Abstract

On average, 5-10% of all cancers occur in patients with hereditary tumors, who may have mutations in tens to hundreds of tumor predisposition genes. The phenotypes in mutation carriers overlap, and parallel analyses with sequencing panels is the method of choice in diagnostics. In our laboratory, we designed a universal panel and a targeted panel for a specific cancer, which allowed us to identify genetic alterations in patients with ovarian cancer, breast cancer, melanoma, and other cancers in the Czech Republic. The results of next generation sequencing (NGS) analyses show that the most frequent genetic alteration in ovarian cancers patients in the Czech Republic are hereditary mutations in *BRCA1* (in 24% of unselected patients) and in malignant melanoma patients *CDKN2A* (in 2% of high risk patients). The presence of hereditary alterations is a clinically significant phenomenon affecting the prognosis and treatment of the disease. However, the interpretation of NGS findings is complicated by the presence of variants of unknown significance (VUS). We participate in the interpretation of VUS in the main predisposing genes *BRCA1* and *BRCA2* within the international consortium ENIGMA (Evidence-based Network for the Interpretation of Germline Mutant Alleles). Our and international results of the most studied group of hereditary cancers – breast and ovarian cancers – identified *CHEK2* as the third most mutated gene, which germline mutations predispose to the development of various cancer types. For its analysis, we prepared a model system based on the targeted deletion of the endogenous *CHEK2* gene using CRISPR/Cas9 in human non-transformed RPE1 cells and the subsequent expression of the fluorescently labeled CHK2 variant. Using this system, we functionally classified VUS found in patients with ovarian cancer and high-risk patients with melanoma. With this approach, we are now analyzing the *CHEK2* variants identified within the ENIGMA consortium. The functional classification of *CHEK2* variants significantly contributed to the classification of benign and pathogenic variants occurring in the Czech population.

Key words: NGS, CZECANCA, panel sequencing, cancer predisposition, functional analysis, *CHEK2*, CHK2 kinase