CHANGES OF SERUM IL-6 AND TNF-Α AND THEIR RELATIONSHIPS WITH DEPRESSION SEVERITY IN PATIENTS WITH POST-STROKE DEPRESSION

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ABSTRACT

Objective: To observe the changes of serum interleukin (IL)-6 and tumor necrosis factor-α (TNF-α) in patients with post-stroke depression (PSD), so as to explore their relationships with the depression severity of patients with PSD.

Methods: A total of 92 patients diagnosed with PSD were selected and divided into mild, moderate and severe depression groups according to Hamilton Depression Scale (HAMD) scores. The expressions of serum IL-6 and TNF-α were detected, the relationships between the levels and normal level of serum IL-6 and TNF-α were compared, and the difference of serum IL-6 and TNF-α in patients with different-degree PSD were analyzed.

Results: The levels of IL-6 and TNF-α were markedly higher in all groups than normal levels, and there were significant differences (P<0.01), which were the lowest in mild depression group and the highest in severe depression group, but there was insignificant difference among different groups (P>0.05).

Conclusion: Serum IL-6 and TNF-α are in close association with the occurrence of PSD, which increase evidently along with the depression severity. However, there is insignificant relationship between the increase range of IL-6 and TNF-α and the depression severity.

Keywords: post-stroke depression, cytokine, interleukin-6, tumor necrosis factor-α, depression severity.

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Introduction

Post-stroke depression (PSD) is the most common emotional disorder after cerebral stroke. PSD belongs to secondary depression which not only impacts patients’ quality of life (QOL), but also postpone the recovery of neurological function and cognitive function of patients with stroke and increase the mortality and disability rates of those patients to a large extent, thus leading to great pain to patients’ body and spirit and increasing the family and social burdens. Most studies have believed that the morbidity of PSD is 30%-50%[1]. PSD not only postpones patients’ recovery of neurological function, cognitive function and activities of daily life (ADL), and increases the length of hospital stay, but also promote the mortality of cerebral stroke[2-5]. The mechanism of PSD is still unclear. At present, most scholars have believed that there are two mechanisms: the first one is the primary endogenous mechanism which believes that cerebral stroke damages brain and the balance between serotonergic neuron and norepinephrine, thus leading to decrease of 5-hydroxytryptamine (5-HT) and norepinephrine levels that further induces depression, and the second one is the theory of reactive mechanism, also named as a reactive depression status, which believed that the PSD is caused by
disorder of psychological balance due to multiple factors from family, society and pathology, etc(6). However, Spalletta et al(7) have proposed a hypothesis of cytokines, in which the occurrence and immunological activation of PSD have been considered to be closely associated with the increased secretion of cytokines. A study has found that interleukin (IL)-6 and tumor necrosis factor-α (TNF-α) are critical cytokines that cause the immunological imbalance. In this study, the expressions of IL-6 and TNF-α in patients with PSD were observed to explore their relationships with depression severity, so as to lay solid foundation for the pathogenesis of PSD and provide basis for the diagnosis and treatment of PSD.

Materials and methods

**General data**

A total of 92 patients diagnosed as PSD hospitalized in Department of Neurology in Guangzhou Hospital of TCM from March 2014 to February 2015 were selected, in whom there were 44 males and 48 females, with average age of (70.45±10.8) years. All patient signed the informed consent forms and conformed to the inclusion criteria. All subjects conformed to the inclusion criteria and signed the informed consent forms. This study had been approved by the Ethic Committee ChiECRCT-2013031.

Diagnostic and inclusion criteria: all enrolled patients conformed to the Key Diagnostic Criteria of Various Cerebrovascular Diseases published in the 4th National Conference for Cerebrovascular Diseases by Chinese Medicine Association in 1995, to the diagnostic criteria of cerebral infarction published in the 4th National Conference for Cerebrovascular Diseases by Chinese Medicine Association in 1995 and to the diagnostic criteria of depression in Chinese Classification Protocols and Diagnostic Criteria of Mental Disorders (Version 3, CCMD-3).

**Inclusion criteria:** patients who conformed to the diagnostic criteria of cerebral infarction by cranial CT or MRI; patients who conformed to the diagnostic criteria of depression, with self-rating depression scale (SDS) ≥41 points, and Hamilton Depression Scale (HAMD) scores (17 programs) >7 points; patients with PSD secondary to acute exacerbation of ischemic cerebral stroke (2 weeks) for 6 months, with depression state for >2 weeks; patients aged 45-85 years; patients with stable vital signs, clear consciousness, high compliance in physical examinations, and certain present and communication ability; patients who signed the informed consent forms by themselves or their families.

**Exclusion criteria:** patients who did not conform to the diagnostic criteria; patients who had acute exacerbation of ischemic stroke within 2 weeks, and sequela for more than 6 months; patients aged >85 years old; patients with severe diabetes complications or severe hepatorenal diseases; patients with unstable vital signs or mental disorders; patients with dementia, disturbance of consciousness or aphasia that had influence on their presentation; patients with anti-depressants within 1 month.

**Methods**

**Collection of blood samples and detection of serum IL-6 and TNF-α:** venous fasting blood 5 mL was collected at an empty stomach in the morning, placed into dry tubes, and sent to the Department of Laboratory in our hospital. Serum was isolated, cooled and sent for detection. Enzyme linked immunosorbent assay was applied, and Guangzhou KingMed Diagnostics Detection Center was in charge of the detection. The relationships of IL-6 and TNF-α with normal levels and with depression severity were observed.

**Depression severity:** HAMD (17 programs) scale was applied, and the depression was classified as follows: non-depression: HAMD score ≤7 points; mild depression: HAMD ≤17 points; moderate depression: HAMD≤24 points; severe depression: HAMD>24 points. In this study, 21 patients had mild depression, 65 had moderate depression and 6 had severe depression.

**Statistical data analysis:** Excel 2003 was used to establish a data library for all clinical data. After data input, SPSS18.0 For Window was applied for data analysis after all data were checked, with inspection level α=0.05, with two-sided test used. Measurement data were expressed by mean ± standard deviation (X±S), and enumeration data by Chi-square test. Measurement data conformed to the requirements of normal distribution and homogeneity of variance. Independent data were analyzed using independent-sample t test, while data among multiple groups were analyzed using one-way analysis of variance and paired comparison using LSD method. Data dissatisfied with the requirements were analyzed with rank sum test.
while data among multiple groups were analyzed by non-parametric test. P<0.05 was considered statistically.

**Results**

**Comparison of baseline data among 3 groups**

There was insignificant difference among 3 groups in general data like gender, age, marital status and the number of nidi (P>0.05), with comparability. Detailed information is shown in Table 1.

<table>
<thead>
<tr>
<th>Group</th>
<th>Case</th>
<th>Male/Female</th>
<th>Age</th>
<th>Married/unmarried/spouses loss/divorced</th>
<th>The number of nidi</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mild depression</td>
<td>21</td>
<td>10/11</td>
<td>70.24±10.82</td>
<td>16/14/0</td>
<td>3.81±1.50</td>
</tr>
<tr>
<td>Moderate depression</td>
<td>65</td>
<td>33/32</td>
<td>70.68±10.85</td>
<td>45/4/14/2</td>
<td>4.12±1.34</td>
</tr>
<tr>
<td>Severe depression</td>
<td>6</td>
<td>1/5</td>
<td>68.67±12.01</td>
<td>3/0/3/0</td>
<td>3.67±1.63</td>
</tr>
</tbody>
</table>

Table 1: Baseline Data of the Three Groups.

In this study, a total of 113 patients with PSD were selected, and only 92 who conformed to the inclusion criteria were enrolled, in whom 21 patients had mild depression, 65 had moderate depression and 6 had severe depression (Figure 1).

**Analysis of IL-6 level in PSD (table 2 and figure 2)**

In this study, the normal upper limit of serum IL-6 was 5.9pg/ml. Single-sample t test was applied, and the result showed a significant difference (t=4.647, P=0.000), indicating that serum IL-6 level in patients with PSD was markedly higher than normal level.

Comparison of serum IL-6 levels in different groups using non-parametric test showed an insignificant difference among groups (X²=2.885, P=0.236), demonstrating that the serum IL-6 was similar in patients with different-degree PSD.

<table>
<thead>
<tr>
<th>Depression severity</th>
<th>n</th>
<th>IL-6</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mild depression</td>
<td>21</td>
<td>10.79±13.07</td>
</tr>
<tr>
<td>Moderate depression</td>
<td>65</td>
<td>24.68±35.83</td>
</tr>
<tr>
<td>Severe depression</td>
<td>6</td>
<td>43.13±60.53</td>
</tr>
</tbody>
</table>

Table 2: Comparison of serum IL-6 with normal levels among 3 groups (±s).

**Analysis of TNF-α level in PSD (table 3 and figure 3)**

In this study, the normal upper limit of serum TNF-α was 8.1pg/ml. Single-sample t test was applied, and the result showed a significant difference (t=6.331, P=0.000), indicating that serum TNF-α level in patients with PSD was markedly higher than normal level. Comparison of serum IL-6 levels in different groups using homogeneity test of variance followed by non-parametric test showed an insignificant difference among groups (X²=0.083, P=0.959), demonstrating that the serum TNF-α was similar in patients with different-degree PSD.

<table>
<thead>
<tr>
<th>Depression severity</th>
<th>n</th>
<th>TNF-α</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mild depression</td>
<td>21</td>
<td>11.95±4.46</td>
</tr>
<tr>
<td>Moderate depression</td>
<td>65</td>
<td>13.92±8.90</td>
</tr>
<tr>
<td>Severe depression</td>
<td>6</td>
<td>15.08±11.48</td>
</tr>
</tbody>
</table>

Table 3: Comparison of serum TNF-α with normal levels among 3 groups (±s).
Discussion

Although there are numerous studies on the pathogenic mechanisms of PSD in recent years, it has not been interpreted from single system. This disease involves multiple systems\(^6\), including neuroanatomy, neurotransmitters, inflammatory reactions, neurotrophic factors, neuroendocrinology, brain neuropeptides, and social psychology, etc. The pathogenic mechanism of PSD is still unclear, which leads to difficulties for the clinical diagnosis and treatment. Cytokine theory is a recognised important hypothesis which explains that inflammatory factors, like IL-6, TNF-\(\alpha\) and IL-1\(\beta\), increase persistently after the occurrence of PSD\(^8\)\(^9\).

Su et al\(^10\) discovered that in patients with PSD, all of IL-6, IL-10, TNF-\(\alpha\) and interferon \(\gamma\) (INF-\(\gamma\)) increased markedly, which indicated that immunological imbalance played an important role in the pathological and physiological process of PSD, while IL-6 and TNF-\(\alpha\) were the critical cytokines that induced the immunological imbalance. The clinical study of Tang X et al\(^11\) revealed that the increase of serum IL-6 and TNF-\(\alpha\) levels in patients with PSD was in close association with patients’ depression severity. Above studies all have proved that serum IL-6 and TNF-\(\alpha\) levels increase notably in patients with PSD, which was consistent with the conclusion of this study. Differently, serum IL-6 and TNF-\(\alpha\) levels all increased along with the aggravation of depression severity, but there was insignificant difference in serum IL-6 and TNF-\(\alpha\) levels among patients with different-degree PSD, which might be caused by the small sample selected in this study. In further study, the sample should be expanded to prove the relationship between depression severity and the levels of IL-6 and TNF-\(\alpha\).

A cytokine is a small-molecule protein or peptide with bioactivity that is secreted by active immune cells, which plays an important regulating role in maintaining in-vivo homeostasis and some pathological status\(^12\). IL-6 and TNF-\(\alpha\) are critical cytokines that induce immunological imbalance, whose increase indicates that the occurrence of PSD is closely associated with immunological balance. Meanwhile, levels of IL-6, TNF-\(\alpha\) and high-sensitivity C-reactive protein (hs-CRP) have certain predictive effects on the occurrence of PSD, and early interventions on PSD is of certain guiding significance in reducing the morbidity of PSD\(^13\)\(^-\)\(^15\).

In this study, the levels of IL-6 and TNF-\(\alpha\) were notably higher than normal levels in all groups, which were the lowest in mild depression group and the highest in severe depression group, showing that levels increased along with the aggravation of depression. However, the study results were limited to certain extent, which might be associated with the limited conditions and small study samples that originated from the same hospital, therefore, multi-center large-scale study is expected to further prove above results.

Recent studies have demonstrated that inflammatory factors have participated in the occurrence and development of PSD, which is marked by the increase of levels of different cytokines, but the detailed mechanism is still unclear. A study\(^16\) has believed that the potential mechanism of TNF-\(\alpha\) participating in the occurrence and development of PSD is that the increase of TNF-\(\alpha\) can activate immunocyte function, which may leads to pathological damage to the immune of central nerve system, thus triggering depression. Pascoe et al\(^17\) believed that the inflammatory response was closely associated with PSD after the occurrence of cerebral stroke, in which inflammation might cause imbalance of homeostasis and metabolic disorder in human body under certain conditions, which directly or indirectly impact the balance of the secretion of intracranial neurotransmitters, thus triggering the occurrence of depression. Cytokine signals will impact the synthesis, release and re-absorption of emotion-associated neurotransmitters after entering into brain. An animal experiment study\(^18\) illustrated that administration of cytokines or cytokine inducers could markedly influence the metabolism of 5-HT, norepinephrine and dopamine. Whether patients’ depression severity could be understood by monitoring the levels of cytokines and whether PSD could be prevented or treated by inhibiting inflammatory reactions should be further studied.
References


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