

GRP78/BiP overexpression in P-glycoprotein-positive L1210 cells is responsible for resistance to tunicamycin-induced endoplasmic reticulum stress

Mário Šereš^{1,*}, Lucia Pavlíková¹, Viera Boháčová¹, Tomáš Kyca¹, Ivana Borovská¹, Boris Lakatoš², Albert Breier^{1,2,*} and Zdena Sulová^{1*}

¹ Institute of Molecular Physiology and Genetics, Centre of Biosciences, Slovak Academy of Sciences, Dúbravská cesta 9, 84505 Bratislava, Slovakia

² Institute of Biochemistry and Microbiology, Faculty of Chemical and Food Technology, Slovak University of Technology in Bratislava, Radlinského 9, 81237 Bratislava, Slovakia.

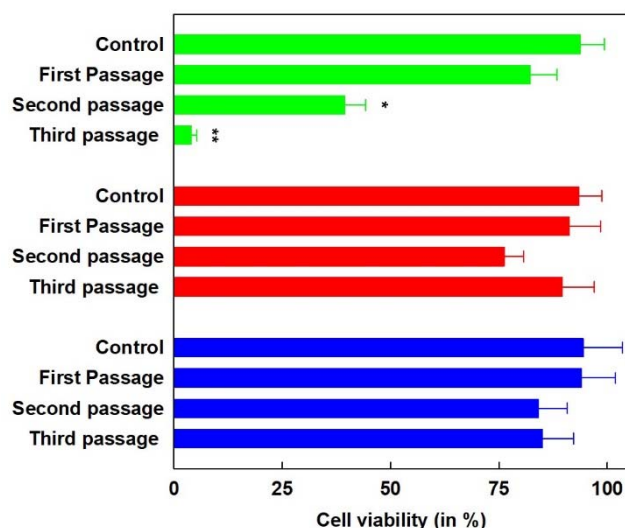


Figure S1. Cell viability of S, R and T cells after 1-3 passage in the presence of tunicamycin (0.1 μ M) by using both propidium iodide and FITC-annexin V staining. The bars represent the mean \pm S.E.M. (of three independent measurements) of the percentage of cells that were not stained with either propidium iodide or FITC-annexin V. S cells: green; R cells: red and T cells: blue. Significance: Data differ from control at the levels: *-P<0.02; **-p<0.01.

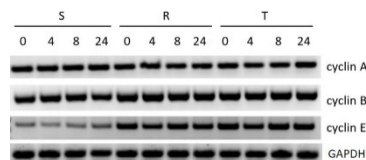


Figure S2. RT-PCR detection of cyclin A, B1 and E expression in S, R and T cells after 0, 4, 8 and 24 hours of incubation in medium containing 0.1 μ M tunicamycin. GAPDH was used as an internal control. Data are representative of three independent measurements.

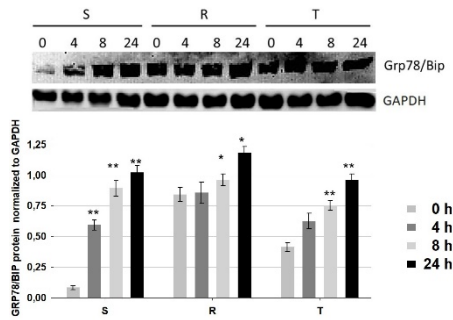


Figure S3. Levels of GRP78/BiP proteins in S, R and T cells after incubation of cells in medium containing 0.1 μ M tunicamycin for 0, 4, 8 and 24 hours. Data are representative of three independent measurements. Protein bands were quantified by densitometry and data were expressed as means \pm S.E.M from three independent measurements. Significance: Data differ from values obtained in cells that were not incubated in the presence of tunicamycin at the levels: *- $p < 0.02$; ** - $p < 0.01$.

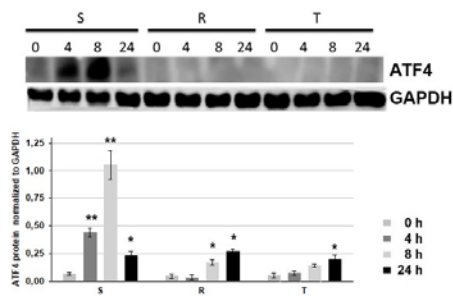


Figure S4. Levels of ATF4 proteins in S, R and T cells after incubation of cells in medium containing 0.1 μ M tunicamycin for 0, 4, 8 and 24 hours. Data are representative of three independent measurements. Protein bands were quantified by densitometry and data were expressed as means \pm S.E.M from three independent measurements. Significance: Data differ from values obtained in cells that were not incubated in the presence of tunicamycin at the levels: *- $p < 0.05$; ** - $p < 0.01$.

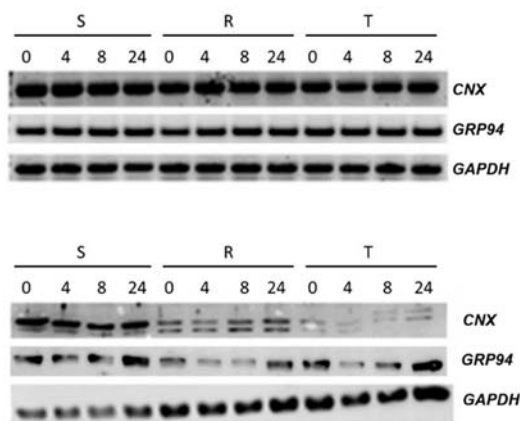


Figure S5. Expression of calnexin and GRP94 using RT-PCR (upper) and Western blotting (lower) in S, R and T cells after incubation of cells in medium containing 0.1 μ M tunicamycin for 0, 4, 8 and 24 hours. Data are representative of three independent measurements.