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Redox regulation of the effect of antitumor drug doxorubicin in HEp-2 human larynx carcinoma cells

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Tumor cells chemoresistance is one of the main challenges for modern antitumor therapy. The investigation of mechanisms of tumor cells respond to stress and the development of methods for overcoming their resistance is important to developing novel therapeutic approaches for cancer treating. Previously in our work, redox regulation of chemoresistance by phenolic antioxidants was studied. It was found that one of the key mechanisms responsible for the formation of tumor cell resistance is the attenuation of apoptosis through increase of redox buffering capacity [1]. In the present study the effects of *para*-benzoquinones, known as effective redox regulators, on antitumor effect of doxorubicin were examined.

HEp-2 cells were cultivated in DMEM supplemented with 8–10% fetal bovine serum at 37°C in 5% CO_2 atmosphere. To determine the effect on the proliferative activity, the compound was added 24 h after the cell passage. Cell counts were determined after 3 days of cultivation. Changes in the mitochondrial membrane potential were monitored using tetramethylrhodamine ethyl ester; the direction of changes was determined using carbonyl cyanide 3-chlorophenylhydrazone. The study also employed inhibition assay (antimycin A – inhibitor of ubiquinol : cytochrome *c* oxidoreductase, cyclosporin A - inhibitor of mitochondrial permeability transition pores assembly).

The combined effect of *para*-benzoquinones and antitumor agent doxorubicin on the proliferative activity of tumor cells was studied. Thymoquinone in non-toxic doses was found to enhance the effect of antitumor drug doxorubicin. The dependence of the effect obtained on a time interval between addition of the quinone and doxorubicin was investigated. It has been established that the most pronounced effect of enhancing the action of doxorubicin by thymoquinone is observed with simultaneous addition of compounds to cell culture. It was shown that 1,4-benzoquinone in non-toxic doses does not cause an increase in the antitumor effect of doxorubicin. To determine the possible mechanisms of thymoquinone effect on doxorubicin action, studies of changes in the mitochondrial membrane potential while adding the compounds have been carried out. It was previously shown that

thymoquinone causes a dose-dependent decrease in mitochondrial membrane potential in a suspension of tumor cells [2]. Doxorubicin has also been found to cause a dose-dependent decrease in mitochondrial potential of tumor cells. After the addition of thymoquinone, a dose-dependent decrease in mitochondrial potential under the action of doxorubicin was not observed. These results suggest that mechanisms of thymoquinone and doxorubicin effects on tumor cells include several common pathways and presumably include participation of mitochondria.

References

1. Mechanisms of redox regulation of chemoresistance in tumor cells by phenolic antioxidants / Martinovich G.G. [et al.] // Biophysics. 2017. Vol. 62. № 6. P. 942-949.
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