OT with worst prognosis presented increased levels of Phosphocholine, Choline, Fatty acids and Alanine. In the same way OT harboring 1p/19q ROH present higher glutathione levels.
Conclusions: Each tumour is singular but alterations detected in this study depict the genetic landscape for oligodendroglial tumours and could reveal the divergent response showed by these molecular subgroups in survival and chemotherapy treatment.

808 Comparative proteomic study of multidrug resistance in chronic myeloid leukemia

S. Corrêa[1], L. Pizzatti[2], B. DuRocher[2], E. Abdelhay[2]. [1]*Universidade Federal do Rio de Janeiro, Ibccf, Rio De Janeiro, Brazil,* [2]*Instituto Nacional de Câncer, Cemo, Rio De Janeiro, Brazil*

Background: Chronic myeloid leukaemia (CML)'s treatment has improved with the advance of Imatinib mesylate (Glivec®, IM, Norvatis). IM is a tyrosine's kinase inhibitor for CML biomarker, the BCR-ABL oncoprotein. Despite this improvement, BCR-ABL dependent and independent mechanisms of IM therapy's resistance are known to occur. The latter has been associated to multidrug resistance (MDR) phenotype emergence. MDR is known as the major cause of failure in cancer treatment, and it is most related with the expression of ABC transporters, such as P-glycoprotein (Pgp – *ABCB1*). Although the identification and the knowledge of ABC transporters, the resulting pathways in drug resistance in leukemic cells remain uncharacterized. In the present work, we investigated the possible relationship between MDR and resistance to IM therapy in CML.
Material and Methods: We screened drug transporters and *BCR-ABL* RNA transcripts levels, by real time Q-PCR, in the multidrug resistant cell line Lucena (K562/VCR) and verified its cellular viability, apoptosis and cell cycle after IM treatment. Then, we compared its proteomic profile to the parental cell line K562. Proteomics results were validated *in vivo* by real time Q-PCR and multivariate statistical analysis were applied.
Results: Our results demonstrate that MDR cell line Lucena has a resistant pattern to IM treatment. The proteomic approach resulted in identification of forty-six differentially expressed proteins. Among them, *LRPPRC, MCM7* and *RBM17*, jointly with *ABCB1* gene, were validated in fourteen CML patients and six donors. We found, through multivariate statistical analysis that, altogether, they were able to categorize patients' status as responsive or resistant to IM therapy.
Conclusions: By the data presented in this work, we showed that MDR can be closely attached to IM's failure, demonstrating its importance as a CML's prognostic factor. Moreover, the proteomic approach pointed out some new possibly markers associated in MDR phenotype, which could lead to additional information of this phenomenon and clinical improvement for MDR detection in patients.
Financial support: CAPES, CNPQ, MS.

809 Prediction of lymph node metastases in small T1 breast cancers by expression profiling

A. Lee[1], J.M. Chua[1], P. Tongyoo[1], W.S. Yong[2], G.H. Ho[2], H.N. Koong[2], P. Ang[3], B.T. Teh[1], Z.W. Wong[3]. [1]*National Cancer Centre Singapore, Division of Medical Sciences, Singapore, Singapore,* [2]*National Cancer Centre Singapore, Department of Surgical Oncology, Singapore, Singapore,* [3]*National Cancer Centre Singapore, Department of Medical Oncology, Singapore, Singapore*

Background: The principal cause of mortality in breast cancer is distant metastases. Tumour size and lymph node positivity were classically used as prognostic factors for relapse risks. However, up to 30% of lymph node negative patients eventually develop metastases. We aimed to study breast cancers less than or equal to 2 cm where metastases to regional lymph nodes is generally uncommon, and hypothesised that these small tumours have acquired the ability to metastasise as an early event in oncogenesis.
Methods: Fresh frozen tissues from breast tumours that were $\leqslant$2 cm, positive (n = 23) and negative (n = 42) for lymph node metastases were expression profiled using the Illumina HumanWG-6 v3.0 Expression BeadChips.
Results: Distinct differences in the expression profiles between oestrogen receptor positive (ER+) tumours as compared to the oestrogen receptor negative (ER−) tumours were obtained by unsupervised clustering. As such, we subsequently performed supervised clustering on ER+ tumours (n = 47) and ER− tumours (n = 16) as separate subgroups using genes that were differentially expressed and with *P* values of <0.05. Our analysis showed segregation of breast cancers that were lymph node positive from those that were lymph node negative in ER− tumours. There were four HER2 positive tumours (defined as having immunohistochemical staining of 3+ or positivity with fluorescence *in situ* hybridisation) in this ER− subgroup. By selecting only genes that had at least 2-fold differences between the node negative and positive tumours, we identified 53 differentially expressed genes which were mostly involved in signal transduction, cell communication, metabolic processes and response to stimuli. Of these 53 genes, 13 were downregulated and 40 were upregulated in those with lymph node metastases.
Conclusion: Our results suggest that in ER− breast cancer, it may be possible to discriminate patients with or without lymph node metastases using gene expression profiling. The details of the differentially expressed genes will be presented at the meeting. We will perform further validation on an independent set of breast tumours.

810 Differential enrichment of pathways in association with TP53 mutation status of breast cancers

H.N. Joshi[1], A. Langerød[2], O.G. Troyanskaya[3], A.L. Børresen-Dale[2], V.N. Kristensen[1]. [1]*Faculty of Medicine University of Oslo, Institute of Clinical Epidemiology and Molecular Biology (EPIGEN), Lørenskog, Norway,* [2]*Institute of Clinical Medicine University of Oslo, Department of Genetics Institute for Cancer Research Division of Surgery and Cancer Oslo University Hospital Radiumhospitalet, Oslo, Norway,* [3]*Princeton University, Department of Computer Science, Princeton NJ, USA*

Background: Various studies have so far tried to explain the biology of breast cancer associated with TP53 mutation status in terms of differential expression of genes. Keeping in view that altered functions of many genes belonging to specific biological pathways might result in a particular tumour phenotype, we attempt to infer the association of key pathways related gene sets that might play important role in development of breast cancers with wild type or mutant TP53 status.
Material and Methods: A single expression dataset based on Agilent whole genome microarrays platform, consisting of total 111 samples with 73 wild-type TP53 and 38 mutant TP53 status, was analysed by fitting a regression model as proposed by (Goeman *et al*., 2004, 2009) with gene expressions being the covariates and phenotypic data (TP53 mutation status of breast cancer) as the response variable. We also applied iterative signature algorithm (Bergmann *et al*., 2003, Csárdi 2009) in order to identify modules with enrichment of specific key pathways in our dataset.
Results: Using multiple test-corrected p-values based ranking, we identified top 20 important biological pathways (KEGG) and their associated genes. We also studied the extent of inter-sample similarity in pathway representation. Apart from p53 signaling pathway, we found differentially expression of purine metabolism; glycine, Serine and threonine metabolism; prostate cancer and vitamin B6 metabolism pathways. The biclustering analysis identified a module showing differential enrichment of key pathways − p53 signaling pathway, cell cycle and DNA replication and differential co-expression of corresponding genesets. Another module identified differential enrichment of immune response related pathways – such as cytokine-cytokine receptor interaction, T cell receptor signaling pathway, natural killer cell mediated cytotoxicity pathway.
Conclusions: Our findings from biclustering algorithm add valuable information to the findings from regression model-based pathways analysis and provide new insights about the potential pathway alterations that might be responsible for breast cancer development in association with TP53 mutation status. However, it remains to be established, which alterations are responsible for the cancer development and which alterations are consequences or mere associations to the TP53 mutation status. Therefore, we propose more detailed studies aiming at investigation of the association and possible role of certain pathways, such as purine metabolism and vitamin B6 metabolism pathway, immune response related pathways including natural killer cell mediated cytotoxicity pathway – in breast cancer development.

811 8q24 amplification in metastatic endometrial cancer

D. Konstantinova[1], H. Edvardsen[2], V. Mitev[3]. [1]*Medical University Sofia, University Hospital of Obstetrics and Gynaecology "Maichin Dom" National Genetics Laboratory, Sofia, Bulgaria,* [2]*Institute for Cancer Research Oslo University Hospital Radiumhospitalet, Department of Genetics, Oslo, Norway,* [3]*Medical University Sofia, Department of Medicinal chemistry and Biochemistry, Sofia, Bulgaria*

Amplification of 8q24 is a hallmark of metastatic cancer. The target genes of 8q24 somatic amplification have not been precisely established. So far, investigations on different (but also on the same) cancer types have produced varying results. The *EIF3S3* and *ASAP1* have been proposed as targets in prostate cancer, the *PTK2* and *EIF3S3* – in hepatocellular carcinoma, the *BOP1* – in colorectal cancer, etc. The *MYC* and *PRL3* are two of genes in this region, known to be overexpressed in many cancer types, however this has not always been associated to gene amplification.
Endometrial tumours are particular for their paucity of genomic amplification, however overrepresentation of the 8q24 region has been described in metastatic endometrial cancer (EC). We compared the amplification profiles of 5 metastatic (MEC) and 20 metastasis-free EC samples (NMEC) by using Illumina 660K SNP-array. Tissue samples were obtained at the University Hospital of Obstetrics and Gynaecology "Maichin Dom", Bulgaria. All patients gave informed consent.

The only region of amplification that differed significantly between MEC and NMEC was 8q24. It was amplified in 2/5 (40%) of MEC versus none (0%) of 20 NMEC tumours (P = 0.03). The region started at 8q24.12, upstream of *EIF3H*, and ended at 8q24.13, downstream of *MYC*. It encompassed the *DEPDC6* (upstream) to *HAS2* (downstream) and contained *COL14A1* (collagen, type XIV), *MTBP* (Mdm2-binding protein, which stabilizes MDM2 and in this way increases p53 degradation) and *SNTB1* (syntrophin, beta 1, which is a dystrophin binding protein).
Of the putative targets of 8q24 amplification identified, the *HAS2* gene merits particular attention. It encodes hyaluronan synthase, and has been found overexpressed in many tumour types. The resulting high concentration of hyaluronan has been used as a tumour marker as its direct measurement in urine and serum samples has shown very good predictive values for cancer detection and grading. Also, in vitro silencing of the *HAS2* gene has reversed the aggressive potential of cancer cells and is hoped to entice pharmacological potential. The gene also encodes for a *cis*-antisense mRNA (HAS2AS gene), which regulates HAS2 transcription. Thus, if confirmed as a target of 8q24 amplification, the mechanism by which this increases cancer growth should also be elucidated. Further investigation of the significance of the 8q24 genes for EC aggressiveness is warranted. If confirmed, their amplification could reveal new knowledge on the mechanism behind the metastatic process.

812 Transcriptional profiling of early onset colorectal cancer identifies CLC as a potential cancer susceptibility gene

T.H. Ågesen[1,2], M. Berg[1,2], L. Cekaite[1,2], E. Thiis-Evensen[3], M. Merok[4,5], A. Nesbakken[4,5], R.A. Lothe[1,2], R.I. Skotheim[1,2]. *[1]Department of Cancer Prevention, Institute for Cancer Research, Oslo University Hospital, The Norwegian Radium Hospital, Oslo, Norway, [2]Centre for Cancer Biomedicine, University of Oslo, Oslo, Norway, [3]Medical Department, Oslo University Hospital, Rikshospitalet, Oslo, Norway, [4]Department of Gastrointestinal Surgery, Oslo University Hospital, Aker, Oslo, Norway, [5]Faculty of Medicine, University of Oslo, Oslo, Norway*

Background: Genetic predisposition is estimated to be the cause of colorectal cancer (CRC) in up to 30% of all cases, including the known inherited CRC syndromes (*e.g.* Lynch syndrome and familial adenomatous polyposis) that accounts for only about 5%. The incidence of CRC increases with age and an early onset of disease is an indication of genetic predisposition. In an attempt to identify cancer susceptibility genes, whole genome transcriptional differences between samples from patients diagnose with CRC at an early age and samples from sporadic/late onset CRC were compared.
Material and Methods: CRC samples were obtained from hospitals in the South-East region of Norway. In total, 24 primary tumours from patients diagnosed at an early age (mean 43 years), 17 sporadic tumours from elderly patients (mean 79 years) and four normal mucosa samples were included. All tumours were microsatellite stable, and samples from both genders and from the different Dukes' stages were represented equally. Applied Biosystems AB1700 microarrays were used, which enables measurements of gene expression using 32,878 unique probes.
Results: Principle component and cluster analysis showed an equal overall expression profile when comparing the early and late onset tumours. Nevertheless, although not all reaching statistical significance, we identified 20 protein coding genes differentially expressed in the early onset tumours compared to those with a late onset. *CLC* was the overall most significant gene with an increased expression in the early onset samples. Gene Set Enrichment Analysis identified chromosome band 19q13 as the most significant region with an enrichment of genes with an increased expression in the early onset samples, a region that includes *CLC*. Supporting these findings, the expression data has in parallel been integrated with corresponding DNA copy number data and chromosome band 19q13.2 was one of the loci identified with concomitantly DNA copy number gain and increased mRNA expression (Berg *et al.*, unpublished).
Conclusions: Minor differences were found when comparing the overall transcriptome profiles of early and late onset CRC. Nevertheless, we have identified several genes which serve as potential cancer susceptibility genes warranting further investigation in the continuing search for inherited genetic alteration in CRC.

813 The scaffolding adaptor GAB2 promotes anchorage independence and drives a transcriptional program associated to metastatic progression of breast cancer

E. Medico[1], C. Isella[1]. *[1]Institute for Cancer Research and Treatment (IRCC), Laboratory of Oncogenomics, Candiolo, Italy*

Background: The ability to grow in the absence of anchorage to the extracellular matrix represents a key oncogenic property of cancer cells. To screen for genes conferring anchorage independence, we exploited a novel functional genomics approach recently developed in our laboratory.
Material and Methods: The screening was based on transduction of MCF10A human breast cells with a murine retroviral expression library. Transduced cells were then selected for anchorage independence by culturing them on polyhema-coated dishes. Exogenous cDNAs enriched after selection were identified by one-shot, species-specific quantitative tracing with murine DNA microarray analysis before and after selection.
Results: Independent infection-selection experiments highlighted significant and reproducible enrichment for murine cDNAs encoding the Gab2 protein, suggesting a role for this scaffolding adaptor in anchorage-independent growth. Gab2 was confirmed to strongly promote anchorage-independent growth when overexpressed. Such effect did not involve protection from detachment-induced apoptosis, but rather the maintenance of a proliferative status also in the absence of the consensus provided by integrin engagement. Interestingly, downregulation by RNA interference of endogenous Gab2 in neoplastic cells did not affect their adherent growth, but abrogated their growth in soft agar. Gab2-driven anchorage independence was found to specifically involve activation of the Src-Stat3 signaling axis. A transcriptional "signature" of 205 genes was obtained from GAB2-transduced, anchorage-independent MCF10A cells, and found to contain two main functional modules, respectively controlling proliferation and cell adhesion/migration/invasion. Notably, the signature was enriched in genes discriminating responsiveness of breast cancer cell lines to Dasatininb, a Src-family kinase inhibitor. Extensive validation on breast cancer datasets showed that the Gab2-signature provides a robust prognostic classifier for breast cancer metastatic relapse, largely independent from existing clinical and genomic indicators and from estrogen receptor status.
Conclusions: This work highlights a pivotal role for GAB2 and its transcriptional targets in anchorage-independent growth and breast cancer metastatic progression. Moreover, it delivers a transcriptional signature capturing metastatic propensity of breast cancer with high sensitivity and accuracy.

814 Men genotyped for BRCA1/2 mutations: how does it affect them?

S. Shiloh[1], E. Dagan[2], I. Friedman[3], N. Blank[1], E. Friedman[4]. *[1]Tel Aviv University, Psychology, Tel Aviv, Israel, [2]Rambam Medical Center, Genetics, Haifa, Israel, [3]Sheba Medical Center, Onco-genetics, Ramat Gan, Israel, [4]Tel Aviv University and Sheba Medical Center, Human Genetics, Tel Aviv and Ramat Gan, Israel*

Background: Men who undergo genetic testing for harboring germline mutations in breast/ovarian cancer associated genes (*BRCA1*/*BRCA2*) represent an interesting and under investigated population. The reasons for testing are often a family history of either neoplasm or finding a mutation in a female relative. Testing for a "feminine" disease may have significant effects on men who are mutation carriers. The purpose of the study was to examine the cognitive, emotional and behavioral impacts that BRCA1/2 testing has on those tested men, by comparing carriers to non-carriers in a follow-up study.
Material and Methods: Fifty-one male carriers of a mutation in either the BRCA1 or BRCA2 genes were compared to a similar group of ethnically matched non-carriers on a questionnaire that measured changes in health behaviors since testing, within family communication about test results, risk perceptions, cognitive representations of breast cancer, and emotional reactions to test results. Participants filled-out the questionnaire in a telephone interview a few months after receiving test results and counseling in an oncogenetic clinic.
Results and Conclusions: Comparisons between carriers and non-carriers will be presented. Undergoing genetic testing for BRCA1/2 mutations may be experienced by some men as a threat to their manhood. Men found to be carriers may need support in adjusting to their genetic status and the impact that it has on their own health and that of their female offspring.

815 Fast statistical analysis of high density CGH and SNP arrays

G. Rigaill[1], T. Dubois[2], E. Barillot[3], S. Robin[4]. *[1]Institut Curie, AgroParisTech, INRA, Département de Transfert, MIA UMR 518, Paris, France, [2]Institut Curie, Département de Transfert, Paris, France, [3]Institut Curie, INSERM U900, Mines ParisTech, Paris, France, [4]AgroParisTech, INRA, MIA UMR 518, Paris, France*

Background: Array-based Comparative Genomic Hybridization (array-CGH) and Single Nucleotide Polymorphism (SNP) arrays are used for studying the genetics of cancer. Their bioinformatical and statistical analysis is a critical step to identify gained and lost regions containing potential oncogenes or tumour suppressor genes.
Material and Methods: The CGHseg method (Picard et al 2005), which uses a dynamic programming algorithm, was shown to be one of the best methods (Lai et al 2006) to analyze CGH arrays and detect DNA copy number alterations. However, its application to very high density CGH and SNP arrays measuring the DNA copy number on more than 1 million loci per patient was limited due to algorithm complexity.
Results: We have found shortcuts in the dynamic programming algorithm and have implemented an improved version of the CGHseg method. The new algorithm recovers exactly the same result as the previous one in a drastically