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ADHESIVE PROPERTIES OF PROBIOTIC STRAINS OF LACTOBACILLI IN VITRO ON THE MODELS OF EPITHELIAL CELLS AND ERYTHROCYTES

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One of the main properties of probiotic strains that contribute to their widespread use for the prevention of diseases and treatment of patients with various infectious-inflammatory diseases is that they provide colonization resistance, i.e. the ability to defense walls of various tracts and organs from bacteria and toxic products of various origins [1]. An important role in the implementation of colonization resistance is played by the ability of microorganisms to adhesion, i.e. the colonization of a particular biotope of the body [2-3].

To study the prospects of potential probiotic strains of lactobacilli, it is recommended to examine their adhesive properties – the ability to attach on epithelial cells *in vitro*. This indicator is one of the most important criteria for the selection of potentially probiotic strains for further creation of probiotic preparation for oral and intravaginal use [2]. There are different methods of study of probiotic strains adhesive properties, the most common research technique is studying adhesiveness to epithelial cells [4]. This method is rather complicated in execution and involves pre-cultivation of cells. A more simple method of investigation of adhesiveness of the strains associated with the use of red blood cells, because pre-cultivation of erythrocytes is not required.

Therefore, the aim of this work was to compare two methods of research of adhesion properties of *Lactobacillus acidophilus* IMV B-7279, *L. casei* IMV B-7280 and *L. delbrueckii* subsp. *bulgaricus* IMV B-7281 potentially probiotic strains *in vitro* using epithelial cells and erythrocytes. *L. acidophilus* IMV B-7279, *L. casei* IMV B-7280 and *L. delbrueckii* subsp. *bulgaricus* IMV B-7281 were allocated from the associated culture during laboratory studies of the fermented biological material from the intestines of humans. The study was performed using bacteria lyophilized in Cuddon Freeze Dryer FD1500 (New Zealand). Before each experiment the viability of lyophilized strains were tested by monitoring their growth on the Man-Rogosa-Sharpe agar media at 37 °C for 24-48 hours.

The adhesive properties of the strains were studied according to the Brilis V.I. et al (1986) method using erythrocytes from 0(I) Rh+ group of human blood and human buccal epithelial cells [5]. The following indicators were determined: the average adhesion index (AAI) – the average number of microbial cells that attached to the surface of one erythrocyte (epitheliocyte); the participation rate of erythrocytes (epitheliocytes) (EPR) – the percentage of cells containing microorganisms on their surface; the index of adhesiveness of microorganisms (IAM) – the average number of microbial cells that attached to the surface of one erythrocyte (epitheliocyte). IAM was calculated according to the formula: IAM = (AAI*100)/EPR. Microorganisms were considered as non adhesive when IAM ≤ 1.75 standard units, low adhesive – when IAM from a 1.76 to 2.5 standard units, medium adhesive – when IAM from 2.51 to 4.0 standard units, high adhesive – when IAM > 4.00 standard units.

The obtained data showed that SPA index on epithelial cells and red blood cells for *L. acidophilus* IMV B-7279 (2.25 ± 0.10 and 2.04 ± 0.19 respectively) and *L. delbrueckii* subsp. *bulgaricus* IMV B-7281 (1.98 ± 0.06 and 2.08 ± 0.22 respectively) had no significant

differences, whereas *L. casei* IMV B-7280 in the study of adhesive properties to epithelial cells and erythrocytes had different SPA index (6.83 ± 0.27 and 4.40 ± 0.30 respectively). No significant differences have been identified when comparing the adhesive properties of all studied strains to epithelial cells and erythrocytes by the EPR index. Analysis of the ability of strains to attach to epithelial cells and erythrocytes by IAM has established a reliable difference in the values for *L. casei* IMV B-7280 (7.81 ± 0.86 and 4.78 ± 0.47 standard units on epithelial cells and erythrocytes respectively). However, according to the IAM criteria, *L. casei* IMV B-7280 strain had a high, *L. delbrueckii* subsp. *bulgaricus* IMV B-7281 – medium, and *L. acidophilus* IMV B-7279 – low adhesive activity based on the results of both studies – on epithelial cells and erythrocytes.

By the results of our research we can make a conclusion that the use of both methods of determination of adhesive properties of potentially probiotic strains give similar results and therefore can be used in the primary researches of microorganisms *in vitro* in the search of bacteria for the subsequent probiotic preparations creation on their basis. It is known that red blood cells contain substances on their surface that are identical to epithelial cells, therefore the method of determining strains adhesive properties on erythrocytes can probably be used by researchers if it is impossible to work with epithelial cells.

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