Results: From both mutation carriers and controls, three RT-PCR products were obtained: one corresponding to the full length transcript with the expected size (214 base pairs (bp)), another with 192bp corresponding to a deletion of 22bp of exon 5 (BRCA1-Δ22ntex5), and a third with 134bp corresponding to the in frame skip of exon 5 (BRCA1-Δex5). Semi-quantitative fragment analysis showed a relative amount of BRCA1-Δ22ntex5 more than eight-fold higher in patients and only the wild type allele was present in the full length transcript. The haplotype identified in the three Portuguese families and in the Galician family is compatible with a common origin of this mutation. The mutation segregates with the disease in the family with two affected members. Of the three breast cancers, one was an atypical medullary carcinoma and two were invasive ductal carcinomas with medullar features. All breast carcinomas were grade III and two of them were hormone receptor negative (data not available from the third case). No LOH was detected.

Conclusions: We conclude that disruption of alternative transcript ratios is the mechanism causing hereditary breast/ovarian cancer associated with the BRCA1 R71G mutation, and segregation and histopathologic data are consistent with its pathogenicity. Furthermore, our findings indicate a common ancestry of the Portuguese and Galician families sharing this mutation.

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Heat Shock Protein 60kDa in breast cancer tissue and cell lines

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Background: Breast cancer has been reported as the most common cancer of women in U.S.A., Western Europe and Korea. Breast cancer is curable with an early diagnosis, and many researchers have made efforts to find a marker for this malady. Heat shock protein (HSP) consists of 6 groups, it is highly preserved throughout both the prokaryotic and eukaryotic cells and it acts as a molecular chaperone that is involved in protein folding. HSPs have been recently reported to be related with breast cancer. In this study, we investigated the changes of expression of HSP60 in breast cancer tissues and cancer cell lines.

Materials and Methods: We obtained breast cancer tissues and normal tissues from twenty breast cancer patients, and we purchased several cancer cell lines from ATCC. We treated the human breast cancer tissues and cancer cell lines with heat shock protein. Proteins and mRNAs were isolated from the tissues and the cancer cell lines and then we performed Western bloting, RT-PCR and FACS on them.

Results: On Western blot, HSP60 was more overexpressed in the cancer tissue and the cancer cell lines than in the normal breast tissue and in the normal cell lines. The Expression of HSP60 showed 2 types of molecular weight differences in both the breast cancer tissues and the cancer cell lines, and specifically, low HSP60 was over-expressed in the cancer tissues. There was no difference between the expression of HSP60 protein and mRNA according to the treatment with heat shock protein in both the breast cancer tissue and the normal cell lines. Also, there was no relationship between phosphorylation and the structural difference of HSP60 protein according to HSP60 protein molecular weight.

Conclusion: We conclude that HSP60 may be used as a diagnostic marker for breast cancer. Detailed investigation of the usefulness and significance of the HSP60 expression as a prognostic factor is required in further studies.

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Is triple negative a prognostic factor in breast cancer?

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Background: Breast cancer is characterized by hormone dependency and endocrine therapy is a key treatment in breast cancer. Recently, targeted therapies such as Trastuzumab treatment for HER2 positive breast cancer has been important. Triple negative breast cancer is characterized by lack of expression of estrogen receptor (ER) and progesterone receptor (PgR), and the absence of HER2 protein overexpression, and so there is no targeted therapy for this subtype. In this study, we examined the biological and prognostic characteristics in triple negative breast cancer.

Patients and Methods: Between January 1998 and September 2006, 1552 patients with primary breast cancer were investigated retrospectively in this study and ER, PgR and HER2 status were evaluated in all cases. Furthermore, p53 overexpression and Ki67 values were examined immunohistochemically.

Results: Patient distribution according to ER, PgR or HER2 status were as follows; ER and PgR positive: 57.9% and ER and PgR negative: 25.1%. With regards to the HER2 status, HER2 positive was 23.3%, and triple negative (TN) was 14.0%. TN breast cancer has a high proliferation rate, high nuclear grade and frequent p53 overexpression. Patients with TN tumors had a significantly poorer disease-free survival (DFS) than those with non-TN tumors. After recurrence the overall survival (OS) rate in TN cases were significantly lower than that of the non-TN cases. Multivariate analysis revealed that TN was a significant factor for DFS and OS after recurrence.

Conclusion: TN breast cancer is a rare subtype and has a high proliferation rate, a high nuclear grade. p53 overexpression, and lower DFS/OS. To improve the prognosis of TN breast cancer, a new effective strategy needs to be developed.

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Simultaneous analysis of HER-2/neu gene amplification and protein overexpression in single cells of pleural and ascitic effusions from patients with breast and ovarian cancer

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Background: HER2/neu protein overexpression is found in 20−30% of breast cancers and correlates with poor clinical outcome. Patients are selected for anti-HER2/neu-therapy by examination of tumour specimens by IHC (immunohistochemistry) and FISH (fluorescence-in-situ-hybridisation). Good correlation between both methods has been found for score 1+ and 3+ samples, but not for score 2+ samples. Combined approaches using FISH and immunofluorescence on the same tumour specimen have been described. In this study we examined pleural and ascitic effusions with a method allowing simultaneous analysis of protein expression by IHC and gene amplification by FISH. We regarded the following aspects: 1. the frequency of HER2/neu protein expression and gene amplification in effusions. 2. the correlation between protein expression, gene amplification and chromosome 17 polyploidy.

Methods: We examined 35 effusions from patients with breast cancer (n = 31) and ovarian cancer (n = 4). The same cytospins were analysed by IHC using two anti-HER2/neu antibodies and by FISH with HER2/neu/CEP17 probes. Amplification was defined as: 1. HER2/neu gene copy number of >4 and 2. HER2/neu/CEP17 ratio $\geqslant$2.0.

Results: 35 tumour-cell-positive effusion specimens were examined. 25 of them were scored HER2/neu positive (score 2+, 3+). All of them contained cells with heterogeneous protein scores. Single cells were analysed for HER-2/neu gene amplification and chromosome 17 ploidy with regard to their scores. 9 of these 25 samples showed mean HER2/neu copy numbers of >4 in cells with a 2+ and 3+ score, but only 12% (n = 3) of these samples were amplified according to HER2/neu/CEP17-ratio. 32% (n = 8) were polyploid (mean CEP17 >4). In some samples we found tumour cells with gene amplification but without protein overexpression (score 1+) and cells without gene amplification but strong protein expression.

Conclusion: The combination of IHC and FISH allows a differentiated analysis of single cells, which is especially important for effusions that often contain heterogeneous cells. In this study only few samples showed HER2/neu amplified cells. Protein overexpression was not always correlated with gene amplification. For the selection of patients for an anti-HER2/neu-therapy protein overexpression might be more important since it might sometimes be caused by CEP17 polyploidy rather than by gene amplification.

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Clinical features of BRCA1/BRCA2 positive hereditary breast cancers

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Background: BRCA1 and BRCA2 mutations cause hereditary breast cancer (BC). Patients (pts) who carry this type of mutations have a significantly cumulative lifetime risk of developing breast and ovarian cancer. The authors review all cases of BRCA1/BRCA2 positive BC in their Institution.

Material and Methods: Retrospective analysis of consecutive pts with BRCA1/BRCA2 positive BC followed at the Portuguese Institute of Oncology, Porto. Clinical data were obtained from medical records. Data were analyzed using the statistical package SPSS 13.0. Survival curves were calculated by the Kaplan–Meier method.

Results: A total number of 30 pts were evaluated.

BRCA1 positive BC (40%): All 12 pts were female. Characteristics at diagnosis of first event: median age – 46 yrs old (range: 26−61); 66.7% of women were premenopausal; histology – 66.7% ductal, 8.3% lobular, 16.7% medullar, 8.3% other; grade – 16.7% II, 66.7% III, 16.7% unknown; hormone receptor status (HR) – 58.3% negative, 25% unknown; HER2 – 8.3% positive, 66.7% unknown; stage of disease – 25% I, 66.7% II, 8.3% III. First treatment: 91.7% surgery; 8.3% neoadjuvant chemotherapy (CT). Adjuvant therapy: 75% CT, 25% hormonal therapy (HT), 50% radiotherapy (RT). Thirty-three percent of pts developed contra-lateral BC and 25% ovarian cancer. Median time until second BC was 1.5 yrs (range: 0.5−4). Prophylactic surgery: 16.7% bilateral salpingo-oophorectomy and 8.3% mastectomy. Median time of follow-up was 7.8 yrs (range: 2.9−24.5). At the time of writing, only one patient died, with cerebral metastasis.

BRCA2 positive BC (60%): Two of the 18 pts were male. Characteristics at diagnosis of first event: median age – 43.5 yrs old (range: 31−61); 81.3 of women were premenopausal; histology – 72.2% ductal, 16.7% mixed, 11.1% other; grade – 5.6% I, 33.3% II, 50% III, 11.1% unknown; HR – 11.1 negatives, 22.2% unknown; HER2 – 38.9% negatives, 61.1% unknown; stage of disease – 27.8% I, 22.2% II, 50% III. First treatment: 88.9% surgery; 11.1% neoadjuvant CT. Adjuvant therapy: 77.8% CT, 66.7% HT, 88.9% RT. Sixteen percent of pts developed contra-lateral BC and 6.3% ovarian cancer. Median time until second BC was 12.1 yrs (range: 9.6−13). Prophylactic surgery: 18.7% of women bilateral salpingo-oophorectomy and 5.6% mastectomy. Median time of follow-up was 6 yrs (range: 1.5−18.3). All pts are still alive at the time of writing.

Conclusions: BRCA1 and BRCA2 positive BC pts have different clinico-pathologic features.

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Target-oriented microarray analysis for detailed characterization of high-risk breast tumors

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Background: Gene expression profiling is widely used to identify new markers for prognosis and prediction of breast cancer. Different sets of genes were reported which are expected to provide a better basis for individualized therapies than routinely measured clinical parameters alone. Different studies have analyzed the utility of expression profiles in breast cancer decision-making, yet little is known about the reliability of this method compared to classical risk factors. Therefore, correlation between clinical characteristics and gene expression is analyzed in this study.

Material and Methods: For target-oriented expression analysis a 70mer oligonucleotide microarray containing 180 breast cancer related genes was designed. Core biopsies were taken before neoadjuvant chemotherapy of 18 patients with primary breast cancer. All were high-risk patients with one or more of the following characteristics: poorly differentiated tumor (G3), negative ER, regional lymph node involvement (N1), inflammatory carcinoma. The gene expression profiles are subject to statistical analysis with respect to correlation to tumor-related data like Her2-neu-, estrogen and progesterone receptor status.

Results: Correlation of gene expression profiles with clinical data is analyzed qualitatively using unsupervised clustering analysis and by means of statistical correlation of protein and gene expressions of relevant prognostic factors. Hierarchical clustering of the tumors yielded two main clusters with a strong correlation to the expression of estrogen receptor alpha. Separate statistical analysis using Spearman's rank correlation coefficient showed significant correlation of gene and protein expression for both hormone receptors and Her2-neu in this sample.

Conclusions: Gene expression results for high-risk tumors showed high agreement with routinely measured clinical data and indicate a good reliability for the method. Both methods together give better tumor characterization and provide a good basis for further analyses due to tumor response in the neoadjuvant setting and disease outcome.

Wednesday, 16 April 2008 12:30–14:30

POSTER SESSION

Nursing

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Nursing intervention prior to breast biopsy – is it necessary?

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Ultrasound guided needle biopsy is a common technique used for diagnosis of breast cancer lesions. The procedure is not well known to the general population, can be frightening and painful, and there is a waiting time till the diagnostic results are available. All these factors together with breast cancer fear cause uncertainty and high levels of anxiety. In light of the above, we have created at our clinic a nursing information providing procedure which includes verbal guidance, assessment and support together with written information at the referral, guidance and supportive nursing and medical staff during the biopsy procedure, instruction including pain management at dismissal and continuous support until final diagnosis is available. Women from other medical centers, who did not get the same instruction and support prior to the procedure are also accepted for biopsy at our clinic. We compared the level of information, knowledge and anxiety between those two populations. A questionnaire was given to every patient coming to biopsy. The questionnaire was aimed to evaluate the information the patient was given at referral to biopsy and to examine the course of the procedure. 238 patients who underwent ultrasound guided biopsy during 2/2006–8/2007 were included, 61% from Lin breast clinic and 39% from other centers. 94% from our clinic population reported having pre biopsy guidance opposed to only 42% from other centers ($p < 0.001$). High level of knowledge about the procedure was found in 72% of our center population but only in 2% of women referred to us. Only 21% of our breast clinic patients reported high level of anxiety as opposed to 87% of the patients referred from other centers. 90% of patients from our clinic reported about pain treatment guidance opposed to only 13% from other centers. In the whole group a strong correlation was found between level of anxiety and information provided, 85% of those found to suffer from high level of anxiety did not get any information about the procedure.

We conclude that patients undergoing proper nursing intervention prior to guided biopsy are more knowledgeable about the examination and show statistically significant less anxiety compared to those who had not. Therefore, nursing intervention is necessary and should be integrated as common practice in this clinical situation.

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National organization for breast care nurses

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The SIG breast care is a national platform for the communication between nurses working in breast care. The SIG breast care is part of the V&VN oncology, it reflects the oncologic department of Nurses and Caretakers in the Netherlands.

From all integral cancer departments of the Netherlands (IKC's) breast cancer nurses and nurse practitioners breast care are represented in the SIG breast care. There are nine IKC's in the Netherlands, these are regional network organizations that support workers in the oncologic and palliative care.

Aims:

- Optimizing the quality of care for patients with breast cancer.
- Professional continuing education.
- Exchange of knowledge.

Goals:

- Communications platform for nurses with the 'sub' specialisation breast care.
- Encouraging, supporting and promoting the network of breast cancer nurses and nurse practitioners breast care at a national level.
- Identify and examine specific care needs within the breast care.
- Develop and implement guidelines.
- Share of education, knowledge and clinical expertise.
- Monitor the quality of care within the sub specialisation breast care.
- To support the mission and goals of the V&VN oncology.

Mission: Nurses & Oncologic Caretakers Netherlands is one of the leading organizations in cancer care and promotes excellent oncologic nursing care.