mon overrepresentations were seen at 8q (11 cases), 4q (9 cases), 7q (8 cases:), 5p (7 cases), and 1p (8 cases). The smallest regions of overlap were narrowed down to 8q23 (10 cases), 4q12–13 (8 cases), 5p13–14 (7 cases), 7q31–32 (7 cases), 8q21 (7 cases), and 4q28–31 (5 cases). This data demonstrates that a number of chromosomal regions and even two distinct loci on 4q and 8q are involved in the pathogenesis of OS. More than 40% of the OS samples displayed low to moderate amplification of the MYCC oncogene, verified by FISH. Interestingly, amplification of the MYYC oncogene had no adverse prognostic impact in the OS cases studied.

735 POSTER

EWS-FLI1 gene rearrangement and CD99 positivity identify a breast tumor as a PNET

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Rearrangements of the *EWS* gene with ETS transcription factor genes as a result of chromosomal translocation and high expression levels of $CD99^{MIC2}$ characterize the Ewing family of tumors which usually affects bone and soft tissue in children and young adults. We report on a case of a $CD99^{MIC2}$ positive small round cell tumor m the breast of a sixty year old woman in which by cytogenetic analysis a t(11;22)(q24;q12) chromosomal aberration was identified. Reverse transcriptase polymerase chain reaction (RT-PCR) followed by sequence analysis revealed expression of a chimeric transcript in which *EWS* Eggs exon 10 was fused to *FLI1* exon 6. The specific gene rearrangement of *EWS* intron 10 was confirmed on Southern blots of genomic DNA. This case further contributes to the growing list of unusual neoplasms in adults that carry genotypic and phenotypic traits of the Ewing family of tumors. After mastectomy, adjuvant chemotherapy was performed with VACA protocol consisting of vincristine, adriamycin, cyclophosphamide and acfmomycin D with G-CSF support and radiotherapy with 50 Gy was given to the thorax wall. The patient has been tumor-free for 1 year now.

736 POSTER

Radiation-induced chromosome aberrations in two cell types of healthy donors and breast cancer patients

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Purpose: In recent studies different DNA-repair deficiencies were identified in lymphocytes of certain cancer patients. Defects in DNA-dsb-repair processes can be detected by analyzing chromosome aberrations after in vitro irradiation. We compared radiation-induced aberrations in lymphocytes and fibroblasts of healthy donors and breast cancer patients.

Material and Methods: Plateau-phase skin fibroblasts and blood G0 lymphocytes obtained from healthy donors or from 5 breast cancer patients were irradiated in vitro with a test dose of 3 Gy of 200 kV X-rays. The genomic yields of dicentrics, acentrics and the partial yields of reciprocal translocations (FISH-method, #4, #7, #9) were scored in 1st and 2nd post-irradiation metaphases.

Results: With respect to reciprocal translocations we found no differences neither between the two cell types, nor between the groups of healthy donors and breast cancer patients. Dicentric chromosomes were slightly increased in fibroblasts from the cancer patient group. Acentric fragments associated with chromosome deletions were significantly increased in both cell types of the cancer patient group.

Conclusion: In agreement with the reported increased levels of chromatid breaks or micronuclei in lymphocytes of certain cancer patients we observed increased levels of deletions in five breast cancer patients. In these patients, increased levels were also measured in skin fibroblasts. These findings indicate that at least two cell types from breast cancer patients display an increased level of unrepaired DNA-dsb's.

737 POSTER

Susceptibility to breast and ovarian cancer: The role of glutathione S-transferase polymorphism

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Purpose: Polymorphism[Γ]s in many xenobiotic metabolizing enzymes occur leading to variation in the level of enzyme expression in vivo. We hypothesize that women who carry deletions (null genotype) in glutathione-s-transferase genes, GSTM1 and GSTT1, may be more susceptible to the effects of environmental carcinogens than women who carry wild type (wt) alleles.

Methods: We studied this genotypes in a total of 105 cases, 85 cases with breast cancer (BC) and 20 cases of women with ovarian cancer (OC), using a differential polymerase chain reaction to simultaneously characterize inactivating mutations responsible for the null alleles of GSTM1 and GSTT1. We also studied 123 healthy controls (HC).

Results: 43 of 85 (50.6%) of BC were GSTM1 null, frequency not significantly different than HC (58%). 24 of 83 (28.9%) of BC were GSTT1 null and not significantly different than the frequency in HC (32.7%). When stratified by age at diagnosis we found a frequency of 46.6% in women diagnosed with BC after age 40. However, in women diagnosed with breast cancer before the age of 40, a tendency for a higher frequency of GSTM1 null genotypes (8 of 10 or 80%) was found and the trend for the differences between the two age groups was significant ($p = 0.048$, Fisher exact test). No association was found for the GSTT1 null genotype. In OC cases, GSTM1 null genotypes were found in 45.0% and GSTT1 null genotypes in 50.0%. Comparison of frequency distributions did not show significant differences from the HC.

Conclusion: Our results may suggest that the alteration in the metabolic pathways of xenobiotics may be associated with an earlier onset of breast cancer. The lack of the glutathione S-transferase M1 (GSTM1 null alele) did not appear to influence susceptibility to BC or OC. Further studies are necessary to understand if there are some differences in the tumor behavior or in the response to chemotherapeutic agents.

738 POSTER

Cytogenetic characteristics of 81 cases of human thyroid tumours

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Thyroid tumors, especialy benign ones like coloido-nodular goiters and follicular adenomas have been rarely analysed genetically. The results of cytogenetic analysis 41 coloido-nodular goiters, 33 follicular adenomas and 7 cancers (2 follicular, 2 papillary and 3 anaplastic) are presented. The method of short-term in vitro tissue culture were used. The chromosomes were G-band stained, In 7 colloido-nodular goiters single clonal structural chromosomal rearrangements were found. In 13 out of 33 follicular adenomas more complex rearrangements were detected – 13 numerical or structural In benign tumor thyroid tumours characteristics chromosal aberrations were not found. All thyroid cancer displayed the presence of sturtural aberration. The number of more complex rearrangements were higher in anaplastic cancers than in follicular and papillary ones. Structural chromosomal abberations of long arms of chromosome 4 were detected in 2 anaplastic and in 1 follicular cancer.

739 POSTER

Familial neoplastic clustering in 81 gastric cancer patients

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Gastric cancer (GC) is rarely due to a genetic syndrome (FGP, HNPCC), but many studies revealed that 15% of GC patients has a positive familial history for GC.

Methods: We asked 81 consecutive italian GC pts (52 male, 29 female; mean age 57) their family history and identified the cases of cancer family syndromes (CFS): FGP; HNPCC; other unspecified. Among the relatives of pts without CFS we assessed the total number of subjects affected by cancer at any site and analized the differences of the number of neoplasms occurred in the families after stratifying the sample by age (pts < 50 vs pts

> 50 years), by histology (intestinal vs diffuse type) and by gastric site of disease (cardial vs body and pylorus neoplasms) of the pts.

Results: We found 4 cases of CFS (of which only in one case a HNPCC was suspected). Among the 1502 relatives of the other 77 pts there were 101 neoplasms: of these, 22 were GC, 12 breast cancer, 9 HCC. No significative differences in the number of affected relatives was detected when the sample was stratified by age (p: 0.70) and by histology (p: 0.95), while the number of affected relatives was higher for pts with a cardial neoplasm (p: 0.05).

Conclusion: Genetics of GC remains largely unknown. Despite the reduction of GC incidence, cardial tumor is increasing and an inherited predisposition could play an important role in its aetiology.

740 PUBLICATION

Absence of point mutations on serine 17 of MDM2 in human primary tumors

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Purpose: Studies *in vitro* have demonstrated that the mutations of the Serina 17 of Mdm2 protein, blokcs its phosphorilation for a serin-treonin kinase that regulates the interaction between Mdm2 and p53. The present study investigates the occurrence of mutations at this corresponding Mdm2 gene position, somaticaly in human tumors and in germline in patients with multiple primary tumors, as a possible mechanism for which the tumoral cells can escape to this regulation.

Methods: DNA extracted from tissue samples provenient of 192 cases of different tumor types: 70 breast carcinomas, 14 bladder tumors, 18 colon cancers, 60 testicle tumors and 30 samples of peripheral mononuclear blood cells of patientes with three primary different tumors, were analyzed by PCR-SSCP method and electrophoresed on polyacrylamide gels. To corroborate the PCR-SSCP results, twenty tumor DNA and five blood DNA samples were selected randomly and also checked by direct sequence of the third exon of Mdm2, independently of the normal pattern of SSCP.

Results: In none of the cases studied, tumor and blood samples, electroforetic mobility shifts of bands after SSCP were observed. No mutations at this position (Ser-17) in the twenty-five cases sequenced, were observed.

Conclusion: Mutations of Mdm2 gene, corresponding at Ser-17, implicated in phosphorilation process of Mdm2, were not demonstrated in human tumors, somaticaly neither at germline level. This fact suggest that mutations at this position of Mdm2 gene is not a pathogenic mechanism of tumorigenesis in some common type of human cancers.

741 PUBLICATION

Study of L-MYC polymorphism in elderly tumour-free individuals, healthy donors, and cancer patients

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Purpose: The earlier reports provided evidence that S allele of L-MYC oncogene may be associated with tumour-resistant genetic constitution. The aim of the study was to assess the involvement of L-MYC polymorphism in the cancer tolerance and susceptibility.

Methods: L-MYC alleles were identified by PCR in 184 elderly tumour-free individuals, 122 healthy middle-aged donors (HD) as well as in 95 breast cancer (BC), 63 colorectal cancer (CC) and 58 lung cancer (LC) patients.

Results: Contrary to previous publications, L:S allele frequencies ratio in elderly donors (ED) did not significantly differ from that in HD (0.49:0.51 and 0.54:0.46, respectively). However, S allele was slightly overrepresented among elderly smokers as compared to middle-aged ones (55% vs. 44%; P = 0.059; OR = 1.57 (0.98–2.50)), that implies it may be linked with smoking-tolerant genetic constitution. Whereas CC and LC were strikingly similar to the controls, specific features were observed in the BC patients. Occurrence of S allele in BC cohort (57%) was significantly higher than in middle-aged healthy females (41%; P = 0.016; OR = 1.92 (1.13–3.25)) and elderly non-affected women (47%; P = 0.050; OR = 1.54 (1.00–2.37)).

Conclusions: 1) S allele of L-MYC indicates an increased risk of breast cancer development; 2) the same allele may be associated with better longevity among smokers.

742 PUBLICATION

DCC and p53 protein expression in early stages of gastric cancer

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Purpose: Oncogenes and tumor suppressor genes may be involved in the pathogenesis and the progression of gastric cancer (GC). We investigated the expression of the p53 and the DCC proteins in the early stages of the disease.

Methods: An immunohistochemical analysis for p53 and DCC was performed on consecutive tumor tissue samples from patients (pts) with stage pT1-2 GC. DCC positivity expresses a normal gene function and its staining is reported an "all or nothing" phenomenon. Nuclear accumulation of p53 is commonly related to gene point mutations and a cut-off value of 30% was selected for positivity.

Results: Thirty-eight consecutive cases of GC were analyzed. All the pts underwent radical surgery with a median time of follow-up of 60 months. Disease stage was pT1 in 23 cases and pT2 in 15 cases. Lymphnode metastases were present in 17 cases. DCC positivity was found in 33/38 cases (87%) and p53 positivity in 23/38 cases (60%). A significative higher number of p53 positive cases was found among pT2 cases (11/15; 73%). Preliminary data on 9 relapsed pts showed a high frequency of p53 positivity (7/9).

Conclusions: The detction of wild-type DCC protein in the majority of cases suggests that mutations of this gene are not early events in the genesis of GC. On the contrary, p53 gene mutations with accumulation of its abnormal protein may be related to GC development and invasiveness.

743 PUBLICATION

Identification of HLA class II in patients with endometrial adenocarcinoma (EA)

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Purpose: We determined the distribution of HLA class II alleles in patients (pts) with EA comparing with healthy control group to find the correlation between the phenotype frequency of HLA class II alleles and early risk of developing of EA in North-West Russian population.

Methods: We typed by PCR-SSP technique (HLA-DRB1*01–16, DQB1*0201–0608, DQA1*0101–0601 alleles) of 37 pts with EA and 78 healthy subjects.

Results: The phenotype frequencies of the DRB1*03 and DQB1*0601 alleles were increased in pts with EA (29.7% and 16.2%) compared with healthy subjects (12.8% and 5.1%), respectively; x2 = 4.81 and 3.89, $p < 0.05$. The phenotype frequencies of the DRB1*01, DRB1*04 and DQB1*0301 were decreased in pts with EA (13.5%, 5.4% and 10.8%) compared with healthy subjects (32.1%, 26.9% and 37.2%), respectively; x2 = 4.47, 7.26 and 8.53, $p < 0.01$. In group of patients with EA there was no alleles of HLA-DQA1*0401, but x2-test was not significant (3.003, $p < 0.1$) compare with control group.

Conclusion: We suppose that increased phenotype frequencies of HLA-DRB1*03 and DQB1*0601 alleles are correlated with early risk of developing of EA in North-West Russian population.