglycaemic ischemic stroke patients compared to normoglycemic ischemic stroke patients. Neuron specific enolase (NSE) is a reliable marker of neuronal damage. So NSE can be used as a diagnostic parameter of stroke thereby to assess stroke severity.

## REFERANCES


1. World Health Organisation Principal Investigators. The World Health Organisation MONICA Project (monitoring trends and determinants in cardiovascular disease): a major international collaboration. J Clin Epidemiol. 1988;41(2):105-114.
2. Butterworth RJ, Wassif WS, Sherwood RA, Gerges A, Poyser KH, Garthwaite J, Peters TJ, Bath PM. Serum neuron-specific enolase, carnosinase, and their ratio in acute stroke: An enzymatic test for predicting outcome?. Stroke. 1996 Nov;27(11):2064-8.
3. Ishiguro Y, Kato K, Ito T, Nagaya M, Yamada N, Sugito T. Nervous system-specific enolase in serum as a marker for neuroblastoma. Pediatrics. 1983 Nov 1;72(5):696-700.
4. Selakovic V, Raicevic R, Radenovic L. The increase of neuron-specific enolase in cerebrospinal fluid and plasma as a marker of neuronal damage in patients with acute brain infarction. Journal of clinical neuroscience. 2005 Jun 1;12(5):542-547.
5. Cunningham RT, Young IS, Winder J, O'kane MJ, McKinstry S, Johnston CF, Dolan OM, Hawkins SA, Buchanan KD. Serum neurone specific enolase (NSE) levels as an indicator of neuronal damage in patients with cerebral infarction. European journal of clinical investigation. 1991 Oct;21(5):497-500.
6. Missler U, Wiesmann M, Friedrich C, Kaps M. S-100 protein and neuron-specific enolase concentrations in blood as indicators of infarction volume and prognosis in acute ischemic stroke. Stroke. 1997 Oct;28(10):1956-1960.
7. Scharschmiolt H, Prange HW, Reiber H. Neuronspecific enolase concentrations in blood as a prognostic parameter in cerebrovascular disease. Stroke. 1994;25:558-65.
8. Pulsinelli WA, Levy DE, Sigsbee B, Scherer P, Plum F. Increased damage after ischemic stroke in patients with hyperglycemia with or without established diabetes mellitus. The American journal of medicine. 1983 Apr 1;74(4):540-544.
9. Melamed E. Reactive hyperglycaemia in patients with acute stroke. Journal of the neurological sciences. 1976 Oct 1;29(2-4):267-275.
10. Vasudevan DM, Sreekumari S. Regulation of Blood glucose, Insulin and Diabetes Mellitus. Textbook of Biochemistry for medical students, 2013; 6, 277.
11. Siesjö BK, Bendek G, Koide T, Westerberg E, Wieloch T. Influence of acidosis on lipid peroxidation in brain tissues in vitro. Journal of Cerebral Blood Flow & Metabolism. 1985 Jun;5(2):253-258.
12. OuYang YB, Mellergård P, Kristian T, Kristianova V, Siesjö BK. Influence of acid-base changes on the intracellular calcium concentration of neurons in primary culture. Experimental brain research. 1994 Sep 1;101(2):265-71.
13. Choi DW, Rothman SM. The role of glutamate neurotoxicity in hypoxic-ischemic neuronal death. Annual review of neuroscience. 1990 Mar;13(1):171-182.
14. Anderson RE, Tan WK, Martin HS, Meyer FB. Effects of glucose and PaO2 modulation on cortical intracellular acidosis, NADH redox state, and infarction in the ischemic penumbra. Stroke. 1999 Jan 1;30(1):160-170.
15. Olsen TS, Larsen B, Herning M, Skriver EB, Lassen NA. Blood flow and vascular reactivity in collaterally perfused brain tissue. Evidence of an ischemic penumbra in patients with acute stroke. Stroke. 1983 May;14(3):332-341.
16. Wunderlich MT, Ebert AD, Kratz T, Goertler M, Jost S, Herrmann M. Early neurobehavioral outcome after stroke is related to release of neurobiochemical markers of brain damage. Stroke. 1999 Jun;30(6):1190-5.
17. Pandey A, Saxena K, Verma M, Bharosay A. Correlative study between neuron-specific enolase and blood sugar level in ischemic stroke patients. Journal of neurosciences in rural practice. 2011 Jan;2(1):50-54.
18. Woo E, Chan YW, Yu YL, Huang CY. Admission glucose level in relation to mortality and morbidity outcome in 252 stroke patients. Stroke. 1988 Feb;19(2):185-91.
19. Myers MG, Norris JW, Hachniski VC, Sole MJ. Plasma norepinephrine in stroke. Stroke. 1981 Mar;12(2):200-4.