Serum neuron-specific enolase and S-100β levels as prognostic follow-up markers for oxygen administered carbon monoxide intoxication cases

Ali Osman Yildirim1, Murat Eroglu1, Umit Kaldirim2, Yusuf Emrah Eyi2, Kemal Simsek3*, Murat Durusu2, Levent Yamanel2, Ibrahim Arziman2, Salim Kemal Tuncer2, Mehmet Toygar4, Arzu Balkan5, Tuncer Cayci6, Seref Demirbas7, Sukru Oter8 and Cumhur Bilgi6

1Department of Emergency Medicine, Gulhane Haydarpasa Training Hospital, Istanbul, Turkey
Departments of 2Emergency Medicine, 3Undersea & Hyperbaric Medicine, 4Forensic Medicine, 5Pulmonary Medicine, 6Biochemistry, 7Internal Medicine, and 8Physiology, Gulhane Military Medical Academy, Ankara, Turkey

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Serum neuron-specific enolase (NSE) and S-100β levels are considered novel biochemical markers of neuronal cell injury. In this study, the initial and post-treatment levels of NSE and S-100β were compared in carbon monoxide (CO) poisoning patients, who received normobaric oxygen (NBO) or hyperbaric oxygen (HBO) therapy. Forty consecutive patients with acute CO poisoning were enrolled in this prospective, observational study. According to their clinical symptoms and observations, twenty patients were treated with NBO, and the other twenty with HBO. Serum S-100β and NSE levels were measured both at time of admission and 6 h later (post-treatment). Serum NSE and S-100β values decreased significantly in both of the therapeutic modalities. The initial and post-treatment values of NSE and S-100β in NBO or HBO patients were comparable. A clear negative correlation was observed between the decrease of NSE and S-100β levels and initial blood carboxyhemoglobin levels. In conclusion, the present results suggested the use of serum S-100β and NSE levels as indicators for brain injury. Due to the significant increase of their values with oxygen therapy, they may also be useful as prognostic follow-up markers. However, the current findings reflected no difference between the efficacy of NBO or HBO therapy.

Keywords: Brain injury, Carbon monoxide poisoning, Hyperbaric oxygen, Neuron-specific enolase, Oxygen therapy, S-100β

Carbon monoxide (CO) is an agent that can cause intoxication, mortality and morbidity worldwide1. CO poisoning is responsible for 40,000 emergency department visits each year in the USA; 600 are attributable to unintentional causes and five to ten-times to suicidal thoughts2-4. CO intoxication accounts for approx. 31% of deaths, resulting from poisoning4. Similarly, a study conducted in Turkey has revealed that 15.7% of all forensic autopsies have been performed due to asphyxial death, of which 8.2% are caused by CO poisoning5. The central nervous system (CNS) is the most vulnerable region to CO inhalation6. Nonetheless, CO poisoning leads also to early and permanent injuries in brain, heart, muscle and kidney that also require high and constant oxygen supplement function properly. Some of the patients surviving after serious CO poisoning can stay asymptomatic, while 67% of them experience neurologic sequelae7-9.

The management of patients with CO poisoning includes supportive and symptomatic treatments and administration of supplemental oxygen. Although hyperbaric oxygen (HBO) treatment may be useful to prevent long-term neurologic sequelae in selected patients, however, there is no biochemical marker available which can be used to decide the usage of HBO, instead of normobaric oxygen (NBO) treatment in CO poisoning10. In fact, there is requirement for prognostic biochemical markers in the evaluation and management of patients with CO intoxication.

S-100β is a calcium-binding protein which is produced and released by astroglial cells in the CNS11. It exerts neuro- and gliotrophic effects and may have an important role in the development and
recovery of CNS after injury. It has been shown that S-100β may be a useful biochemical marker of brain damage in cardiac arrest, stroke, traumatic head injury and subarachnoid hemorrhage. Neuron-specific enolase (NSE) is a glycolytic enzyme localized in neuronal cytoplasm, which is not secreted physiologically; its elevated level in serum and cerebrospinal fluid can be associated with structural damage to neuronal cells, indicating a traumatic brain injury, cardiac arrest or Parkinson’s disease.

In the medical literature, only few clinical studies are available on the possible association of serum NSE and S-100β levels with CO poisoning-dependent brain injuries. In the present study, we have aimed to investigate the relationship between NSE and S-100β with regard to carboxyhemoglobin (HbCO) levels and the clinical outcome of CO intoxication patients. In addition, the follow-up values for NSE and/or S-100β have been compared in the two different therapeutic modalities, i.e. HBO and NBO.

**Materials and Methods**

**Patients**

A total of 40 CO poisoning cases admitted to the emergency service of the Gulhane Military Medical Academy Hospital, Ankara, Turkey over a period of 4 months were included in the study. In relation to the cause of intoxication, duration of exposure, symptoms and findings at admission, the estimated Glasgow coma scale (GCS) and blood HbCO levels. Twenty of these patients (12 male, 8 female; mean age 33.95 ± 16.83) were treated with NBO and the other 20 (11 male, 9 female; mean age 37.05 ± 12.78) were administered HBO. The selection for NBO or HBO treatment was assessed according to universally accepted guidelines as shown in Table 1. All patients enrolled in the study gave their consent for the usage of their data. The study was performed in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki and with the approval of the Institutional Ethical Committee.

**Therapeutic interventions**

All cases were treated with pure oxygen via a non-rebreather face-mask. NBO was administered at a flow rate of 15 l/min throughout the therapy period with 5-min air break intervals after every 25 min of oxygen breathing. The HBO pressure/duration range was set as 2.4 atm/90 min during which the patients breathe pure oxygen with the same scheme of NBO patients; after HBO exposure, the patients continued receiving NBO until end of the treatment procedure (6 h).

**Biochemical measurements**

Blood samples were collected from the patients twice; at the time of arrival in the emergency service and at 6 h later (after therapeutic approach). Serum NSE and S-100β values were estimated along with the routine hematologic parameters, such as blood cell count, hematocrit and hemoglobin. Enzyme-linked immunoassay (ELISA) was used for S-100β and NSE measurements.

**Statistical analysis**

Data analysis was performed with the Statistical Package for the Social Sciences (SPSS version 15.0) software. The mean plus standard deviation and median values were used for data defining. Normality analysis for data distribution was done by the Kolmogorov-Smirnov test; since the test reflected non-normal distribution, non-parametric tests were used. The Mann-Whitney U test was used for group-to-group comparisons of the same parameter, while the Wilcoxon test was used for comparing continuous variables within the same group and the Chi-square test was used to compare discrete variables. In addition, the possible relation between variables was evaluated by using the Pearson's correlation test. P levels less than 0.05 were considered statistically significant.

**Results**

The baseline symptoms and their values in selection of patients to HBO or NBO therapies are presented in Table 1. In brief, demographic characteristics, such as age and gender were similar between the two groups. As expected, headache and syncope were significantly more in the HBO treatment group (P = 0.006 for both). Although statistically insignificant, dizziness was also more often observed in the HBO patients. The patients who received HBO treatment showed a relative longer exposure time to CO and their Glasgow coma scale was also slightly lower than the NBO patients (not significant).

Table 2 represents the HbCO, NSE and S-100β levels at time of arrival and 6 h post-treatment. The patients of HBO group had significantly higher mean blood HbCO levels (P = 0.017, HBO vs NBO group); the HbCO levels of both HBO and NBO groups were diminished after the therapy (P < 0.001 and P = 0.008...
respectively, compared with levels at arrival). The serum NSE and S-100β levels were found to be slightly (but not significant) higher in the HBO treated patients. At 6 h after treatment, both NSE and S-100β values were significantly lowered in HBO and NBO groups (HBO group, \( P = 0.002 \) for NSE and \( P = 0.018 \) for S-100β; NBO group, \( P = 0.004 \) for NSE and \( P = 0.001 \) for S-100β). There were no differences for the post-treatment values of these markers between HBO and NBO patients. A negative moderate (\( r = -0.324 \)) and statistically significant (\( P = 0.008 \)) correlation was found between blood HbCO values at arrival and serum NSE level (Fig. 1). Similarly, a negative moderate (\( r = -0.398 \)) and significant (\( P = 0.032 \)) correlation was observed among HbCO and S-100β levels (Fig. 2).

### Table 1—Basic demographic data of cases and selection criteria for the therapeutic modality

<table>
<thead>
<tr>
<th></th>
<th>HBO</th>
<th>NBO</th>
<th>( P )</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (Mean ± SD)</td>
<td>37.05 ± 12.78</td>
<td>33.95 ± 16.83</td>
<td>0.285*</td>
</tr>
<tr>
<td>Gender (Male/Female)</td>
<td>11/9</td>
<td>12/8</td>
<td>0.749*</td>
</tr>
<tr>
<td>Headache (n) (%)</td>
<td>17 (89)</td>
<td>8 (47.1)</td>
<td>0.006**</td>
</tr>
<tr>
<td>Syncope (n) (%)</td>
<td>9 (52.9)</td>
<td>2 (10.5)</td>
<td>0.006**</td>
</tr>
<tr>
<td>Dizziness (n) (%)</td>
<td>11 (57.9)</td>
<td>6 (33.3)</td>
<td>0.175**</td>
</tr>
<tr>
<td>Exposure time (Mean ± SD) (Median)</td>
<td>4.34 ± 5.18 (1)</td>
<td>4.22 ± 2.5 (4)</td>
<td>0.036*</td>
</tr>
<tr>
<td>Glasgow coma scale (Mean ± SD) (Median)</td>
<td>13.93 ± 2.12 (15)</td>
<td>14.95 ± 0.23 (15)</td>
<td>0.067*</td>
</tr>
</tbody>
</table>

*Mann-Whitney U test; **Chi-square test

### Table 2—Carboxyhemoglobin (HbCO), neuron-specific enolase and S-100β levels at time of arrival and 6 h post-treatment

<table>
<thead>
<tr>
<th></th>
<th>HBO</th>
<th>NBO</th>
<th>( P )</th>
</tr>
</thead>
<tbody>
<tr>
<td>HbCO (Mean ± SD) (Median)</td>
<td>29.6 ± 10.2 (30.9)</td>
<td>21.7 ± 8.6 (20.95)</td>
<td>0.017</td>
</tr>
<tr>
<td>( p^{**} )</td>
<td>&lt; 0.001</td>
<td>0.008</td>
<td></td>
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<tr>
<td>NSE (Mean ± SD) (Median)</td>
<td>6.82 ± 5.84 (5.14)</td>
<td>5.51 ± 4.64 (4.27)</td>
<td>0.31</td>
</tr>
<tr>
<td>( p^{**} )</td>
<td>0.002</td>
<td>0.425</td>
<td></td>
</tr>
<tr>
<td>S100β (Mean ± SD) (Median)</td>
<td>4.57 ± 3.07 (2)</td>
<td>3.61 ± 1.63 (3)</td>
<td>0.461</td>
</tr>
<tr>
<td>( p^{**} )</td>
<td>0.018</td>
<td>0.545</td>
<td></td>
</tr>
</tbody>
</table>

*Mann-Whitney U test; **Wilcoxon test

**Fig. 1**—Relation between blood HbCO values and difference between serum NSE levels at admission and 6 h post-treatment (Chi-square test; \( r = -0.324 \), \( r^2 = 0.105 \), \( P = 0.008 \))

**Fig. 2**—Relation between blood HbCO values and difference between serum S-100β levels at admission and 6 h post-treatment (Chi-square test; \( r = -0.398 \), \( r^2 = 0.159 \), \( P = 0.032 \))
Discussion

Oxygen therapy is the cornerstone for the management of CO intoxication; in selected cases, the use of HBO is also considered. One of the main targets affected by CO poisoning is the central nervous system (CNS). HBO therapy is recommended to the patients presenting with syncope, coma, seizures, focal neurologic deficits, and patients having a HbCO level higher than 25% (or 15% for pregnant women). The majority of the HBO therapy suggested symptoms, such as headache, dizziness or syncope are clinical presentations caused by the brain. Thus, biochemical markers demonstrating the brain damage can provide additional evidence in determining neurological response and can guide for HBO therapy in patient selection.

In literature, only a limited number of studies are available comparing NSE and S-100β and their levels in CO poisoning. In an experimental CO poisoning model, Brvar et al. have reported that serum S-100β levels are significantly increased after exposure and that this difference is particularly more pronounced in patients with loss of consciousness. Another study of the same group has suggested that S-100β may be a compatible marker for the brain injury in CO poisoning. By comparing S-100β and NSE levels in 70 CO poisoning cases and 20 control individuals, Yardan et al. have reported significantly increased S-100β levels in CO intoxication cases; the significant rise of S-100β levels is found to be significantly higher in patients with loss of consciousness, similar to the report of Brvar et al. On the other hand, NSE levels are increased and a positive correlation has been found between S-100β and NSE levels. In another study, where NSE and S-100β levels have been compared at 0, 3 and 6 h in 30 CO poisoning cases, a marked decrease of their levels is reported with oxygen treatment; the decrease rate is more pronounced in S-100β levels.

The significantly decreased levels of serum S-100β and NSE compared to their initial values in the present study was in line with the previous findings of other groups. Both HBO and NBO therapies resulted in lowering HbCO rates, as well as S-100β and NSE levels; however, the present findings could not provide an advantage of one therapy modality to the other. Since the values of NSE and S-100β at admission were comparable, using these biomarkers to make a decision between HBO and NBO therapies could not be confirmed.

In conclusion, the present results provided evidence for the usefulness of serum S-100β and NSE levels as indicators for brain injury in CO poisoning. These markers could also be used as follow-up parameters during the therapeutic process, i.e. lower their values, the better the clinical outcome. But, they seemed unable to provide information regarding the decision for using NBO or HBO therapy in CO intoxication cases. However, due to the relative low number of patients in the present work, further large-scale studies are needed to confirm the possible benefits of S-100β and NSE as prognostic markers for CO poisoning.

References