THE MORPHOMETRIC CHARACTERISTICS OF THE PEYERS' PATCHES LYMPHATIC NODULES OF THE SMALL INTESTINE IN RATS AFTER ADMINISTRATION OF CYCLOPHOSPHAMIDE

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Abstract

The morphometric features of the Peyers' patches lymphatic nodules of the small intestine in rats after cyclophosphamid injection were studied. In the experimental group the length and width of the lymphatic nodules and germinal centers were smaller than those in the control group on the 7 and the 30 day with the most evident changes on the 30 day of the experiment. The morphometric characteristics in the experimental group as well as in the control one were identical by the 90 day. Basing on that we can assume these changes show adaptive abilities of the small intestine immune system after acute immunosuppression.

Key words: rat, small intestine, Peyer's patches, lymphatic nodules, cyclophosphamid, morphometric features.

Introduction

Today the problem of immunodeficiency states has become extremely important [1, 2]. The excessive influence of the factors which depress or stimulate the immune system causes the development of the immune deficiency. The combined influence of toxic substances characterizes most of cases in general. The such substances can accumulate in products both of phytogenic and animal origin [3]. Considering the fact that the intestine directly communicates with a great number of various nutrients, coming from environment and serves as an entrance for many pathogens, mucous associate lymphoid tissue (MALT) plays a very important part in general and local immune protection [4-7]. Cyclophosphamid, commonly used as an anti-neoplastic drug, can cause adverse side-effects including immunotoxicity, urotoxicity and immunosuppression [8]. Immunosuppression is a state of temporary or permanent immunity dysfunction and can make organism more sensitive to pathogens due to the damage of immune system [9]. Thus, the aim of the research was to study the morphometric peculiarities of the Peyers' patches lymphatic nodules of small intestine in rats after cyclophosphamid injection.
Methods

The study was carried out on 36 white mature male rats. Animals were divided into 2 groups. The rats of Group 1 were injected cyclophosphamide intramuscularly at a dose of 200 mg/kg body weight. The rats of Group 2 were out of toxic influence. The rats were taken out of the experiment on 7, 30 and 90 day after cyclophosphamide injection. The formalin-fixed, paraffin-embedded blocks obtained from all rats were performed for routine histopathological examination. The specimens of small intestine (5-6 microns) were stained with hematoxylin-eosin and examined by automated morphometric complex comprising a light microscope Olympus CX41, a digital camera and a personal computer with a set of application programs. Using a licensed program "Morpholog" the height and width of the lymphatic nodules and germinal centers were measured [10]. The resulting digital data were processed by using the program “STATISTICA”.

Main part

In the intact Group the patches surface was smooth and facing into the intestinal lumen. The adjacent areas were covered with villi, which partially closed its surface. One row of the lymphatic nodules was found in the lamina propria and submucosa. Germinal centers were predominantly located at the base of lymphatic nodules (Fig. 1a, b).

Figure-1. The lymphatic nodules of the Peyers' patches in the small intestine (a - on the 30 day after the cyclophosphamide injection, b – intact rats): 1 - the dome of the lymphatic nodule, 2 - germinal center of lymphatic nodule, 3 - peripheral zone, 4 - internodular zone. Hematoxylin & Eosin. ×400.

The height and width of the Peyers' patches lymphatic nodules of the small intestine in Group 1 were smaller than those in Group 2 by 7.7% and 19.0% and the height and width of the germinal centers - by 21.6% and 15.0% respectively on the 7 day of the experiment (Table 1). The same tendency was observed on the 30 day of the experiment: the height and width of the lymphatic nodules decreased by 23.5% and 0.5%, the height and width of the
germinal centers - by 30.5% and 1.8% respectively. The morphometric parameters of the lymphatic nodules of Group 1 became equal to these in Group 2 on the 90 day of the experiment.

**Table 1: The morphometric parameters of the Peyers' patches lymphatic nodules of the small intestine in the mature rats of Groups 1 and 2. M±m (n=36).**

<table>
<thead>
<tr>
<th>Parameters</th>
<th>The morphometric parameters of the Peyers' patches lymphatic nodules (in microns)</th>
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<tbody>
<tr>
<td></td>
<td>7 day Group 1</td>
</tr>
<tr>
<td>Height of the lymphatic nodule</td>
<td>903.0±14.1*</td>
</tr>
<tr>
<td>Width of the lymphatic nodule</td>
<td>888.0±28.9*</td>
</tr>
<tr>
<td>Height of the germinal center</td>
<td>518.0±21.4*</td>
</tr>
<tr>
<td>Width of the germinal center</td>
<td>627.0±9.6*</td>
</tr>
</tbody>
</table>

Note: * - significant differences from the control data if p<0.05.

Minimal measure of the morphometric parameters of the internodular zones in Group 1 in comparison with Group 2 were detected on the 30 day of the experiment. These changes prove that cyclophosphamide has a great influence on the T-dependent zone (internodular zone) and B-dependent zone (lymphatic nodules with germinal center) on the 7 and the 30 days of the experiment. Throughout the experimental period, the lymphatic nodules in rats of Group 1 preserved the main morphological and morphometrical features. However, the boundary between the germinal center and the peripheral zone of the nodules became indistinct on the 30 day. This phenomenon can be explained by cell quantity decreasing in the peripheral zone. Frequently enough there were identified Peyer’s patches lymphatic nodules of the small intestine with double domes (Fig. 2). These changes may be connected with the too large doses of cyclophosphamide.

Fig. 2. The dome of the lymphatic nodules in rats of Group 1 on the 30 day of the experiment: 1 - epithelium, 2 - lamina propria of the mucosa, 3 - lymphocytes. Hematoxylin & Eosin. ×600.
According to the data of S.Y.C. Rolph et al., T. Nakahara et al. the use of high-dose cyclophosphamide in clinical practice is based on the cytostatic effects of the drug, leading to inhibition of actively proliferating cells [11, 12]. Lymphocytes are the first to undergo the cyclophosphamide influence because they refer to the most proliferative type of cells [13]. It can explain mentioned above lesions of the Peyer's patches of the small intestine.

Conclusions

1. A high degree reactivity manifested in evident changes of morphometric characteristics of the Peyer’s patches lymphatic nodules of the small intestine was observed after cyclophosphamide injection (200 mg/kg).

2. The most significant changes of the morphometric parameters were detected on the 7 and 30 days of the experiment.

3. The usual morphometric parameters found at the latest stages of the experiment characterize the immune system ability for adapting to the acute immunosuppression.

References


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