in MBC for the last fifteen years and second to explore its association to prognostic factors affecting outcome including therapeutic regimen.
Material and Methods: This meta-analysis uses individual patient data collected from all ten trials on MBC (6 non randomized, 4 randomized) conducted by HeCOG from 1991 through 2006. Four 4-year time periods (1991–1994, 1995–1998, 1999–2002 and 2003–2006) were constructed for exploration of time trends in survival according to the patient's date of metastatic diagnosis. Different first line regimens in the 10 trials include anthracycline monotherapy (epirubicin, in the early 90s) and taxane-containing regimens either as monotherapy or in different combinations with anthracyclines or other drugs. In two phase II studies and the last randomized study, trastuzumab was administered in all patients with HER2 overexpressing tumors.
Results: Information is based on a total of 1365 patients with a median follow up of 3.7 years and median survival of 1.9 years (median survival 1.3, 1.7, 2.2 and 2.6 years for 1991–1994, 1995–1998, 1999–2002, and 2003–2006, respectively). Survival improved significantly across diagnosis time periods, by 26%, 44% and 52% respectively in each time period as compared to the first (1991–1994), (1995–1998: HR = 0.74, p = 0.002; 1999–2002: HR = 0.56, p < 0.001; 2003–2006: HR = 0.48, p < 0.001). The effect of metastatic diagnosis time period remains almost unchanged in the presence of the following significant prognostic factors: performance status, hormonal receptor status, previous adjuvant treatment, visceral metastasis at entry and number of metastatic sites. When exploring the effect of new treatment introduction, taking into account the same significant prognostic factors, the effect of time period disappears and the same effect magnitude is explained directly by the introduction of taxanes or trastuzumab (taxanes at 1st line: yes vs. no: HR = 0.73, p = 0.004; trastuzumab at 1st line: yes vs. no: HR = 0.64, p < 0.001).
Conclusions: The results provide significant evidence of improvement in prognosis of MBC patients within the last 15 years, taking into account all important significant prognostic factors, and this improvement could be explained almost fully by the use of new agents in the management of the disease.

5007 ORAL

MicroRNA profiling of circulating tumor cells (CTC) present in large quantities of leukocytes

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Background: The CellSearch Circulating Tumor Cell Test (Veridex) is the only FDA approved diagnostic test for the detection and enumeration of CTCs. CTC enumeration by this technique has proven clinical relevance in metastatic prostate, colorectal and breast cancer. Next to enumeration, there is great interest in the molecular characterization of CTCs, which may yield better prognostic and predictive factors and models. Although this system allows capture of CTCs in blood of cancer patients by selectively isolating EpCAM-positive cells followed by visual quantification of DAPI- and CK-8/18/19-positive cells [1], there are still considerable quantities of contaminating leukocytes (DAPI+/CD45+) present after enrichment. Previously, we optimized a method to determine mRNA expression of up to 96 genes in as little as a single breast cancer cell [2]. By using a set of genes with no or minor expression by leukocytes, we succeeded to specifically determine gene expression profiling in a small number (frequently less than 5) of CTCs present in a CTC-enriched blood sample typically containing over 800 contaminating leukocytes. In this study we set out to similarly characterize these CTCs at the miRNA level. MiRNAs are naturally occurring non-coding RNAs that play a role in gene regulation. Expression of various miRNAs have been associated with outcome in breast cancer.
Methods: We screened healthy blood donors (HBDs), breast cancer tissues, breast cancer cell lines spiked in blood from HBDs, and breast cancer patients for miRNA expression specific for breast cancer tumor cells with the TaqMan human MicroRNA assay v1 set (Applied Biosystems) containing 446 miRNAs.
Results: Of the 446 miRNAs, ~300 could be measured reliable in human breast cancer tissue specimens. Out of these, 60 appeared to be specific for the breast cancer tumor cells, i.e expression was over 10-fold higher when compared with the levels measured in the healthy blood donors. Next, the potential clinical applicability of these 60 differentially expressed miRNAs was validated on CTCs from a cohort of breast cancer patients with metastatic disease as detected by the CellSearch CTC test.
Conclusion: We consider our approach of great interest for the further characterization of CTCs, thereby improving insight into biological processes involved in cancer progression and ultimately patient management.

References

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Oral presentations (Tue, 22 Sep, 09:00−11:00)
Breast cancer II − Early disease

5008 ORAL

Minimal axillary lymph node involvement in breast cancer has different prognostic implications according to the staging procedure

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Purpose: It is still controversial whether the identification of micrometastases and isolated tumor cells in the axillary lymph nodes of patients with breast cancer has any prognostic value.
Patients and Methods: We evaluated the prognostic role of isolated tumor cells and micrometastases in the axillary lymph nodes in 3,158 consecutive patients (pT1−2 pN0-N1mi (with a single involved lymph node) and M0, referred to the Division of Medical Oncology after surgery performed at the European Institute of Oncology from April 1997 to December 2002. Median follow-up was 6.3 years (range 0.1−11 years).
Results: Sentinel lymph node biopsy (SLNB) and axillary lymph node dissection (ALND) were performed in 2,087 and 1,071 patients respectively. A worse metastasis-free survival (MFS) was observed for patients with micrometastatic disease compared to node negative patients, if staged with ALND (log-rank p < 0.0001; HR 3.17; 95% CI: 1.72−5.83 at multivariate analysis), but not for patients who underwent SLNB (log-rank p = 0.36).
Conclusion: The presence of a single micrometastatic lymph node is associated with a higher risk of distant recurrence as compared to node negative disease only for patients undergoing ALND for staging purposes. Treatment recommendations for systemic therapy should not take into account the presence of a single micrometastatic lymph node identified during complete serial sectioning of sentinel node(s).

5009 ORAL

Influence of isolated tumor cells in sentinel nodes on outcome in early pT1N0M0 breast cancer

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Aim: The aim of the study was to evaluate the prognostic significance of isolated tumor cells found in a sentinel node biopsy.
Patients and Methods: The study is based on a prospectively followed-up cohort of 1,865 consecutive patients diagnosed with invasive pT1 (tumor size $\leqslant$20 mm) breast cancer in one university breast unit between February 2001 and August 2005. Of the 1,390 patients who had received no neoadjuvant therapy and who underwent a sentinel node biopsy, 63 had isolated tumor cells in the sentinel nodes (stage pT1N0i+M0, verified by axillary node dissection), and 868 had not (pT1N0i-M0). The median follow-up time was 55 months.
Results: Patients with pN0i+ disease were treated more often with systemic adjuvant therapy than those with pN0i-disease (87% vs. 51%; $P < 0.0001$). There was no significant difference between the groups in 5-year recurrence-free survival (90.3% vs. 93.2%, respectively; $P = 0.32$) or overall survival, but patients with pN0i+ cancer had less favorable 5-year breast cancer-specific survival (95.2% vs. 98.4%; $P = 0.035$), and they were more frequently diagnosed with distant metastases from breast cancer (8.1% vs. 1.9%) during the first 5 years of follow-up ($P = 0.001$). Several

traditional prognostic factors, such as histological grade, steroid hormone receptor status, and the cell proliferation rate were more strongly associated with outcome than the pN0i status.
Conclusions: The findings suggest that presence of isolated tumor cells in the sentinel nodes is an adverse prognostic factor in early breast cancer, but its prognostic significance in association with the standard factors may be limited.

5010 ORAL

Survival and safety post study treatment completion: an updated analysis of the Intergroup Exemestane Study (IES) – submitted on behalf of the IES Investigators

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Background: The IES trial demonstrated a benefit in disease outcome of switching adjuvant therapy to exemestane (E) after 2−3 years tamoxifen (T) in postmenopausal patients with early breast cancer (Coombes RC et al, Lancet 2007). At that time the trial steering committee agreed to conduct an updated survival and safety analysis when at least 90% of surviving patients had a minimum of 6 years follow-up available. The emphasis of this presentation will focus on disease free survival (DFS) outcome after study treatment completion and a more mature analysis of overall survival.
Materials and Methods: 4724 postmenopausal women with histologically confirmed, completely resected, ER positive/unknown unilateral breast cancer, disease free after 2−3 years T, were randomised to continue T or switch to E to complete a total of 5 years adjuvant endocrine therapy. This study is an investigator-led study, sponsored by Pfizer Ltd and is registered as an International Standard Randomised Controlled Trial, number ISRCTN11883920.
Results: Previous results based on 55.7 months follow-up reported an un-adjusted hazard ratio (HR) for DFS of 0.75 (95%CI: 0.65, 0.87); p = 0.0001 in favour of E in the ER+/unknown group (n = 4602, excluding 122 patients with ER-disease). Similarly, with E = 210 versus T = 251 deaths, a modest improvement in overall survival was demonstrated (HR = 0.83, 95%CI (0.69, 1.00); p = 0.05). We expect to reach the minimum data cut-off requirement in May 2009 after which the database will be frozen for analysis. An expected median follow-up of 88 months and just over 770 deaths will allow a detailed analysis of DFS partitioned at time of treatment completion to further characterise post treatment effects and allow a more robust estimate of the effect on overall survival. A detailed safety analysis will further clarify the post study treatment safety profile of E.
Conclusions: Additional follow-up information on patients in IES will quantify the extent of any carryover effect of exemestane and provide stronger evidence of any continuing overall survival benefit. The safety analysis will determine any longer term effects of exemestane when used in this setting.

5011 ORAL

ZORO: Prospective randomized multicenter-study to prevent chemotherapy induced ovarian failure with the GnRH-Agonist Goserelin in young hormone insensitive breast cancer patients receiving anthracycline containing (neo-) adjuvant chemotherapy (GBG 37)

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Background: Premature ovarian failure due to chemotherapy (ChT) leads to premature menopause and infertility. To prevent related health problems and to maintain fertility after ChT in patients (pts) with hormone insensitive breast cancer, ovarian function might be protected by using a GnRH-Agonist.
Methods: Pts ⩾18 and >46 years, with a hormone receptor neg primary breast cancer, regular and spontaneous menstruation with premenopausal values for FSH and no evidence of distant metastasis were randomised to receive an anthracycline containing ChT with (G+) or without 3.6 mg goserelin (G−). Goserelin was applied at least 2 weeks before ChT start and every 4 weeks thereafter until the end of the last ChT cycle (EOC). The primary endpoint was normal ovarian function after ChT defined as 2 consecutive menstrual periods within 21−35 days within 5−8 months after last application of goserelin. Secondary objectives are treatment compliance, toxicity, quality of life, menopausal symptoms score, ovarian function at 0, 6, 12, 18 and 24 months by menstruation and endocrine function (estradiol, progesterone, FSH, LH, SHBG), duration until recovery of regular menstrual period, pregnancy rate. A clinically relevant difference between the treatment arms is to be detected at $\alpha = 0.05$ (two-sided) with 80% power. The rate of intact ovarian function will be increased to 80% for patients receiving goserelin. To show an absolute increase of intact ovarian function at 6 months by 30% from 50% without goserelin to 80% with goserelin protection, a total of 62 pts is required.
Results: Between March 2005 and August 2007, 63 pts in 16 sites were enrolled. 60 pts are evaluable, 3 pts withdrew their informed consent. All pts had a regular menstruation and premenopausal FSH. The median age was 35 years in the G+ group and 38.5 years in the G− group. 29 pts received a taxane free ChT (15 in G+, 14 in G−). One woman in each group became pregnant. One pt died within 6 months after EOC due to progression. 6 women started with regular menstruation within 5−8 months after the last application of chemotherapy, 1 in G+ and 5 in G-. 9 serious adverse events occurred, which were mainly chemotherapy related (3 febrile neutropenias, 2 neutropenias, 2 nauseas, 1 infection of port-catheter and 1 psychogenic hyperventilation).
Conclusions: These data in pts with primary breast cancer who are treated by (neo)adjuvant ChT do not support the use of goserelin to protect ChT induced ovarian failure.

5012 ORAL

Persistent pain following breast cancer surgery: a nationwide study of predictors and consequences

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Context: Persistent pain and sensory disturbances following treatment for breast cancer remain a significant clinical problem. The pathogenic mechanisms are complex and may be related to patient characteristics, surgical technique and adjuvant therapy.
Objective: To examine risk factors and consequences of persistent pain after treatment for breast cancer in Denmark.
Design, Setting, and Patients: Nationwide cross-sectional questionnaire study of women operated on for primary breast cancer in Denmark between 2005 and 2006 with well-defined principles for surgery and adjuvant therapy.
Main Outcome Measures: Prevalence, location, and severity of persistent pain and sensory disturbances in 12 well-defined treatment groups with an average follow-up of 26 months. Odds ratio (OR) of reported pain and sensory disturbances in relation to surgical technique, chemo- and radiotherapy.
Results: 3253 women returned the questionnaire (response rate 87%). Overall, 1543 patients (47%) reported pain, of whom 13% reported severe pain, 39% moderate pain and 48% light pain. Pain was located in the breast area (86%), axilla (63%), arm (57%), or on the side of thorax (56%). Factors associated with increased risk of experiencing pain were young age (OR = 3.62; CI: 2.25−5.82, p < 0.0001) and adjuvant radiotherapy (p < 0.05), but not chemotherapy (p = 0.95). Axillary lymph node dissection increased risk of pain (OR = 1.75; CI: 1.41−2.17, p < 0.0001) compared with sentinel lymph node dissection. Mastectomy increased the risk of moderate to severe pain (OR = 1.37; CI 1.00−1.87; p < 0.05) compared to breast-conserving surgery. Pain complaints from other parts of the body were associated with increased risk of pain in the surgical area (p < 0.0001). 10% of pain patients had contacted a physician with the last 3 months for pain complaints in the surgical area. Risk of sensory disturbances was associated with young age (OR = 5.14; CI: 3.13−8.48, p < 0.001) and axillary lymph node dissection (OR = 4.97; CI: 3.92−6.29, p < 0.0001)
Conclusions: Persistent pain in the surgical area after breast cancer treatment remains a significant clinical problem in about 25−60% of patients. Although breast-conserving surgery and sentinel node dissection have reduced the number of complaints, future strategies for further improvement should include nerve-sparing axillary dissection and attention to patients with other chronic pain complaints.