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# INCREASED EXPRESSION LEVEL OF ADAPTOR PROTEIN RUK/CIN85 IN DOXORUBICIN-RESISTANT HUMAN NON-SMALL LUNG ADENOCARCINOMA MOR CELLS IS ASSOCIATED WITH THEIR METABOLIC REPROGRAMMING

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One of the most lethal types of cancer in the world is non-small-cell lung carcinoma (NSCLC). This is due to diagnosing of lung cancer mainly at advanced stages associated with increased resistance of cancer cells to traditional anticancer chemotherapeutic drugs as well as their high metastatic potential [1]. We previously showed that overexpression of the adaptor protein Ruk/CIN85 in breast cancer cells is associated with their malignancy and increased chemoresistance [2, 3]. To promote survival under the treatment with anti-cancer drugs, cancer cells dynamically change their metabolism.

*Aim*. The purpose of the present study was to find out the role of Ruk/CIN85 in modulation of activities/content of key enzymes/components of glycolysis and hydrogen peroxide using as a model human NSCLC MOR wild type and resistant to drugs MOR/0.2R cells.

*Materials and Methods*. MOR (ECACC 84112312) and MOR/0.2R (ECACC 96042335), drug-resistant cell line, were cultured in the complete RPMI medium under standard conditions. Enzymes activity, content of metabolites and protein in cell extracts and the conditioned cell culture medium were estimated by spectrophotometric and fluorometric assays.

*Results and Discussion*. First of all, by using $RT^2$-PCR it was revealed that the level of Ruk/CIN85 mRNA in drug-resistant MOR cells was 10 times higher than in parental MOR cells. As can be seen from Fig. *A, B*, the high expression level of Ruk/CIN85 in MOR/0.2R cells was related to the increased activity/content of markers of aerobic glycolysis (known as the Warburg effect), lactate dehydrogenase A (LDHA) and lactate as well as marker of chemoresistance of cancer stem cells, aldehyde dehydrogenase (ALDH), as compared to control. The activities of lysyl oxidase (LOX) and diamine oxidase (DAO) were also significantly higher in resistant cells (Fig. *C*). It has been shown that these enzymes are associated with aggressiveness of tumor cells. In particular, the activity of LOX is related to the increased degree of stiffness of the extracellular matrix, while the activity of DAO is involved in the control of cell proliferation [4, 5]. Significant increase in the level of hydrogen peroxide in the MOR/0.2R cells (Fig. *D*) may be, at least partially, associated with increased activities of amine oxidases studied as one of the products of the enzymatic reaction. Based on the obtained results, we draw a conclusion that observed changes in the intensity of glycolysis, amine oxidases activities and content of hydrogen peroxide in doxorubicin-resistant MOR/0.2R cells positively correlate with the expression level of the adaptor protein studied.

*Conclusions*. In conclusion, it can be assumed that the adaptor protein Ruk/CIN85 is involved in metabolome reprogramming and may function as an important component of regulatory networks required for the acquisition of drug resistant phenotype by NSCLC cells.

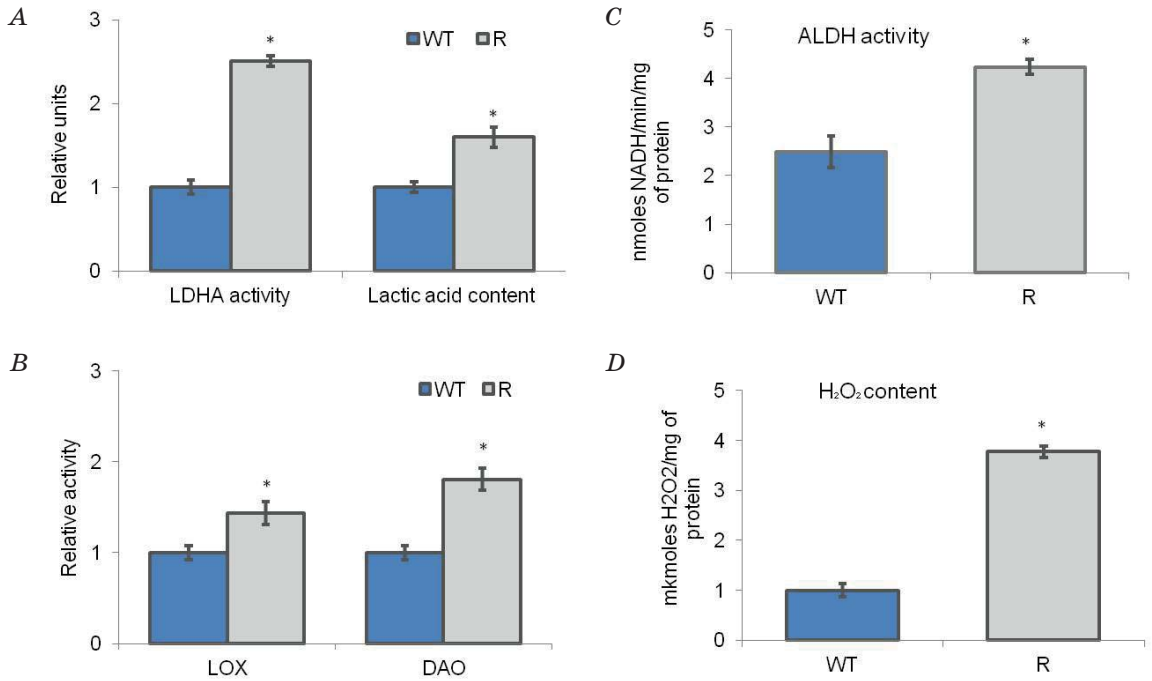


*Figure.* **Increased level of adaptor protein Ruk/CIN85 in MOR/0.2R cells positively correlates with metabolic parameters that potentially ensure their chemoresistance:**
$M \pm m$, $n = 3$, * — $P < 0.05$ compared to the corresponding control

***Key words:*** non-small-cell lung carcinoma (NSCLC); adaptor protein Ruk/CIN85; chemoresistance; metabolome reprogramming; metabolic enzymes; hydrogen peroxide.

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