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To study the BRCA1 and BRCA2 mutations in Chinese patients with early onset breast cancer and affected relatives, 489 high-risk breast cancer patients from five breast clinical centers in China, were analyzed by using PCR-DHPLC or SSCP-DNA sequencing analysis. Allelotype analysis was done at five short tandem repeat (STR) markers in or adjacent to BRCA1 on the recurrent mutation carriers. In the 489 patients, 447 patients received mutation detection in both genes and 41 cases were analyzed only in BRCA1 gene. Twenty-three BRCA1 mutations and 21 BRCA2 ones were detected in our study. Two recurrent mutations in BRCA1, 1100delAT and 5589del8, were identified. The recurrent mutations account for 34.8% BRCA1 mutations in our series. Furthermore, 2 cases of BRCA1 5589del8 (0.5%) and one case of BRCA1 1100delAT (0.2%) was detected in 426 sporadic breast cancer patients and 564 healthy controls, respectively. Similar allelotypes were detected in most STR status for those harboring the same mutations. The frequency of BRCA1 and BRCA2 mutation in our study was both 4.7%. For those analyzed both genes, 8.7% of early-onset breast cancer cases and 12.9% of familial breast cancer cases had a BRCA1 or BRCA2 mutation, as compared with the 26.1% of cases with both early-onset breast cancer and affected relatives. When we stratified with number of breast cancer patients in family, the frequency had no statistical significance. However, the average age of disease onset in the families carrying BRCA1/2 mutations was significantly younger than the families without mutations ($P = 0.003$), and more the younger patients in family, higher the mutation frequency was. For those reporting malignancy family history other than breast/ovarian cancer, the prevalence of BRCA1/2 mutation is about 20.5%, and it was significantly higher than the patients only with family history of breast/ovarian cancer ($P = 0.02$). The family history of ovarian cancer (26.7% vs. 11.9%) and stomach cancer (23.8% vs. 11.8%) doubled the incidence of BRCA1/2, but the difference did not reach the statistical significance. We recommended the BRCA1 and BRCA2 genetic analysis could be done for high-risk breast cancer patient in Chinese population, especially for those with both early-onset breast cancer and affected relatives.

P18

Toremifene as a reference drug in the treatment of breast cancer

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Tamoxifen has opened the era of oestrogen receptor blockers. However, Tamoxifen has turned out to be a genotoxic and teratogenic drug as it has been reported by numerous authors and mentioned in the Statements of St. Gallen Consensus Conference.

During the past 15 years, we have been administering Tamoxifen instead of Toremifene. One thousand five hundred seventy one cases of administration of the drug in postmenopausal patients with breast cancer of above 50 years of age have been analysed. No cases of cancer of the liver or endometrium have been detected. Endometrium hyperplasia eliminated by a temporal cancellation of the administration of the drug was reported in 23 % of cases.

A study of the hormonal homeostasis has shown that administration of Toremifene makes it possible to achieve a fourfold reduction of follicle-stimulating hormone levels (reduction by 1/3 is observed on administration of Tamoxifen), which is important from an etiopathogenetic point of view. Moreover, Toremifene acts in both oestrogen-receptor-positive and oestrogen-receptor-negative patients (5 year survival is similar in both subpopulations).

Toremifene is advantageous as compared with Tamoxifen in the combined therapy of patients with advanced breast cancer: full remission has been achieved in 8.7 % of patients taking Toremifene and in 3.4 % of those taking Tamoxifen and time to progress is 5.1 months in patients on Toremifene and 3.9 months in patients on Tamoxifen.

The above data suggest that Toremifene should be a reference drug for the treatment of breast cancer.

P19

Multivariate discrimination functions composed with amino acid profiles (Amino Index®) as a novel diagnostic marker for breast and colon cancer

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Introduction: Amino acids balance is changed in patients of various diseases due to metabolic transition while it is maintained in healthy human. For example, Fisher ratio (ratio of branched amino acids to aromatic amino acids) is used as a primary index of hepatic failure. It is also known that the metabolism cancer cells are totally altered. Therefore, it is expected that the detection of metabolic transition using amino acid profiles is promising biomarker of various cancers. In this study, significant changes of plasma amino acid profiles of breast cancer and colon cancer patients were detected by discriminants (Amino Index®) by conducting with multivariate analysis.

Material and Methods: Blood samples were collected and cooled down from 50 colon cancer patients, 30 breast cancer patients, and age-gender adjusted healthy controls. Then, plasma samples were collected by centrifuge (3000rpm, 15min) and amino acid concentration was measured using LC/MS. The most appropriate discrimination functions for each cancer were estimated. The discrimination ability of discriminants was estimated as the area under the receiver-operator curve (ROC_AUC) of function values. The performances of the discriminants were also estimated using the test data randomly divided in advance. All the statistical and multivariate analysis was performed with MATLAB and GraphPad Prism.

Results: For discrimination of colon cancer, discriminant-1 composed with six amino acids (phenylalanine, threonine, alanine, glutamine, alpha-aminobutyric acid, and citrulline) was obtained. For breast cancer, discriminant-2 composed with composed with six amino acids (glutamine, arginine, isoleucine, alanine, ornithine, and histidine) was obtained. Both of them showed high performance for discrimination; 0.849 of ROC_AUC for discriminant-1 and 0.882 for discriminant-2, respectively. Then, the validation of performances of them using test data resulted 0.943 of ROC_AUC for discriminant-1 and 0.897 for discriminant-2, respectively, suggesting the reliability of these discriminants. Furthermore, the possibility of classification between these three groups was investigated. By conducting with discrimination analysis, it was suggested that the three statuses were

able to distinguish each other at high resolution using all the data obtained from women. According to these results, monitoring the plasma amino acid balance is a promising biomarker to diagnosis of various cancers.

GI-cancer prevention

P21
Sensitivity of colorectal cancer cell lines to cytostatics is not modulated by the calcium-binding protein calretinin

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One of the major obstacles for successful chemotherapy is that tumors frequently are resistant to certain forms of chemotherapy. Calretinin is an EF-hand calcium-binding protein mostly expressed in specific neurons and additionally in a variety of normal tissues, but is also observed in certain tumors derived from mesothelial and colon cells. Recent studies have postulated that treatment of colon cancer cells with either 5-fluorouracil (5-FU) or oxaliplatin leads to a resistant subpopulation of cells characterized by elevated calretinin expression levels. It was hypothetized that over-expression of calretinin may be one of the reasons leading to increased resistance of cells to treatment with either 5-FU or oxaliplatin. To test this, we investigated the sensitivity of calretinin-expressing HT-29 and calretinin-negative CaCo-2 colon cancer cells to 5-FU and cisplatin using the MTT assay. To directly assess the role of calretinin, the sensitivity of CaCo-2 cells stably transfected with either calretinin or the alternative splice variant, calretinin-22k, was determined. Our results have shown that HT-29 cells are more sensitive to 5-FU than CaCo-2 cells, while CaCo-2 cells were more sensitive to cisplatin. However, we did not observe any significant differences in sensitivity to either 5-FU or cisplatin of calretinin and calretinin-22k transfected CaCo2 cells when compared to mock-transfected CaCo-2 control cells. Although both calretinin variants affect adenocarcinoma cell differentiation, expression of either protein does not modulate sensitivity of CaCo-2 cells to the investigated cytostatics. Supported by grant aAV/1106/2004 from Ministry of Education of Slovak Republic.

P22
Bone mass density, subsequent risk of colon cancer and survival in postmenopausal women

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Objectives: To test the hypothesis that high bone mass density (BMD), a potential marker for cumulative exposure to endogenous estrogen, calcium and vitamin D intake, is associated with a lower risk of colon cancer, and that women with a lower BMD are likely to develop a more aggressive form of colon cancer, as defined by mortality.
Study design and Setting: BMD was measured in three different sites (Ward's triangle, trochanter, femoral neck) in 1,471 women 60 years of age. All incident cases of colon cancers were identified through record-linkage of cancer registry. The women were followed for a mean of 9.5 years.
Results: Overall 31 cases of colon cancer were observed among 28.6 expected (standardized incidence ratio (SIR) = 1.09, 95 percent confidence interval: 0.79–1.25). The SIR decreased with increasing bone mass density showing a significantly decreasing risk of 20% for women who were at the higher bone mass density comparatively to women who were at the lower bone mass density in all the skeletal sites. The 10-year survival rates showed that survival was increasing with increased BMD, but not significantly.
Conclusion: The findings suggest that postmenopausal women with lower BMD have an increased risk of colon cancer, but not more aggressive cancer.

P23
No association between VDR gene polymophisms (FoqI, TaqI and Apa1) and colorectal cancer in Romanian patients?

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Introduction and Purpose of the paper: Apart from the regulation of calcium metabolism, vitamin D3 plays an essential role in cell proliferation and differentiation in several tissues, including colonic epithelium. Polymorphisms in the vitamin D receptor (VDR) gene have been reported in several studies to be associated with colorectal cancer. o The goal of this study was to analyse the association between VDR gene polymorphisms (Foq, TaqI and Apa1) and colorectal cancer in Romanian patients.
Means and Methods: Blood samples were obtained, after informed consent from individuals, the test group including 70 colorectal cancer patients and 65 healthy persons. Genomic DNA was extracted from peripheral blood leukocytes using Promega Wizard kits. Three restriction fragment length polymorphisms (RFLPs) were genotyped: FoqI in exon 2, ApaI in intron 8 and TaqI in exon 9 of VDR gene. These polymorphic regions were amplified by standard unlabeled oligonucleotides followed by restriction enzyme digestion corresponding to each RFLP.
Results:

- These polymorphisms for the case-control association studies were conform to Hardy-Weinberg equilibrium expectation ($p > 0.05$) in both case and control groups.
- For FoqI polymorphism, distribution of genotype frequencies and allele frequencies do not differ significantly between all cancer patients and control (p 0.3983, respectively p 0.6113).
- For ApaI polymorphism, distribution of genotype frequencies and allele frequencies do not differ significantly between all cancer patients and control (p 0.2770, respectively p 0.4811).
- For TaqI polymorphism, distribution of genotype frequencies and allele frequencies do not differ significantly between all cancer patients and control (p 0.1095, respectively p 0.4118).

Conclusions:

- In our study, we do not find any association between analyzed polymorphisms and colorectal cancer patients.
- The role of the VDR gene polymorphisms should be further studied, by increasing the power of the study and the number of polymorphisms.