

understood and our interest was to compare the gene expression pattern of mammary osteosarcomas to that of osteosarcomas on the head and the extremities. We have previously reported that genes related to the embryonic development of the head are overrepresented in canine mammary sarcomas compared to mammary carcinomas [1].

**Material and Methods:** The study investigates gene expression profile in canine mammary osteosarcomas as compared to normal mammary tissues – based upon the earlier Affymetrix-assay [1] –, with that of head osteosarcomas and extremity osteosarcomas.

Analysis of the gene expression data was carried out in the statistical computing language R (<http://www.r-project.org>) using packages available from the Bioconductor project ([www.bioconductor.org](http://www.bioconductor.org)). The raw data were normalized using the (RMA) method [2]. In order to search for the differentially expressed genes, an empirical Bayes moderated t-test was applied [3]. To address the problem with multiple testing, the *p*-values were adjusted according to Benjamini and Hochberg [4]. The hierarchical clusterings were performed in the program Genesis, version 1.7.1 [5].

**Results:** Preliminary results of the Affymetrix data with adjusted *p* value <0.05 gave about 2500 differentially expressed genes (DEG) in mammary osteosarcomas compared to the control of normal mammary tissue. About half of these genes were similarly expressed in the osteosarcomas of the head and the trunk (1140 and 1211 respectively), whereas 970 genes did overlap. Genes of interest will be further studied, described and be concluded.

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## 5103

## POSTER

### Status of Granulocyte Colony-Stimulating Factor Receptor in Tumour Tissue of Patients With Breast Cancer

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**Objectives:** Granulocyte colony-stimulating factor receptor has been observed on the surface of not only hematopoietic cells but also several cancer cells, including bladder cancer, epithelial skin tumours, primary ovarian carcinomas, ewing sarcom and head and neck squamous cell carcinomas. There was no study to show the expression of GCSF-R in breast cancer. The aim of this study was to investigate the expression of granulocyte colony-stimulating factor receptor in tumour tissue of patient with breast cancer, and also to evaluate the relationship between GCSF-R with prognostic factors and prognosis of patients.

**Patients and Method:** 57 patients, were diagnosed with breast cancer at Ankara University Faculty of Medicine in 2004–2005 and taken monitoring after treatment, were studied. Tumour tissues were received from paraffin-embedded blocks of patient. GCSF-R detected by immunohistochemical staining method. Results were compared with prognostic factors and prognosis of patients.

**Results:** Fifty five cases of the 57 breast cancers showed positive stainings of GCSF-R (61.4%), and twenty two cases showed negative (38.6%). There was no relationship between GCSF-R staining and prognostic factors. The prognosis of 49 patients was known. We found 7 relapsing cases(12.2%). 5 cases was in negative group and 2 cases was in positive group. Relaps (*p*=0.041) and disease-free survival (*p*=0.036) were more in negative group. There was no relation between overall survival and GCSF-R.

**Conclusion:** There was no study that showing the relationship between breast cancer and GCSF-R. Our study is the first about this. In this study, we showed that there is GCSF-R expression in tumour tissues of patients with breast cancer. There was no association between prognostic factors and GCSF-R staining. Overall survival was similar. But there was significant association between GCSF-R staining and patient prognosis. GCSF-R negative group relapsed more than positive. This was very different from the results of studies in other cancers.

## 5104

## POSTER

### Vitamin D Analogs Enhance the Anastrozole Activity in Human Breast Cancer Models

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**Background:** Breast cancer is the most common malignant cancer among women in Poland. The growth and invasion of breast cancers are mostly estrogen dependent and estrogens play a key role in cancer progression. Calcitriol is known to evaluate a therapeutic effect against breast cancer, mainly by lowering the expression of estrogen receptors and aromatase activity. Low-calcemic vitamin D<sub>3</sub> analogs; PRI-2191 and PRI-2205 were previously tested for their antiproliferative activity against different cancer cell lines. In our latest studies, the influence of calcitriol analogs on the activity of anastrozole against breast cancer has been evaluated (*in vitro* and *in vivo*).

**Materials and Methods:** *In vitro:* Cells (MCF-7, T47D, and SKBR-3) were placed in 96-well or 24-well flat-bottom plates at a density of 5x10<sup>3</sup> or 1x10<sup>5</sup> cells per well 24 hours before addition of the tested compounds. Cancer cells were exposed for 120 hours to calcitriol or its analogs and anastrozole. The cytostatic effect was measured by the SRB assay. The cell cycle changes were evaluated by flow cytometry.

*In vivo:* The antitumour effect of combined treatment was evaluated as tumour growth inhibition (TGI), tumour volume and body weight changes were monitored.

**Results:** *In vitro:* PRI-2191 and PRI-2205 showed synergy or an additive effect in proliferation inhibition when combined with anastrozole on T47D, MCF-7 and SKBR-3 cancer cells. The cell cycle observations showed an increase in the percentage of cells in G2/M stage or an induction of apoptosis after calcitriol analogs and anastrozole treatment.

*In vivo:* Both analogs showed synergy or an additive effect in inhibiting MCF-7 tumour growth. PRI-2191 was found to be more active in inhibiting tumour growth than anastrozole alone.

**Conclusion:** It is supposed that the calcitriol analogs may be of potential use in anti-cancer therapy when combined with anastrozole.

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## 5105

## POSTER

### Assessing the Cognitive Function Among the Breast Cancer Patient After Chemotherapy Treatment

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**Introduction:** The previous research has shown the growing evidence of cognitive impairment among the breast cancer patients after receiving adjuvant or local chemotherapy. However the measuring of cognitive function requires additional time and application of complex battery. Therefore, clinical researchers focus their interest on particular aspects of cognitive function. In our research, we have decided to measure the executive function in breast cancer patients after receiving chemotherapy. Executive functions are very important for patients' daily life, but they are also most vulnerable. For this purpose, we used the Trial Making Test, part A (TMT-A) and part B (TMT-B), since they both can be applied easy and in short period of time.

**Methods:** After they have received chemotherapy treatment, 42 female patients, non-homogeny by age, with breast cancer were examined. We have used TMT-A to measure attention and TMT-B to measure executive function. Their results were compared with results of referent subgroups in general population. The highest subgroup was between 40–49 years old, the before-menopause group.

**Results:** The results from the TMT-A which measure attention was in average level according to normal population or less than average. Results from the TMT-B which measure executive function were on high level according to normal population 7 (16.7%) patients demonstrated high level results compared to normal population.

**Conclusion:** The largest subgroup showed the best results as expected. Low score on TMT-A could be explained in view of setting. Patients have completed the tests while they have been waiting for ambulatory examination, so they were excited to hear the state of their health condition.

However, in a small population in this pilot study we have seen good results in TMT-B, which means without impairment in executive function. This result is different in comparison with previous researches. The future examination with larger sample and with application of different cognitive tests, is highly recommended.

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POSTER

#### Toll Like Receptor 4 – Weapon or Shield – a Murine Breast Cancer Study

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**Background:** TLR4 is a member of a family of pattern recognition receptors that are involved in host defence against infection. Few studies established links between TLR4 and cancer. Thus far, studies have predominantly focused on the effect of TLR4 in cancer cells; however this study focuses on TLR4 in both the host immune system and cancer cells and its effect on breast cancer progression and metastasis.

**Materials and Methods:**  $3 \times 10^5$  murine mammary adenocarcinoma 4T1 cells and TLR4-knocking down 4T1 cells were inoculated into female wild-type (WT) and C.3-Tlr4<sup>LP8-d/J</sup> (TLR4<sup>-/-</sup>) mice by mammary fat pad injection and divided into a control and a primary tumour resection groups. Tumour growth, tumour body weight ratio, lung metastasis and survival of tumour bearing mice were assessed.

**Results:** There was a significant increase in tumour volume on the 2<sup>nd</sup>, 3<sup>rd</sup> and 4<sup>th</sup> week after inoculation of the TLR4<sup>-/-</sup> mice with 4T1 cells compared to WT mice ( $P < 0.05$ ). The numbers of metastatic lung nodules were significantly higher in the TLR4<sup>-/-</sup> mice ( $P < 0.05$ ). Survival of TLR4<sup>-/-</sup> mice was dramatically reduced compared to WT mice ( $P < 0.05$ ). Knock down of the TLR4 from 4T1 cells led to a synergistic increase in tumour volume and reduction in lung nodules ( $P < 0.001$ ).

**Conclusions:** TLR4 exerts a protective role in this murine metastatic breast tumour model at the host level and an adverse role at the cancer cell level. Further evaluation of the role of TLR4 in breast cancer is warranted.

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POSTER

#### Association of Serum Level of 25 Hydroxy-vitamin D With Prognostic Factors of Breast Cancer

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**Background:** Experimental studies have shown that vitamin D promotes cell differentiation and retards or terminates proliferation of breast cancer cells. However, there is little evidence supporting the association of vitamin D and prognosis of breast cancer. This study aimed at evaluating possible association of serum 25 hydroxy-vitamin D with prognostic factors of breast cancer.

**Materials and Methods:** In this analytic-descriptive study, 119 female patients with histologically proven breast cancer were recruited in Tabriz oncology clinics in a 15-month period of time. Serum level of 25 hydroxy vitamin D (25(OH)D) was measured in all patients. Patients' demographics, stage and grade of cancer, status of lymph node involvement, metastasis, and biomarkers such as ER, PR, HER2, Ki-67 and P53 were documented.

**Results:** One hundred and nineteen patients with a mean age of  $50.4 \pm 12.6$  (26–76) years were enrolled in the study. The mean serum level of 25(OH)D was  $15.7 \pm 17.8$  (4–122) ng/ml, deficient in 66 cases, insufficient in 36 cases and normal level in 17 cases. The median level of 25(OH)D was lower in the P53+ group in a borderline trend (17.3 vs. 13.6 ng/ml;  $p = 0.07$ ). The median level of 25(OH)D was significantly higher in the patients with metastasis, as well (27.7 vs. 12.0 ng/ml;  $p = 0.03$ ). There was no significant association between the serum level of 25(OH)D and other studied parameters.

**Conclusions:** Based on our findings, there may be an association between the serum level of 25(OH)D and prognosis of breast cancer.

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POSTER

#### Association of Decreased NK Cell Activity and IFN $\gamma$ Expression With pSTAT Dysregulation in Breast Cancer Patients

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**Background:** Impaired IFN $\gamma$  production in peripheral blood lymphocytes (PBL) and their subsets reflects immunosuppression and inadequate antitumour immune response in cancer patients. Decreased function of natural killer (NK) cells has not been investigated in breast cancer with respect to altered pSTAT signaling pathways.

**Materials and Methods:** PBL of breast cancer patients and healthy controls were analyzed for IFN $\gamma$  and pSTAT1 expression and NK cell activity using flow cytometry and <sup>51</sup>Cr-release assay, respectively. The level of pSTAT1, 3 and 5 was investigated by Western blotting.

**Results:** Our results indicated that PBL and CD3<sup>+</sup>CD16<sup>+</sup> NK cells of patients had significantly lower level of IFN $\gamma$ . The patients had a significantly decreased NK cell cytotoxicity compared to controls, with the decrease being dependent on the stage of disease. Positive correlation between IFN $\gamma$  level in PBL and NK cytotoxicity in controls and patients was also shown. The PBL of patients, compared to controls, expressed lower level of pSTAT1, 3 and 5. The patients' T and NK cell subsets had lower pSTAT1 level.

**Conclusions:** This study indicates that pSTAT1 in PBL of breast cancer patients could be a biomarker of decreased NK cell cytotoxicity and IFN $\gamma$  level that are associated with progression of this disease.

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POSTER

#### The Beliefs, Knowledge, Understanding, Attitudes and Treatment Access to Breast Cancer Amongst Rural Women in Northern Nigeria

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**Goal:** The goal of this study is to ascertain the beliefs, knowledge, understanding, attitudes and treatment access to breast cancer among rural women in Nigeria.

**Background:** Breast cancer has become a popular topic in recent years with several thousands of women diagnosed to be positive every year. The availability of care/treatment upon early detection is key to survival.

**Methods:** An interview guide was designed specifically for this study in which 200 rural women in Northern Nigeria, age 45 and over took part in. It contained questions about beliefs, knowledge, understanding and attitudes about Breast Self-Examination (BSE), Clinical Breast Examination (CBE) and mammogram. In addition, questions assessing the variables of the Health Belief Model and health motivations also were included. The data were obtained during face-to-face interviews in the primary language of the participating woman. The interviews were transcribed and translated into English.

**Results:** Out of the 200 women who participated, only 1% two (2) of the participants practiced BSE monthly, 8% had undergone at least one CBE during their lives, and 91% had never had a mammogram. There were little or no access to treatment even at early detection in these rural areas causing thereby vulnerability to loss of life. Majority of these rural women (95%) said they knew little or nothing about breast cancer. While 15% of the women said detecting cancer early was important, only 3% reported that cancer could be cured. Age, education, or mother tongue showed no statistically significant relationship with the breast health practice scores. However, proficiency with the English language ( $p = 0.009$ ) and number of years exposed to awareness and education ( $p = 0.009$ ) had a significant relationship with the breast health practice scores. The significant explanatory factor for the variable breast health practices was a cue to action ( $p = 0.009$ ).

**Conclusions:** The level of awareness and treatment access to breast cancer amongst Northern Nigeria's rural women is extremely low thereby making them not to engage in screening and/or detection practices. This alarming situation calls for urgent intervention of medical/health organizations to provide immediate breast cancer awareness, screening and care so as to reduce incidences or threat at early detection.