1.36, 95% CI = 1.13-1.62, P = 0.001). In stratified analyses, this combined effect was more evident among premenopausal women, ER and/or PR positive women, and subjects with an older age at first live birth. Furthermore, there is a significant additive interaction between risk genotypes and menopausal status (P for multiplication interaction/additive interaction: 0.083/0.037).

Conclusions: These findings showed that genetic variants in FGFR2 may contribute to the susceptibility of breast cancer in Chinese women, possibly through mediating estrogen/progesterone related pathways.

179 Poster

Detection of novel biomarkers by plasma proteomic profiling of oesophageal cancer mouse xenografts using three human cell lines in response to chemotherapy

P. Kelly[1], V. Appleyard[1], K. Murray[1], F. Paulin[1], S. Fong[1], A. Thompson[1]
[1]University of Dundee, Surgery and Molecular Oncology, Dundee, United Kingdom

Background - Oesophageal cancer is the 9th most common malignancy worldwide with poor survival rates and an increasing incidence in recent years. Use of neoadjuvant chemotherapy in locally advanced cancer prior to surgery has been shown to improve outcomes, but the response to therapy is variable. Hence, the effective use of chemotherapy could be greatly improved by the availability of biomarkers that predict response to therapy. The purpose of this study was to identify candidate biomarkers in mouse xenograft models of oesophageal cancer. Materials and methods - OE19, OE21 and OE33 xenografts were established in SCID immune-deficient mice and tumour growth rates recorded. A clinical dose of epirubicin, cisplatin or 5-fluorouracil was administered to xenografts/or controls, by once weekly peritoneal injection for up to 3 weeks. Plasma collected from treated and untreated xenografts/ controls was analysed by SELDI-TOF MS using Ciphergen CM10 (weak cationic) and Q10 (strong anionic) protein chips. Panels of markers were identified that distinguish between treatment groups using pattern recognition software and class prediction tested using k-nearest neighbours and support vector machine algorithms. Samples containing statistically significant markers were fractionated on anion exchange spin columns and peaks of interest identified by MS/MS analysis of SDS-PAGE gel pieces.

Results - Tumour growth was suppressed in treated compared with untreated xenografts. Protein peaks (m/z) were identified that differed significantly ($p<0.05$) between the treatment groups for each cell line with each drug. Peaks were identified that were both common and unique to each cell line and drug combination. Biomarker panels could correctly identify xenograft versus control or predict drug treatment in 95% or 70% respectively of a test set (n=20) using the support vector machine. Two statistically significant peaks (m/z 28303 and 29162) were identified by MS/MS as apolipoprotein A1.

Conclusion - These experiments have established a response to chemotherapy in oesophageal xenografts by proteomic profiling of plasma. Biomarker panels have been identified that can accurately distinguish xenografts from controls or between treatment groups. Identification of the proteins in these peaks is in progress, with one protein identified to date. A follow-up clinical study is being established.

180 Poster

Genotypes and haplotypes in the insulin-like growth factors, their receptors and binding proteins in relation to plasma metabolic levels and mammographic density

M. Biong[1], I. Brill[2], F. Johansen[1], Y. Bremnes[3], L. Burdette[4], M. Yaeger[4], G. Ursin[5], I.T. Gram[3], V. Kristensen[1]
[1]The Norwegian Radium Hospital, Department of Genetics, Oslo, Norway;
[2] University of Alabama, Department of Epidemiology, Birmingham, USA;
[3] University of Tromsø, Institute of Community Medicine, Tromsø, Norway;
[4] National Cancer Institute, Core Genotyping Facility, Bathesda, USA;
[5] University of Oslo, Institute of Nutrition Research, Oslo, Norway

Increased mammographic density is one of the strongest independent risk factors for breast cancer. It is believed that one third of breast cancers are derived from breasts with more than 50% density. Variation in breast density may be due to lifestyle factors such as alcohol intake and diet as well as polymorphisms in genes involved in steroid hormone biosynthesis, metabolism and signalling genes.

Exposure to endogenous and exogenous steroid hormones and growth factors has been linked to increased mammographic density and breast cancer risk. Insulin-like growth factor 1 (IGF1) is one such growht factor. IGF1 is a mitogen in various cell types, and predicted to be involved in the development of several human cancers, including breast cancer. Furthermore the circulating levels of IGF1 are strongly associated with breast density. The levels of IGF1 are related to age and menopause status, young women tend to have higher levels of IGF than postmenonpausal women. In this study we have looked at the involvment of genetic polymorphisms in the IGF genes in postmenopausal women, and their influence on mammographic density and ciculating levels of IGF, the binding protein IGFBP3, and their ratio.

Samples from 964 postmenopausal women were genotyped on ABI 7900HT. The statistical analyses of the genotypes were perfomed by the use of SAS/GENETICS™ (SAS 9.1.3) and PROC HAPLOTYPE.

The haplotype analysis yielded six haploblocks within the genes IGF1, IGF2, IGF1R, IGF2R, IGFals and IGFBP3. Of the six haploblocks, four had significant associations to the parameters IGF level, IGFBP3 level and mammographic density. In IGF1 one haplotype variant is associated with mammographic density. Within the IGF2 gene one haplotype variant is associated with the level of both IGF1 and IGFBP3. The IGF2 receptor had two haplotype variants associated to the levels of IGF1. Both variants of the IGFBP3 haplotype are associated with the level of the IGFBP3 level and indicate cis regulation.

These result shows that polymorhisms within the IGF gene itself and related genes have an impact on IGF1 and IGFBP3 levels and are associated with mammographic density in postmenopausal women.

POSTER SESSION

Carcinogenesis

181 Poster

The effect of p53 on the DNA repair enzyme Thymine-DNA Glycosylase

N. Meireles da Costa[1,2], A. Hautefeuille[1], M. Pierre Cros[1], L.F. Ribeiro Pinto[2], P. Hainaut[1]
[1]Molecular Carcinogenesis and Biomarkers Group, International Agency for Research on Cancer (WHO), Lyon, France; [2]Toxicology and Molecular Biology Laboratory, Department of Biochemistry, University of the State of Rio de Janeiro (UERJ), Rio de Janeiro, Brazil

Esophageal cancer is a highly frequent and fatal malignancy presenting a remarkable geographical variation in its incidence worldwide. A characteristic of high incidence areas is the large proportion of double mutations at CpG sites of TP53 gene. The relevance to the cancer problem comes from the possibility that this cancer may be associated with acquisition of a defect in DNA repair - specifically in the DNA repair enzyme Thymine-DNA-Glycosylase (TDG) which is responsible for this repair - following an initial mutation in TP53, according to the concept of a “mutator phenotype”. The aim of this work is to analyse the effect of p53 on the expression and activity of TDG. TP53 was inhibited by using small interfering (si) RNA in TE-1 (esophageal cell line wich presents a temperature-sensitive mutation in TP53 resulting in inactive protein when cultured at 37°C and active at 32°C); increase of p53 levels was induced by transfecting an expression vector in TE-13 (a p53-deficient esophageal cell line); DNA damage was achieved by treating MN1 and MDD2 (derived from MCF-7 breast cancer cell line. MDD2 expresses an inhibitory peptide that blocks p53 oligomerization and MN1 is transfected with the same peptide without the coding sequence) with doxorubicin. Levels of TDG mRNA and protein expression were analysed by Quantitative - RT PCR and Western blotting, respectively. Moreover, DNA damage was induced in TE-1 by treatment with Methyl methanesulfonate (MMS). Analysis of expression and subcelular localization of TDG followed the treatment with MMS in TE-1 and doxorubicin in MN1 and MDD2 and were accessed by Confocal Microscopy. In TE-1 cells, a decrease of 80% was detected in cells kept at 37°C compared to cells at 32°C. Treatment with p53 siRNA decreased as the TDG mRNA levels by 45% at 32°C when compared to control. The transfection of 1,0 g of p53 expression vector in TE-13 increased (4,6-fold) the levels of TDG mRNA. TDG protein levels showed the same pattern with an induction following transfection. Doxorubicin treatment resulted in 2,2 - fold induction in MN1 TDG mRNA levels but not in MDD2 cells. Surprisingly, TDG was detected in the cytoplasm of TE-1 cultured at 37 ºC and 32°C. After the treatment with MMS, TDG was found into the nucleus of the samples treated and kept at 32°C, but not at 37°C. MN1 and MDD2 showed a similar result, presenting TDG in the cytoplasm and its migration into the nucleus after the treatment with doxorubicin in MN1 samples, but not for MDD2. The results suggest a role of active p53 on the expression and in the migration of TDG from cytoplasm to the nucleus and in its activity. These results provide evidence that wild-type p53 may regulate TDG activity, and that this property is lost after functional inactivation of p53 in cancer cells.