Significance and changes of peripheral blood, placental tissue cytokines and NO in patients with hypertensive disorders of pregnancy

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ABSTRACT

Objective: To investigate significance and changes of placental tissue cytokines and NO levels in peripheral blood of patients with hypertensive disorders of pregnancy and their relationship.

Methods: A total of 75 cases of hypertensive disorders of pregnancy were selected as researching objects, including 26 cases of pregnancy-induced hypertension subgroup, 29 cases of patients with mild preeclampsia and 20 cases of subgroups severe preeclampsia subgroups; another 45 cases of healthy pregnant women were also selected as the control group. The maternal blood and placental tissue factor (TNF-α, IL-6, hs-CRP) and NO levels of the four groups were compared.

Results: Peripheral blood and placental tissue cytokine levels of patients with hypertensive disorders of pregnancy were significantly higher, NO levels were significantly lower than the control group (P<0.01). In subgroups peripheral blood and placenta cytokine levels were gradually increased, NO levels were increased with gestational hypertension disease degree (P<0.05); NO was significantly negatively correlated with TNF-α, IL-6, hs-CRP levels (P<0.01).

Conclusions: During pregnancy, monitoring of TNF-α, IL-6, hs-CRP and NO in placental tissue is helpful in prediction and evaluation of early hypertensive disorders of pregnancy, and is also helpful in treatment.

1. Introduction

Hypertensive disorders of pregnancy (HDP) is the disease with coexistence of pregnancy and raised blood pressure, and the clinical morbidity rate is 5%-12%[1,2]. HDP carries very serious effect on the mother-infant prognosis, and is the major explanation for increased mortality of pregnant and lying-in women as well as perinatal infants[3]. At the present, it remains clinically unclear about the pathogenesis of HDP, but many experts believe that the unusual levels of peripheral blood, placental tissue cytokines and nitric oxide (NO) are the major explanations for the incidence of HDP[4]. The present research aims to analyze the monitoring results of peripheral blood, tumor necrosis factor (TNF)-α, interleukin (IL)-6, high-sensitivity C-reactive protein (hs-CRP) in placental tissue and NO in pregnant women, in order to provide the scientific basis for the early diagnosis as well as treatment, and prognosis judgment of HDP.

2. Materials and methods

2.1. General data

A total of 75 cases of HDP partal patients received in hospital for treatment from July 2011 to October 2014 were taken as HDP Group, in accord with clinical diagnostic standards and classification criteria for HDP formulated in Obstetrics and Gynecology (Version 7) edited by Yue Jie. According to the severity criteria for HDP, the patients in HDP Group were divided into 3 groups, namely, (1) gestational hypertension group (n=26), aged 22-35 years, weighed 63-80 kg and pregnant for 35-39 weeks, with age of (28.72 ± 3.27) years, weight of (72.09 ± 4.39) kg and gestational age of (37.83 ± 2.08) weeks, on average; (2) mild preeclampsia group (n = 29), aged 22-34 years, weighed 61-82 kg and pregnant for 35-39 weeks, with age of (28.15 ± 3.00) years, weight of (73.54 ± 4.92) kg and gestational age of (37.56 ± 2.13) weeks, on average; (3) severe...
preeclampsia group \((n=20)\), aged 21-33 years, weighed 60-79 kg and pregnant for 35-39 weeks, with age of \((27.25 \pm 3.11)\) years, weight of \((71.16 \pm 4.59)\) kg and gestational age of \((37.72 \pm 2.32)\) weeks, on average. In addition, 45 cases of healthy pregnant women expecting the delivery in the same term were taken as Control Group, aged 22-33 years, weighed 61-77 kg and pregnant for 36-40 weeks, with age of \((27.09 \pm 2.37)\) years, weight of \((70.00 \pm 5.03)\) kg and gestational age of \((38.09 \pm 1.57)\) weeks, on average. All of the groups were singleton pregnancy and no premature rupture of fetal membranes, placenta previa, placental abruption, hemorrhagic diseases, renal disease, diabetes, malignant tumors, immune diseases, etc. were observed. The differences in age, gestational age and weight in all groups were not statistically significant \((P > 0.05)\), and therefore, were comparable.

2.2. Severity classification criteria for HDP[5-6]

(1) Gestational hypertension: high blood pressure only, with or without edema, without albuminuria;
(2) Mild preeclampsia: high blood pressure with albuminuria;
(3) Severe preeclampsia: blood pressure 160/110 mmHg; albuminuria 3 g/24 h, accompanying with headache, blurred vision, nausea, emesis, pain in right upper quadrant; spasm, exudation or bleeding in eye ground; liver and renal dysfunction, or abnormal clotting mechanism; heart failure and/or pulmonary edema.

2.3. Methods

Five milliliter peripheral venous blood was taken from pregnant women in all of the groups, went through the anticoagulation process with heparin, and then was centrifuged at 2 000 r/min for 20 min. After that, the supernatant liquor was taken out and preserved at -20 °C for test. After delivery, 500 mg of the very central tissue of maternal surface of placenta was immediately taken from pregnant women in all groups, to which 2 mL phosphate buffer at concentration of 25 mmol/L (pH = 7.4) was added. The mixture was put into refiner to make homogenate and was centrifuged at 2 000 r/min for 20 min. The supernatant liquor was also taken out and preserved at -20 °C for test. The determination of NO, TNF-α and IL-6 was conducted by radioimmunoassay while the hs-CRP was determined by immunoturbidimetry. All the determination was conducted in strict accordance with the instruction on kits[7-9].

2.4. Observation indicators

The levels of peripheral blood, cytokines in placental tissue (TNF-α, IL-6, hs-CRP) and NO in pregnant women of all groups were determined.

2.5. Statistical processing

Data were processed and analyzed by SPSS 14.0 software and expressed by Mean±SD. t test was conducted on differences between groups. If \(P<0.05\), the differences were considered statistically significant.

3. Results

3.1. Comparison of peripheral blood, cytokines in placental tissue and NO between HDP Group and Control Group

In HDP Group, the level of NO was significantly lower than that in Control Group while the levels of TNF-α, IL-6 and hs-CRP were significantly higher than in Control Group, with statistically significant differences \((P < 0.01)\). Detailed information was in Table 1.

<table>
<thead>
<tr>
<th>Groups</th>
<th>n</th>
<th>TNF-α (μmol/L)</th>
<th>IL-6 (pg/mL)</th>
<th>hs-CRP (mg/L)</th>
<th>NO (μmol/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control Group</td>
<td>45</td>
<td>25.58 ± 3.38</td>
<td>89.28 ± 11.10</td>
<td>8.09 ± 0.59</td>
<td>44.02 ± 5.56</td>
</tr>
<tr>
<td>HDP Group</td>
<td>75</td>
<td>51.27 ± 10.38</td>
<td>108.18 ± 14.39</td>
<td>17.02 ± 1.32</td>
<td>28.01 ± 5.09</td>
</tr>
<tr>
<td>t value</td>
<td></td>
<td>12.298</td>
<td>8.209</td>
<td>11.299</td>
<td>14.398</td>
</tr>
<tr>
<td>P value</td>
<td></td>
<td>0.000</td>
<td>0.0078</td>
<td>0.000</td>
<td>0.000</td>
</tr>
</tbody>
</table>

Table 2.

Comparison of peripheral blood, cytokines in placental tissue and NO within HDP Group (Mean±SD).

<table>
<thead>
<tr>
<th>Groups</th>
<th>n</th>
<th>TNF-α (μmol/L)</th>
<th>IL-6 (pg/mL)</th>
<th>hs-CRP (mg/L)</th>
<th>NO (μmol/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gestational hypertension group</td>
<td>26</td>
<td>43.30 ± 5.45</td>
<td>96.59 ± 11.01</td>
<td>13.09 ± 2.22</td>
<td>31.29 ± 4.45</td>
</tr>
<tr>
<td>Mild preeclampsia group</td>
<td>29</td>
<td>50.28 ± 6.17</td>
<td>117.08 ± 14.39</td>
<td>16.27 ± 3.77</td>
<td>27.28 ± 3.88</td>
</tr>
<tr>
<td>Severe preeclampsia group</td>
<td>20</td>
<td>70.09 ± 8.72</td>
<td>144.3 ± 22.38</td>
<td>28.02 ± 5.58</td>
<td>24.39 ± 2.17</td>
</tr>
<tr>
<td>F value</td>
<td></td>
<td>4.309</td>
<td>6.282</td>
<td>4.029</td>
<td>3.927</td>
</tr>
<tr>
<td>P value</td>
<td></td>
<td>0.032</td>
<td>0.015</td>
<td>0.034</td>
<td>0.035</td>
</tr>
</tbody>
</table>
TNF-α, IL-6, hs-CRP in placental tissue were increased gradually while the NO level was gradually decreased. There were statistically significant differences among the three groups in HDP Group ($P<0.05$). Detailed information was in Table 2.

### 3.3. Relevance of peripheral blood, cytokines in placental tissue and NO in HDP Group

In HDP Group, the significantly negative correlation was shown between NO level and TNF-α, IL-6, hs-CRP levels ($r = -0.728, -0.613, -0.578, P<0.01$).

### 4. Discussion

HDP has great adverse effect on life and health of pregnant women and perinatal infants, with unclear inducing factor and very limited therapy options for early prevention and treatment, to which close attention has been paid domestically and overseas at the present[10]. Basically, HDP occurs 20 weeks after pregnancy, with clinical symptoms of high blood pressure, albuminuria, edema and so forth; twitch and coma would happen in severe case and it can be life-threatening to both mother and infant[11]. By far, it has been clinically believed that the factors leading to HDP morbidity may be the damage in immunologic function and blood vessel endothelium of pregnant women’s, shallow placental implantation, ateliosis of the damage in immunologic function and blood vessel endothelium of pregnant women’s, shallow placental implantation, ateliosis of placenta and decoated vascular network, etc.

Endothelium dysfunction is the result of broken dynamic balance in damage and repair of endothelium, and one important factor that represents the function of blood vessel endothelium is NO[12]. NO is the most important endogenous vasodilatory substance secreted by vascular endothelial cell. Under physiological condition, the moderate release of NO in pregnant women plays a key role in regulation of placental blood circulation. Whether the integrated function of endothelial cells can be maintained, depends on, to some extend, the dynamic balance of vasoactive substances like NO, whose imbalance can be caused by the damage in endothelial cells. The decreasing secretion of NO plays a significant role in clinical pathogenesis of HDP.[13]

TNF-α is the polypeptide cytokine produced by macrophages, and the important intermediate substance involved in kinds of physiological and immune responses. There are many macrophages in uterine deciduas and placental tissue, and TNF-α plays a very key role in maintenance of normal pregnancy and initiation of secretion[14]. In normal case, TNF-α possesses the antitumor and anti-infection functions, being beneficial to the organism. By making damage to vascular endothelial cells, TNF-α gets involved in the pathogenetic process of HDP. High-level TNF-α can do damage to vascular endothelial cells by combination with receptor in vascular endothelial cells or direct activation of oxygen free radical, collagenase and protease, and then the balance of secretion of vasoactive substance is broken, arteriospasm happens and sequentially the HDP occurs or the condition is aggravated.

IL-6 is a sort of glycoprotein, whose molecular weight is 21-26 kD. It can be secreted by many kinds of histocytes, with extensive biological function, and can damage vascular endothelial cells by activating blood platelet to make aggregative response happen. According to the relevant literature, it is said that immune imbalance and vascular endothelial damage are the major reasons for HDP. In the case of HDP, as the monocytes and lymphocytes are activated, the gene of IL-6 mRNA are “switched on” so that more IL-6 can be transcript synthesized by IL-6 mRNA and get involved in the pathogenetic process of HDP. Relevant researches found that the levels of peripheral blood, IL-6 in placental tissue in patients with gestational hypertension, mild preeclampsia and severe preeclampsia were significantly higher than in normal pregnant women and they were elevated with worse condition[15]. The results of the present research are in accordance with that.

CRP shows extremely low appearance in the serum of healthy persons, but significantly elevated CRP levels in the serum of patients with acute inflammatory response, trauma, rheumatoid arthritis, etc., with high sensitivity and accuracy. Hs-CRP is the main protein involving in inflammatory response or acute phase response. When inflammatory response occurs in the organism, the hs-CRP level would be significantly elevated. The levels of peripheral blood, CRP in placental tissue cells in women with normal pregnancy are also significantly higher than in early pregnancy. The mechanism of action for higher hs-CRP in HDP patients than normal pregnant women with the same gestation age could be the excessive activation of inflammatory response, as well as the significantly increasing secretion of inflammatory medium, inflammatory cytokines and oxidative stress product, caused by many factors; due to the involvement of high-concentration CRP in inflammatory response, and damage of vascular endothelial cells, the intima is thickened, vascular resistance is increased and diastolic function of blood vessels is reduced so that high blood pressure happens[16]. The results of present research are in accordance with that. Moreover, the present research finds that the NO level and TNF-α, IL-6, hs-CRP levels are negatively correlated ($P<0.01$).

In conclusion, the monitoring of peripheral blood, TNF-α, IL-6, hs-CRP in placental tissue and NO in patients with HDP can make an early prediction and evaluation of HDP severity and provide
certain reference frame for the efficient therapeutic approach.

References


