P54 MicroRNA for differentiation low and high grade of noninvasive bladder cancer

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Introduction & Objectives: MicroRNAs are a class of small 17 - 25-nucleotide, single-stranded, non-coding RNAs, whose main function is post-transcriptional regulation of gene expression. The purpose of the study was identify microRNA which can accurately predict the presence of BC and differentiate low grade tumors from high grade bladder cancer (BC).

Materials & Methods: The tested group consisted of 55 tumour tissue samples and 30 samples of normal urothelium from healthy individuals. Majority of patients were males (45/55). 37 patients of the group suffered from non-invasive bladder cancer, among whom 19 patients were in stage Ta and 18 patients were in stage T1. The rest of 18 patients were diagnosed with invasive bladder cancer in stage T2. The tumors selected for RNA isolation were submerged in RNA later solution and stored at - 20°C until the isolation time. Purity of the samples were verified using Qubit ssDNA HS assay kit. miRs from 85 samples were reverse transcribed using Taqman MicroRNA Reverse Transcription Kit and the expression level of the selected microRNAs was assessed with the use of SYBR Green and CFX96 Biorad. The expression level of each patient was normalized against the endogen control – miR-103a-5p. After that, the difference of the tested and control microRNAs (ΔCt) was calculated for individual samples. The calculations of fold change were made under the formula: $FC = 2^{-\Delta\Delta Ct}$. Association of microRNA expression with clinical parameters were analysed using the U Mann-Whitney test. The area under the receiver operating characteristics curve (AUC) was also calculated.

Results: Significant differences between tumour and normal tissue have been observed for miR-205 and miR-20 expression. No correlation was found between the expression level of the tested microRNAs and other clinical parameters. Using backward stepwise elimination we found that mir-182 and mir-20 are able diferrentaite low and high grade tumors.

Conclusions: Three microRNA can accurately identify BC and differentiate low and high grade noninvasive disease. Such test can be useful in detection and assessment of biological potential of tumor.