pPR = persistence of less than 10 microscopic foci of invasive tumor cells in the breast or in the axilla; pNR = any larger amount of residual cancer cells in the breast or in the axilla.

Results: Gene expression was plotted toward pathological response. Low tubulin levels in isoforms III, IVa, IVb were associated with pathological response.

Conclusion: The patterns of beta-tubulin isoforms distribution may predict for pathological response to paclitaxel and paclitaxel/radiation regimens.

544 POSTER

Proliferation, apoptosis and related markers in invasive ductal breast carcinoma

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Purpose & Methods: Expression of bcl-2, bax, p53 and PCNA genes was studied immunohistochemically in 170 invasive ductal breast carcinomas (median follow-up time 91 months, range 24–186 months). In addition the mitotic activity index (MAI) and apoptotic cell death (Tunnel technique) were scored. Classic histopathological features and steroid receptor status of the tumours, and clinical patient characteristics were incorporated in the database.

Results: No relationship could be observed between bcl-2, bax or p53 status and tumour grade, pTNM staging and menopausal status. A strong positive relationship was demonstrated between bcl-2 immunoreactivity and steroid receptor status (ER and PR: $p < 0.001$). There was an inverse relationship between bcl-2 expression and p53, but not with bax or PCNA. Multivariance analysis demonstrated absence of bcl-2 expression and the MAI to be independently related to shortened disease-free survival ($p < 0.01$) and shortened overall survival ($p < 0.001$).

Conclusions: Our data suggest that bcl-2 expression plays a crucial role in the behaviour of invasive ductal carcinoma and may be an important modulator of response to adjuvant therapy.

545 POSTER

Incidence of breast cancer associated with use of hormone replacement therapy and other risk factors in 1709 patients

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Background: Various studies have suggested an association between hormone replacement therapy (HRT) and breast cancer but the reports have been conflicting and controversial. A meta-analysis of 52 epidemiological studies in 21 countries by the Imperial Cancer Research Fund (ICRF) in 1997 concluded that the relative risk for breast cancer was 1.023 for each year of use.

Aims:

(1) To assess the incidence of HRT related breast cancer in our patient population.
(2) To study other risk factors in the women who developed breast cancer while on HRT.

Patients & Methods: A retrospective study of 1709 patients seen between January 1987 and December 1997 was made. The mode of presentation, duration of HRT, age at diagnosis, other "risk" factors (alcohol, smoking, family history, past history of breast biopsy) and pathological features of the cancer were analysed.

Results: Sixty-five per cent of the 62 patients (mean age 60 years) with HRT associated breast cancer presented with symptomatic disease while 19% were detected within the UK Breast Screening Programme. The duration of HRT intake was <2 years in 9, 2–5 years in 24, 6–10 years in 17, 10–15 years in 5 and >15 years in 5 patients. In 38.7% a family history of breast cancer (11 first degree and 13 second degree relatives) was noted. Smoking and alcohol intake was average in 87% of patients. Twenty patients had a history of a previous benign breast biopsy. Histological review showed that 55.8% had a T1 carcinoma and 57% were N0. Invasive ductal carcinoma was found in 82.3% with 19.5% being Grade I (Bloom & Richardson) and 61% Grade II.

Conclusion: The highest risk in relation to HRT appears to be in the first five years suggesting that HRT may stimulate the growth of an undetected cancer. The high association with a strong family history and previous benign biopsies has implications for women seeking advice about starting HRT.

546 POSTER

Breast cancer and missense mutations in the transactivation region of the BRCA1 gene

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Purpose: The implication of missense mutations in germline of BRCA1 gene in the pathogenesis of breast cancer is not well understood. When a missense mutation occurs in a specialized region of the gene, the functional impact of this change can be meaningful.

Methods: We investigated missense mutations located in the transcriptional transactivation gene region (amino acids 1528–1863), and correlate them with clinical and familial characteristics of the patients. We studied 192 patients, 87 with a definite family history of the disease, and 105 without antecedents and considered to have sporadic breast cancer. The entire coding region of the BRCA1 gene was analyzed by the PCR-SSCP method. Specimens showing a differential band were amplified and used for direct DNA sequencing.

Results: Two mutations were detected, Glu1735Lys and Asp1778His. The first mutation was identified in a family with five breast cancer patients in first-degree, distributed between two generations. One patient showed bilateral tumor. The second missense mutation appeared in a 44-year-old patient with a sporadic invasive ductal carcinoma with axillary involvement and negative steroid receptors.

Conclusions: The two new mutations detected may represent a functional change in the transactivation potential of the BRCA1 gene.

547 POSTER

The expression of new protein taking part in cancerogenesis, p65, and its correlation with steroid receptors in ductal carcinoma of female breast

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A 65-KD phosphoprotein (p65) was isolated from human breast carcinoma cell line MCF7. The aminoacid sequence analysis of N-terminal part of the p65 molecule was similar to the steroid receptor protein. This raises the possiblility that the p65 gene may belong to the family of genes which encode nuclear receptors for various hydrophobic ligands of steroid hormones. Paraffin-embeded tissue slides from 89 infiltrating ductal carcinoma specimens were assessed immunohistochemically with the usage of monoclonal antibodies against human p65 antigen. The p65 expression was correlated with oestrogen receptor (OR) and progesterone receptor (PR) levels and grade of malignancy according to Bloom and Richardson scale. It is suggested that the high OR and PR levels are accompanied by the presence of p65 in breast cancer tissue. The funcion of p65 and its ligands is still unknown. However, p65 may be important in the process of development of tumours. It is probable that the conserved cysteine-rich domains found in the human p65 and which are also common to human OR provide an important biological funcion.

548 POSTER

A prospective study on genetic risk factors in an unselected sample of breast cancer patients who receive adjuvant radiotherapy

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5–8% of all breast cancers (BCs) are due to hereditary factors. Detection of women at high risk for BC and offering genetic counselling (GC) to them might be worthwhile. In high risk women regular mammography (started at younger age) and preventive surgery (in some cases) may result in substantial gains in health and in life expectancy. Women at high risk for BC can be identified by systemetically searching for presumed risk factors. The Radiotherapy Department of the University Hospital (UH) in Utrecht together with the Clinical Genetics Center and the Comprehensive Cancer Center prepared a study to prospectively evaluate the prevalence of risk factors for hereditary BC in 1.000 patients. All BC patients referred for radiotherapy as part of curative treatment for their disease are included (60% of all newly