O-86. Trastuzumab and CerbB2 positive brain metastases – treatment synergy leading to longer survival?

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Trastuzumab (T) in combination with chemotherapy or as single agent is proven to extend survival in metastatic breast cancer (MBC) overexpressing c-erbB2. Studies have noted a higher than expected incidence of brain metastases (BM) in patients (pts) with MBC receiving T.

We have performed a retrospective review of pts with MBC receiving T in our institution between May 2002 and the present day.

12 of 54 (22%) of pts treated with T have developed BM. 2 pts with multiple BM and poor performance status (WHO>2) died before planned whole brain radiotherapy (WBRT) could be given. Those remaining have continued T (8 pts) or started T at time of diagnosis of BM (2 pts with brain as first site of metastasis) and have received WBRT plus further treatment with neurosurgery (1 pt), stereotactic radiosurgery (1 pt), and systemic chemotherapy (6 pts). Median survival for these pts is 12 months (range 3–36 months) with 7 pts alive at present. Notably, one woman with symptomatic progression of BM 6 months after WBRT showed partial response (PR) of BM to carboplatin chemotherapy maintained for 6 months and at progression a further PR to capecitabine. Another patient with multiple BM was treated with T and WBRT and is currently asymptomatic on maintenance T with no radiological evidence of BM 36 months after BM were diagnosed.

The favourable findings in our patients raise the possibility that continuation of T in women with BM may result in clinical improvements in survival. Although T is a large molecule that does not cross an intact blood-brain barrier (BBB) the disturbed BBB noted in BM may allow sufficient concentrations of T within BM to synergise with radiotherapy and chemotherapy, a phenomenon well demonstrated in pre-clinical work. Our findings require confirmation in prospective clinical trials.

O-87. Seeing through "chemo-fog" – myth or reality

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The debate continues over the degree to which chemotherapy for the treatment of breast cancer affects cognition because previous studies have lacked pre treatment performance scores and used different analyses to calculate impairment. We report the cognitive performance on 93 women with breast cancer following treatment with standard chemotherapy compared to 43 healthy controls together with self report measures of quality of life, psychological well being and cognitive failure. At baseline patients and controls had similar cognitive functioning. There was an overall improvement in performance on four tasks (2 working memory, 1 processing speed and 1 executive function) however the patient group performed significantly worse than the control group on three tasks (AVLT supraspan $p = 0.045$; AVLT total score $p = 0.024$; (verbal memory) Stroop $p = 0.01$ (executive function). Using a statistical method that calculates change in individual performance (reliable change analysis) 12% of patients and 5% of controls were reliably impaired at the second assessment relative to baseline whereas 15% patients and 9% of controls reliably improved. The evidence suggests that only a small proportion of women experience objective measurable change in their concentration and memory following standard chemotherapy.

O-88. Effect of grape procyanidins on intracellular calcium levels in human breast cancer MCF 7 cells

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The effect of grape procyanidins (GPC) on intracellular calcium ($[Ca^{2+})]_i$) as well as cell proliferation and apoptotic levels was investigated in human breast cancer MCF 7 cell line. Cells were pre-treated with 0, 5, 50, 100 and 200 mg/L of GPC, and Fura 2AM, MTT assay and cell death detection ELISA were subsequently applied to detect $[Ca^{2+})]_i$, proliferation and apoptosis. GPC induced a significant concentration dependent increase in $[Ca^{2+})]_i$ levels: 3.2 folds with 50 mg/L, 7.6 folds with 100 mg/L and 10.3 folds with 200 mg/L of GPC compared with the cells without GPC treatment. GPC also showed to increase the release of calcium by endoplasmic reticulum. The proliferation of MCF 7 cells treated with GPC at the dose range of 50–200 mg/L was significantly inhibited by 21.4–56.2%. Furthermore, elevated apoptotic levels were observed with GPC treated cells. Apoptotic level was increased by GPC from 0.105± 0.003 (0 mg/L) to 0.537± 0.005 (50 mg/L), 0.717± 0.003 (100 mg/L) and 0.954± 0.006 (200 mg/L). All the increase was significant ($P < 0.01$). The massive increase in $[Ca^{2+})]_i$ and apoptosis as well as the reduction of cell proliferation suggest that grape procyanidins exert cytotoxic effect in MCF 7 cells. Clinically this implies that grape procyanidins intake might play a beneficial role in human breast cancer treatment.

O-89. Biochemical properties of breast cancer derived exosomes

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Exosomes are vesicles secreted by tumour cells, which are enriched in tumour specific antigens, costimulatory and MHC class I molecules. They have been used to stimulate cytotoxic T cell responses against breast cancer cells *in vitro*, and have great potential in immunotherapy. A successful phase I clinical trial in melanoma has been completed. Tetraspanins are proteins expressed on exosomes which have important roles in cell signalling and adhesion. Both CD9 and CD82 expression have been shown to correlate with metastasis in breast cancer.

We isolated exosomes from established breast cancer cell lines and normal breast epithelial cells, and examined the expression of various tetraspanins under different culture conditions. Using SOS-PAGE and Western blotting we detected the presence on exosomes of MHC class I and tetraspanins CD9, CD63, CD81, CD82 and CD151. When cells were cultured with interferon gamma for 3 days, the expression of MHC class I increased markedly. In addition, novel tetramers of CD9 and CD151 were detected, and CD63 expression also increased. We

also analysed exosomes from breast cancer lines which were stably transfected to overexpress CD82. These were shown to express higher levels of MHC class I, which supports previous studies showing an association between CD82 and MHC class I.

These findings show that exosomal proteins can be modified by altering cell culture conditions. This may allow optimisation of exosome targeting to antigen presenting cells, in order to stimulate a cytotoxic T cell response against breast cancer *in vivo*.

O-90. Is mammographic spiculation an independent good prognostic factor in screen detected invasive breast cancer?

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Objective: The aim of this study was to look at the prognostic significance of pathologic and radiologic factors for screen detected invasive breast cancers of any size.

Material and Methods: The patient group was a consecutive series of 470 screen detected invasive breast cancers diagnosed between 1988 and 1998. Data regarding tumour type, grade, maximum invasive diameter, lymph node status and the presence or absence of vascular invasion was recorded as were the mammographic features of the lesion. Survival was ascertained from hospital records and cancer registry. Differences in survival were assessed using Kaplan-Meier survival curves with log-rank test for difference. The significance of any correlations was assessed using Chi square and Chi-square for trend. Multivariate analysis used a Cox proportional hazards model.

Results: At univariate analysis, large invasive size, the presence of definite vascular invasion, high histological grade and nodal involvement were associated with poorer breast cancer specific survival. Mammographic spiculation (the presence of either a spiculate mass or distortion) was associated with more prolonged breast cancer specific survival. The presence or absence of mammographic comedo calcification did not influence breast cancer specific survival. In a Cox multivariate analysis which included those factors significant in univariate analysis, size, grade, nodal stage and mammographic spiculation maintained their prognostic significance.

Conclusion: Mammographic spiculation is an independent good prognostic factor for screen detected invasive breast cancer. The mechanism of how mammographic spiculation confers a beneficial prognostic effect is not clear.

O-91. Nipple biopsies in Paget's disease of the breast: an 8 year retrospective study

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Background: Paget's disease of the breast is a relatively uncommon malignant process that can mimic benign diseases. It is a pathohistological diagnosis and different breast surgeons use different biopsy methods. This audit aims to assess whether there are benefits in a particular biopsy method being used.

Methods: A retrospective case audit was performed using the records of the Pathology department to gather the case records of patients who had undergone nipple biopsy over the last eight years at a Glasgow teaching hospital. The audit took into account: age, gender, presenting complaint, biopsy, biopsy result and definitive diagnosis.

Results: Over the last eight years in the Victoria Infirmary 121 nipple biopsies were performed, and of those 24 cases of Paget's disease were detected. Only two biopsy methods were used. Punch biopsies had a stronger predictive value than excision (100 v 90) but a lower negative predictive value (95 v 98.3). In seven cases overall there was more than one biopsy taken, six of these involved malignant disease. 9% of punch biopsies needed repeating compared with 1% of excision biopsies. These biopsies needed confirming with another to verify diagnosis

Conclusions: There appears no significant difference between the two main biopsy modalities in Paget's disease. Given the similar outcomes it may be postulated that punch biopsies be the first line as they can be performed in the outpatient setting with a smaller incidence of morbidity and allowing the patient to return home that day. However, punch biopsies often needed repeating and maybe more than one punch should be taken.

O-92. The frequency of breast cancer screening: results of a randomised trial

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This randomised trial in 110,000 women between 1989 and 1996 compared screening at the standard interval of 3 years (Controls – C) with screening annually (Trials – T), in women aged 50–64 who had undergone a prevalent screen.

A previous analysis used the Nottingham Prognostic Index (NPI) to predict outcomes of invasive carcinoma diagnosed; these predictions were based on observed survivals in cancers prior to 1988.

However survival within each NPI group has improved, due to better therapy. Recalculation is based on these new figures of outcomes within each NPI group.

1. Predicted outcomes for, are compared with the observed outcomes at, six years:

Invasive cancers

	Diagnosed		Predicted surviving at 10 years		Observed surviving (actuarial) at 10 years	
	C	T	C	T	C	T
	n	*n*	*n*	*n*	*n*	*n*
GPG	92	113	87	108	89	107
MPG	87	96	68	76	66	82
PPG	22	20	11	10	14	14
Total	201	229	166 (82%)	194 (85%)	169 (84%)	203 (89%)

There is good agreement between the predicted and observed 10 year survivals. Neither show significant difference between C and T groups.

2. Although in the Trial group there were more cases in the GPG and less in the PPG, this was not large enough to significantly improve survival and the absolute difference is