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Дослідження життєздатності штамів *Escherichia coli* після тривалого низькотемпературного зберігання у кріозахисних середовищах

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Viability of *Escherichia coli* Strains After Long Term Storage in Cryoprotective Media

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Maintenance of bacterial strain collections are part of standardized microbiological laboratory's procedures for clinical, epidemiological, and scientific needs. Low-temperature preservation in presence of different cryoprotective compounds has been considered as an effective method for long term storage with high rate of microorganism survival after recultivation [Ning Guo, 2020; Anaïs Biclot, 2022].

This study aimed to evaluate the effect of using different cryoprotective media on viability of *E.coli* strains following long term low-temperature storage.

The study was conducted in 5 *E.coli* clinical strains. Two cryoprotective media were used: peptone-meat broth with 1.0% glucose and peptonemeat broth with 20% glycerol. Freezing was achieved by direct immersion of cryotube containing culture suspension (10^9 CFU/ml in 1.0 ml of cryoprotective medium) to liquid nitrogen (−196°C). Viability testing was assessed by Koch's method after 24 and 36 months of storage. Statistical analysis of the results was performed using the software package STATISTICA 6.1 (StatSoft).

The results of *E.coli* viability testing showed that the cryoprotective medium with glycerol yielded higher survival rates: (82.4 ± 5.3)% after 24-month and (74.6 ± 5.0)% after 36-month storage. Survival rates of cultures suspended in media with 1.0% glucose was (23.3 ± 4.7)% and (18.0 ± 2.9) % respectively ($p < 0.05$). Accordingly, in comparison with medium with glucose, medium with glycerol has protective effect nearly 3 times higher after 24-month and nearly 4 times higher after 36-month storage.

Suitability of cryopreservation as a method for bacterial strain collection long-term storage has been confirmed. Survival rates depend on composition of cryoprotective medium. Glycerol was found to be the optimal cryopreserving compound for freezing *E.coli* strains.

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Вплив ядерних клітин кордової крові людини на показники імунної системи тварин з атопічним дерматитом

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Influence of Nucleated Cells of Human Cord Blood on Indices of Immune System of Animals with Atopic Dermatitis

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Atopic dermatitis (AD) is classified as a chronic inflammatory disease characterized by the appearance of irritation and rashes on the dermis. Attempts to create new methods of treating AD have led to the development of a fundamentally new strategy for the therapy of this pathology with preparations of human cord blood – HCB.

The aim of the work is to experimentally substantiate the possibility of using cryopreserved (cNC HCB) and lyophilized nucleated cells of human cord blood (lNC HCB) for the purpose of correcting the state of the immune system of animals with induced autoimmune pathology in the form of AD.

The experiments were performed in 6-month-old Wistar rats, weighing 180–200 g. The AD was initiated by rubbing a 5% alcohol-acetone solution of dinitrochlorobenzene into the skin of the back (3x3 cm^2) for 21 days. Rats were divided into groups: 1 – healthy (control); 2 – AD; 3 – AD + prednisolone; 4 – AD + cNC HCB 0.5 ml each, 5×10^6 cells; 5 – AD + lNC HCB in the same volume. On the 15th day in rats with AD and after treatment with cNC HCB, lNC HCB in the spleen, the phenotypic characteristics of cells were determined using Mab to CD3, CD4, CD8, CD16, CD25 antigens. Histological examination of the spleen and lymph nodes was performed.

On the 15th day after application of a neutralizing dose of allergen in rats with AD, in comparison with a group of healthy animals, a decrease in the number of all studied subpopulations of T-lymphocytes, natural killer cells ($CD16^+$) was observed against the background of an increase in the number of T-reg cells. After introduction of cNC HCB or lNC HCB, the restoration of the content of T-lymphocytes and $CD16^+$ cells was observed. In AD, the predominance of white pulp over red pulp was observed in the spleen, and in lymph nodes – the predominance of follicles of the II and III stages of development. The positive corrective effects of cNC HCB and lNC HCB regarding the morphological characteristics of lymphohematopoietic complex were shown.

The use of lNC HCB and cNC HCB is possible for stimulation of skin regeneration processes and immunomodulation of the immune system under conditions of AD development.