Abstract

Colorectal cancer is a serious malignant disease with an incidence of over 1.8 million new cases per year worldwide. There are about 8 000 patients diagnosed with CRC in the Czech Republic each year, and about half of them present with an advanced disease. Screening program identifies patients in the early stages of CRC resulting in overall better prognosis and survival. There is also a lack of biomarkers of early CRC detection and of response to treatment.

The first aim of our project was to conduct a national multicentre prospective observational study to evaluate the impact of CRC screening within the framework of a Czech population screening programme. Between March 2013 and September 2015, a total of 265 patients were enrolled in 12 centres across the Czech Republic. Patients were divided into screening and control groups and compared for pathology status and clinical characteristics. Screening was defined as a primary screening colonoscopy or a colonoscopy after a positive FOBT in an average-risk population. The distribution of CRC stages was significantly favourable in the screening group compared with the control group (stages 0, I and II, 63% versus 43.3%; $p < 0.001$). The presence of distant (M1) and local metastases (N1 and N2) was significantly less prevalent in the screening group (0%, 28.8%) than in the control group (18.2%, 45.3%) ($p < 0.001$). In both groups, patients had tumour localized in left colon (screening – 91.9%; control – 74.9%). CRC diagnosed by screening disclosed less advanced clinical-pathological characteristics and results in patients with higher probability of radical surgery (R0) than diagnoses established based on symptoms, with subsequent management differing accordingly between groups. These results advocate the implementation of a suitable worldwide screening programme.

The second aim of our project was to evaluate microRNA (miRNA) as a promising source of cancer-related biomarkers since miRNA signatures are specific for each cancer type and subgroups of patients with diverse treatment sensitivity. Yet this miRNA potential has not been satisfactorily explored in rectal cancer (RC). The aim of the study was to identify the specific miRNA signature with clinical and therapeutical relevance for RC. Expression of 2 555 miRNAs were examined in 20 pairs of rectal tumours and matched non-malignant tissues by 3D-Gene Toray microarray. Candidate miRNAs were validated in an independent

cohort of 100 paired rectal tissues and in whole plasma and exosomes of 100 RC patients. To study the association of miRNA profile with therapeutic outcomes, plasma samples were taken repeatedly over time period of one year reflecting thus patients' treatment responses. Finally, the most prominent miRNAs were investigated *in vitro* for their involvement in cell growth. We identified RC specific miRNA signature that distinguishes responders from non-responders to adjuvant chemotherapy. A predominant part of identified miRNAs was represented by members of miR-17/92 cluster. Upregulation of miRNAs 17, 18a, 18b, 19a, 19b, 20a, 20b and 106a in the tumour was associated with higher risk of tumour relapse and their overexpression in RC cell lines stimulated cellular proliferation. Examination of these miRNAs in plasma exosomes showed that their levels differed between RC patients and healthy controls and correlated with patients' treatment response. MiR-17/92 cluster miRNAs represent a non-invasive biomarker to predict post-treatment prognosis in RC patients.

The third aim of our project was to evaluate expression of quantitative trait loci (eQTL) variants in ABC transporter and their possible role in CRC development or treatment response. We have identified 14 single nucleotide polymorphisms (SNPs) in 11 ABC transporter genes acting as eQTL. We enrolled 1098 CRC patients and 1442 healthy controls. We did not find any significant association between SNPs and risk of CRC. The SNP rs3819720 was significantly associated with shorter overall survival. The allele rs3819720 affected the expression of 36 downstream genes. Screening for eQTL polymorphisms in genes, for example gen for ABC transporter, could help to elucidate the genetic background of individual response to treatment.