Effect of probiotics combined with zinc preparation treatment on serum related indicators in children with protracted diarrhea

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Objective: To analyze the effect of probiotics combined with zinc preparation treatment on serum related indicators in children with protracted diarrhea. Methods: A total of 70 children with protracted diarrhea were included in the study and randomly divided into observation group and control group (n=35). Control group accepted probiotics treatment alone, observation group received probiotics combined with zinc preparation treatment, and differences in serum related indicator values were compared between two groups. Results: Serum NO, MDA, GAS and sIL-2R values of observation group after treatment were lower than those of control group, and SOD, RGE2, IgA, IgM, IgG and zinc values were higher than those of control group (P<0.05); serum inflammatory markers IL-6, IL-8, hs-CRP, PCT and HMGB1 values were lower than those of control group (P<0.05). Conclusions: Probiotics combined with zinc preparation treatment can optimize the overall conditions and restore homeostasis in children with protracted diarrhea, and has positive clinical significance.

1. Introduction

Infantile diarrhea is clinically common, the diarrhea caused by pathogenic bacteria infection has the highest incidence, and others also include food allergy, deficiency of trace elements and so on. Diarrhea that lasts for more than 2 weeks may develop into protracted diarrhea and lead to dehydration and decomposition of more nutrients, and severe cases can show a series of complications[1,2]. Early active treatment of diarrhea and prevention of complications is the principle of protracted diarrhea treatment, probiotics is the most commonly used clinical drug that uses normal microorganisms or the substances that promote the growth of microorganisms, can promote normal microbiota growth and reproduction and inhibit pathogenic bacteria growth and reproduction, and has positive effect on promoting diarrhea rehabilitation. Long-term diarrhea can result in excessive zinc decomposition and malabsorption as well as low serum zinc in children, this is one of the important causes of intestinal mucosal dysfunction in children with diarrhea, and so many clinical scholars suggest adding zinc preparation in children with diarrhea[3,4]. In the study, the effect of probiotics combined with zinc preparation treatment on serum related indicators in children with protracted diarrhea was mainly analyzed, hereby reported as follows.

2. Materials and methods

2.1. General information

A total of 70 children with protracted diarrhea who received treatment in our hospital from December 2014 to December 2015 were the research subjects, all patients conformed to the diagnostic criteria for diarrheal disease in our country and the course of disease was more than 2 weeks, and those with parasites (eggs) in stool cultures were excluded.
According to different treatment, the included patients were randomly divided into observation group and control group (n=35). Control group included 19 male cases and 16 female cases, they were 6 months to 5 years old, the average age was (18.26±4.05) months, 5 cases were with severe dehydration, 11 cases were with moderate dehydration and 19 cases were with mild dehydration; observation group included 18 male cases and 17 female cases, they were 5 months to 5 years old, the average age was (18.76±4.82) months, 4 cases were with severe dehydration, 13 cases were with moderate dehydration and 18 cases were with mild dehydration. The two groups showed no significantly difference in gender, age and disease severity (P>0.05), and they were comparable.

2.2. Treatment regimens

Observation group received probiotics combined with zinc preparation treatment, specifically as follows: probiotics (Siliankang) 1 tablet/time for patients <1 year old, 2 times a day, and 2 tablets/time for patients ≥1 years old, 2 times a day; zinc gluconate particles (Hebei Shijiazhuang Shineway pharmaceutical) for patients 1-6 years old, 10-21 kg, 35 mg (0.5 bag)/1 time/day, and for patients < 1 year old, weight <10 kg, reducing zinc preparation appropriately. Control group accepted probiotics treatment alone, and the specific usage and dosage were the same as those of observation group.

2.3. Specimen collection

Before treatment (within 24 hours after admission) and after one course of treatment, 3ml of fasting peripheral venous blood was collected respectively, put in heparin anticoagulation centrifuge tubes and centrifuged for 5 min at 2 500 r/min at low temperature, and the supernatant was collected and saved in -20 ℃ refrigerator for testing.

2.4. Serum related indicators

Nitrate reductase method was used to determine serum nitric oxide (NO) levels, thiobarbituric acid method was used to determine malondialdehyde (MDA) levels, and xanthine oxidase method was used to determine superoxide dismutase (SOD) levels.

RIA method was used to determine serum gastrin (GAS) and prostaglandin E2 (RGE2), and enzyme-linked immunosorbent assay (ELISA) was used to determine serum IgA, IgM, IgG, blood zinc, soluble interleukin-2 receptor (sIL-2R), interleukin-6 (IL-6), interleukin-8 (IL-8), hypersensitive C-reactive protein (hs-CRP), procalcitonin (PCT) and high mobility group box-1 (HMGB1).

2.5. Statistical methods

Data obtained in the study was analyzed by SPSS23.0 software, measurement data comparison was by t test, count data comparison was by chi-square test and P<0.05 was set as the standard of statistical significant differences.

3. Results

3.1. Serum redox-related indicators

Serum redox-related indicators of two groups were not statistically different before treatment (P>0.05), serum NO and MDA values of both groups after treatment were lower than those before treatment while SOD value was higher than that before treatment (P<0.05), serum NO and MDA values of observation group after treatment were lower than those of control group, and SOD value was higher than that of control group (P<0.05), shown in Table 1.

3.2. Serum illness-related indicators

After observation group received probiotics combined with zinc preparation treatment, serum GAS and sIL-2R values were lower than those of control group, and RGE2, IgA, IgM, IgG and zinc values were higher than those of control group (P<0.05), shown in Table 2.

3.3. Serum levels of inflammatory factors

Serum levels of IL-6, IL-8, hs-CRP, PCT and HMGB1 of observation group after treatment were significantly lower than those of control group, and the differences between two groups were significant (P<0.05).

4. Discussion

Protracted diarrhea refers to the diarrheal disease with the course of disease between two weeks and two months, and most scholars have argued that intestinal infection is the main cause of pediatric chronic and protracted diarrhea, among which bacteria, fungi and parasites are the common pathogeny[5]. With the development of social and medical environment, the protracted diarrhea caused by infectious factors has become less and less, and non-infectious factors such as
food allergies, lactose intolerance and deficiency of trace elements have increasingly become the main reasons for protracted diarrhea. Long-term diarrhea can lead to dehydration and the deficiency of a variety of essential elements in children with diarrhea and cause a series of complications, children have poor tolerance, and severe diarrhea can lead to their death.[6,7]

Probiotics is the living microbial preparation made from normal microorganisms or substances that promote the growth of microorganisms, can regulate intestinal function and quickly build intestinal microecological balance, and is the most commonly used drug in the clinical treatment of diarrhea and constipation.[8] Many studies have proved that probiotics can promote Th1 cytokine expression and down-regulate inflammatory factor levels in patients with diarrhea, maintain the integrity of the intestinal mucosa as a whole, and effectively control diarrhea conditions. Studies in many countries have shown that zinc deficiency can easily cause diarrhea, zinc supplementation for children with zinc deficiency can reduce the incidence of diarrhea, so the occurrence and development of diarrheal disease is closely related to zinc deficiency, and children with diarrhea should receive appropriate zinc supplementation[9,10].

In the study, observation group received probiotics combined with zinc preparation treatment and were compared with control group who received probiotics treatment alone in order to make clear the significance of zinc supplement to protracted diarrhea treatment.

Hydroxyl radicals are one of the main causes of intestinal mucosa damage in all kinds of diarrheal diseases, and they can induce nitric oxide synthase expression and its product NO level increase.[11] NO easily reacts with reactive oxygen species to generate active nitrogen metabolites, which leads to oxidized desmin and tubulin, destroyed cytoskeleton and intestinal barrier dysfunction. In the study, after observation group received one course of probiotics combined with zinc preparation treatment, serum NO and MDA values decreased and SOD value increased. Both NO and MDA belong to oxidation products while SOD represents the body's antioxidant capacity, and the above results showed that after combined treatment, the oxidation-antioxidation balance in patients was effectively corrected, the peroxidation state in the body was contained, and thus the intestinal mucosa function gradually restored.[12,13]

Protracted diarrhea can lead to zinc malabsorption or excessive decomposition, and the overall effect is reduced zinc levels in the circulating blood, which influences intestinal mucosa regeneration and the integrity of intestinal mucosa structure, continuously damages the small intestinal mucosa, further reduces the zinc absorption and forms a vicious circle. For children with protracted diarrhea, timely zinc supplementation is very meaningful for rapid recovery and effective rehabilitation of the disease. Zinc deficiency, in particular, will lead to inhibited activity of immunoglobulin receptors on the surface of B cells and reduced serum levels of IgA, IgG, IgM, etc, which is not conducive to the maintenance of normal immune function in children. In the study, serum RGE2, IgA, IgM, IgG and zinc values of observation group after treatment were higher while GAS and sIL-2R values were lower. sIL-2R is the antagonistic protein of interleukin-2, and high concentration of sIL-2R is one of the symbols of abnormal immune response[14]. Both PGE2 and

### Table 1
Comparison of serum values of redox-related indicators between two groups before and after treatment.

<table>
<thead>
<tr>
<th>Groups</th>
<th>NO (μmol/L) Before treatment</th>
<th>NO (μmol/L) After treatment</th>
<th>MDA (μmol/L) Before treatment</th>
<th>MDA (μmol/L) After treatment</th>
<th>SOD (U/L) Before treatment</th>
<th>SOD (U/L) After treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td>Observation</td>
<td>131.37±19.72</td>
<td>74.35±8.11</td>
<td>2.63±0.34</td>
<td>1.57±0.23</td>
<td>0.021±0.003</td>
<td>0.073±0.008</td>
</tr>
<tr>
<td>Control group</td>
<td>128.54±17.38</td>
<td>101.86±15.92</td>
<td>2.71±0.39</td>
<td>2.18±0.34</td>
<td>0.024±0.002</td>
<td>0.041±0.005</td>
</tr>
<tr>
<td>t</td>
<td>0.215</td>
<td>8.394</td>
<td>0.183</td>
<td>5.372</td>
<td>0.114</td>
<td>5.783</td>
</tr>
<tr>
<td>P</td>
<td>&gt;0.05</td>
<td>&lt;0.05</td>
<td>&gt;0.05</td>
<td>&lt;0.05</td>
<td>&gt;0.05</td>
<td>&lt;0.05</td>
</tr>
</tbody>
</table>

### Table 2
Comparison of serum values of illness-related indicators between two groups after treatment.

<table>
<thead>
<tr>
<th>Groups</th>
<th>GAS (ng/mL)</th>
<th>RGE2 (μg/mL)</th>
<th>IgA (g/L)</th>
<th>IgM (g/L)</th>
<th>IgG (g/L)</th>
<th>Zinc (μmol/L)</th>
<th>sIL-2R (pg/mL)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Observation</td>
<td>26.36±2.77</td>
<td>55.13±6.17</td>
<td>1.21±0.25</td>
<td>1.37±0.21</td>
<td>9.72±1.04</td>
<td>64.37±7.12</td>
<td>342.74±41.92</td>
</tr>
<tr>
<td>Control group</td>
<td>33.92±3.75</td>
<td>48.85±5.32</td>
<td>0.82±0.09</td>
<td>0.96±0.11</td>
<td>7.34±0.86</td>
<td>56.52±6.05</td>
<td>511.38±58.35</td>
</tr>
<tr>
<td>P</td>
<td>&lt;0.05</td>
<td>&lt;0.05</td>
<td>&lt;0.05</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
Serum levels of inflammatory factors of two groups after treatment.

<table>
<thead>
<tr>
<th>Groups</th>
<th>IL-6 (ng/L)</th>
<th>IL-8 (ng/L)</th>
<th>hs-CRP (mg/L)</th>
<th>PCT (ng/mL)</th>
<th>HMGB1 (μg/mL)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Observation</td>
<td>112.74±15.83</td>
<td>153.85±20.17</td>
<td>5.42±0.49</td>
<td>0.53±0.04</td>
<td>3.27±0.45</td>
</tr>
<tr>
<td>Control group</td>
<td>219.63±28.75</td>
<td>316.83±37.04</td>
<td>9.81±0.94</td>
<td>1.39±0.12</td>
<td>8.92±0.91</td>
</tr>
<tr>
<td>t</td>
<td>8.392</td>
<td>11.274</td>
<td>7.384</td>
<td>5.374</td>
<td>7.242</td>
</tr>
<tr>
<td>P</td>
<td>&lt;0.05</td>
<td>&lt;0.05</td>
<td>&lt;0.05</td>
<td>&lt;0.05</td>
<td>&lt;0.05</td>
</tr>
</tbody>
</table>
GAS are gastrointestinal hormones, and PGE₂ is a strong vaso-active substance that can promote the arterial smooth muscle relaxation and reduce the vascular resistance, and can inhibit the secretion of gastric acid in intestinal tract; GAS has an effect on the whole gastrointestinal tract, and can promote gastric acid secretion and nourish parietal cells. In the study, combined treatment could fully optimize the state of intestinal mucosa function and promote disease rehabilitation.

Chronic infectious diarrhea is the main cause of protracted diarrhea and will cause the activation of systemic inflammation, and in the study, serum levels of related inflammatory factors of both groups were detected after treatment. hs-CRP is the inflammatory factor earliest and massively secreted after intestinal inflammation, and has a very high sensitivity. IL-6 and IL-8 are important inflammatory and proinflammatory factors that can further stimulate other inflammatory mediator generation and increase intestinal mucosal inflammation[15]. PCT is the protein secreted by thyroid C cells, its level can rise sharply in 6h after infection, it is not influenced by other hormones in the body, and the degree of increase is positively correlated with the severity of infection. HMGB1, also known as amphiphysin, is a new kind of regulator in late inflammation, can activate the endogenous cells or endothelial cells and thus produce pro-inflammatory cytokines, is an important proinflammatory factor, and participates in starting and maintaining a “waterfall” inflammation[16]. The research results showed that probiotics combined with zinc preparation treatment of children with protracted diarrhea could alleviate intestinal mucosal inflammation state, and this is one of the direct signs of intestinal functional recovery.

To sum up, it is concluded as follows: probiotics combined with zinc preparation treatment can optimize the overall conditions and restore homeostasis in children with protracted diarrhea, and it’s worth popularization and application in clinical practice in the future.

References


