Research Paper www.ijcps.com

International Journal of Chemical and Pharmaceutical Sciences
2013, Mar., Vol. 4 (1)

ISSN: 0976-9390
IJCPS

Evaluation of diagnostic importance of serum NSE levels in patients after cardiac arrest for early predication of outcome

[1] Junaid Mahmood Alam*, [2] Sikandar Khan Sherwani, [3] Tahseen Kazmi, [1]Amna Hussain, [4]Syed Riaz Mahmood, [1] Ishrat Sultana ,[5] Sarah Sughra Asghar and [6] Muhammad Ajmal Shah.

[1] Department of Biochemistry and Laboratory Services-Liaquat National Hospital and Medical College, Karachi. Pakistan.

[2] Department of Microbiology, Federal Urdu University of Science and Technology, Karachi, Pakistan.

[3] Department of Community Medicine, Liaquat National and Medical Hospital, Karachi, Pakistan.

[4] Department of Pathology, Govt Lyari General Hospital, Karachi, Pakistan.

[5] Department of Anatomy, Basic Medical Science Institute, Jinnah Postgraduate Medical center-Karachi. Pakistan.

[6] Department of Pharmacognosy, Federal Urdu University of Arts, Science and Technology, Karachi, Pakistan

***Corresponding Author:** E- sikander_biology@hotmail.coml.

ABSTRACT

Present study describes the evaluation of diagnostics value of Neuron Specific Enolase (NSE) in patients with cardiac arrest of related cardiac dysfunction. **Objectives:** To examine the diagnostic and prognostic value of serum NSE for early prediction of outcome in patients after cardiac arrest. **Study Design**: Cross-Sectional study. Place and duration of the study. The Emergency Department and Biochemistry Lab, Liaquat National Hospital, Karachi from June 2006 to December 2007. *Patients and Methods*: A total of 92 patients brought to the Emergency Department of LNH with cardiac arrest, dysfunction and abnormalities, were included in the study. Each of these patients was successfully resuscitated and shifted to either ICU or CCU. Blood samples were taken to compare levels of NSE along with other markers such as AST, LDH, CK and CKMB iso-enzyme. **Results:** increasing levels of NSE significantly ($P<0.01$) correlated with the increasing value of CK and CKMB. Moreover, high AST levels also show borderline significance with high NSE level, when gender-wise independent t-test for equality of variance was performed. The results depict further diagnostic importance, at par with three out of four biochemical cardiac markers (CK, CKMB, AST except LDH). **Conclusion:** In present study, estimated NSE levels, determined in a group of males and females patients suffering from cardiac arrest or related cardiac dysfunction, was found to be a significant diagnostic tool, statistically evaluated to be a par with the diagnostically important CK and CKMB (inclusive of AST when correlated cumulatively), the two tests of choice in cardiac abnormalities.

Key Words: Neuron Specific Enolase, Cardiac Arrest, Diagnostic Significance.

1. INTRODUCTION

In spite of all the advances in cardiac pulmonary managements, morbidity and mortality associated with cardiac arrest remain extremely high [1]. The prognosis is also not very promising with mild to moderate disability to persistent vegetative state [1]. It is reported that 80% of sudden death survivors remain in a coma for various lengths of time, and a full neurological recovery is still rare [1, 2]. It has been postulated that an irreversible anoxic brain damage might have occurred soon after the arrest. It has been suggested that to facilitate evaluation of cardiac arrest patients, an accurate prognostic tool may have major ethical and economic consequences [1].

Presently, prognosis based on several clinical, neuro-imaging and electrophysiological methods [1, 3-6], which sometimes difficult to administer and manage. In this regard biochemical markers, in comparison, are a low cost-alternative that may be more suitable both economically and management-wise. Neuron-specific enolase (NSE) is a known marker of ischemic brain damage and already been evaluated in traumatic brain injury [7], stroke [8] and anoxic encephalopathy after cardiac arrest [9-10]. NSE, the neuronal form of the glycolytic

enzyme enolase, is found almost exclusively in neurons and cells of neuroendocrine origin [1]. It is a di-meric form compounded of two subunits that converts 2-phosphoglycerate into phosphoenolpyruvate, measurable in blood and cerebrospinal fluid [11]. There are several studies carried out on the prognostic value of NSE in patients surviving in-hospital cardiac arrest [1], post-anoxic coma [12], long-term outcome [13] out of hospital cardiac arrest [14] and cardiac surgery [15]. To the best of our knowledge, there have been no studies in our setting to evaluate diagnostic value of NSE in patients with cardiac arrest or related cardiac dysfunction.

2. MATERIALS AND METHODS

We prospectively evaluated 92 patients (Males 68, Females 24) who came to Emergency Ward (ER) of our hospital in the state of cardiac arrest in the period from June 2006 to December 2007 at Liaquat National Hospital, a tertiary-care hospital of Karachi. We included patients who were successfully resuscitated after coming to ER for cardiac arrest or related cardiac dysfunction, pulseless ventricular tachycardia, pulseless electrical activity and asystole or premature ventricular contraction, ventricular tachycardia, arterial flutter, ST segment evaluation, development of abnormal Q-waves. We excluded patients under the age of 25 years, those presenting drug intoxication, accidental or therapeutic hypothermia, those with neoplastic diseases known to increase NSE levels, stroke ischemic and/or hemorrhagic) or traumatic brain injury and patients subjected to extracorporeal circulation.

Patients were evaluated in term of signs and symptoms and time interval to blood sampling for NSE measurement. Every resuscitated patient was admitted to an intensive care unit or Coronary unit ant the care provided followed routine of the units, without interference from the investigators. Blood sampling took place for NSE measurement between 12 and 36 hours after cardiac crest. Attending physicians and the critical care team were unaware of the result of NSE measurements. None of the patients had a do-not-resuscitate order and there was no limitation of life support.

2.1. Procedure

Protocol [1] was followed. Blood samples were withdrawn by peripheral vein puncture and centrifuged for 10 minutes at 2500 rotations per minutes. Serum (1 ml) was frozen and stored at -86^{o}C. Hemolyzed sample were considered lost. NSE measurements were performed with an electrochemiluminescence immunoassay (ECLIA), using a sandwich technique, in duplicate, with NSE kits (Roche, Diagnostic, Basil) and the Elecsys 2010 analyzer (Roche Diagnostics, Basil). NSE measurements were also performed in twenty control individuals. Values will be considered normal if is it less than the reported (Roche Diagnostics, Basil) value of 1.3 ng/ml. other parameters that were routinely performed were AST, LDH, CK, CKMB on Hitachi 912 (Roche Diagnostic, Basil). Normal reference ranges of AST, LDH, CK and CKMB are, upto 40 U/L, 230-460 IU/L, upto 270 U/L and < 25 U/L, respectively.

2.2. Statistical Analysis

Continuous data are presented as means and SD, and nonparametric data as medians range. Student's t test and the Pearson's correlation (2 tailed), were used to compare the data. The discriminative power of NSE to predict an unfavorable outcome was determined by analysis of all cardiac parameters (AST, LDH, CK, CKMB). The significance level was set at $p < 0.05$ and $P < 0.01$. Statistical analysis was performed with statistical package for the Social Sciences (SPSS) version 15.

3. RESULTS

All 92 patients included were grouped as 68 males and 24 females and were brought to ER with complaints of suspected cardiac arrest and related cardiac dysfunction. Table 1 shows NSE levels measured cumulatively in males and females as a single group as well as individual gender-wise. Moreover NSE levels were measured between 10 to 15 hours after admittance of the patients in ER. Cumulative NSE mean level of all patients was 36.45 ng/ml ± 1.5 with median of 33.50 ng/ml (minimum 18.91 ng/ml; maximum 61.0 ng/ml) (Table 1), whereas that of AST = 104.04 ± 12.41 IU/L; LDH = 688.90 ± 153.349 IU/L; CPK = 632.31 ± 193.884 IU/L and CKMB = 39.40 ± 16.20 IU/L. NSE estimated individually gender wise shows mean level of 37.55 ng/ml ± 11.8 in males and mean level of 33.41 ng/ml ± 12.8 in females (Fig 1).

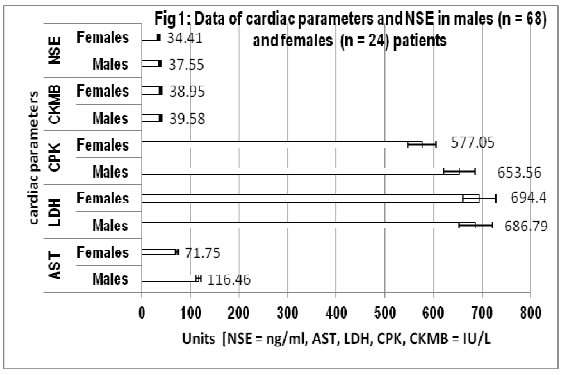

Table - 1: Statistical analysis of biochemical markers per Frequencies in cumulative group of both males (68) and females (24) patients

Parameters	AST(IU/L)	LDH(IU/L)	CPK(IU/L)	CKMB(IU/L)	NSE(ng/ml)
N	92	92	92	92	92
Mean	104.04	688.90	632.31	39.40	34.38
Std. Deviation ±	± 12.41	± 153.349	± 193.884	± 16.208	± 11.088

Table - 2: Pearson's correlation analysis of NSE with other diagnostically important cardiac biomarkers in cumulative group of 92 patients [males 92; females = 24]

Test	Statistical analysis	CKMB IU/L	AST IU/L	LDH IU/L	CPK IU/L
NSE (ng/ml)	Pearson's Correlation	.337 (**)	0.060	0.121	.344 (**)
	Sig. (2-tailed)	.004	.615	.310	0.003

**** Correlation is significant at the 0.01 level (2-tailed)**

Table - 3:Pearson's correlation analysis of NSE with other diagnostically important cardiac biomarkers in individual group of 68 male patients

Test	Statistical analysis	CKMB IU/L	AST IU/L	LDH IU/L	CPK IU/L
NSE (ng/ml)	Pearson's Correlation	.295 (**)	0.021	0.066	.286 (**)
	Sig. (2-tailed)	.034	.881	.640	0.040

**** Correlation is significant at the 0.05 level (2-tailed)**

Table - 4: Pearson's correlation analysis of NSE with other diagnostically important cardiac biomarkers in individual group of 24 female patients

Test	Statistical analysis	CKMB IU/L	AST IU/L	LDH IU/L	CPK IU/L
NSE (ng/ml)	Pearson's Correlation	.486 (**)	0.187	0.347	.457 (**)
	Sig. (2-tailed)	.030	.429	.134	0.043

**** Correlation is significant at the 0.05 level (2-tailed)**

All patients with higher NSE values > 30.0 ng/ml (normal reference value < 3.0 ng/ml) showed equally high CK and CKMB levels with coherent clinical and cardiac characteristics of moderate to severe potency, where those with NSE levels < 30.0 ng/ml depicts milder clinical signs and symptoms and subsequent mild to moderate levels of CK and CKMB. Interestingly, when CKMB level estimated in patients were compared with NSE (Pearson's correlation, two tailed, $p < 0.05$), a significant correlation was noted ($p < 0.04$) for the group (Table 2). Similarly, CK comparison with NSE also showed significant correlation ($P < 0.03$) for all 92 patients. Individual gender-wise (Pearson's correlation $P < 0.05$) estimation of NSE with CKMB and CK in males ($P< 0.034$; $P < 0.04$, respectively) (Table 3) and females ($P< 0.043$; $P< 0.034$, respectively) (Table 4) also showed mild to moderate significance. Such significant correlation of raised NSE levels with CK and CKMB depicts its diagnostic importance in cardiac problems as both CK and CKMB raised values were directly linked with MI, cardiac arrest and related cardiac dysfunction.

Correlation of NSE with AST and LDH were found to be non-significant in all estimations in cumulative group as per Pearson's correlation estimations. However when cumulative-independent sample t-test was performed as per Levene's test for equality of variance, AST and NSE values were statistically significant. The diagnostic significance of NSE in comparison with CK and CKMB was also estimated regression-correlation graph. NSE showed moderate correlation linearity with CK ($R^2 = 0.119$) and CKMB ($R^2 = 0.114$) in all patients evaluated, where as AST and LDH did not ($R^2 = 0.004$ and $R^2 = 0.015$, respectively).

4. DISCUSSION

In the present study NSE levels, estimated in males and females 10-15 hours after the occurrence of MI, cardiac arrest or related cardiac dysfunction, was found to be a significant diagnostic tool, statistically evaluated to be at par with the diagnostically important CK and CKMB, the two tests of choice in cardiac abnormalities. Previous and recent studies reported the predictive and prognostic importance of NSE in cardiac arrest patients [12-14, 16-19]. It is also reported that NSE levels remain significantly higher after 24 and 48 hrs in patients who didn't regain consciousness even after CPR than in patients who regained consciousness [12]. Moreover, it is also noted that patients who underwent cardiac arrest, and exhibited NSE concentrations > 33.0 ng/ml, depicts persistent coma [17]. Similarly, serum NSE levels greater than or equal to 65 ng/ml in non-traumatic cardiac arrest patients suggest increased risk of death or persistent vegetative state [16]. Furthermore, estimation of NSE level was also reported to be effective prognostic indicator, along with CKMB in hypoxic brain injury after cardiac arrest [20]. In our study, collective as well group (gender-wise) evaluation of NSE showed similar significant level of diagnostic correlation with clinical characteristics. It has also shown significant correlation ($P < 0.05$) with the established cardiac arrest markers of CK and CKMB, in all patients. NSE estimation and resultant increased level of it, in cardiac arrest patients also predicts long-term outcome in these patients along with S-100 protein, an established biochemical marker of CNS injury [13]. It was asserted that the prognostic value of brain damage marker was comparable with that of traditional clinical parameters [13]. High levels of NSE predict a poor outcome, according to the GOS (Glasgow Outcome Scale) and GCS (Glasgow Coma Scale) [1, 13, and 16]. Moreover, increased NSE levels along with S-100 also reported to provide additional information, when estimated to predict cognitive dysfunction after resuscitation from out of hospital cardiac arrest [15]. In addition, few recent studies also suggests significance of NSE level assessment that correlated with cardiac arrest [21], poor outcome, death or vegetative state [22] and other markers of brain injuries and electrographic status epilepticus (ESE) [23].

In present study, increasing level of NSE (18.91-61.0 ng/ml; mean 36.45 ngml) also correlated with the increasing value CK (134-997 IU/L, mean 632.31 IU/L) and CKMB (13-96 IU/L; 39.40 IU/L), both cumulatively and gender-wise, thus suggesting its diagnostic importance in cardiac abnormality and therefore asserting a poor outcome after cardiac arrest or dysfunction. Interestingly, high AST levels (51-664 IU/L; mean 104 IU/L) also depicts borderline significance with high NSE level, when cumulative independent t-test for equality of variance was performed as per Levene's test, thus depicting a further diagnostic importance, at par with three out of four biochemical cardiac markers, i.e. CK, CKMB, AST except LDH.

It was reported that the decreasing NSE values (< 25.0 µg/l) at 6 months intervals in cardiac arrest patients, treated with hypothermia is associated with good outcome ($P < 0.005$), recovery of consciousness ($P < 0.001$) and survival for atleast 6 months after cardiac arrest ($P< 0.012$) [18]. Examination of the prognostic vale of NSE for early prediction of outcome in patients at risk for anoxic encephalopathy after cardiac arrest, also results in high specificity and a

positive predictive value [17]. It was suggested that patients who have been resuscitated after cardiac arrest, high levels of NSE (> 33 ng/ml) predicts persistence coma with high specificity [17]. The combination of GCS and serum value of NSE greater than or equal to higher value (> 60 ng/ml) predicts increased risk of death and persistent vegetative state [13]. It was conclude that combined evaluation of GCS with neuro-proteins NSE and S-100 protein at 72 hr after CPR (cardiac pulmonary resuscitation) permits more reliable prediction of outcome in post arrest coma patients [13].

In conclusion, present study represents estimated NSE levels, determined in a group of male and female patients suffering from cardiac arrest or related cardiac dysfunction, that was found to be a significant diagnostic tool, statistically evaluated to be at par with the diagnostically important CK and CKMB (inclusive of AST when correlated cumulatively), the two tests of choice in cardiac abnormalities.

5. REFERENCES

1. Rech TH, Vieira SRR, Nagel F, Brauner JS and Scalco R. Serum neuron-specific enolase as early predictor of outcome after in-hospital cardiac arrest: a cohort study. **Critical Care,** 2006; 10: 1-6
2. Madl C and Holzer M. Brain function after resuscitation from cardiac arrest. **Curr Opin Crit Care,** 2004;10**:** 213-7
3. Madl C, Karmer L, Domanovits H, Woolard RH, Gervais H, Gendo A, Eisenhuber E, Grimm G and Sterz F. Improved outcome prediction in unconscious cardiac arrest survivors with sensory evoked potentials compared with clinical assessment. **Crit Care Med,** 2000; 28: 721-6.
4. Ballew KA, Philbrick JT, Caven DE and Schorling JB. Predictors of survival following in-hospital cardio-pulmonary resuscitation. A moving target. **Arch Intern Med,** 1994; 154**:** 2426-32.
5. Nakabayashi M, Kurokawa A and Yamamoto Y. Immediate prediction of recovery of consciousness after cardiac arrest. **Intensive Care Med,** 2001; 27: 1210-4.
6. Zandbergen EG, de Haan RJ, Stoutenbeek CP, Koelman JH and Hijdra A. Systemic review of early prediction of poor outcome in anoxic-ischaemic coma. **Lancet,** 1998; 352: 1808-12.
7. Pleines UE, Morganti-Kossmann MC, Rancan M, Joller H, Trentz O and Kossmann T. S-100 beta reflects the extent of injury and outcome, whereas neuronal specific enolase is a better indicator of neuro-inflammation in patients with severe traumatic bran injury. **J Neurotrauma,** 2001; 18: 491-498.
8. Anand N and Stead LG. Neuron-specific enolase as a marker for acute ischemic stroke: a systemic review. **Cerebrovasc Dis,** 2005; 20: 213-9.
9. Martens P, Raabe A and Johnsson P. Serum S-100 and neuron specific enolase for prediction of regaining consciousness after global cerebral ischemia. **Stroke,** 1998; 29: 2363-6.
10. Pfeifer R, Borner A, Krack A, Sigusch HH, Surber R and Figulla HR. Outcome after cardiac arrest: predictive values and limitations of the neuro-proteins neuron-specific enolase and protein S-100 and the Glasgow Coma Scale. **Resuscitation,** 2005; 65: 49-55.
11. Johnsson P, Blomquist S, Luhrs C, Malmkvist G, Alling, C, Solem JO and Stahl E. Neuron-specific enolase increases in plasma during and immediately after extracorporeal circulation. **Ann Thorac Surg,** 2000; 69: 750-4.
12. Meynaar IA, Oudemans-van Straaten HM, an der Wetering J, Verlooy P, Slaats EH, Bosman RJ, van der Spoel JI and Zandstra DF. Serum neuron-specific enolase predicts outcome in post-anoxic coma: a predictive cohort study. **Intensive Care Med,** 2003; 29(2): 189-95.
13. Rosen I, Sunnerhagen SK, Herlitz J, Blomstrand C and Rosengren L. Serum levels of the brain-derived proteins S-100 and NSE predict long-term outcome after cardiac arrest. **Resuscitation,** 2001; 49: 183-191.
14. Grubb NR, Simpson C, Sherwood RA, Abraha HD, Cobb, SM, O'Carroll RE, Deary I and Fox KAA. Prediction of cognitive dysfunction after resuscitation from out of-hospital cardiac arrest using serum neuron-specific enolase and protein S-100. **Heart,** 2007; **93**: 1268-73.
15. Schmitt B, Bauersfeld U, Schmid ER, Tuchschmid P, Molinari L, Fanconi S. and Bandtlow, C. Serum and CSF levels of NSE in cardiac surgery with cardio-pulmonary bypass: a marker of brain injury? **Brain and Development,** 1998; 20: 536-9.
16. Almaraz AC, Bobrow BJ, Wingerchuk DM, Wellik KE and Demaerschalk BM. Serum neuron specific enolase to predict neurological outcome after cardiopulmonary resuscitation: a critically appraised topic. **Neurologist,** 2009; 15 (1): 44-48.
17. Fogel W, Krieger D, Veith M, Adams HP, Hund E, Storch-Hagenlocher B, Buggle F, Mathias D.

and Hacke W. Serum NSE as early predictor of outcome after cardiac arrest. **Crit Care Med,** 1997; 25: 1133-8.

18. Tiainen M, Roine RO, Pettila V and Takkunen O. Serum NSE and S-100B protein in Cardiac arrest patients treated with hypothermia. **Stroke**, 2003; 34: 2881-6.
19. Zingler VC, Krumm B, Bertsch T, Fassbender K and Pohlmann-Eden B. Early prediction of neurological outcome after cardio-pulmonary resuscitation; a multi-model approach combining neuro-biochemical and electrophysiological investigations may provide high prognostic certainty in patients after cardiac arrest. **Eur Neurol,** 2003; 49: 79-84.
20. Karkela J, Bock E and Kaukinen S. 1993. CSF and serum brain-specific creatine kinase iso-enzyme (CK-BB), neuron-specific enolase (NSE) and neural cell adhesion molecule (NCAM) as prognostic marker for hypoxic brain injury after cardiac arrest in man. **J Neurol Sci,** 1993; 116: 100-9
21. Sulaj M, Saniova B, Drobna E and Schudichova J. Serum neuron specific enolase and malondialdehyde in patients after out-of-hospital cardiac arrest. **Cell Mol Neurobiol,** 2009; 29: 807-10.
22. Daubin C, Quentin C, Allouche S, Etard O, Gaillard C, Seguin A, Valette X, Parienti JJ, Prevost F, Ramakers M, Terzi N, Charbonneau P and du Cheyron D. Serum neuron-specific enolase as predictor of outcome in comatose cardiac-arrest survivors: a prospective cohort study. BMC C**ardiovasc Disord,**2011; **8;** 11:48.
23. Cronberg T, Rundgren M, Westhall E, Englund, E, Siemund R, Rosén I, Widner H and Friberg H. Neuron-specific enolase correlates with other prognostic markers after cardiac arrest. **Neurology,** 2011; 16:623-30.