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Selectivity studies of the drug fs-1 in mixture of eukariotic and microbial cells

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Abstract

A comparative study of the degree of influence of a drug being developed on microbial and eukaryotic cells is one of the important stages in the drug development.

This study examined the selective effect of a novel drug FS-1 on bacterial cells in the presence of eukaryotic cells.

The drug FS-1 is a nanocomplex of iodine and lithium and magnesium halides with dextrin and polypeptides, the molecular weight of dextrin enables penetration of active centers of the FS-1 molecule into the cells. Mycobacterium smegmatis ATCC 607 was used as a model system.

Data were obtained that indicate the effect of FS-1 predominantly on bacterial cells.

Materials and Methods

Studies were performed on the MDCK cell line (canine kidney epithelial cells) and rapidly growing atypical mycobacterium, Mycobacterium smegmatis ATCC 607 (ATCC, USA). The preparation 131I-FS-1 was used at a concentration of 0.5 μg/mL. A combination of eukaryotic and prokaryotic cells was cultivated together in the medium for 1 hour in the presence of FS-1 labeled with the 131I isotope (experimental samples) and in the absence of the preparation (control samples). 4.5 mL of a suspension MDCK cell line (5×106 cells/mL) and 4.5 mL of M. smegmatis (2×109 CFU/mL) were taken for studies. DNA was isolated using the commercial PureLink Genomic DNA Mini Kit (Invitrogen, United States) according to the manufacturer's protocol. DNA content in the isolated samples and its purity were determined spectrophotometrically using the NanoDrop 2000 instrument (Thermo Scientific, USA). The radioactivity of the samples was measured with the HIDEX 300SL β-spectrometer (HIDEX Oy, Finland) using MicroWin software, with further conversion to the amount of DNA. The measurement parameters used were 500 keV and measuring time of 5 min. To obtain background values, the natural radioactivity of control DNA samples was determined using a β-spectrometer. The values obtained by measuring the background samples were subtracted from the parameters of the experimental samples. The data obtained from studies were evaluated with GraphPad Prism software version 6.0 using the non-parametric One-way ANOVA method and ranking criteria. P- values higher than 0.05 ($p > 0.05$) were considered non-significant.

Results and Discussion

The quantitative DNA content ranged from 72.1 to 98.4 ng/mL in samples isolated from the MDCK cell line, from 102 to 123.8 ng/mL DNA in samples isolated from the bacterial cell. The purity index of the isolated samples varied in the 1.8-2.2 range, which gave evidence of sufficiently high degree of purification of DNA molecule from impurities.

As a result of studying the effect of FS-1 on the genomic DNA of a pathogen and a eukaryotic cell under their combined exposure by the isotope analysis method, it was established that the specific radioactivity of bacterial DNA samples was 27 times higher than that of DNA isolated from eukaryotic cells (MDCK).

The concentration of 131I-FS-1, used in the experiment (0.5 μg/mL) was 0.25 MBC for Mycobacterium smegmatis and 0.0025 CC50 for eukaryotic cells.

Since FS-1 does not possess genotoxic properties and does not damage DNA of eukaryotic cells, the specific radioactivity of DNA may indicate the formation of a labile donor-acceptor bond between FS-1 and DNA of eukaryotic cells. At the same time, the high specific radioactivity of DNA of bacterial cells indicates a more profound effect on DNA in bacteria, leading to a sharp decrease in their viability.

Biography:

In 2008 he graduated from the Al-Farabi Kazakh National University, Faculty of Biology, Department of Genetics and Molecular Biology, Master's degree in Biology. My general scientific experience exceeds 12 years. The primary research interests are focused on cell and molecular biology. I am currently working as a Senior Researcher at the Scientific Center for Anti-Infectious Drugs, Immunology Laboratory. The number of published papers is over 13.

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