Serum homocysteine level in patients with acute cerebral infarction and its correlation with inflammatory factors, nerve factors and NO metabolism

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ABSTRACT

Objective: To study the serum homocysteine (Hcy) level in patients with acute cerebral infarction and its correlation with inflammatory factors, nerve factors and NO metabolism.

Methods: A total of 118 patients with acute cerebral infarction were selected as observation group and 50 healthy volunteers who received physical examination in our hospital during the same period were selected as the normal control group. Serum levels of Hcy in two groups were detected, and the median of Hcy levels in observation group was referred to further divide them into high Hcy group and low Hcy group, 59 cases in each group. Serum contents of inflammatory factors, nerve factors and NO metabolism markers were compared between acute cerebral infarction patients with different levels of Hcy. Results: Serum Hcy level in observation group was higher than that in control group. Serum contents of inflammatory factors such as IL-1β, IL-6, IL-17 and hs-CRP in high Hcy group were higher than those in low Hcy group, contents of nerve factors such as Copeptin, NT-proBNP, NSE and S-100B in high Hcy group were higher than those in low Hcy group, and contents of NO metabolism indexes such as NO and NOS in high Hcy group were higher than those in low Hcy group. Conclusion: Serum Hcy level increases in patients with acute cerebral infarction, and the level of Hcy is directly related to inflammatory factors, nerve factors and NO metabolism.

1. Introduction

Acute cerebral infarction is the most common clinical cardio-cerebrovascular disorder, it is the brain tissue necrosis caused by sudden cerebrovascular interruption, the worldwide incidence at present is about 200/100 000, and its incidence and mortality rates are rising along with the growth of age[1,2]. Early judgment of acute cerebral infarction severity is the key to to set up reasonable therapy and improve the prognosis of patients, head CT/MRI is the gold standard in the diagnosis of cerebral infarction, but it is unable to monitor patients’ condition change in real time, and looking for the serum indexes with high sensitivity and specificity to guide clinical treatment is the current research hotspot. Homocysteine (Hcy) is the intermediate of methionine metabolism, it is related to the active expression of cystathionine β-synthase, hyperhomocysteinemia is considered to be the independent risk factor for acute cerebral infarction[3], but less research has been carried out at present about the intrinsic relationship between specific Hcy levels and acute cerebral infarction severity. In the study, the serum Hcy levels in patients with acute cerebral infarction and normal subjects were compared, and the differences in the levels of inflammatory factors, nerve factors, nitric oxide (NO) metabolism and other disease indexes in patients with different Hcy were further explored, now reported as follows.

2. Information and methods

2.1 Case information
A total of 118 patients with acute cerebral infarction who were treated in our hospital between January 2013 and July 2016 were selected as observation group, 50 healthy volunteers who received physical examination in our hospital during the same period were selected as the normal control group, and the included research subjects them or their families signed informed consent. Normal control group included 26 male cases and 24 female cases that were 41-78 years old; observation group included 62 male cases and 56 female cases that were 40-74 years old. The two groups of research subjects were not significantly different in gender and age distribution (P>0.05), and the hospital ethics committee approved the study.

2.2 Diagnostic criteria for acute cerebral infarction

(1) With acute disease attack, and illness reaching its peak in a short period of time (within hours or 1-2 d); (2) associated with clinical manifestations such as headache, dizziness, incoherent speech, nausea and vomiting as well as hemiplegia, and severe cases with coma; (3) the head CT showing cerebral ischemia lesions and peripheral edema.

2.3 Serum sample collection and serum preparation

Immediately after admission, 2.0 mL of cubital venous blood was extracted from two groups of research subjects, joined by anticoagulant, then let stand at room temperature for stratification and centrifuged at 3 000 r/min for 10 min, and the supernatant was collected and cryopreserved in -70 °C low-temperature environment for testing.

2.4 Serum Hcy level detection

The Hcy levels in serum of two groups were determined by the fully automatic biochemical analyzer (Roche instrument center, model C111). The median of serum Hcy level of observation group was used as boundary to divide them into high Hcy group and low Hcy group, 59 cases in each group.

2.5 Illness-related indexes

Enzyme-linked immunosorbert assay (ELISA) was used to detect serum contents of inflammatory factors of observation group, including interleukin-1 β (IL-1 β), interleukin-6 (IL-6), interleukin-17 (IL-17) and hypersensitive C-reactive protein (hs-CRP). ELISA was used to determine serum levels of nerve factors, including Copeptin, N-terminal pro-B-type natriuretic peptide (NT-proBNP), neuron-specific enolase (NSE) and S-100B protein (S-100B). Nitrate reductase method was used to detect serum nitric oxide (NO) content and chemical colorimetry was used to detect nitric oxide synthase (NOS) content.

2.6 Statistical methods

Statistical software was SPSS 21.0, and the statisticians were with professional statistical knowledge and researcher qualification. Hcy levels, inflammatory factors, nerve factors, NO metabolism indexes and other measurement data were in terms of mean ± standard deviation, and comparison between groups was by grouping t test. \( P<0.05 \) was the standard of statistical significance in differences in the study.

3. Results

3.1 Serum Hcy levels

Mean serum Hcy level in observation group was (30.17±4.85) μmol/L and mean serum Hcy level in control group was (10.16±1.85) μmol/L. Serum Hcy level in observation group was significantly higher than that in control group, and differences were statistically significant (\( P<0.05 \)). The median of serum Hcy level in observation group was 25.37 μmol/L and used as boundary to divide them into high Hcy group and low Hcy group, 59 cases in each group.

3.2 Serum inflammatory factors

Comparison of serum inflammatory factors IL-1 β (μg/L), IL-6 (ng/L), IL-17 (μg/L) and hs-CRP (mg/L) contents between acute cerebral infarction patients with different Hcy levels was as follows: serum IL-1 β, IL-6, IL-17 and hs-CRP contents in high Hcy group were higher than those in low Hcy group. Differences in serum

Table 1.

Comparison of serum inflammatory factor contents between acute cerebral infarction patients with different Hcy levels.

<table>
<thead>
<tr>
<th>Groups</th>
<th>n</th>
<th>IL-1 β (μg/L)</th>
<th>IL-6 (ng/L)</th>
<th>IL-17 (μg/L)</th>
<th>hs-CRP (mg/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Low Hcy group</td>
<td>59</td>
<td>9.65±0.98</td>
<td>22.67±3.05</td>
<td>12.67±1.68</td>
<td>5.16±0.63</td>
</tr>
<tr>
<td>High Hcy group</td>
<td>59</td>
<td>13.27±1.98</td>
<td>32.18±4.36</td>
<td>18.23±2.75</td>
<td>7.48±0.91</td>
</tr>
<tr>
<td>T value</td>
<td></td>
<td>9.231</td>
<td>13.274</td>
<td>10.382</td>
<td>6.583</td>
</tr>
<tr>
<td>P value</td>
<td></td>
<td>&lt;0.05</td>
<td>&lt;0.05</td>
<td>&lt;0.05</td>
<td>&lt;0.05</td>
</tr>
</tbody>
</table>
inflammatory factors IL-1β, IL-6, IL-17 and hs-CRP contents were statistically significant between acute cerebral infarction patients with different Hcy levels (P<0.05), shown in Table 1.

3.3 Nerve factors

Comparison of serum nerve factors Copeptin (pmol/mL), NT-proBNP (ng/mL), NSE (ng/mL) and S-100B (ng/mL) contents between acute cerebral infarction patients with different Hcy levels was as follows: serum Copeptin, NT-proBNP, NSE and S-100B contents in high Hcy group were higher than those in low Hcy group. Differences in serum nerve factors Copeptin, NT-proBNP, NSE and S-100B contents were statistically significant between acute cerebral infarction patients with different Hcy levels (P<0.05), shown in Table 2.

3.4 NO metabolism indexes

Comparison of serum NO metabolism indexes NO (mg/L) and NOS (U/mL) contents between acute cerebral infarction patients with different Hcy levels was as follows: serum NO and NOS contents in high Hcy group were higher than those in low Hcy group. Differences in serum NO metabolism indexes NO and NOS contents were statistically significant between acute cerebral infarction patients with different Hcy levels (P<0.05), shown in Table 3.

4. Discussion

Many studies show that hypertension is synergistic with hyperhomocysteinemia, and they promote the occurrence and development of acute cerebral infarction together. At the same time, Hcy can activate platelet function and accelerate thrombosis, further causing brain cell ischemia hypoxia and intracellular mitochondrial dysfunction[4,5]. In the study, serum Hcy levels were first compared between patients with acute cerebral infarction and normal subjects, and it was found that serum Hcy level in observation group was significantly higher than that in control group, it confirms that unusually high Hcy levels participate in the occurrence and progression of acute cerebral infarction, and this is in accordance with the previous research conclusion. At present, there is not much research on the inner link between Hcy levels and acute cerebral infarction, and the differences in illness-related indexes in patients with acute cerebral infarction under different Hcy conditions were further explored in this study. Inflammation is a recognized cause of acute cerebral infarction, and studies have shown that local inflammatory responses triggered by cerebral infarction can lead to adverse outcomes[6,7]. The activated neutrophils in ischemic lesions after acute cerebral infarction are combined with vascular endothelium and lead to brain damage, and the damaged neurons and axons release IL-1β, IL-6 and other chemotaxis factors, and prompt more neutrophils to migrate to ischemic tissue[8,9]. IL-17 is the newly named Th17 cell-derived inflammatory factor, which can be combined with the receptor to produce strong anti-inflammatory effects. Hs-CRP is a sensitive marker to reflect the presence and severity of inflammation, its high expression can be detected in serum early after cerebral infarction, and hs CRP levels rise with the aggravation of brain damage degree[10]. In the study, the inner link between serum Hcy levels and inflammatory factor contents in patients with acute cerebral infarction was explored, and it was found that serum IL-1β, IL-6, IL-17 and hs-CRP contents in high Hcy group were higher than those in low Hcy group, indicating that the serum Hcy levels in patients with acute cerebral infarction are positively correlated with the degree of inflammation.

Nerve injury is the primary manifestation of acute cerebral infarction, and it is manifested as abnormal contents of a number of neuron injury-related indicators in serum[11]. Copeptin is a part of the C terminal of pro-arginine vasopressin, which was released into the blood early after brain injury, and is consistent with the severity of cerebral infarction and the size of the lesion[12]. NT-proBNP is the N-terminal fragment from pro-brain natriuretic peptide decomposition, it was mostly used for judging the severity of myocardial infarction in the past, the latest research suggests that it is also closely related to the occurrence and development of acute cerebral infarction, and increased levels of NT-proBNP can be

### Table 2.
Comparison of serum nerve factor contents between acute cerebral infarction patients with different Hcy levels.

<table>
<thead>
<tr>
<th>Groups</th>
<th>n</th>
<th>Copeptin</th>
<th>NT-proBNP</th>
<th>NSE</th>
<th>S-100B</th>
</tr>
</thead>
<tbody>
<tr>
<td>Low Hcy group</td>
<td>59</td>
<td>4.11±0.53</td>
<td>139.48±16.77</td>
<td>3.85±0.42</td>
<td>2.85±0.32</td>
</tr>
<tr>
<td>High Hcy group</td>
<td>59</td>
<td>5.83±0.67</td>
<td>183.94±22.75</td>
<td>4.93±0.56</td>
<td>3.54±0.46</td>
</tr>
<tr>
<td>T value</td>
<td></td>
<td>7.092</td>
<td>15.437</td>
<td>6.883</td>
<td>6.623</td>
</tr>
<tr>
<td>P value</td>
<td></td>
<td>&lt;0.05</td>
<td>&lt;0.05</td>
<td>&lt;0.05</td>
<td>&lt;0.05</td>
</tr>
</tbody>
</table>

### Table 3.
Comparison of serum NO metabolism index contents between acute cerebral infarction patients with different Hcy levels.

<table>
<thead>
<tr>
<th>Groups</th>
<th>n</th>
<th>NO</th>
<th>NOS</th>
</tr>
</thead>
<tbody>
<tr>
<td>Low Hcy group</td>
<td>59</td>
<td>98.63±10.19</td>
<td>41.58±5.32</td>
</tr>
<tr>
<td>High Hcy group</td>
<td>59</td>
<td>121.37±15.85</td>
<td>52.37±6.09</td>
</tr>
<tr>
<td>T value</td>
<td></td>
<td>11.283</td>
<td>7.092</td>
</tr>
<tr>
<td>P value</td>
<td></td>
<td>&lt;0.05</td>
<td>&lt;0.05</td>
</tr>
</tbody>
</table>
detected in serum of patients with cerebral stroke. NSE specifically exists in neurons and axons cells, nerve cell function is damaged after local cerebral ischemia hypoxia, and NSE is released from inside to the outside of the cells and enters the peripheral blood through the blood brain barrier. S-100B exists in the glial cells, it can be released into the blood early after acute cerebral infarction and be detected, and therefore, it is considered to be the reliable index to objectively reflect the cerebral infarction disease[13]. In the study, differences in above serum nerve factor contents were compared between acute cerebral infarction patients with different Hcy levels, and it was found that serum Copeptin, NT-proBNP, NSE and S-100B contents in high Hcy group were higher than those in low Hcy group, illustrating that the Hcy levels in patients with acute cerebral infarction are positively correlated with the degree of nerve damage, and the contents of Hcy can objectively reflect the degree of nerve dysfunction.

Current studies show that NO is involved in acute brain infarction progression, and vascular endothelial cell NOS in ischemic part can prompt the NO synthesis, promote the collateral circulation establishment and inhibit platelet aggregation[14-16]. NO and NOS content rise is the body’s self protective behavior to deal with cerebral ischemia hypoxia injury, the more severe the damage, the more the reactive release of NO and NOS, so the serum NO and NOS content can indirectly reflect the illness severity. In the study, differences in serum NO metabolism index contents were compared between acute cerebral infarction patients with different Hcy levels, and it was found that serum NO and NOS contents in high Hcy group were higher than those in low Hcy group, it indicates that serum Hcy levels in patients with acute cerebral infarction are positively correlated with NO metabolism index content, and the contents of Hcy can directly reflect the NO metabolism activity, and indirectly reflect the severity of cerebral infarction.

It shows that there is massively secreted Hcy in serum of patients with acute cerebral infarction, and Hcy levels are positively correlated with local or systemic inflammatory response, nerve injury and NO metabolism activity. Serum Hcy levels in patients with acute cerebral infarction may objectively and accurately reflect the illness severity, and be used as one of the bases of subsequent clinical scheme selection, treatment effect judgment and prognosis evaluation.

References