THE EFFECT OF GLUCOSE ON THE SURVIVAL AND METABOLIC PLASTICITY OF LEWIS LUNG CARCINOMA CELLS DURING THEIR ADHESIVE AND DE-ADHESIVE GROWTH

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High proliferative activity of tumor cells requires a sufficient amount of energy and plastic substrates, in particular glucose [1]. Intensive consumption of this metabolite by tumor cells leads to its depletion in the intercellular environment and creates an uneven distribution of glucose in the tumor [2-3]. As a result, some cells can survive and adapt to glucose deficiency by reprogramming their metabolism and/or by migrating to the vasculature. Tumor cells that lose adhesive contact, become resistant to anoikis and acquire metastasis potential [4]. The ability to undergo such reprogramming may significantly depend on the characteristics of the deficient microenvironment preceding the transition of tumor cells from an adhesive to a de-adhesive state.

The work aimed to investigate the impact of glucose at different concentrations on the survival and metabolic plasticity of Lewis lung carcinoma (LLC) cells during their adhesive and de-adhesive growth.

Methods. High-metastatic variant of Lewis lung carcinoma (LLC) cells was used. The methods of cell biology, experimental oncology, biochemical, cytological, flow cytometry, optical spectrophotometry, and mathematical and statistical methods were used. Comparative studies were conducted on the proliferative potential, distribution of tumor cells by cell cycle phases, the level of apoptosis, the rate of glucose consumption, and analysis of the cytological structure of LLS cells during their adhesive and de-adhesive growth in a glucose-non-deficient (11 mM) medium, which was preceded by a one-day incubation of these cells with glucose concentration in it of 0.0 mM, 2.5 mM, and 11 mM.

Results. The conducted studies showed that on the first day of adhesive growth
of LLC cells, the number of cells incubated in a medium with a starting glucose concentration of 11 mM was statistically significantly higher than the corresponding indicator of cells incubated in a medium with 2.5 mM glucose. On the second and third days, the rate of cell growth in the non-deficit medium increased significantly, which led to an increase in the number of cells by 35-40% (p<0.01). The rate of glucose absorption progressively decreased in the process of adhesive growth of cells, and it was significantly more pronounced at the starting glucose concentration of 11 mM. Despite the significantly different starting levels of glucose in the incubation medium, the dynamics of changes in glucose concentration were practically the same.

Comparative studies of the proliferative potential and intensity of glucose metabolism of LLC cells during their adhesive and de-adhesive growth in a glucose-deficient environment showed no significant difference in the number of cells, although a tendency to decrease the number of cells during de-adhesive growth was observed.

It was shown no differences in the cell cycle phase distribution of tumor cells on the 3rd day of LLC cells adhesive growth after a one-day incubation in a medium with a glucose content of 0 mM, 2.5 mM, and 11.0 mM. However, during the de-adhesive growth, the percentage of LLC cells in the G2 and S phases of the cell cycle incubated in medium with 0 mM and 2.5 mM was significantly different from the corresponding indicators of cells incubated with 11 mM glucose. Notably, there were no significant differences in the distribution of cells incubated in a medium with 0 mM compared to 2.5 mM glucose. It is worth noting that the transition from one-day adhesive growth of LLC cells in 2.5 mM glucose medium to de-adhesive growth in 11 mM glucose resulted in statistically significant (p<0.05) changes (compared to corresponding indicators during adhesive growth): a 33% decrease in the number of viable cells, a 12% decrease in the percentage of cells in the G1 phase of the cell cycle, an almost two-fold increase in the percentage of cells in the G2 phase, a six-fold increase in the percentage of apoptotic cells, a 22% decrease in the number of round cells, a 48.1% increase in the number of elongated cells, including large multinucleated cells appear (2.1%), and an almost two-fold increase in the number of multicellular spheroids.

**Conclusions.** 1. It has been demonstrated that the proliferative activity of Lewis lung carcinoma cells during both adhesive and de-adhesive growth is significantly dependent on the glucose concentration in the incubation medium: higher initial concentration results in more intense tumor cell growth. However, regardless of the different starting levels of glucose in the incubation medium, the dynamics of glucose uptake by the cells are practically the same.

2. It was found that in the absence of glucose influence at the investigated concentrations on the distribution of cells across the phases of the cells cycle during adhesive growth of LLC cells, during de-adhesive growth the percentage of cells in the G2 and S phases, which were incubated in medium with 0 mM and 2.5 mM glucose was significantly different from the corresponding indicators of cells incubated with 11 mM glucose.

3. During the transition from the adhesive growth of LLC cells with a glucose level of 2.5 mM to a de-adhesive growth in the incubation medium with a glucose
level of 11 mM, there is a decrease in the number of viable cells due to apoptosis, changes in cycle phase distribution resulting in a decrease in the percentage of cells in G1 phase and an increase in the percentage of cells in the G2 phase, as well as cytological changes in tumor cells, which is manifested in a decrease in the number of small round cells and an increase in the number of elongated cells of various sizes and spheroids.

References: