development. Mol. Cell. Biol. V. 26. P. 789. https://doi.org/10.1128/MCB.26.3.789-809.2006

Kim J., Tchernyshyov I., Semenza G., Dang, C. 2006. HIF-1-mediated expression of pyruvate dehydrogenase kinase: a metabolic switch required for cellular adaptation to hypoxia. Cell Metab. V. 3. P. 177. https://doi.org/10.1016/j.cmet.2006.02.002

Li M., Zhou Z.P., Sun M., Cao L., Chen J., Qin Y.Y., Qin Z.H. 2015. Reduced nicotinamide adenine dinucleotide phosphate, a pentose phosphate pathway product, might be a novel drug candidate for ischemic stroke. Stroke. V. 47. P. 187. https://doi.org/10.1161/STROKEAHA.115.009687

Lukyanova L., Kirova Y. 2015. Mitochondria-controlled signaling mechanisms of brain protection in hypoxia. Front. Neurosci. V. 9. P. 320. https://doi.org/10.3389/fnins.2015.00320

Meijer T., Kaanders J., Span P., Bussink J. 2012. Targeting hypoxia, HIF-1, and tumor glucose metabolism to improve radiotherapy efficacy. Clin. Cancer Res. V. 18. P. 5585. https://doi.org/10.1158/1078-0432.CCR-12-0858

Semenza G. 2001. Hypoxia-inducible factor 1: oxygen homeostasis and disease pathophysiology. Trends Mol. Med. V. 7. P. 345. https://doi.org/10.1016/s1471-4914(01)02090-1

Sheldon R., Lee C., Jiang X., Knox R., Ferriero D. 2014 Hypoxic preconditioning protection is eliminated in HIF-1α knockout mice subjected to neonatal hypoxia-ischemia. Pediatr. Res. V. 76. P. 46. https://doi.org/10.1038/pr.2014.53

Spencer N.Y., Stanton R.C. 2017. Glucose 6-phosphate dehydrogenase and the kidney. Curr. Opin. Nephrol. Hypertens. V. 26. P. 43. https://doi.org/10.1097/MNH.0000000000000294

Stincone A., Prigione A., Cramer T., Wamelink M.M., Campbell K., Cheung E., Ralser M. 2014. The return of metabolism: biochemistry and physiology of the pentose phosphate pathway. Biol. Rev. Cambridge Philos. Soc. V. 90. P. 927. https://doi.org/10.1111/brv.12140

Sun Y., Chen X., Zhang X., Shen X., Wang M., Wang X., Liu W., Liu C., Liu J., Liu W., Jin X. 2017. β2-adrenergic receptor-mediated HIF-1α upregulation mediates blood brain barrier damage in acute cerebral ischemia. Front. Mol. Neurosci. V. 10. P. 257. https://doi.org/10.3389/fnmol.2017.00257

Tang B.L. 2019. Neuroprotection by glucose-6-phosphate dehydrogenase and the pentose phosphate pathway. J. Cell. Biochem. V. 20. P. 14285. https://doi.org/10.1002/jcb.29004

Vetrovoy O., Sarieva K., Galkina O., Eschenko N., Lyanguzov A., Gluschenko T., Tyulkova E., Rybnikova E. 2019. Neuroprotective mechanism of hypoxic post-conditioning involves HIF1-associated regulation of the pentose phosphate pathway in rat brain. Neurochem. Res. V. 44. P. 1425. https://doi.org/10.1007/s11064-018-2681-x

Vetrovoy O., Sarieva K., Lomert E., Nimiritsky P., Eschenko N., Galkina O., Lyanguzov A., Tyulkova E., Rybnikova E. 2020. Pharmacological HIF1 inhibition eliminates downregulation of the pentose phosphate pathway and prevents neuronal apoptosis in rat hippocampus caused by severe hypoxia. J. Mol. Neurosci. V. 70. P. 635. https://doi.org/10.1007/s12031-019-01469-8

Transcription Factor HIF1 Negatively Regulates Glucose-6-Phosphate Dehydrogenase Content in HEK293T Cells

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Using cobalt chloride ($CoCl_2$), a pharmacological inducer of hypoxia-induced transcription factor-1 (HIF1), and topotecan, a translation blocker of HIF1α, the previously demonstrated on the rat brain phenomenon of the HIF1-dependent suppression of the glucose-6-phosphate dehydrogenase (G6PD), key enzyme of the pentose phosphate pathway, was estimated. On the primary culture of rat skin fibroblasts, non-toxic doses of $CoCl_2$ (50 and 100 μM) and topotecan (0.2 and 1 μM) were determined. Transfection with a vector containing luciferase under the control of the HIF1-dependent promoter (HRE) was performed on the cell line of the human embryonic kidney HEK293T (Human Embryonic Kidney 293T). The efficacy of $CoCl_2$ and topotecan dose-dependent HIF1-mediated transcription was analyzed by chemiluminescence method. Quantitative analysis of HIF1α and G6PD was performed by Western Blot. $CoCl_2$ causes dose-dependent accumulation of HIF1α and increased luciferase activity, contributing to a decrease in the amount of G6PDH. Topotecan causes a decrease in the accumulation of HIF1α, which is accompanied by a weakening of the luciferase activity and normalization of the content of G6PDH. The results confirm the suppressive effect of the transcription factor HIF1 on the pentose phosphate pathway, and demonstrate the universality of this mechanism for cells of various rat and human tissues.

Keywords: HEK293T, HIF1, $CoCl_2$, topotecan, pentose phosphate pathway, glucose-6-phosphate dehydrogenase