

ABSTRACT

Historically pig is allocated to a group of animals which use certain parts of their small intestine to acquire a fully developed primary B cell repertoire. Development of such primary repertoire is independent on the antigen presence and resembles the primary lymphopoietic activity of avian bursa of Fabricius. However, some findings concerning the pig's alignment in the above mentioned group suggest otherwise. This graduation thesis is focused on the investigation of lymphocyte populations and subpopulations in the small intestine of germ-free and conventional piglets. The aim is to determine whether the percentage amounts of lymphocyte populations is dependent on the intestinal colonization. Using Flow Cytometry the significant differences between individual samples were assessed allowing us to conclude which parts of the small intestine could possibly be used for the development of B cell repertoire. Moreover, the status of isotype switching of B lymphocytes isolated from different intestinal parts was determined by the means of PCR analysis. Our data suggest that the small intestine colonization has a crucial role in development of all the main lymphocyte populations as well as some of their subpopulations. The greatest influence of colonization was observed concerning B lymphocytes and their subpopulations. Oppositely, significant differences of lymphocyte distribution of individual parts of the small intestine were not discovered. Though we can say that noticeable deviations occur in the region of terminal ileum. These deviations are likely to be caused by the highly probable interaction of lymphocytes and bacteria of the large intestine. Our data also impose the predominance of $\alpha\beta$ T lymphocytes in *lamina propria* and, in contrary, the predominance of $\gamma\delta$ T lymphocytes in the intestinal epithelium. In agreement with general findings B lymphocytes in both *lamina propria* and epithelium occur after colonization. Analysis of B cell genome and transcripts regarding the Ig isotypes suggests that while there is a change of Ig isotype in Peyer's patches by the means of alternative splicing, the class-switch in *lamina propria* occurs as secondary genome rearrangement. We speculate that B lymphocytes begin the class-switch process in Peyer's patches using alternative splicing and then migrate to *lamina propria* where they settle as IgA^+IgM^- B cells and finish the DNA rearrangement.

Keywords

Pig, intraepithelial lymphocytes, *lamina propria* lymphocytes, Peyer's patches, distribution of lymphocytes, class-switch