Serum Neuron Specific Enolase (NSE) as a biomarker for Hypoxic Ischemic Encephalopathy in a tertiary care hospital- A prospective study.

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Abstract: Background: The cause for disease and mortality throughout the infant was hypoxic ischemic encephalopathy secondary to perinatal asphyxia. It leads to permanent neuropsychological disability. Identification of a biomarker for hypoxic insult can not only help in earlier implementation of neuro-protective strategies but also in prognosticating the long term outcome.

Method: A prospective observational study conducted in IMS and SUM hospital over a period of 2 years in which 60 newborns with clinical evidence of HIE were recruited as case group. A group of 20 newborns with no evidence of HIE served as control. The serum neuron specific enolase (NSE) levels at 4 hours and 48 hours of birth of both the groups were tested and compared.

Results: The mean serum NSE levels at 4 hours and 48 hours was significantly higher in the case group (42.43 ng/ml, 28.97 ng/ml) as compared to the control group (18.51 ng/ml, 15.62 ng/ml). Additionally, the mean serum NSE stages increased by the harshness of HIE in the various subgroups as per the clinical staging. ROC curve for this study has shown a good predictability for neurological outcome. For the cut off value of 40.4 mcg/l, the sensitivity was 80 % and specificity was 81.2 %.

Conclusion: Serum NSE levels can be a reliable biomarker for neurological insult in Hypoxic ischemic Encephalopathy insult in order to implement early neuroprotective strategy

Keywords: Neuron precise enolase, Hypoxic ischemic encephalopathy,
1. INTRODUCTION:

The cause for neonatal deaths globally is HIE secondary, owing toward perinatal asphyxia. HIE is occurrence between 1 and 3 in 1000 filled-term babies, with around 60% of pre-advanced children. About 15 to 20 per cent of post-natal infection are impaired by neuropsychological impairments, include cerebral paralysis, mental retardation, intellectual disabilities and serious and irreversible neuropsychological impairments.

The capacity of a physician to determine the future of HIE newborns isn't really easy. The Sarnat grading scale (Sarnat, 1976) uses clinical methods to evaluate the HIE level. This theory classifies neonates in and out of mild, modest, or serious classifications to anticipate the diagnosis for a Neonate (Finer et al., 1981) as well as indicators the advance of neurological slur. The Sarnat score system however is arbitrary but over period evolving. A new bedside device, incorporated electroencephalographic amplitude (aEEG), can help to assess accident intensity and model forecasting (Hellstrom-Westas etc., 1995). Sadly, the ratings of Sarnat and aEEG remain not quite as efficient in forecasting results in hypothermia in newborns (Thoresen et al., 2010). Don't give the time of the accident details. Brain MRI may help decide whether the injuries happened, but MRI to dysfunctional individuals isn't really feasible.

The very next objective is to establish bio-markers which can help clinically decide the neural endocrinology-protection. Biomarker's main objective is to consider lesion in long-term effects.

NSE is among the enolase proteins among all glycolytic tissues and species. NSE throughout the nervous system is significant, and platelets, RBC and kidneys are substantially lower. It could be distinguished in brain, liver, muscular as well as platelet stages of non-neuronal enolase current in RBC and at a much reduced rate. As platelet and red blood cell contain NSE, haemolysed sample cannot be tested for NSE. NSE is biologically important in the blood plasma only at small quantities. CSF-NSE is 17.3±4.6 ng / mL; serum seems to be 8.7±3.9 ng / mL NSE saturation.

Serum NSE is indeed a proxy for neuronal damage following brain injuries, neurological including neurodegenerative illnesses. The serum NSE saturation associated by HIE is a measure of neuronal death and can be utilized to determine the harseness and result of HIE Serum NSE.

2. MATERIAL AND METHOD:-

The study is a prospective observational study conducted in IMS and SUM hospital over a period of 2 years with proper ethical committee clearance.

INCLUSION CRITERIA: - 1.Gestational age >37wk
   2. postnatal age <6hr
   3. Need of resuscitation with positive pressure ventilation >3mins.

EXCLUSION CRITERIA: - 1.Gestational age <37wk
   2. Post natal age > 6hr
   3. Syndromic babies

We have recruited 60 newborn babies satisfying the inclusion and exclusion criteria. These newborns were labelled as Cases divided into subgroups of HIE level I, HIE level II besides HIE level III based on Sarnath & Sarnath staging. We have also recruited 20 newborns without any history of perinatal asphyxia to serve as a Control.

Blood sample as well as examination: Blood samplings were taken by aseptic methods. These blood samples were transferred to laboratory on same day maintaining proper
temperature. Electrochemiluminescence immuno assay principal was used to assay the NSE.

3. RESULTS:

Out of 60 newborns selected as cases, 32 cases were diagnosed as HIE I, 18 cases as HIE II and 10 cases as HIE III. During the process of blood collection and storage some blood sample were haemolysed from case group, so 72 participants (52 cases and 20 controls) were included in the final analysis.

Table 1 shows that of the 72 cases 39 were males (54%) and 33 were females (46%). In the various subgroups also males had greater incidence as compared to females.

Table 1:

<table>
<thead>
<tr>
<th>GENDER</th>
<th>CONTROL GROUP</th>
<th>HIE I</th>
<th>HIE II</th>
<th>HIE III</th>
<th>TOTAL</th>
</tr>
</thead>
<tbody>
<tr>
<td>MALE</td>
<td>11</td>
<td>16</td>
<td>9</td>
<td>6</td>
<td>39</td>
</tr>
<tr>
<td>FEMALE</td>
<td>9</td>
<td>11</td>
<td>7</td>
<td>3</td>
<td>33</td>
</tr>
<tr>
<td>TOTAL</td>
<td>20</td>
<td>27</td>
<td>16</td>
<td>9</td>
<td>72</td>
</tr>
</tbody>
</table>

Table 2 shows that in both groups, serum NSE was collected at both 4hr and 48 hrs. All measurements were made in ng/mL. In control group mean serum NSE value at 4hrs was 18.51, which decreased to 15.62 at 48hrs. In case group serum NSE value at 4hr was 42.43, which was decreased to 28.97 at 48hrs which was expressively greater than the regulator group.

Table 2:

<table>
<thead>
<tr>
<th>Groups</th>
<th>Serum NSE at 4 hr</th>
<th>Serum NSE at 48hr</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control group</td>
<td>18.51ng/ml</td>
<td>15.62 ng/ml</td>
</tr>
<tr>
<td>Case group</td>
<td>42.43ng/ml</td>
<td>28.97 ng/ml</td>
</tr>
</tbody>
</table>

Table 3 shows the comparison between serum NSE levels in the different subgroup of cases at 4hours and 48hrs respectively. It was found that NSE concentration at 4hrs was 27.11ng/ml, 42.36ng/ml and 60.69 ng/ml in HIE I, HIEII and HIE III respectively. Similarly at 48hrs it was 18.44 ng/ml, 27.72 ng/ml and 13.25 ng/ml in HIE I, HIEII and HIE III respectively.

Table 3

<table>
<thead>
<tr>
<th>Groups</th>
<th>Serum NSE at 4hrs</th>
<th>Serum NSE at 48hr</th>
</tr>
</thead>
<tbody>
<tr>
<td>HIE I</td>
<td>27.11</td>
<td>18.44</td>
</tr>
<tr>
<td>HIE II</td>
<td>42.36</td>
<td>27.72</td>
</tr>
<tr>
<td>HIE III</td>
<td>60.69</td>
<td>13.25</td>
</tr>
</tbody>
</table>

Thus the levels increased correspondingly with the severity of HIE.

The neonates were followed up till 1 year with respect to neurodevelopment. Table 4 shows the comparison between children with normal development and those with neurodevelopmental delay with respect to mean serum NSE levels at 4 hours and 48 hours of life. The mean NSE levels at 4hours and 48 hours were significantly higher in developmentally delayed child as compared to developmentally normal children in each of the subgroups.
Serum NSE at 4hrs

<table>
<thead>
<tr>
<th>GROUPS</th>
<th>Normal child</th>
<th>Developmental delay</th>
</tr>
</thead>
<tbody>
<tr>
<td>HIE I</td>
<td>25.60</td>
<td>35.16</td>
</tr>
<tr>
<td>HIE II</td>
<td>38.21</td>
<td>52.32</td>
</tr>
<tr>
<td>HIE III</td>
<td>49.47</td>
<td>71.91</td>
</tr>
</tbody>
</table>

Serum NSE at 48hrs

<table>
<thead>
<tr>
<th>GROUPS</th>
<th>NORMAL CHILD</th>
<th>DEVELOPMENTAL DELAY</th>
</tr>
</thead>
<tbody>
<tr>
<td>HIE I</td>
<td>16.86</td>
<td>26.86</td>
</tr>
<tr>
<td>HIE II</td>
<td>24.76</td>
<td>34.81</td>
</tr>
<tr>
<td>HIE III</td>
<td>36.25</td>
<td>49.34</td>
</tr>
</tbody>
</table>

Figure 1 shows the ROC curve for serum NSE levels. Area under the curve is 0.884 which is suggestive of high predictability of NSE for neurological outcome. It was also seen that for the cut off value of 40.4 mcg/l, the sensitivity was 80 % and specificity was 81.2 %

Fig 1: ROC CURVE

4. DISCUSSION:

Notwithstanding advancements of therapeutic and technical progress, the higher mortality rates of HIE remain being a topic of significant worry. The new emergence of neuroprotective approaches demonstrates its need for rapid and precise guidance as well as for the treatment and outcomes of HIE. Biochemical features may help studies into neuroprotection. Biomarkers were compounds that are emitted or those that are unique to a certain organ and it may provide an indication including its physiologic or pathological state of the organism. NSE is a member including its enzyme system susceptible of glycolysis. NSE recognition is
predicted to occur in the peripheral serum following neuronal mortality and blood brain obstructed destruction.

As per ROC lines, serum NSE tier above 40 mg / l among 4 a.m. and 48 hr. can differentiate infants without or mild HIE from newborns to severe or profound HIE [11], which was researched by Ceutiket et al. particular enolase as an HIE intensity indicator. Serum NSE is provided with such a 45.4 mcg / l cutting point for good performance. The mean serum NSE values were meaningfully better throughout the cluster compared than those throughout the treatment cluster throughout the current study at 240 minutes and 48 hours. In addition, mean serum NSE stages in various subsets have expanded only with intensity of HIE for every clinical stage. ROC curve for our study has shown a good predictability for neurological outcome. It was also seen that for the cut off value of 40.4 mcg/l, the sensitivity was 80 % and specificity was 81.2 %. Several studies measuring NSE level in serum besides CSF of newborns by HIE has shown similar result. Thonberg et. al discovered great NSE level in 2 circumstances of position II HIE as well as in all mark III HIE [12].

In numerous neurological disorders, NSE was already formed as a valid and effective indicator of brain loss and diagnosis. Nevertheless, few findings are available on its importance for diagnosing and forecasting HIE in the case of perinatal asphyxiation. Several of these have utilized NSE as an indicator to neurological impairedness as the CSF benefit. That is mainly because of the facility of blood processing relative to a number of lateral punctures. The findings of this investigation shows serum NSE by HIE degrees including neurological tests Predictive potential. However, studies need to be conducted in a larger population so as to corroborate the result in a larger scale in order to implement early neuroprotective strategies.

REFERENCES


