Abstract

Colorectal carcinoma (CRC) is one of the most prevalent malignancies in the Czech Republic. In general, there are two molecular pathways leading to CRC: one is characterized by chromosomal instability, the other by the deficiency in DNA mismatch repair (MMR) genes. *MutL homologue 1 (MLH1)* gene, a member of the MMR gene-family, represents a key component of the MMR system, responsible for recognition of nucleotide mismatches occurring during DNA replication, and for the recruitment of repair proteins to correct the replication errors. According to literature, somatic mutations in MMR genes, and *MLH1* in particular, hallmark sporadic, MMR deficient, CRC cases.

We aimed at analyzing somatic events in *MLH1* gene and the determination of microsatellite instability (MSI) status in 99 DNA samples from 96 patients with sporadic CRC. Mutations were screened by high resolution melting (HRM) curve analysis. Positive cases in each run were subsequently verified by automated sequencing.

Mainly gene variants were found in *MLH1* gene: We discovered two new variants, one in exon 2 at position c. 204 C>G, p. Ile68Met (98 C/C, 1C/G) and the other in exon 11 at position c. 973 C>T, p. Arg325Trp (98 C/C, 1 C/T). Only the latter variant c. 973 C>T was identified as somatic mutation. All other variants found in *MLH1* gene were germ-line variants. Most predominant was polymorphism c. 655 A>G, p. Ile219Val in exon 8, found in 50 DNA samples (49 A/A, 34 A/G and 16 with G/G). The other polymorphisms were detected in intron 13 at position c. 1558+14 G>A (93 G/G, 6 G/A), in intron 14 at position c. 1668-19 A>G (23 A/A, 56 A/G, 20 G/G), in exon 17 c. 1959 G>T, p. Leu653Leu (94 G/G, 5 G/T) and in exon 19 c. 2146 G>A, p. Val716Met (97 G/G, 2 G/A). We also detected mutation in exon 16 at position c. 1733 A>G, p.Glu578Gly in one patient (98 A/A, 1 A/G), which was previously described in hereditary non-polyposis colorectal cancer (HNPCC). Microsatellite instability-high (MSI-H) status was determined in 9 DNA samples (8 patients), the other 90 samples (88 patients) were considered as microsatellite stable (MSS).

We may conclude that somatic mutations in *MLH1* gene in investigated patients apparently play minor role in the development of sporadic form of CRC.

**Key words:** *MLH1* gene, colorectal cancer, mismatch repair, mutations