Effect of (Huai) Rehmanniae Radix Praeparata Polysaccharides on Immune Organs of Rats with Qi and Blood Both Deficiency

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Keywords: Rehmanniae Radix Praeparata polysaccharides, Blood deficiency model, Immune organs, Pathological changes.

Abstract. Purpose: Investigate the protective effect of (Huai) Rehmanniae Radix Praeparata polysaccharides on immune organs of rats with qi and blood both deficiency. Methods: Sixty rats were randomly divided into blank group, angelica blood tonic oral liquid group, and a large, medium and small dose of Rehmanniae Radix Praeparata polysaccharides groups. In addition to the blank group, the model of qi and blood both deficiency in rats was established by tail bloodletting and intraperitoneal injection of cyclophosphamide. After the final administration for 2h, the rats were sacrificed with decervical vertebra, and the thymus and spleen were taken for pathological sections. The thickness of thymus cortex and the number of lymphocytes, the size of splenic nodules and the number of lymphocytes were observed under the microscope. By measuring the widest and narrowest areas of the thymic cortex and averaging its thickness, count the number of lymphocytes loaded on the micrometer scale. The number of lymphocytes pressed on both sides of the line was calculated by taking the central artery of the spleen as the midpoint, the mean is the number of lymphocytes in the surrounding lymphatic sheath of a central arteriole. The other three number of lymphocytes in the surrounding lymphatic sheath of the central splenic artery was calculated by the same method, taking the mean as the number of lymphocytes. Results: In the model group, thymus thickness and spleen nodules decreased significantly, the numbers of lymphocytes in thymic cortex and spleen decreased significantly (P<0.01). Rehmanniae Radix Praeparata polysaccharides groups with different dose could increase the thickness of thymic cortex, enlarge the splenic nodule and increase the number of lymphocytes in thymic cortex and spleen (P<0.01 or P<0.05). Conclusion: (Huai) Rehmanniae Radix Praeparata polysaccharides can improve the pathological changes of immune organs in rats with qi and blood both deficiency.

Introduction

Blood deficiency syndrome is a basic syndrome in TCM syndrome differentiation system of qi, blood, body and fluid. Blood deficiency syndrome of traditional Chinese medicine contains two basic aspects: reduction of substance and disorder of function. Blood deficiency syndrome of traditional Chinese medicine is closely related to blood diseases in modern diseases (common anemia, aplastic anemia, hemorrhagic anemia, hemolytic anemia, etc.), but it is not limited to modern blood diseases. Blood deficiency syndrome of traditional Chinese medicine also contains other diseases caused by abnormal blood function, such as heart palpitations, insomnia, immune dysfunction and coronary heart disease [1-2]. Because blood deficiency syndrome involves a wide range of diseases and can be mixed with other diseases to promote the occurrence and development of diseases, it is necessary to take certain measures to timely intervene and prevent it. Cooked rehmannia is made by cooking the root of rehmannia from scrophulariaceae. Modern studies have shown that cooked rehmannia glutinosa contains catalpa, sugars and glycosides, and has pharmacological effects such as regulating immunity, anti-tumor, promoting hematopoiesis and anti-aging. And after processing, the sugar content of rehmannia increased[3]. Cooked rehmannia has a certain effect on hematopoietic deficiency mice models induced by cyclophosphamide and phenylhydrazine hydrochloride, and
rehmannia polysaccharides can improve the cellular activity in the peripheral blood of mice with qi and blood both deficiency [4-5]. However, there are few reports on the effect of rehmannia glutinosa on the immune organ function of the animal model of blood deficiency. In order to further clarify the mechanism of the improvement of blood deficiency, the effect of (Huai) rehmannia glutinosa on the immune organ of the rats with blood deficiency was investigated.

**Experimental Materials**

**Drugs**

(Huai) Rehmanniae Radix Praeparata, Henan wuzhi county medicinal materials company; Rehmanniae Radix Praeparata polysaccharides, provided by the college of pharmacy, the department of phytochemistry; Cyclophosphamide, Shanghai hualian pharmaceutical co., LTD., batch no. 000116; Anggui blood-enriching oral liquid, produced by zhenzhou union pharmaceutical factory, batch no. 010301.

**Animal**

Wistar rats, SPF, male, weighing 180-200g, Hebei experimental animal center provided, license number: SCXK (hebei) 2003-1-0003, certificate number: 610085.

**Experimental Method [6-7]**

Ten rats were randomly selected as the blank group, and the remaining 50 rats were divided into the model group, the danggui blood-enriching oral liquid group and the large, medium and small rehmannia polysaccharide group. At day 1, 3, 5, 7 and 9 of the experiment, 1ml/180g of blood was discharged from each mouse tail, 40mg/kg of the normal saline solution of cyclophosphamide was injected intraperitoneally on day 2, and 20mg/kg of cyclophosphamide was injected intraperitoneally on day 4, 6, 8, 10 and 12, respectively. On the first day of mold making, the administration group was given water solutions of rehmannia polysaccharide in large, medium and small doses (doses: 200mg/kg, 100mg/kg, 50mg/kg) and danggui blood-enriching oral liquid (3.3ml/kg) once a day for 14 consecutive days. 2h after the administration on the 14th day, the rats were sacrificed by decolling their necks, and the thymus gland and spleen were taken and set in 10% formalin for general slicing. The paraffin-embedded sections were cut and stained, and the thickness of thymus cortex, lymphocyte number, size of splenic nodule and lymphocyte number were observed under the microscope.

**Experimental Results**

**Effect of (Huai) Rehmannia Polysaccharides on Thymus Tissue in Rat Model of Qi and Blood both Deficiency**

Effect of (Huai) rehmannia polysaccharides on thymus tissue: In the blank group, the thymus was divided into distinct leaves, cortex and medullary were demarcated clearly, and cortex was thick and dense with cells. In the model group, the thymus was significantly atrophied, the lobes were not clear, the cortex was significantly atrophied and thinned, and the cells were sparse and reduced. In the danggui blood-enriching oral liquid group, thymus was significantly atrophied and the lobes were not clear, but the cortex was thicker and the cell density was higher than the model group. In the high-dose group of (Huai) rehmannia polysaccharides, the thymus was severely atrophied, the cortex became thin and the lymphocytes were sparse. In the medium dose group of (Huai) rehmannia polysaccharides, there were obvious atrophy, thin cortex and sparse cells. In the small dose group of (Huai) rehmannia polysaccharides, the thymus also atrophied, the cortex became thin and the cells were reduced.
Table 1. Effect of (Huai) rehmannia polysaccharides on thymus tissue in rat model of qi and blood both deficiency (X ± s).

<table>
<thead>
<tr>
<th>Group</th>
<th>Dose (mg/kg)</th>
<th>Thymus thickness (um)</th>
<th>Cortical lymphoid number</th>
</tr>
</thead>
<tbody>
<tr>
<td>Blank</td>
<td></td>
<td>47.3±7.4**</td>
<td>120.5±20.6**</td>
</tr>
<tr>
<td>Model</td>
<td>18.6±4.7</td>
<td></td>
<td>48.2±9.7</td>
</tr>
<tr>
<td>danggui blood-enriching oral liquid</td>
<td>3.3</td>
<td>22.8±3.6</td>
<td>56.6±10.2**</td>
</tr>
<tr>
<td>(Huai)rehmannia polysaccharides</td>
<td>200</td>
<td>20.2±3.2</td>
<td>56.2±5.4**</td>
</tr>
<tr>
<td>100</td>
<td>31.6±5.8**</td>
<td></td>
<td>58.9±8.7**</td>
</tr>
<tr>
<td>50</td>
<td>34.8±6.2**</td>
<td></td>
<td>56.7±116**</td>
</tr>
</tbody>
</table>

Compared with the model group: * means P<0.05 ** means P<0.01

As shown in table 3, compared with the blank group, the thymus thickness of the model group was significantly thinner and the number of thymic cortical lymphocytes was significantly reduced (P<0.01). Compared with the model group, (Huai) rehmannia polysaccharide in the high-dose group significantly increased the number of cortical lymphocytes in rats with blood deficiency (P<0.01). The thickness of thymus and the number of cortical lymphocytes were significantly increased in the middle and low dose groups (P<0.01).

Effect of (Huai) Rehmannia Polysaccharides on Spleen Tissue in Rat Model of Qi and Blood both Deficiency

Effect of (Huai) rehmannia polysaccharides on spleen tissue: In the blank group, the red and white pulp of spleen was clearly demarcated, and the spleen nodules were large and the cells were dense. In the model group, the border of red and white pulp of the spleen was not clear, the splenic nodules were significantly reduced, and the cell density was significantly reduced. In the danggui blood-enriching oral liquid group, the red and white pulp of the spleen was clearer, the intrapleural nodules were not significantly enlarged, and the cell density was higher than that of the model group. In the high-dose group of (Huai) rehmannia polysaccharides, the spleen nodules were significantly reduced and lymphocytes were sparse. In the medium dose group of (Huai) rehmannia polysaccharides, the spleen nodules were reduced and lymphocytes were sparse. In the small dose group of (Huai) rehmannia polysaccharides, the spleen nodules were not significantly reduced, and the linbai cells were not sparse.

Table 2. Effect of (Huai) rehmannia polysaccharides on spleen tissue in rat model of qi and blood both deficiency( X ± s ).

<table>
<thead>
<tr>
<th>Group</th>
<th>Dose (mg/kg)</th>
<th>Size of splenium (um)</th>
<th>Lymphocyte number</th>
</tr>
</thead>
<tbody>
<tr>
<td>Blank</td>
<td></td>
<td>26.3±2.4**</td>
<td>42.8±7.6**</td>
</tr>
<tr>
<td>Model</td>
<td>17.6±3.5</td>
<td></td>
<td>26.4±5.4</td>
</tr>
<tr>
<td>danggui blood-enriching oral liquid</td>
<td>3.3</td>
<td>22.8±3.6*</td>
<td>30.2±2.7*</td>
</tr>
<tr>
<td>(Huai)rehmannia polysaccharides</td>
<td>200</td>
<td>17.8±4.6</td>
<td>23.8±5.8</td>
</tr>
<tr>
<td>100</td>
<td>28.4±4.7**</td>
<td></td>
<td>47.3±10.2**</td>
</tr>
<tr>
<td>50</td>
<td>31.6±5.2**</td>
<td></td>
<td>49.6±12.4**</td>
</tr>
</tbody>
</table>

Compared with the model group: * means P<0.05 ** means P<0.01

As shown in table 4, compared with the blank group, the splenic nodule was significantly smaller and the number of spleen lymphocytes was significantly reduced in the model group (P<0.01). Compared with the model group, the medium and small doses of (Huai) rehmannia polysaccharides can significantly increase the splenic nodule and the number of splenic lymphocytes (P<0.01).

Summary

Dandelion has a long medicinal history, was contained in the New herbal of Tang Dynasty. Dandelion is widely used in folk medicine for the treatment of inflammation and skin diseases. In recent years, many studies have been made on its clinical applications and pharmacological. The recent studies indicate that dandelion has the pharmacological effects of protecting liver, gallbladder, resisting
stomach injury, anti-tumor, anti-virus, antibacterial, anti infection and anti free radicals, and gradually applied to the clinical. But compared to Chinese patent medicine, herbal drugs has great limitations in clinical practice to some extent. In the 2015 edition Chinese Pharmacopoeia records 41 kinds of dandelion Chinese patent medicine, can be used for the treatment of 50 kinds of diseases.

The following problems still exist in the spread and application of Chinese patent medicines containing dandelion. China has a vast territory, and the distribution of medicinal dandelion is limited, which affects its spread and application. The majority of domestic residents’ awareness of the dandelion stay in food hunger level, lack of the knowledge of medical treatment level. Just 12 kinds of dandelion Chinese patent medicine are made of 5 medicinal herbs, only occupy 29.2% which lead to the mechanism of dandelion is not clear. The material basis of efficacy is ambiguous, studies on the single active constituents of Taraxacum mongolicum is lack. Dandelion Chinese patent medicine is seldom used alone, and quite a half of it is combined with Western Medicine. There are only two kinds of topical preparations of dandelion Chinese patent Medicine.

In combination with the above questions, the following suggestions are made. Optimize the transport chain of traditional Chinese medicine in the process of transmission and application, so that the Chinese patent medicines are sold at all levels of pharmacies; Strengthen the importance of Chinese herbal medicines and Chinese patent medicines in education, so that promote their dissemination and popularization; Based on the theory of Chinese medicine, expand the study and application of dandelion Chinese patent medicine in external use; Try to apply dandelion Chinese patent medicine separately to clinical research, excluding the impact of Western medicine and other treatments. Above all, dandelion Chinese patent medicine has broad prospects for development and economic benefits, both in the field of medical, diet therapy, and education.

Acknowledgement
This research was financially supported by the Henan Provincial Production and Research Project (142107000039) and National international cooperation base (2016-151).

References


