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Influence of Colostral Growth Factors on Bone Marrow Colony-Formation *in Vitro*

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Bovine colostrum has been shown to contain an array of growth factors and hormones influencing the growth and recovery of numerous cells and tissues. The aim of the present study was to investigate the influence of bovine colostrum on colony-forming ability of murine bone marrow cells.

Bone marrow cells were isolated from Balb/c mice and tested in semi-solid agar cultures for colony formation. Bovine colostrum was added to the cultures at concentrations of 0.1, 1 and 10%.

Bovine colostrum at concentration of 1% showed very good colony-forming ability. It stimulated the formation of erythroid and myeloid bone marrow cell colonies. As controls we used cultures containing epithelial growth factor (EGF) and fibroblast growth factor (FGF) at concentrations similar to those found in the bovine colostrum. The latter growth factors did not promote significant colony-forming ability of murine bone marrow cells.

Key words: bone marrow, colostrum, epithelial growth factor, fibroblast growth factor, colony formation.

Introduction

Colostrum is the milk produced during the first few days after parturition. It is a complex fluid rich not only in nutrients but also with high level of bioactive components: immunoglobulins, growth factors, growth hormone, lactoferrin, lysozyme, lactoperoxidase, cytokines, vitamins [2]. Lactoferrin, lactoperoxidase, lysozyme and immunoglobulins provide immunity transfer to the newborn and growth factors's control of some fundamental life processes such as cell division, differentiation and apoptosis.

The major peptide growth factor constituents of bovine colostrums are: transforming growth factor α and β (TGF- α and TGF- β), insulin-like growth factors I and II (IGF I and IGF II), epithelial growth factor (EGF), platelet-derived growth factor (PDGF), bovine colostral growth factor (BCGF) and several other peptides which structure and function are less clearly defined.

During the last two decades interest in beneficial physiological effects of bovine colostrum and the possibility to utilize its components have increased. Its role in eliminating infection and stimulating growth and development of neonatal organs and tissues was widely discussed [1, 5, 6]. However, the effect on adult health after applying some

of these components has not been thoroughly examined and its value in the prevention and treatment of adult injury is less explored.

The aim of the present study was to investigate the influence of bovine colostrum on colony-forming ability of murine bone marrow cells.

Materials and Methods

The bone marrow cells were isolated from murine femur bones by flushing with RPMI 1640 medium and the agar cultures were prepared after Z v et k o v a et al. [7]. Bovine colostrums was added to the cultures at concentrations of 0.1, 1 and 10 %. As controls we used cultures containing EGF at concentration of $3\mu g$ /ml and 3T3 culture medium – rich in FGF. The cultures were grown for 14 days at 37°C, 5%CO₂. After drying they were stained with eosin-methylene blue. The colonies were observed under light microscope.

Results

All the applied concentrations of bovine colostrums showed colony-formation activity most pronounced at 1%. The bovine colostrums promoted the growth of huge number of small erythoid (Fig. 1) and myeloid (Fig. 2) colonies. At day 14^{th} we observed significant number of relatively big myeloid (Fig. 3) and erythroid (Fig. 4) colonies. The cultures stimulated with recombinant EGF (Fig. 5) did not show any colony-forming ability. FGF activated the bone marrow cells only to a certain extent, so they formed small colonies for 14 days.



Fig. 1. Small bone marrow myeloid colonies. Originally $\times 150$



Fig. 2. Small bone marrow erythroid colonies. Originally \times 150



Fig. 3. Big bone marrow myeloid colonies. Originally \times 150



Fig. 4. Big bone marrow erythroid colonies. Originally $\times 150$



Fig. 5. Influence of EGF on bone marrow colony formation. Originally $\times\,150$

Discussion

The colostrums and milk provide a link between the mother and the newborn in the extrauterine environment in a manner similar to the placental links between mother and fetus in utero. They help to prevent age-related diseases affecting the gastrointestinal tract during the newborn period. Colostrum and milk contain factors that may be missing in inherited diseases of inborn errors in metabolism and provide passive protective factors that lessen the expression of neonatal allergic and infectious diseases. In some instances, by providing the missing factor in an inherited disease, the newborn may be protected from serious damage even to its developing brain [4]. Colostrum feeding affects the cell proliferation rates of B lymphocytes in fillicles of Payer's patches, follicular associated epithelium and interfollicular areas of Payer's patches. It also affects the cell proliferation in the thymus [3]. These effects of colostral growth factors gave us ground to test their influence on colony forming ability of the precursors of the lymphoid cells – bone marrow cells. Our results showed that unseparated colostrum possesses colony forming ability and stimulated big as well as small colonies of erythroid and myeloid origin. One of the growth factors contained in colostrums, EGF, was unable to exerted such stimulation. The FGF stimulated but only to a certain extent the colony formation of murine bone marrow cells. We may therefore conclude that the colony forming ability of bovine colustrum to other but not the above tested growth factors which are our further task to separate and test.

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