Effect of folic acid and vitamin supplement on endothelial injury and placental blood perfusion in patients with hypertensive disorder complicating pregnancy

Wei Lin¹, Sha Ma²*, Lin Yang³

¹. Department of Obstetrics and Gynecology, Wuhan Red Cross Hospital (Wuhan 11th Hospital) in Hubei Province, Wuhan City, Hubei Province, 430015
². Department of Obstetrics and Gynecology, Wuhan Women and Children’s Health Care Center in Hubei Province, Wuhan City, Hubei Province, 430015
³. Branch of Allergic Reactions, Tongji Hospital in Wuhan Hubei Province, Wuhan City, Hubei Province, 430030

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ABSTRACT

Objective: To study the effect of folic acid and vitamin supplement on endothelial injury and placental blood perfusion in patients with hypertensive disorder complicating pregnancy.

Methods: A total of 62 patients who were diagnosed with preeclampsia in Wuhan Red Cross hospital between March 2015 and February 2017 were selected as the research subjects and randomly divided into two groups, the observation group received folic acid, vitamin and conventional symptomatic treatment, and control group accepted conventional symptomatic treatment. Serum contents of endothelial injury markers and oxidative stress indexes were detected before treatment and 2 weeks after treatment; the expression of apoptosis genes in the placenta were detected after delivery. Results: 2 weeks after treatment, serum ET-1, tTG, sFlt-1, sEng, MPO, IMA, MDA and AOPP levels of both groups of patients were significantly lower than those before treatment, and serum ET-1, tTG, sFlt-1, sEng, MPO, IMA, MDA and AOPP levels of observation group were significantly lower than those of control group; after delivery, Fas, Bax and Caspase-3 mRNA expression levels in placenta tissue of observation group were significantly lower than those of control group while Bcl-2, Survivin and Livin mRNA expression levels were significantly higher than those of control group. Conclusion: Folic acid and vitamin supplement can significantly improve the endothelial injury and placental blood perfusion in patients with hypertensive disorder complicating pregnancy.

1. Introduction

Hypertensive disorder complicating pregnancy is the common complication during pregnancy, preeclampsia (PE) is the most common in it, it is with unknown etiology so far and short of effective treatments, and the termination of pregnancy and delivery of placenta are still the most basic therapies for preeclampsia. The maternal endothelial function damage, decreased placental blood perfusion and abnormal placental development are the important pathological features of pre-eclampsia[1,2]. High homocysteine is an independent risk factor for cardiovascular and cerebrovascular diseases, which has received more and more attention and has been proven to be closely related to the onset of preeclampsia in recent years. In the course of preeclampsia, the abnormally accumulated homocysteine can affect endothelial function and placental development by means of inflammatory response, oxidative stress and apoptosis, etc[3,4]. Folic acid and vitamin B12 are the necessary prothetic groups in the homocysteine metabolism pathways, and supplementation of folic acid and vitamins can improve the homocysteine metabolism and reduce homocysteine content. In the following studies, we analyzed the effects of folic acid and vitamin supplementation on endothelial injury and placental blood perfusion in patients with hypertensive disorder complicating pregnancy.
2. Case information and methods

2.1 General information of preeclampsia cases

A total of 62 patients who were diagnosed with preeclampsia in WuHan Red Cross Hospital between March 2015 and February 2017 were selected as the research subjects, all the patients were in accordance with the diagnostic criteria for preeclampsia, and the patients complicated by hypertension before pregnancy as well as those combined with gestational diabetes mellitus and intrahepatic cholestasis of pregnancy were ruled out. Random number table method was used to divide the 62 patients with preeclampsia into two groups, 31 cases in each group. Observation group were 24-34 years old, and the gestational age at delivery was 36-39 weeks; control group were 23-34 years old, and the gestational age at delivery was 36-38 weeks. There was no statistically significant difference in general information between the two groups ($P>0.05$).

2.2 Preeclampsia therapy

Control group received routine symptomatic treatment, and the method was as follows: 25% magnesium sulfate injection 60 mL in 5% glucose liquid 1 000 mL, intravenous drip, 1 time/d; Nifedipine Tablets 10 mg, taken orally, 3 times/d. Observation group of patients, on the basis of conventional symptomatic treatment, received folic acid and vitamin therapy, and the method was as follows: 400 μg folic acid, 50 μg vitamin B12 tablets, 1.0 g vitamin C tablet and 400 IU vitamin E, taken orally, 1 time/d, for continuous use until delivery.

2.3 Serum index detection

Before treatment and 2 weeks after treatment, 3-5 mL of cubital venous blood was collected from two groups of patients and centrifuged to separate serum, and enzyme-linked immunosorbent assay kit was used to determine the levels of endothelin-1 (ET-1), tissue transglutaminase (tTG), soluble fms-like tyrosine kinase-1 (sFlt-1), soluble endoglin (sEng), myeloperoxidase (MPO), ischemia-modified albumin (IMA), malondialdehyde (MDA) and advanced oxidation protein products (AOPP).

2.4 Gene expression detection in placenta

Moderate amount of placental tissue was collected within 30 minutes after delivery, the calcified area and great vessels should be avoided, the tissue was washed with saline repeatedly until the blood was basically removed, then shortly frozen in liquid nitrogen and added in Trizol lysis buffer, total RNA in tissue was extracted and synthesized into cDNA by reverse transcription, fluorescence quantitative PCR kit was used for cDNA amplification, the amplified target genes included factor associated suicide (Fas), Bcl-2-associated X protein (Bax), cysteinyl aspartate specific proteinase-3 (Caspase-3), B-cell lymphoma-2 (Bcl-2), Survivin and Livin, and the mRNA expression was calculated according to the amplification curve.

2.5 Statistical methods

SPSS 17.0 software was used to input and analyze data, data analysis between two groups was by t test and $P<0.05$ indicated statistical significance in differences.

3. Results

3.1 Serum endothelial injury marker levels

Before treatment and 2 weeks after treatment, analysis of serum endothelial injury markers ET-1 (μg/mL), tTG (ng/mL), sFlt-1 (ng/mL) and sEng (ng/mL) between two groups of patients was as follows: serum ET-1, tTG, sFlt-1 and sEng levels were not significantly different between two groups of patients before treatment ($P>0.05$); 2 weeks after treatment, serum ET-1, tTG, sFlt-1 and sEng levels of both groups of patients were significantly lower than those before treatment ($P<0.05$), and serum ET-1, tTG, sFlt-1 and sEng levels of observation group were significantly lower than those of control group ($P<0.05$).

Table 1.

<table>
<thead>
<tr>
<th>Groups</th>
<th>n</th>
<th>Time</th>
<th>ET-1 (μg/mL)</th>
<th>tTG (ng/mL)</th>
<th>sFlt-1 (ng/mL)</th>
<th>sEng (ng/mL)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Observation</td>
<td>31</td>
<td>Before</td>
<td>4.51±0.67</td>
<td>10.38±1.56</td>
<td>7.62±0.89</td>
<td>4.23±0.52</td>
</tr>
<tr>
<td></td>
<td></td>
<td>treatment</td>
<td>1.93±0.24</td>
<td>7.61±0.83</td>
<td>2.55±0.34</td>
<td>1.87±0.24</td>
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<tr>
<td>Control</td>
<td>31</td>
<td>Before</td>
<td>4.47±0.69</td>
<td>10.81±1.49</td>
<td>7.81±0.92</td>
<td>4.41±0.59</td>
</tr>
<tr>
<td></td>
<td></td>
<td>treatment</td>
<td>2.77±0.35</td>
<td>7.61±0.83</td>
<td>3.89±0.52</td>
<td>2.77±0.36</td>
</tr>
</tbody>
</table>

*: comparison between observation group and control group, $P<0.05$; #: comparison between before and after treatment, $P<0.05$. 

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Pre-eclampsia (PE) is the most common type of hypertension
4. Discussion
Pro-apoptosis gene expression in placenta of two groups of patients.
Table 3.
Survivin and Livin expression in placenta tissue were statistically
significant between two groups of patients (P<0.05), and serum MPO, IMA, MDA and AOPP levels of observation group were significantly lower than those of control group (P<0.05).
3.3. Apoptosis gene expression in placenta
After treatment, analysis of apoptosis genes Fas, Bax, Caspase-3,
Bcl-2, Survivin and Livin expression in placenta between two
groups of patients was as follows: serum MPO, IMA, MDA and AOPP levels were significantly different between two groups of patients before treatment (P>0.05); 2 weeks after treatment, serum MPO, IMA, MDA and AOPP levels of both groups of patients were significantly lower than those before
treatment (P<0.05), and serum MPO, IMA, MDA and AOPP levels of observation group were significantly lower than those of control
group (P<0.05).
3.2 Serum oxidative stress index levels
Before treatment and 2 weeks after treatment, analysis of serum oxidative stress indexes MPO (pg/mL), IMA (ng/mL), MDA (nmol/mL) and AOPP between two groups of patients was as follows: serum MPO, IMA, MDA and AOPP were not significantly different between two groups of patients before treatment (P>0.05); 2 weeks after treatment, serum MPO, IMA, MDA and AOPP levels were not significantly different between two groups of patients before treatment (P>0.05), and serum MPO, IMA, MDA and AOPP levels of observation group were significantly lower than those of control group (P<0.05).
5. Discussion
Pre-eclampsia (PE) is the most common type of hypertension
in pregnancy. Although the etiology of PE is not clear at present,
the close relationship of the maternal endothelial injury, the
placental ischemia hypoxia and other pathological links with
preeclampsia has been unanimously approved[5,6]. In recent years, the relationship between hyperhomocysteinemia and preeclampsia has attracted more and more attention. Homocysteine is a by-product in methionine metabolic cycle, and its metabolism pathways consist of the following two ways[7,8]: (1) under the participation of vitamin B6, about 50% of homocysteine is decomposed into cysteine and then gradually degraded into pyruvic acid and water; (2) under the participation of vitamin B12 and folic acid, about 50% of homocysteine reacts with 5-methyltetrahydrofolic acid and synthesize methionine. Lack of folic acid and vitamin in pregnant women can affect the homocysteine metabolism and cause elevated homocysteine levels in the body, which influences endothelial function and placenta development through inflammatory reaction, oxidative stress, apoptosis and other pathways. Based on the close relationship between hyperhomocysteinemia and the pathogenesis of preeclampsia, some scholars have used folic acid and vitamin to treat preeclampsia and achieved positive effect, which can effectively reduce homocysteine levels and blood pressure levels[9]. However, there is no report about the effects of folic acid and vitamin therapy on endothelial injury, placental hypoxia and other pathological links in preeclampsia.
Maternal endothelial injury is an important pathological feature of preeclampsia patients, and it is also an important cause of small vasospasm, microthrombosis, and glomerular dysfunction. In the process of endothelial function injury, various endothelial markers such as ET-1, tTG, sFlt-1 and sEng are massively released into the blood circulation. ET-1 is a vasoconstrictor synthesized by endothelial cells, and the endothelial injury will increase the release of ET-1 and cause vasospasm[10]; tTG is a type of protein with catalytic activity, which is synthesized by placenta trophocytes and falls off into the blood circulation, and mainly mediates NF-
κB-mediated inflammation and causes endothelial damage by inflammatory mediators; sFlt-1 and sEng are the products when extracellular Flt-1 and Endoglin structures fall off into the blood circulation respectively, which can be combined with VEGF, PLGF, TGFB-β and so on to antagonize their biological functions and cause endothelial injury[11,12]. In order to define the effect of folic acid and vitamin on endothelial damage degree in patients with preeclampsia, serum levels of endothelial injury markers were analyzed in the study before and after treatment, and the results showed that serum

<table>
<thead>
<tr>
<th>Groups</th>
<th>n</th>
<th>Time</th>
<th>MPO (pg/mL)</th>
<th>IMA (ng/mL)</th>
<th>MDA (nmol/mL)</th>
<th>AOPP</th>
</tr>
</thead>
<tbody>
<tr>
<td>Observation group</td>
<td>31</td>
<td>Before treatment</td>
<td>13.41±1.85</td>
<td>57.41±7.61</td>
<td>8.31±0.93</td>
<td>142.41±19.45</td>
</tr>
<tr>
<td></td>
<td></td>
<td>After treatment</td>
<td>8.34±1.03*</td>
<td>33.25±4.57*</td>
<td>3.44±0.52*</td>
<td>83.41±10.24*</td>
</tr>
<tr>
<td>Control group</td>
<td>31</td>
<td>Before treatment</td>
<td>13.87±1.91</td>
<td>58.12±7.94</td>
<td>8.24±0.98</td>
<td>144.10±17.64</td>
</tr>
<tr>
<td></td>
<td></td>
<td>After treatment</td>
<td>10.93±1.42*</td>
<td>42.94±5.65*</td>
<td>5.73±0.61*</td>
<td>113.48±14.48*</td>
</tr>
</tbody>
</table>

*: comparison between observation group and control group, P<0.05; #: comparison between before and after treatment, P<0.05.
stress and aggravate the endothelial oxidation damage but also increases the formation of free radicals, activate oxidative homocysteine in the body not only directly injures the endothelium, and inhibit the over-activation of apoptosis induced by hypoxia.

In the process of oxidative stress, MPO, IMA, MDA and AOPP are the oxidative reaction products of lipid and protein with free radicals. The accumulation of homocysteine in the body not only directly injures the endothelium, but also increases the formation of free radicals, activate oxidative stress and aggravate the endothelial oxidation damage[14,15]. In order to define the effect of folic acid and vitamin supplementation on oxidative stress in patients with preeclampsia, the changes in serum levels of oxidative stress reaction indexes before and after treatment were analyzed in the study, and the results showed that serum MPO, IMA, MDA and AOPP levels of both groups of patients after treatment were significantly lower than those before treatment, and serum MPO, IMA, MDA and AOPP levels of observation group after treatment were significantly lower than those of control group while Bcl-2, Survivin and Livin mRNA expression levels in placenta tissue showed that Fas, Bax and Caspase-3 mRNA expression levels in placenta tissue of observation group were significantly lower than those of control group. This indicates that supplementation of folic acid and vitamins on the basis of conventional treatment can improve the placental blood perfusion and inhibit the over-activation of apoptosis induced by hypoxia.

Supplementation of folic acid and vitamin can significantly improve the endothelial injury, also reduce oxidative stress response and inhibit apoptosis in placenta of patients with gestational hypertension.

References


