Correlation of serum S-100 protein level with involvement of territory and size of lesion in acute ischemic stroke

Harish Kumar, Manoj Lakhotia, Hansraj Pahadiya, Jagdish Singh, Jainapur Ravi Sangappa, Akanksha Choudhary

Department of Medicine, Dr. S N Medical College and Associated Group of Mahatma Gandhi Hospital, Jodhpur, Rajasthan, India

ABSTRACT

Background: Cerebrovascular accident is the most common and devastating disorders in old age group. The diagnosis of stroke remains a clinical one, with confirmatory evidence obtained through neuroimaging. Neurobiochemical markers have gained special importance in the determination of brain damage resulting from acute stroke. Aim: In this study, we aimed to evaluate serum S-100 protein in blood samples from patients with acute ischemic stroke and investigate the relationship of serum S-100 protein level with the involved territory and size of the lesion. Methods: This was a prospective observational study conducted among 94 patients of acute ischemic stroke admitted to the Medicine Department within 48 h. Serum sample was collected within 48 h and was sent for measurement of serum S-100 protein level. Patients were classified according to involved territory as anterior cerebral artery (ACA), middle cerebral artery (MCA), posterior cerebral artery (PCA), and more than one territory and correlate it with S-100 protein level. All patients had cranial computerized tomography scan and magnetic resonance imaging in the first 48 h. Neurological examination was done with National Institute of Health Stroke Scale in acute stage and Rankin scale at the time of discharge. Results: Serum S-100 protein levels were significantly higher and maximum in multiple territory involvements followed by MCA, PCA, and ACA infarct. Conclusions: As serum S-100 protein level correlates with the involved territory or infarct size, we can predict the involved territory with the level of S-100 protein.

Key words: Brain damage, infarct size, National Institute of Health Stroke Scale, Rankin scale, S-100 protein, serum marker, stroke, territory

INTRODUCTION

Early detection of brain damage is the demand of recent years for rapid diagnosis and prognosis in stroke patients. Therefore, neurobiochemical serum markers of brain damage increase attention to neurologist. Easy availability, low cost, and easy to investigate make it valuable. While computerized tomography

This is an open access article distributed under the terms of the Creative Commons Attribution-NonCommercial-ShareAlike 3.0 License, which allows others to remix, tweak, and build upon the work non-commercially, as long as the author is credited and the new creations are licensed under the identical terms.

Address for correspondence: Dr. Harish Kumar, B-3, Shastri Nagar, Bikaner, Rajasthan, India. E-mail: drharishsgnr@gmail.com
(CT) and magnetic resonance imaging (MRI) may take some time to perform, result is normal in most cases in CT, even in confirmed ischemic stroke.\cite{1,2} Hence, the availability of brain damage markers support the diagnosis and severity. S-100 protein is a dimeric protein which is secreted from astroglial cells of the brain.\cite{3,4} These proteins are called S-100 because of their solubility in a 100% saturated solution with ammonium sulfate at neutral pH. They were first identified by Moore in 1965.\cite{5}

Elevation of S-100 protein level in cerebrospinal fluid (CSF) has been reported in various studies previously, but the difficulty of collecting CSF sample by lumber puncture attracts the researcher to study such biochemical marker in blood.\cite{6-8} In our study, we correlate the serum S-100 protein level in blood with infarct size and involved territory.

**METHODS**

This was a cross-sectional study where data are collected prospectively, conducted among 94 patients of acute ischemic stroke admitted to Mahatma Gandhi Hospital, Jodhpur, in June 2013-January 2014. Age-and sex-matched healthy subjects not suffering from ischemic heart disease, cerebrovascular disease, hypertension, or any other disease which could alter the value of S-100 protein level selected as a control.

The inclusion criteria were as follows: Patients above 18 years old of acute ischemic stroke admitted in our hospital within 48 h of onset, diagnosis was confirmed by CT/MRI scan of brain, and patients above the age of 18 years. Exclusion criteria were as follows: Patients other than ischemic stroke such as haemorrhage, subarachnoid haemorrhage, and cortical vein thrombosis; patients with peripheral vascular disease, systemic inflammatory disorders, or tumors; patients who had acute cardiac disease, melanoma, neurofibroma, glioblastoma and malignancy; and patients whose National Institute of Health Stroke Scale (NIHSS) score could not be calculated at the time of admission. This study was approved by the Local Research Ethics Committee.

Detailed history and examination was done by the attending physician and diagnosis was confirmed by CT/MRI scan of the brain depending on accessibility. Neurological evaluation was made by NIHSS in the acute stage and by Rankin scale at the time of discharge.\cite{9,10}

Venous samples were drawn within 48 h of the onset of symptoms and sent for routine blood examinations including measurement of S-100 protein level. The concentration of S-100 protein in blood samples was measured in electrochemiluminescence immunoassay is intended for use on Elecsys and cobas e-immunoassay analyzers. Patients were classified according to infarct size and involved territory as anterior cerebral artery (ACA) (n = 4), middle cerebral artery (MCA) (n = 67), posterior cerebral artery (PCA) (n = 17), and more than one territory (n = 6). MCA territory further divided with cortical (stump territory) and subcortical infarct (lenticulostriate branch).

Perametric data are expressed as mean value ± standard deviation and categorical variables as percentages. The Chi-square test was used for the comparison of dichotomous variables and the Student’s t-test for continuous variables. One-way ANOVA was used to test differences on multiple levels by a single factor (independent) variable. A P < 0.05 was considered statistically significant. All statistical analyses were performed using SPSS Windows, Version 17.

**RESULTS**

The mean value of serum concentration of S-100 proteins in stroke patients (1.1231 ± 1.5864) was higher than the mean value found in the control group (0.159 ± 0.168), which was statistically significant (P < 0.001).

Patients are divided into four groups according to involved territory and two subgroups in MCA territory. Groups are according to localization and size of the lesion with involved territory seen in CT and MRI. Group were as follows: ACA territory (small size), PCA territory (medium size), MCA territory (large size), and more than one territory (largest size). In MCA territory, two subgroups have been made with cortical (stump, n = 42) and subcortical (lenticulostriate branch, n = 25).

S-100 protein level was significantly higher and maximum in multiple territory involvement (more than one territory) (mean S-100 protein level 4.04 ± 3.473, P < 0.0001). Control mean S-100 protein was 0.1782 ± 0.1622 (0.0509-0.901) μg/L. Size of lesion was maximum in more than one territory involvement. In large size infarction, MCA territory S-100 protein level was 0.9917 ± 1.254 μg/L (P < 0.0001) followed by protein levels of 0.754 ± 0.854 μg/L (P < 0.0001) and 0.51 ± 0.6678 μg/L (P < 0.0001) for PCA and ACA territory, respectively [Table 1 and Graph 1].

Relationship of S-100 protein level in MCA territory is given in Table 2 and Graph 2.
Kumar, et al.: S-100 protein level predict the involved territory and size of lesion

**DISCUSSION**

S-100 protein is an astroglial protein in cytosol secreted by brain damage.\(^3,4\) As mentioned earlier, it can be investigated both in serum and CSF. However, easy and safe to measure from blood make it more useful than CSF. Many biochemical neuronal markers are associated with stroke such as neuron-specific enolase, glial fibrillary acidic protein (GFAP), and protein S-100B, which are elevated in many conditions of brain damage such as head trauma, cerebral hypoxia, cerebral bleeding, and ischemic stroke.\(^6,8\) In our study, we investigated the correlation of S-100 protein level with the involved territory and size of infarct in 94 patients of acute stroke patients. Persson et al.\(^{11}\) were the first to observe elevation of S-100 protein in two patients. Herrmann et al.\(^{12}\) studied the comparison of serum S-100 protein and GFAP with the volume of lesion and neurological status at the time of discharge in acute stroke patients. Foerch et al.\(^{13}\) studied the correlation of serum S-100 protein with acute MCA occlusion predict malignant course of infarction. Kenangil et al.\(^{14}\) also studied the relation of S-100 protein to infarct size in MCA territory occlusion. No one studied the correlation of S-100 protein with multiple territories. Here is the first study which reports the association of single biochemical marker in blood in the acute stage of stroke with the involved territory. By this study, we can also predict the involved territory and size of the lesion. Büttner et al.\(^{15}\) studied serum S-100 protein levels after MCA infarctions in relation to clinical data and prognosis. The studies which have been done earlier associated with MCA territory only; the present study is the first which correlates the S-100 protein with territorial involvement other than MCA only. Kumar et al.\(^{16}\) studied serum S-100 protein level was more significantly higher in the ischemic group than hemorrhage and transient ischemic attack group and highest in expired patients. They studied the role of S-100 protein as a co-predictor of outcome in patients with acute stroke.

In our study, we evaluated the infarction in multiple territories and divided them into four groups according to their size and involved territory. In our study, S-100 protein level was significantly higher and maximum in multiple territory involvements (4.04 ± 3.4) followed by MCA (0.9917 ± 1.2), PCA (0.7540 ± 0.854), and ACA (0.51 ± 0.66) infarct, which indicate that S-100 level increases with infarct size and involvement of territories. There are no studies in the literature, relating S-100 level with territorial involvement. Involvement of multiple territory leads to larger size of the lesion; hence, there is high level of S-100 protein. Size of the lesion was confirmed by brain imaging. Thus, S-100 protein directly correlated to involved territory, infarct size, and clinical prognosis, calculated by NIHSS score on admission and modified Rankin scale score on discharge.\(^3,10\) We divided MCA territory in our study to stump MCA (cortical) and lenticulostriate branches (subcortical). With stump MCA involvement, the S-100 levels were high indicating

### Table 1: Relation of S-100 with involvement of territory of vessels

| Involved territory | Mean S-100 ± SD | P versus control | Mean NIHSS ± SD |
|--------------------|-----------------|------------------|-----------------|
| ACA (n=4)          | 0.51±0.6678     | <0.0001          | 74.24           |
| MCA (n=67)         | 0.9917±1.254    | <0.0001          | 14.68±5.86      |
| PCA (n=17)         | 0.754±0.854     | <0.0001          | 13.54±7.13      |
| >1 territory (n=6) | 4.04±3.473      | <0.0001          | 26.16±3.7       |

Control mean S-100 protein 0.1782 ± 0.1622 (0.0509–0.901) μg/L. SD = Standard deviation, ACA = Anterior cerebral artery, MCA = Middle cerebral artery, PCA = Posterior cerebral artery, NIHSS = National Institute of Health Stroke Scale

### Table 2: Relationship of S-100 levels in cortical and subcortical infarct of involved middle cerebral artery territory

| Involved territory | Cortical (stump MCA) (n=42) | Subcortical (lenticulostriate MCA) (n=25) | P     |
|--------------------|------------------------------|------------------------------------------|-------|
| Male/female (n)    | 25/17                        | 16/9                                     |       |
| S-100 mean±SD      | 1.3819±1.395                 | 0.3362±0.537                            | <0.0006|

Control mean S-100 protein 0.1782 ± 0.1622 (0.0509–0.901) μg/L. SD = Standard deviation, MCA = Middle cerebral artery
involved area of the brain, whereas in the lenticulostrate branches of MCA, PCA, and ACA, the S-100 levels were low indicating smaller size of the infarct. Similar levels were found in the study of Kenangil et al.,[14] who found that size of infarct in MCA territory correlated with levels of S-100 protein. Patients with large MCA infarctions had the highest serum S-100 levels and their short- and long-term prognosis was the worst. Similar report was presented by Büttner et al.[15]

CONCLUSION

Finally, we conclude that serum S-100 B measurement can be used as an early marker of brain damage. It is correlated with the size of the lesion not only with MCA infarctions but also with PCA and ACA.

S-100 protein level correlated with the severity by NIHSS which was maximum in multiple territory involvements.

Limitation

The limitation of this study was the volume of infarct size which was not included in this study.

This study only improves the diagnosis of stroke and severity of patients. Thus, by S-100 protein level, we can only predict the involved branch, with high sensitivity and low specificity.

Financial support and sponsorship

Nil.

Conflicts of interest

There are no conflicts of interest.

REFERENCES

1. Hand PJ, Wardlaw JM, Rowat AM, Haisma JA, Lindley RL, Dennis MS. Magnetic resonance brain imaging in patients with acute stroke: Feasibility and patient related difficulties. J Neurol Neurosurg Psychiatry 2005;76:1525-7.
2. Kane I, Whiteley WN, Sandercock PA, Wardlaw JM. Availability of CT and MR for assessing patients with acute stroke. Cerebrovasc Dis 2008;25:375-7.
3. Martens P, Ranbe A, Johnsson P. Serum S-100 and neuron-specific enolase for prediction of regaining consciousness after global cerebral ischemia. Stroke 1998;29:2363-6.
4. Heizmann CW. Ca2+-binding S100 proteins in the central nervous system. Neurochem Res 1999;24:1097-100.
5. Moore BW. A soluble protein characteristic of the nervous system. Biochem Biophys Res Commun 1965;19:739-44.
6. Härdemark HG, Ericsson N, Kotwica Z, Rundström G, Mendel-Hartvig I, Olsson Y, et al. S-100 protein and neuron-specific enolase in CSF after experimental traumatic or focal ischemic brain damage. J Neurosurg 1989;71(5 Pt 1):727-31.
7. Mokuno K, Kato K, Kawai K, Matsuoka Y, Yanagi T, Sobue I. Neuron-specific enolase and S-100 protein levels in cerebrospinal fluid of patients with various neurological diseases. J Neurol Sci 1983;60:443-51.
8. Lamers KJ, van Engelen BG, Gabreëls FJ, Hommes OR, Borm GF, Wevers RA. Cerebrospinal neuron-specific enolase, S-100 and myelin basic protein in neurological disorders. Acta Neurol Scand 1995;92:247-51.
9. Brott T, Adams HP Jr., Olinger CP, Marler JR, Barsan WG, Biller J, et al. Measurements of acute cerebral infarction: A clinical examination scale. Stroke 1989;20:864-70.
10. Meyer BC, Hemmen TM, Jackson CM, Lyden PD. Modified national institutes of health stroke scale for use in stroke clinical trials: Prospective reliability and validity. Stroke 2002;33:1261-6.
11. Persson L, Härdemark HG, Gustafsson J, Rundström G, Mendel-Hartvig I, Esscher T, et al. S-100 protein and neuron-specific enolase in cerebrospinal fluid and serum: Markers of cell damage in human central nervous system. Stroke 1987;18:911-8.
12. Herrmann M, Vos P, Wunderlich MT, Brujin CH, Lamers KJ. Release of glial tissue-specific proteins after acute stroke. A comparative analysis of serum concentrations of protein S-100 B and glial fibrillary acidic protein. Stroke 2000;31:2670-7.
13. Foerch C, Otto B, Singer OC, Neumann-Haefelin T, Yan B, Berkefeld J, et al. Serum S100B predicts a malignant course of infarction in patients with acute middle cerebral artery occlusion. Stroke 2004;35:2160-4.
14. Kenangil G, Yağcı AD, Haklar G. Relation of serum S-100 protein to infarct size and clinical prognosis. Marmara Med J 2004;17:105-8.
15. Büttner T, Weyers S, Postert T, Sprengelmeyer R, Kuhn W. S-100 protein: Serum marker of focal brain damage after ischemic territorial MCA infarction. Stroke 1997;28:1961-5.
16. Kumar H, Lakhotia M, Pahadiya H, Singh J. To study the correlation of serum S-100 protein level with the severity of stroke and its prognostic implication. J Neurosci Rural Pract 2015;6:326-30.