Influence of bifidobacterium quadruple preparations on immunity function and cytokine of neonatal hyperbilirubinemia

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ABSTRACT

Objective: To investigate the influence of bifidobacterium quadruple preparations on immunity function and cytokine of neonatal hyperbilirubinemia. Methods: A total of 92 neonates with hyperbilirubinemia were randomly divided into observation group and control group with 46 cases of each group. The control group received conventional therapy, and the observation group was treated with bifidobacterium quadruple preparations on the basis of the control group. The influence of bifidobacterium quadruple preparations on immunity function and cytokine of the two groups were compared. Results: There was no significantly difference in Bilirubin before the treatment between the two groups (P>0.05), but the observation group was significantly lower than the control group 3, 5, 7 d after the treatment (P<0.01); CD3, CD4, CD4/CD8, IL-6, IL-8 of the observation group were higher than the control group after the treatment (P<0.01), but CD8 was lower than the control group (P<0.01). Conclusions: Bifidobacterium quadruple preparations can improve the immune function, and reduce serum bilirubin levels in neonates with hyperbilirubinemia.

1. Introduction

Hyperbilirubinemia is one of the most common diseases during neonatal period. The incidence could be as high as 30%-50% or higher, and showed increasing trend in recent years[1]. Bilirubin deposited in the nuclei of central nervous system could easily cause bilirubin encephalopathy, of which the mortality rate was as high as 1/3, and the majority of other 2/3 survivors had permanent neurological sequelae[2]. Other related drugs were included in the treatment of hyperbilirubinemia neonatal on the basis of phototherapy so far[3]. To investigate the influence of bifidobacterium quadruple preparations on immunity function of the neonates in the treatment process of neonatal hyperbilirubinemia, we used the bifidobacterium quadruple preparations to treat the 46 cases of the neonatal hyperbilirubinemia, and observed the serum bilirubin levels and cytokine levels before and after the treatment. The results showed as follow:

2. Materials and methods

2.1. General data

A total of 92 neonates with hyperbilirubinemia who were treated from July 2013 to June 2015 were selected as the materials, which included 56 cases of male and 36 cases of female in fetus age of 34 to 42 weeks and the average fetus age of (38.6±4.7) weeks. The birth weights were 2.8 kg-4.1 kg, and average weight was (3.2±0.7) kg. The ages were 13 h-21 d, and average age was (8.6±5.7) d. All the neonates were complying with standard of diagnosis of hyperbilirubinemia in the 4th version of Practical neonatology[4]. 92 neonates were randomly divided into observation group and control group with 46 cases of each group. The gender, age, weight and clinical symptom of the neonates in two groups had no statistically difference (P>0.05), which were comparable. The patients of neonatal hyperbilirubinemia in the control group were given active therapy, meanwhile they were given conventional therapy with the liver enzyme inducer, phototherapy, albumin; the patients of observation group were given the bifidobacterium tetravaccine tablets with 0.25 g of warm water or milk for continuously 10 days.
based on the treatment of the control group (Name: SiLianKang, approved by S20060010, Hangzhou Longda Xinke Biological pharmacy Co. Ltd.).

2.2. Instruments and reagent.

The instruments of XL-4 flow cytometer and the Innova905 argon-ion lasers with power of 200 mW and wavelength of 488 nm were produced by BeckmanCoulter Company; The microplate spectrophotometer of 550 type standard was provided by BioRad Company; quantitative ELISA reagent boxes of IL-6 and IL-8 were provided by the company of PBM.

2.3. Observation indicator

The changes and dynamic of serum bilirubin of the neonates were observed instantly in two groups. 3-5 mL of the venous blood was extracted before and after the treatment and divided into two equally. One of them was dealt with EDTA anti-freezing to separate the lymphocyte, label and test the t lymphocyte subsets (CD3, CD4, CD8, CD4/CD8); The other one was performed to separate the serum by the centrifugal machine, and cryopreserved in the refrigerator of -20 °C for the test of interleukin -6 (IL-6) and interleukin-8 (IL-8).

2.4. Statistical analysis

SPSS19.0 was used for Statistical analysis. The measurement data were expressed as mean±SD. t-test was applied for intergroup comparison of average data, with $P<0.05$ as statistically significant difference.

3. Results

3.1. Serum bilirubin levels

The serum bilirubin of the neonates with hyperbilirubinemia in two groups were not statistically different ($P>0.05$) before the treatment; 3 d, 5 d, and the 7 d after treatment, the serum bilirubin levels of the neonatal hyperbilirubinemia in two groups were all decreased ($P<0.01$). The decreasing speed of the observation group was faster than that of the control group. The details showed in Table 1.

3.2. Lymphocyte subsets and cytokines after treatment

After the above treatment, the lymphocyte subsets, CD3 and CD4 of the observation group were all higher than that of the control group ($P<0.01$), but CD8 was significantly lower than that of the control group ($P<0.01$), meanwhile the value of the CD4 and CD8 was significantly higher than that of the control group ($P<0.01$). The IL-6 and IL-8 of the observation group were all significantly higher than that of the control group after treatment ($P<0.01$). Details showed in Table 2.

4. Discussion

Neonatal hyperbilirubinemia is the most common disease during neonatal period. Because of the short life-span and the overmuch destroy of the erythrocyte of the neonates, the bilirubin in blood was elevated. However, the ability of neonates livers to intake, integrate and excrete the bilirubin was relatively low, and as the lack of the relevant bacterium in the intestinal tract, thus the obstacle was formed in urobilinogen[5]. It was believed that there was a closed relation between the occurrence and development of the neonatal hyperbilirubinemia and the hemolytic jaundice, glucose-6-phosphate dehydrogenase deficiency, breast-feeding jaundice, infection, and the increases of bilirubin in hepatoenteral circulation caused by perinatal factors, in maternal fetal blood group incompatibility[6]. The bilirubin could penetrate the blood brain barrier, which would probably induce the neonatal hyperbilirubinemia, and even the disability and death of the neonates. Therefore, it was of great significance to prevent and cure the cerebral injury and improve the prognosis in early treatment. Regarding the therapeutic principle of the hyperbilirubinemia neonatal, it was undoubtedly effective to smooth bile flow and promote the gallbladder function, improve the excretion of bilirubin, and reduce its repeated absorption. Bifidobacteria could accelerate the bilirubin to restore to the urobilinogen and the stercobilinogen then excreted out of the body.

### Table 1

<table>
<thead>
<tr>
<th>Groups</th>
<th>Number of cases</th>
<th>Before treatment</th>
<th>3 d after treatment</th>
<th>5 d after treatment</th>
<th>7 d after treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td>Observation group</td>
<td>46</td>
<td>234.5±42.1</td>
<td>112.3±41.4</td>
<td>94.2±27.7</td>
<td>41.1±12.7</td>
</tr>
<tr>
<td>Control group</td>
<td>46</td>
<td>232.7±41.6</td>
<td>148.6±42.2</td>
<td>132.6±28.3</td>
<td>48.7±13.2</td>
</tr>
<tr>
<td>$t$</td>
<td></td>
<td>0.206</td>
<td>4.179</td>
<td>6.577</td>
<td>2.814</td>
</tr>
<tr>
<td>$P$</td>
<td></td>
<td>0.837</td>
<td>0.000</td>
<td>0.000</td>
<td>0.006</td>
</tr>
</tbody>
</table>

### Table 2

<table>
<thead>
<tr>
<th>Groups</th>
<th>Number of cases</th>
<th>CD3 (%)</th>
<th>CD4 (%)</th>
<th>CD8 (%)</th>
<th>CD4/CD8</th>
<th>IL-6 (pmol/mL)</th>
<th>IL-8 (pmol/mL)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Observation group</td>
<td>46</td>
<td>67.2±7.1</td>
<td>44.3±5.3</td>
<td>26.5±4.6</td>
<td>1.62±0.33</td>
<td>9.1±3.7</td>
<td>13.3±3.5</td>
</tr>
<tr>
<td>Control group</td>
<td>46</td>
<td>56.3±6.7</td>
<td>34.2±4.8</td>
<td>34.7±4.4</td>
<td>0.99±0.31</td>
<td>15.2±3.6</td>
<td>18.1±3.9</td>
</tr>
<tr>
<td>$P$</td>
<td></td>
<td>0.000</td>
<td>0.000</td>
<td>0.000</td>
<td>0.000</td>
<td>0.000</td>
<td>0.000</td>
</tr>
</tbody>
</table>
It could also transform oligosaccharide into acetic acid and lactic acid, lower the activity of beta GP, and prevent the resolving of the bilirubin directly. Besides, bifidobacteria could also decompose the combined cholic acid to be the freecholic acid, and reduce the repeated absorption of the combined cholic acid, which was beneficial to remove the intrahepatic cholestasis[7,8]. The bifidobacterium quadruple preparations, which was used in this study, was the tetravaccine tablets consisted of oral bifidobacterium, streptococcus thermophilus, bacillus lactis and bacillus cereus. The bacillus lactis and the streptococcus thermophilus were all the probiotics of the bacillus bifidus, which could produce the growth factor that could accelerate and be good to the growth of the bifidobacterium, and then further lower the level of bilirubin. The above four bacterial communities would grow and multiply in the intestinal tract after oral administration and absorption, which made the intestinal tract absorb the normal state of the profitable bacterial communities within a short time, and established the normal intestinal bacterial communities, thus contributed double positive effects to inhibit the growth of the pathogenic bacterium and stimulate the advance of the organisms immunity, participate to the immune function as well, which showed as the increases of the CD3, CD4 and the value of CD4 and CD8, and the raising of the IL-6 and IL-8 on the observed indicators.

In conclusion, we believed that the bifidobacterium quadruple preparations can effectively improve the immune function on the basis of reducing serum bilirubin levels in neonates with hyperbilirubinemia, which is worth recommending and popularizing.

References