The importance of $G2677T/A$ and $C3435T$ polymorphisms of the $MDR1$ gene in the aetiology of colorectal cancer

Grzegorz Stańko, Marek Kamiński, Anna Bogacz, Agnieszka Seremak-Mrozikiewicz, Bogusław Kosiński, Joanna Bartkowiak-Wieczorek, Daniel Kotrych, Bogusław Czerny

1Department of General and Gastroenterological Surgery, Pomeranian Medical University, Szczecin, Poland
2Laboratory of Experimental Pharmacogenetics, Department of Clinical Pharmacy and Biopharmacy, Poznan University of Medical Sciences, Poznan, Poland
3Department of Pharmacology and Phytochemistry, Institute of Natural Fibres and Medicinal Plants, Poznan, Poland
4Division of Perinatology and Women’s Diseases, Poznan University of Medical Sciences, Poznan, Poland
5West Pomeranian Centre of Oncology, Division of Surgical Oncology, Szczecin, Poland
6Department of Orthopaedics and Traumatology, Pomeranian Medical University, Szczecin, Poland
7Department of General Pharmacology and Pharmacoeconomics, Pomeranian Medical University, Szczecin, Poland
8Department of Stem Cells and Regenerative Medicine, Institute of Natural Fibres and Medicinal Plants, Poznan, Poland

Prz Gastroenterol 2016; 11 (1): 35–40
DOI: 10.5114/pg.2015.51185

Key words: colorectal cancer, P-glycoprotein, polymorphism, xenobiotics.

Address for correspondence: Anna Bogacz PhD, Laboratory of Experimental Pharmacogenetics, Department of Clinical Pharmacy and Biopharmacy, Poznan University of Medical Sciences, 14 Św. Marii Magdaleny St, 60-861 Poznan, Poland, phone: +48 61 668 78 37, fax: +48 61 668 78 55, e-mail: aniabogacz23@o2.pl

Abstract

Introduction: Colorectal cancer (CRC) is the most common cancer among patients, and its aetiology is still not precisely known. It is believed that 15–30% of colorectal cancers are genetically determined. P-glycoprotein (P-gp) encoded by the $MDR1$ gene in normal conditions plays an important role in the action of colon epithelial cells. However, the $MDR1$ polymorphism influences the P-gp expression and can weaken its effect against xenobiotics (procarcinogens) and increase the frequency of CRC.

Aim: To evaluate the correlation between the $MDR1$ C3435T and $G2677T/A$ polymorphisms and the risk of colorectal cancer.

Material and methods: The study group with colorectal cancer included 47 women and 60 men while the control group consisted of 110 healthy patients. The diagnosis in patients suffering from CRC was confirmed by histopathological report. Genetic analysis was performed using PCR-RFLP method.

Results: We showed only a correlation between the frequency of CT and TT genotypes of $C3435T$ polymorphism and the risk of colorectal cancer in younger age. There was no correlation between the $C3435T$ and $G2677T/A$ polymorphisms of the $MDR1$ gene and other clinical parameters.

Conclusions: Our findings suggest that T allele carriers of $C3435T$ polymorphism have an increased risk of CRC. However, further studies are needed on a much larger number of patients and genes associated with metabolism and transport of xenobiotics including procarcinogens.

Introduction

Colorectal cancer (CRC) is the most common newly diagnosed cancer and the third most common cause of cancer death among men and women. The risk of CRC is increased by several factors such as gene mutations, lifestyle factors, age, heredity, and polyps of the colon [1, 2]. The relationships between risk factors and colorectal cancer development are not exactly known. About 15–30% of all colon cancer cases are associated with genetic changes. The most common hereditary forms of CRC are familial adenomatous polyposis (FAP) and hereditary nonpolyposis colorectal cancer (HNPPC), although there are other syndromes associated with predisposition [3, 4]. The adenomatous polyposis coli (APC) gene encodes the APC protein that plays a substantial role in the regulation of the Wnt signalling pathway.
Pathway participating in the activation of many genes (MDR1, c-Myc) [5, 6]. Hence, the genetic variability in genes associated with metabolism and transport of procarcinogens, DNA repair system, and activation of major signalling pathways may contribute to the appearance and progression of various cancers including CRC [7–10].

In addition, dietary factors that determine the environment of the intestinal epithelium are thought to play an important role in the process of oncogenesis because their toxic metabolites can cause genetic damage [11]. There are several mechanisms providing protection against toxic agents, such as P-glycoprotein (P-gp) belonging to the ATP binding cassette transporter superfamily, which functions as a transmembrane drug efflux pump, decreasing intracellular xenobiotic accumulation, and eliminates toxic agents from the cell. P-glycoprotein encoded by the MDR1 gene, outside the protection of the body against exogenous compounds, plays a role in immune regulation of cell death [12–15]. In normal conditions, colon epithelial cells have a high concentration of P-glycoprotein, which plays an important role in their action. However, the MDR1 gene polymorphism influencing the P-gp expression can weaken its effect against xenobiotics (procarcinogens) and increase the incidence of CRC [16, 17]. Furthermore, it is believed that the overexpression of the MDR1 gene encoding the P-gp contributes significantly to the phenomenon of multidrug resistance (MDR) responsible for the failure of the pharmacotherapy of cancer. Multidrug resistance is one of the most important causes of reduced efficacy of cancer therapy. Among the best known and most significant polymorphisms of the MDR1 gene are C3435T and G2677T/A [16, 18]. Many studies have shown a significant role of these polymorphisms in the pathogenesis of colorectal cancer [19–21].

Aim

The aim of our study was to determine the frequency of C3435T and G2677T/A polymorphisms of the MDR1 gene in the group of colorectal cancer patients, in relation to healthy patients. In addition, we studied the impact of these polymorphisms on the CRC development and the correlation between the frequency of particular genotypes and the clinical factors.

Material and methods

Patients

In the present study 107 patients with diagnosed colorectal cancer were evaluated. The patients were diagnosed and treated between 2010 and 2012 in the Department of General and Gastroenterological Surgery of SPSK1 Hospital in Szczecin. The study group included 47 women and 60 men. The age range of patients suffering from CRC was 47–83 years. In every case, the diagnosis of CRC was confirmed by histopathological report. The clinical data from the patients was collected, including age, gender, tumour localisation, staging, grading, and clinical symptoms (anaemia, weight loss, bowel obstruction). The control group consisted of 110 healthy patients of similar age. The Bioethical Committee of the Pomeranian Medical University approved the study. All patients were informed about the aim of the study and gave written consent to perform genetic testing.

Among the 53 patients in the study group, the most common histological type of colorectal cancer was adenocarcinoma (the degree of differentiation – G2) constituting 78% (41 cases), adenocarcinoma G1 – 10% (5 cases), adenocarcinoma G3 – 6% (3 cases), carcinoma mucinosum – 4% (2 cases), and carcinoma gelatinosum – 2% (1 case). The 54 remaining patients did not agree to make available data with histopathological examination.

Genetic analysis

Genetic testing was performed at the Laboratory of Experimental Pharmacogenetics, Department of Clinical Pharmacy and Biopharmacy, Poznan University of Medical Sciences. The C3435T and G2677T/A polymorphisms of the MDR1 gene were determined using polymerase chain reaction and restriction fragment length polymorphism (PCR-RFLP) methods. The primers (TibMolBiol, Poland) used in the PCR reaction, length of amplified products, and the conditions of the PCR reaction were applied as previously described [22, 23].

The results of PCR-RFLP were analysed on agarose gels by visualisation in UV light using a documentation system (KS 4000/Image PC, Syngen Biotech Molecular Biology Instruments).

Statistical analysis

The statistical significance of the difference between the control and study groups was assessed by SPSS 17.0 software using one-way ANOVA test (SPSS Inc.). Values of $p < 0.05$ were considered to represent a statistically significant difference.

Results

No differences were found in the MDR1 C3435T polymorphism frequencies between colorectal patients and the control group ($p = 0.94$) (Table I). Among the study group, the most common genotype was heterozygous CT (49.5%) then homozygous TT genotype (34.6%) while homozygous CC genotype occurred in 17 (15.9%) cases. The frequency of the individual alleles also showed no statistically significant differences between the study groups.
The importance of G2677T/A and C3435T polymorphisms of the MDR1 gene in the aetiology of colorectal cancer

Przegląd Gastroenterologiczny 2016; 11 (1)

The importance of G2677T/A and C3435T polymorphisms of the MDR1 gene in the aetiology of colorectal cancer

Przegląd Gastroenterologiczny 2016; 11 (1)

In the control group, the genotype distribution was very similar (CT – 51.8%, TT – 32.7%, CC – 15.5%). Also, the risk of developing colorectal cancer has been studied in three models: recessive, dominant, and additive. None of these models showed a statistically significant increase in risk of colorectal cancer (TT homozygotes OR = 1.03; CT heterozygotes OR = 0.93) (Table II).

No statistically significant impact of C3435T polymorphism on the appearance of CRC was found. The increase of risk of colorectal cancer was only observed for CT and TT genotypes of C3435T polymorphism in younger age. There were no significant differences between the clinical factors of colorectal cancer patients and genotypes frequencies (Table III).

For G2677T/A polymorphism, statistically significant differences between colorectal cancer patients and the control group were observed (p = 0.02) (Table I). The most common genotype in the study group and the control group was heterozygous GT (50.5% vs. 42.7%).

Table I. The frequency of genotypes and alleles of the MDR1 C3435T and G2677T/A polymorphisms in the study group with colorectal cancer and in the control group

| Variable | Study group (n = 107) | Control group (n = 110) | Value of p |
|----------|----------------------|------------------------|------------|
| C3435T Genotype: | | | |
| CC | 17 (15.9) | 16.5 | 17 (15.5) | 17.1 | 0.94 |
| CT | 53 (49.5) | 48.3 | 57 (51.8) | 48.5 |
| TT | 37 (34.6) | 35.2 | 36 (32.7) | 34.4 |
| Allele: | | | |
| C | 87 (40.7) | – | 91 (41.4) | – | 0.88 |
| T | 127 (59.3) | – | 129 (58.6) | – |
| G2677T/A Genotype: | | | |
| GG | 39 (36.4) | (38.6) | 34 (30.9) | 27.3 | 0.02 |
| GT | 54 (50.5) | (45.9) | 47 (42.7) | 50.0 |
| TT | 12 (11.2) | (13.6) | 29 (26.4) | 22.8 |
| TA | 1 (0.9) | (0.7) | 0 (0) | 0 |
| GA | 1 (0.9) | 1.2 | 0 (0) | 0 |
| AA | 0 (0) | 0 | 0 (0) | 0 |
| Allele: | | | |
| G | 133 (62.2) | – | 115 (52.3) | – | 0.03 |
| T | 79 (36.9) | – | 105 (47.7) | – |
| A | 2 (0.9) | – | 0 (0) | – |

*p < 0.05.

Table II. The odds ratio (OR) and 95% confidence intervals (95% CI) for the developing colorectal cancer

| Variable | OR (95% CI) | Value of p |
|----------|-------------|------------|
| C3435T polymorphism: | | |
| Recessive model | TT vs. CT + CC | 1.09 (0.62–1.90) | 0.77 |
| Dominant model | TT + CT vs. CC | 0.97 (0.47–2.01) | 0.93 |
| Additive model | CT vs. CC | 0.93 (0.43–2.01) | 0.85 |
| TT vs. CC | 1.03 (0.46–2.32) | 0.95 |
| G2677T/A polymorphism: | | |
| Recessive model | TT vs. GT + GG | 0.36 (0.17–0.75) | 0.007 |
| Dominant model | TT + GT vs. GG | 0.76 (0.43–1.33) | 0.34 |
| Additive model | GT vs. GG | 1.00 (0.55–1.83) | 1.00 |
| TT vs. GG | 0.36 (0.16–0.82) | 0.01 |

Przegląd Gastroenterologiczny 2016; 11 (1)
Table III. The selected clinical parameters in patients with colorectal cancer compared to different polymorphic variants of the MDR1 gene

| Parameter                              | C3435T polymorphism | G2677T/A polymorphism |
|----------------------------------------|---------------------|-----------------------|
|                                        | CC      | CT      | TT      | Value of p | GG     | GT      | TT      | Value of p |
| Gender:                                |         |         |         |            |         |         |         |            |
| Male                                   | 10      | 31      | 19      | 0.77       | 21      | 31      | 8       | 0.73       |
| Female                                 | 7       | 22      | 18      |            | 18      | 23      | 4       |            |
| Age [years]:                           |         |         |         |            |         |         |         |            |
| Mean Range                             | 73      | 68      | 79      | 0.013      | 73      | 69      | 73      | 0.40       |
|                                         | 57–82   | 54–81   | 68–83   |            | 57–82   | 54–83   | 65–80   |            |
| pT                                     |         |         |         |            |         |         |         |            |
| Tis                                    | 0       | 2       | 0       |            | 0       | 2       | 0       |            |
| T1                                     | 0       | 0       | 0       |            | 0       | 0       | 0       |            |
| T2                                     | 1       | 2       | 2       |            | 2       | 3       | 0       |            |
| T3                                     | 7       | 22      | 5       | 0.64       | 12      | 18      | 3       | 0.92       |
| T4                                     | 2       | 7       | 3       |            | 4       | 6       | 2       |            |
| pN                                     |         |         |         |            |         |         |         |            |
| N0                                     | 5       | 16      | 7       |            | 8       | 18      | 1       |            |
| N1                                     | 2       | 10      | 2       | 0.80       | 5       | 6       | 3       | 0.50       |
| N2                                     | 2       | 7       | 1       |            | 4       | 5       | 1       |            |
| M                                      |         |         |         |            |         |         |         |            |
| M0                                     | 8       | 23      | 9       | 0.14       | 15      | 22      | 3       | 0.20       |
| M1                                     | 1       | 9       | 0       |            | 1       | 6       | 2       |            |
| Location:                              |         |         |         |            |         |         |         |            |
| Colon ascended                         | 2       | 6       | 3       |            | 3       | 7       | 1       |            |
| Colon transverses                      | 1       | 6       | 2       |            | 2       | 5       | 2       |            |
| Colon descends                         | 0       | 3       | 1       |            | 2       | 2       | 0       |            |
| Colon sigmoidal                       | 1       | 15      | 3       | 0.10       | 6       | 12      | 1       | 0.86       |
| Rectum                                 | 6       | 3       | 0       |            | 5       | 3       | 1       |            |
| Histological type:                    |         |         |         |            |         |         |         |            |
| Adenocarcinoma                         | 9       | 32      | 9       | 0.48       | 17      | 27      | 5       | 0.81       |
| Others                                 | 1       | 1       | 1       |            | 1       | 2       | 0       |            |
| Degree of differentiation:            |         |         |         |            |         |         |         |            |
| G1                                     | 0       | 4       | 1       |            | 2       | 3       | 0       |            |
| G2                                     | 8       | 26      | 8       | 0.72       | 15      | 22      | 4       | 0.56       |
| G3                                     | 1       | 2       | 0       |            | 0       | 2       | 1       |            |
| Symptoms of anaemia:                  |         |         |         |            |         |         |         |            |
| No                                     | 5       | 8       | 2       | 0.07       | 7       | 6       | 2       | 0.52       |
| Yes                                    | 5       | 25      | 8       |            | 11      | 23      | 3       |            |
| Body weight loss:                     |         |         |         |            |         |         |         |            |
| No                                     | 5       | 14      | 6       | 0.79       | 11      | 12      | 1       | 0.28       |
| Yes                                    | 5       | 19      | 4       |            | 7       | 17      | 4       |            |
| Obstruction:                           |         |         |         |            |         |         |         |            |
| No                                     | 7       | 18      | 7       | 0.72       | 9       | 20      | 3       | 0.33       |
| Yes                                    | 3       | 15      | 3       |            | 9       | 9       | 2       |            |
| Radical surgery:                      |         |         |         |            |         |         |         |            |
| No                                     | 2       | 6       | 1       | 0.82       | 3       | 4       | 1       | 0.89       |
| Yes                                    | 8       | 27      | 9       |            | 15      | 25      | 4       |            |

TNM classification of malignant tumours (T – size or direct extent of the primary tumour (Tis: carcinoma in situ, T0: no signs of tumour, T1, T2, T3, T4: size and/or extension of the primary tumour), N – degree of spread to regional lymph nodes (N0: tumour cells absent from regional lymph nodes; N1: regional lymph node metastasis present; N2: tumour spread to an extent between N1 and N3), M – presence of distant metastasis (M0: no distant metastasis, M1: metastasis to distant organs)).
A protective role of TT genotype of G2677T/A polymorphism against the development of CRC was observed (OR = 0.36, 95% CI = 0.17–0.75) (Table II). No dependence between colorectal cancer clinical factors and G2677T/A polymorphism was found (Table III).

Discussion

In the present study we showed no effect of C3435T polymorphism of the MDR1 gene on the occurrence of colorectal cancer. We only observed an increase of risk of CRC for TT and CT genotypes of C3435T polymorphism in younger age. This suggests that the T allele carriers have an evaluated predisposition to the occurrence of colorectal cancer. Similar results were obtained by Petrova et al. [24]. The authors investigated the frequencies of MDR1 C3435T polymorphism in patients with colorectal cancer and a control group in a Bulgarian population. They did not show the relationship between the studied polymorphism and the occurrence of colorectal cancer. The obtained values of the risk were not statistically significant and amounted to OR = 0.81 (95% CI: 0.43–1.52) for CT heterozygotes and OR = 1.33 (95% CI: 0.77–2.30) for TT homozygotes in comparison to CC genotype.

Kurzawski et al. also obtained similar results [25] analysing the frequency of C3435T polymorphism of the MDR1 gene in the Polish population. The authors did not show significant differences in the presence of polymorphisms between the study groups and the patients with colorectal cancer. However, they showed a statistically significant increased risk of colorectal cancer in carriers with 3435TT genotype younger than 50 years (OR = 2.74, 95% CI: 1.02–7.53). The authors explain the reduced amount of P-gp in the cell membrane of the intestine, which leads to decreased removal of substances from the body that affect carcinogenesis.

According to Osswald et al. [26], the obtained results showed statistically significant decreased risk of colorectal cancer in carriers with TT and CT genotypes. Moreover, they noted an increased risk of developing colorectal cancer in patients with chronic smoking (OR = 3.9, 95% CI: 1.4–10.6) compared to the same polymorphism in the MDR1 gene. Another study demonstrated the influence of environmental factors on the correlation with C3435T polymorphism and the occurrence of colorectal cancer [20]. The authors analysed the effect of eating meat, smoking, administration of non-steroidal anti-inflammatory drugs (NSAIDs), and hormone replacement therapy. They showed a slightly increased incidence of colorectal cancer among homozygous CC patients consuming an increased amount of red meat (OR = 1.08, 95% CI: 1.00–1.16). Also, the CC genotype correlated with the intake of NSAIDs characterised by a more than two-fold increase in the risk of colorectal cancer (OR = 2.34, 95% CI: 1.22–4.48). Other variants had no effect on the incidence of CRC in correlation with NSAIDs. Furthermore, the authors have not proved the impact of smoking on the risk of colorectal cancer in relation to the MDR1 C3435T polymorphism. Additionally, Andersen et al. demonstrated statistically significantly reduced risk of colorectal cancer among homozygous TT patients (OR = 0.69, 95% CI: 0.48–1.00) [20]. The lack of a direct effect of the MDR1 gene C3435T polymorphism on the risk of colorectal cancer has also been shown in a meta-analysis conducted by Wang et al. [27]. Similar results on the C3435T polymorphism and the occurrence of CRC were obtained in the meta-analysis by Sheng et al. [28].

By analysing polymorphism G2677T/A we showed a statistically significant difference between patients with colorectal cancer and the control group (p = 0.02). In the individual comparative model we demonstrated a significant reduction in the risk of colorectal cancer in the additive model for the homozygous TT patients (OR = 0.36, 95% CI: 0.16–0.82, p = 0.01) and recessive model (OR = 0.36, 95% CI: 0.17–0.75, p = 0.007). Osswald et al. [26] obtained similar results in a study of the Russian population. The obtained results showed a statistically significant reduction in the risk of CRC in the case of heterozygotes GT compared to the homozygous GG (OR = 0.65, 95% CI: 0.45–0.96) and confirmed a protective effect of G2677T/A polymorphism of the MDR1 gene for the development of colorectal cancer. Furthermore, other studies among the populations of Bulgaria [24], Japan [29], and Italy [30] showed no significant association between G2677T/A polymorphism and the development of colorectal cancer.

The results of the presented studies do not provide a clear answer about the relationship between the investigated polymorphisms and the occurrence of colorectal cancer. This may be because of the need for performance analysis of the multi-gene polymorphisms in combination with existing environmental factors that may lead to malignant tumours.

Conclusions

Our results showed that the C3435T polymorphism of the MDR1 gene has no direct impact on the development of colorectal cancer. However, the relationship was found between the frequency of CT and TT genotypes and the risk of colon cancer in younger age. On the other hand, the TT genotype carrier of G2677T/A polymorphism has a protective effect on the incidence of CRC. However, further studies are needed on a much larger number of patients and genes associated with the metabolism and transport of xenobiotics (procarcinogens).
Acknowledgments
The study was supported by a statutory project from the Pomeranian Medical University in Szczecin (Poland).

Conflict of interest
The authors declare no conflict of interest.

References
1. Markowitz SD, Bertagnolli MM. Molecular origins of cancer: Molecular basis of colorectal cancer. N Engl J Med 2009; 361: 2449-60.
2. Park Y, Hunter DJ, Spiegelman D, et al. Dietary fiber intake and risk of colorectal cancer: a pooled analysis of prospective cohort studies. JAMA 2005; 294: 2849-57.
3. King JE, Dozois RR, Lindor NM, et al. Care of patients and their families with familial adenomatous polyposis. May Clin Proc 2000; 75: 57-67.
4. Katballe N, Christensen M, Wikman FP, et al. Frequency of hereditary non-polyposis colorectal cancer in Danish colorectal cancer patients. Gut 2002; 50: 43-51.
5. Lustig R, Behrens J. The Wnt signaling pathway and its role in tumor development. J Cancer Res Clin Oncol 2003; 129: 229-232.
6. Cruz-Bustillo Claren D. Molecular genetics of colorectal cancer. Rev Esp Enferm Dig 2004; 96: 48-59.
7. Romanowicz-Makowska H, Samulak D, Michalska M, et al. RADS1 gene polymorphisms and sporadic colorectal cancer risk in Poland. Pol J Pathol 2012; 3: 193-8.
8. Romanowicz H, Smolarz B, Baszczynski J, et al. Genetics polymorphism in DNA repair genes by base excision repair pathway (XRCC1) and homologous recombination (XRCC2 and RADS1) and the risk of breast carcinoma in the Polish population. Pol J Pathol 2010; 61: 206-16.
9. Markowitz SD, Bertagnolli MM. Molecular origins of cancer: molecular basis of colorectal cancer. N Engl J Med 2009; 361: 2449-60.
10. Zheng B, Wang Z, Chai R. NQO1 C609T polymorphism and colorectal cancer susceptibility: a meta-analysis. Arch Med Sci 2014; 10: 651-60.
11. Park Y, Hunter DJ, Spiegelman D, et al. Dietary fiber intake and risk of colorectal cancer: a pooled analysis of prospective cohort studies. JAMA 2005; 294: 2849-57.
12. Johnstone RW, Ruefl AA, Smyth MJ. Multiple physiological functions for multidrug transporter P-glycoprotein? Trends Biochem Sci 2000; 25: 1-6.
13. Mizutani T, Masuda M, Nakai E, et al. Genuine functions of P-glycoprotein (ABCB1). Curr Drug Metab 2008; 9: 167-74.
14. Maeda K, Sugiyama Y. Impact of genetic polymorphisms of transporters on the pharmacokinetic, pharmacodynamic and toxicological properties of anionic drugs. Drug Metab Pharmacokinet 2008; 23: 223-35.
15. Meijer GA, Schneijers AB, Flens MI, et al. Increased expression of multidrug resistance related proteins Pgp, MRP1, and LRP/MVP occurs early in colorectal carcinogenesis. J Clin Pathol 1999; 52: 450-4.
16. Dudarewicz M, Barańska M, Rychlik-Sych M, et al. C3435T polymorphism of the ABCB1/MDR1 gene encoding P-glycoprotein in patients with inflammatory bowel disease in a Polish population. Pharmacol Rep 2012; 64: 343-50.
17. Kerb R. Implications of genetic polymorphisms in drug transporters for pharmacotherapy. Cancer Lett 2006; 234: 4-33.
18. Cascorbi I, Gerloff T, Johne A, et al. Frequency of nucleotide polymorphisms in the P-glycoprotein drug transporter. Clin Pharmacol Ther 2001; 69: 169-74.
19. Kimchi-Sarfaty C, Oh JM, Kim JW, et al. A “silent” polymorphism in the MDR1 gene changes substrate specificity. Science 2007; 315: 525-8.
20. Andersen V, Ostergaard M, Christensen I, et al. Polymorphisms in the xenobiotic transporter Multidrug Resistance 1 (MDR1) and interaction with meat intake in relation to risk of colorectal cancer in a Danish prospective case-cohort study. BMC Cancer 2009; 9: 407-18.
21. Mickley LA, Lee JS, Weng Z, et al. Genetic polymorphism in MDR-1: a tool for examining allelic expression in normal cells, unselected and drug-selected cell lines, and human tumors. Blood 1998; 91: 1749-56.
22. Cascorbi I, Gerloff T, Johne A, et al. Frequency of single nucleotide polymorphisms in the P-glycoprotein drug transporter MDR1 gene in white subjects. Clin Pharmacol Ther 2001; 69: 169-74.
23. Bogacz A, Mrozekiewicz PM, Deka-Pawlik D, et al. Frequency of G2677T/A and C3435T polymorphisms of MDR1 gene in pre-eclamptic women. Ginekol Pol 2013; 84: 781-7.
24. Petrova DT, Nedeva P, Maslyankov S, et al. No association between MDR1 (ABCB1) 2677G>T and 3435C>T polymorphism and sporadic colorectal cancer among Bulgarian patients. J Cancer Res Clin Oncol 2008; 134: 317-22.
25. Kurzawski M, Drozdzik M, Suchy J, et al. Polymorphism in the P-glycoprotein drug transporter MDR1 gene in colon cancer patients. Eur J Clin Pharmacol 2005; 61: 389-94.
26. Osswald E, Johne A, Laschinski G, et al. Association of MDR1 genotypes with susceptibility to colorectal cancer in older non-smokers. Eur J Clin Pharmacol 2007; 63: 9-16.
27. Wang J, Wang B, Bi J, et al. MDR1 gene C3435T polymorphism and cancer risk: a meta-analysis of 34 case-control studies. J Cancer Res Clin Oncol 2012; 138: 979-89.
28. Sheng X, Zhang L, Tong N, et al. MDR1 C3435T polymorphism and cancer risk: a meta-analysis based on 39 case-control studies. Mol Biol Rep 2012; 39: 7237-49.
29. Komoto C, Nakamura T, Sakaeda T, et al. MDR1 haplotype frequencies in Japanese and Caucasian, and in Japanese patients with colorectal cancer and esophageal cancer. Drug Metab Pharmacokinet 2006; 21: 126-32.
30. De Iudicibus S, De Pellegrin A, Stocco G, et al. ABCB1 gene polymorphisms and expression of P-glycoprotein and long-term prognosis in colorectal cancer. Anticancer Res 2008; 28: 3921-8.

Received: 28.08.2014
Accepted: 22.01.2015