DOI 10.18699/SBB-2022-234

Polymorphism in genes involved in glutathione metabolism is associated with the risk of uterine fibroids

Kudryavtseva O.[1, 2]*, Kobzeva K.[1], Vdovina I.[1, 2], Ponomareva L.[1], Polonikov A.[1], Bushueva O.[1]

[1] *Kursk State Medical University, Kursk, Russia*

[2] *Kursk Regional Clinical Hospital, Kursk, Russia*

** kudrolga.75@mail.ru*

Key words: uterine fibroids, glutathione metabolism, candidate susceptibility genes, *GSR*, *SLC7A11*

Motivation and Aim: Uterine fibroids (UF) is a monoclonal benign tumor derived from the smooth muscle cells of the myometrium. UF is diagnosed in 35-70 % of women of reproductive age, while a third of patients with UF have severe clinical symptoms, which often leads to a significant deterioration in their quality of life and a decrease in reproductive potential [1]. Currently, a significant role in the development of UF is attributed to disturbances in redox homeostasis in myometrial tissues, leading to chronic oxidative stress [2]. One of the most powerful natural antioxidants is glutathione [3]. Depletion of its intracellular pool due to the inheritance of functionally defective alleles of genes encoding enzymes involved in its metabolism leads to free radical damage to the myometrium, initiation and maintenance of the activity of proliferative processes in the tissues of the uterus. The aim of our study was to analyze the role of polymorphic variants of genes involved in glutathione metabolism in the development of UF.

Methods and Algorithms: A total of 889 unrelated Russian individuals from Central Russia were examined: 552 patients with UF and 337 healthy females, without clinical and ultrasound signs of uterine fibroids. Written informed consent was obtained from all participants prior to entering the study. The study was approved by the Regional Ethics Committee of Kursk State Medical University.

Genotyping of single nucleotide polymorphisms of rs713041 *GPX4*, rs4902346 *GPX2*, rs41303970 *GCLM*, rs17883901 *GCLC*, rs1050450 *GPX1*, rs1799811 and rs1695 *GSTP1*, rs2551715 *GSR*, rs1801310 *GSS*, rs4820599 *GGT1*, rs7674870 *SLC7A11* was performed by TaqMan-based PCR on a CFX96 amplifier (Bio-Rad). Genotyping of deletion polymorphisms (+/0 *GSTM1* and +/0 *GSTT1*) was carried out by multiplex PCR. All the genes selected for the study are involved in the metabolism of glutathione and are expressed in myometrium. To analyze the associations of genotypes with the development of UF, we used a log-additive regression model calculated in the SNPStats program (https://www.snpstats.net/start.htm) with an adjustment for age and smoking.

Results: Comparative analysis of genotype frequencies revealed a protective effect of rs2551715 *GSR* on the risk of UF: OR = 0.83, 95 %CI = 0.70–0.99, P = 0.04. It is well-known that the *GSR* gene encodes the enzyme glutathione reductase involved in the reduction of the active form of glutathione [5]. As shown by functional studies, the C allele, compared with the minor T allele, reduces the activity of the GSR enzyme, which can lead to the accumulation of free radicals and increased oxidative stress in the tissues of the uterus due to the low rate of recovery of the active form of glutathione. We also found an association of rs7674870 *SLC7A11* with an increased risk of UF: OR = 1.25, 95 %CI = 1.03-1.50, P = 0.02. We found no functional studies of rs7674870 *SLC7A11*.

However, since this SNP is located in the 3'-untranslated region of the gene, it may affect transcriptional regulation, mRNA stability, or microRNA binding. For interpretation of the functional effects of rs7674870 *SLC7A11* bioinformatic analysis has been done. Analysis of expression quantitative trait loci (eQTL) using the GTEx Portal resource (https://gtexportal.org/home/) revealed that the risk allele G rs7674870 is associated with a decrease in the expression of *SLC7A11* in the blood (P = 0.012, NES = –0,10). Transcription factor binding analysis found that the protective allele A rs7674870 *SLC7A11* binds to 16 transcription factors (TFAP2A, NFIC, SP2, PAX9, RUNX1, RUNX3, RUNX2, PAX1, TFAP2, TFAP2C, NFIB, TFAP2A, DMRTC2, RAD21, RUNX3, E2F4), which are jointly involved in the regulation of cell population proliferation (GO:0042127; FDR = 6.57×10^{-3}). At the same time, the risk allele G binds to 8 transcription factors (MAFK, ATF1, AP1, SPDEF, SMAD3, MAFB, ATF1, AP1), however, no common biological processes were found for this transcription factors.

Conclusion: Thus, we have carried out a comprehensive analysis of the involvement of glutathione metabolism genes in predisposition to uterine fibroids. An association of single nucleotide polymorphisms rs7674870 *SLC7A11* and rs2551715 *GSR* with the risk of uterine fibroids was found for the first time.

Acknowledgements: The study is supported by Kursk State Medical University.

References

1. Stewart E.A., Cookson C.L., Gandolfo R.A., Schulze-Rath R. Epidemiology of uterine fibroids: a systematic review. *BJOG.* 2017;124(10):1501-1512.
2. Fletcher N.M., Abusamaan M.S., Memaj I. et al. Oxidative stress: a key regulator of leiomyoma cell survival. *Fertil Steril.* 2017;107(6):1387-1394.e1.
3. Bansal A., Simon M.C. Glutathione metabolism in cancer progression and treatment resistance. *J Cell Biol.* 2018;217(7):2291-2298.
4. Kudryavtseva O.K., Barysheva E.M., Sorokina M.V., Polshvedkina O.B., Ivanova N.V., Polonikov A.V., Bushueva O.Y. The analysis of relationship between the A/G (rs1801310) polymorphism of the GSS gene and the development of uterine myoma: a pilot study. *Med Genet.* 2017;16(2):37-39. (in Russian)
5. Kelner M.J., Montoya M.A. Structural organization of the human glutathione reductase gene: determination of correct cDNA sequence and identification of a mitochondrial leader sequence. *Biochem Biophys Res Commun.* 2000;269(2):366-368. doi: 10.1006/bbrc.2000.2267.