Parallel Symposia

SY-4. Prognostic Factors: Methodology of Evaluation and Impact on Treatment (September 12)

SY-4-1 Proliferation Markers in Breast Cancers

F. Darro*, R. Kiss. *Laboratoire d'Histologie, Faculté de Médecine, Université Libre de Bruxelles, Bruxelles, Belgique*

The ability to divide subsets of patients with breast cancers into prognostic groups is currently limited. With respect to the determination of proliferation markers in breast cancers, a major problem relies in the confusion which is systematically made between tumour growth and tumour proliferative activity, which are not synonyms. Tumour growth represents the balance between cell gain (number of cells produced per unit of time, i.e. mitoses) and cell loss (number of cell deaths during the same unit of time, for example by apoptosis). Proliferative activity, which can be determined by numerous types of markers (including Ki-67 (or MIB-1), PCNA, S-phase, etc...), relates only to the cell gain compartment. A number of methods are useful in the determination of the cell loss compartment, but are too complex or too tedious to be applied in pathological routines. We therefore developed an original methodology which makes it possible to assess both proliferative activity and cell density (the balance between cell gain and cell loss) in a given breast cancer. Cell density is assessed on Feulgen-stained sections by means of computer-assisted microscopy. Proliferative activity is quantitatively (computer-assisted microscopy) assessed by means of the anti-MIB1 antibody. The relationship between proliferative activity and tissue differentiation was established by quantitatively determining the immunohistochemical amounts of estrogen and progesterone receptors in the series of 20 breast cancers studied here. The present methodology has been validated at the clinical level with respect to sarcomas and astrocytic tumours.

SY-4-2 Invasiveness Markers

E. Roos. *Division of Cell Biology, The Netherlands Cancer Institute, 121 Plesmanlaan, 1066 CX Amsterdam, The Netherlands*

We have studied adhesion mechanisms in metastasis, hoping to detect molecules that may be used as marker for metastatic capacity. For three carcinoma cell lines, including two variants of the same murine mammary carcinoma, we found that they use three different adhesion molecules to bind to hepatocytes, an important step in the formation of liver metastases. The importance of these molecules was demonstrated with knockout mutants of an embryonal carcinoma, lacking $\beta 1$ integrins, which had a strongly reduced metastatic capacity, in the liver probably because they lack the $\alpha 5\beta 1$ integrin fibronectin receptor.

However, some carcinoma cells express the alternative fibronectin receptor alpha;V$\beta 6$, which may take over this function. We are testing this by expressing $\alpha V\beta 6$ in the mutants. The $\alpha V\beta 6$ integrin is only found in carcinomas and in healing wounds, and may be involved in migration. Also $\alpha 5\beta 1$ is normally not present in epithelial cells, but upregulated in wounds. These are a few examples of the similarities between tumors and wounds, that will be discussed. A systematic comparison may yield additional markers.

SY-4-3 Cell Surface Mucins in the Biology and Treatment of Breast Cancer

D.W. Miles, J. Burchell, R.D. Rubens, J. Taylor-Papadimitriou. *ICRF Clinical Oncology Unit, Guy's Hospital, London, SE1 9RT*

Cell surface mucins are complex glycoproteins normally expressed on the apical surface of breast epithelium. In malignancy, the MUC-1 gene which encodes polymorphic epithelial mucin (PEM) is upregulated and its apical distribution is lost. Premature sialation of core-region carbohydrates and reduced expression of glucosyl transferases lead to reduced glycosylation of the peptide backbone and consequent exposure of novel peptide and carbohydrate epitopes. Expression of the novel carbohydrate epitope sialyl-Tn (STn), has been noted in ductal carcinoma *in situ* (DCIS) where correlation with grade of DCIS has been noted. In infiltrating carcinoma expression of STn did not correlate with established prognostic features though expression was associated with a poor prognosis and appeared to predict relative resistance to adjuvant chemotherapy. Humoral and T-cell responses to PEM have been noted in patients with breast cancer and though their prognostic significance remains unclear, preclinical studies have demonstrated that the ability to generate an immune response to peptide or carbohydrate epitopes on PEM can lead to tumour rejection. More recent clinical studies have confirmed that the immune response to such immunogens may be influenced by pretreatment with low dose cyclophosphamide and that ability to develop a specific immune response may influence outcome in patients with metastatic disease.

SY-4-4 Immunohistochemistry as a Tool for New Prognostic Markers

G. Mac Grogan. *Department of Pathology. Institut Bergonié, 33076 Bordeaux, France*

Technical improvements of the IHC method, including antigen unmasking methods enable utilization of new antibodies and the study of fixed and paraffin-embedded breast cancer fragments. At Institut Bergonie we tested antibodies to ER, PR, P53, c-erbB2, PS2, GSTp and mib1.

- First in a series of 942 invasive ductal carcinomas treated by primary surgery between 1980 and 1986, we found very good correlation, and equivalent prognostic value for the IHC and DCC methods of assessing ER and PR. By multivariate analysis, P53 and c-erbB2 were found to be independent markers of poor prognosis, in the whole group for P53 and the node negative group for c-erbB2. However they were of lesser importance than classical prognostic factors.
- Second in a series of 128 pretherapeutic core biopsies of breast invasive carcinomas treated by primary chemotherapy. Tumor regression was highly correlated to IHC-ER negativity and mib1 strong positivity (> 40%). C-erbB2 was the only independent marker of poor prognosis in this series.
- Last in a series of 208 pretherapeutic core biopsies of breast invasive carcinomas, in post menopausal women treated by tamoxifen. Tumor regression was only linked to PS2 and IHC-ER expression.

Our results show that classical factors are still the most important prognostic factors for breast cancers treated by primary surgery, whereas IHC factors are reliable predictors of tumour response and subsequent outcome, following primary hormono- and chemotherapy.

SY-5. Adjuvant Treatments for Low Risk Patients (September 12)

SY-5-1 Risk Determination for Good Prognosis Patients

M. Namer. *Centre A-Lacassagne, 06054 Nice, France*

Analysis of prognostic factors for low risk breast cancer patients allows identification of those who should be given adjuvant treatment. Classification in this group is based on absence of metastatic nodes in axillary resection. Tumor size is the 1st clinical parameter used to select patients with a very low risk. Analysis of 1018 breast cancers operated at our institute from 1975–1995 revealed that 88% of the tumors < 10 mm size were N-. Disease-free survival for these tumors at 10 and 15 yr was 83% and 79%; overall survival was 87% and 85%. The 2nd parameter influencing management is tumor differentiation (histologic grade, hormone receptor status, DNA ploidy); the 3rd step is assay of proliferative markers (thymidine labelling index, S-phase fraction). Determination of primary tumor cells' potential for invasion and metastasis by search for and assays of certain proteases needed to catalyze or degrade basement membrane directly is now essential. For 246 N– patients at our institute, Cathepsin D did not give us any useful data but the Tumor Angiogenesis Count identified a subgroup with an excellent prognosis regardless of other parameters. Association of urokinase Plasminogen Activator and Plasminogen Activator