Friday, 18 April 2008 16:00–17:15

CLINICAL SCIENCE SYMPOSIUM

New insights into the biology of metastatic breast cancer

393 Invited

Detection and characterization of disseminated tumor cells present in bone marrow of cancer patients

K. Pantel[1], V. Müller[2]. [1]*University Medical Center Hamburg Eppendorf, Department of Tumorbiology, Hamburg, Germany;* [2]*University Medical Center Hamburg Eppendorf, Department of Gynecology University Breast Center, Hamburg, Germany*

Early tumor cell dissemination occurs even in patients with small breast cancer tumors and bone marrow (BM) is a common homing organ for blood-borne disseminated tumor cells (DTC) derived from primary tumors. Immunocytochemical or molecular assays allow the detection of single DTC in BM at a frequency of one tumor cell in one million surrounding hematopoetic cells, and tumor cells are frequently detected in the bone marrow of breast cancer patients without clinical or even histopathologic signs of metastasis. Evidence has emerged that the detection of DTC can provide important prognostic information and in addition might help to monitor efficacy of therapy. However, it is crucial to improve and standardize methods for the detection of DTC. Moreover, the characterization of DTC has shed new light on the complex process underlying early tumor cell dissemination and metastatic progression in cancer patients. Characterization of DTC should help to identify novel targets for biological therapies aimed to prevent metastatic relapse and to monitor the efficacy of these therapies.

394 Invited

Circulating tumor cells and breast cancer

D.F. Hayes[1], J. Smerage[1], G. Luker[1], J.P. Eliane[1], G.N. Doyle[2]. [1]*Breast Oncology Program, University of Michigan Comprehensive Cancer Center, Ann Arbor, MI, USA;* [2]*Immunicon, Inc, USA*

Circulating tumor cells (CTC) have been observed for over 100 years. However, technology has been insufficient to reliably and accurately detect and quantify CTC until recently[1]. Several techniques have been employed to do so, based on differences between CTC and normal hematopoetic constituents, such as size, density, and/or expression of epithelial or "tumor-specific" genes.

In a prospective pilot trial, CTC were monitored at baseline and serially during systemic therapy in 177 women with metastatic breast cancer using the CellSearch™ assay[2]. Approximately 50% of these women had elevated CTC (⩾5 CTC/7.5 ml whole blood), which declined to approximately 30% after a single cycle of treatment[2]. Patients with elevated CTC at baseline had a worse prognosis (shorter time to progression and survival) than those without elevated CTC. At first follow-up this distinction was even more striking, suggesting that disappearance of CTC associated with therapy is highly suggestive of benefit from that therapy, regardless of whether all patients were considered or only those starting first line therapy[2,3]. CTC levels appeared to be a better predictor of long term survival than classic measures of disease course, including history, physical examination, or radiographic evaluation, either by the treating physicians or by an independent panel[4]. Furthermore, CTC levels at any time point were associated with a subsequently high risk of rapid progression[5]. The clinical utility of these findings are now being prospectively addressed in a randomized trial conducted within the North American Breast Cancer Intergroup, led by the Southwest Oncology Group[6].

CTC may also be important in stage early breast cancer. Using rt-PCR, investigators have suggested that up to 30% of patients with stage I–III breast cancer have elevated CTC levels[7], while a large prospective study has shown that approximately 10% are positive using CellSearch™ (unpublished). Elevated CTC at baseline and after adjuvant therapy appear to be prognostic, but the precise clinical utility of these findings has not been determined.

Although enumeration of CTC appears to be clinically important, it is not perfectly accurate. Therefore, several investigators have reported attempts to phenotype and genotype CTCs. These include measures of aneusomy and apoptosis as well as quantitative levels of gene amplification and/or expression of a variety of markers, including HER2, bcl-2, NOTCH, and IGFR-1[6]. The precise clinical utility of these assays will depend on careful technological development as well as prospective, hypothesis-based clinical trials. We have recently developed a pre-clinical murine xenograft model in which human CTCs can be identified and further characterized with the CellSearch system. This model should facilitate development of new markers that can then be applied in clinical studies.

References

[1] Smerage JB, Hayes DF. The measurement and therapeutic implications of circulating tumour cells in breast cancer. Br J Cancer 2006;94:8–12.
[2] Cristofanilli M, Budd GT, Ellis MJ, et al. Circulating tumor cells, disease progression, and survival in metastatic breast cancer. N Engl J Med 2004;351:781–91.
[3] Cristofanilli M, Hayes DF, Budd GT, et al. Circulating tumor cells: a novel prognostic factor for newly diagnosed metastatic breast cancer. J Clin Oncol 2005;23:1420–30.
[4] Budd GT, Cristofanilli M, Ellis MJ, et al. Circulating tumor cells versus imaging – predicting overall survival in metastatic breast cancer. Clin Cancer Res 2006;12:6403–9.
[5] Hayes DF, Cristofanilli M, Budd GT, et al. Circulating tumor cells at each follow-up time point during therapy of metastatic breast cancer patients predict progression-free and overall survival. Clin Cancer Res 2006;12:4218–24.
[6] Smerage JB, Doyle G, Budd GT, et al. Circulating Tumor Cells in Breast Cancer Patients Before and During NeoAjuvant Chemotherapy. Proceedings of the American Society of Clinical Oncology, 2006.
[7] Xenidis N, Perraki M, Kafousi M, et al. Predictive and prognostic value of peripheral blood cytokeratin-19 mRNA-positive cells detected by real-time polymerase chain reaction in node-negative breast cancer patients. J Clin Oncol 2006;24:3756–62.

395 Invited

Expression profiles predicting metastatic relapse at different sites

Abstract not received.

396 Proffered Paper Oral

HER2 status correlation between Circulating Tumor Cells (CTC) and corresponding primary tumor in advanced breast cancer patients (pts)

M. Pestrin[1], S. Bessi[1], M. Truglia[1], F. Galardi[1], S. Cappadona[1], C. Biagioni[1], W. Claudino[1], L. Biganzoli[1], A. Giannini[1], A. Di Leo[1]. [1]*Sandro Pitigliani Medical Oncology Unit, Translational Research Unit Dept. of Oncology Hospital of Prato Istituto Toscano Tumori Italy, Prato, Italy*

Background: CTC detection and biocharacterization in the peripheral blood of breast cancer pts might represent a real-time biopsy with minimal pts discomfort. We aimed to assess HER2 *status* on CTC from breast cancer pts using immunomagnetic-capture technology and correlate these results with HER2 status evaluated on primary tumor.

Material and Method: 20 ml blood sample was drawn from breast cancer pts. CTC were immunomagnetically separated and fluorescently stained using the CellSearch® System.

CTC were defined as nucleated cells positive for cytokeratins and negative for CD45. Cases were defined as CTC positive if >1 CTC/7.5 ml of blood were identified.

The HER2 status on CTC was assessed by immunofluorescence (IF) and when feasible by fluorescence in-situ hybridization (FISH). A case was defined as HER2 positive if ⩾50% of CTC expressed the protein and/or if HER2/CEP17 ratio was ⩾2.2.

Results: 84 pts were enrolled in this study and 54 (64%) were CTC positive (48/54 pts with advanced disease).

Concordance in terms of the HER2 status between the primary tumor and corresponding CTC was evaluated in 40 cases. We found non-concordant results in 32% (13/40) of cases. 29% (8/28) of HER2 negative primary tumors had HER2 positive CTC and 42% (5/12) of HER2 positive primary tumors had HER2 negative CTC (table).

We have found that the two methods (IF and FISH) had an agreement rate of 88% in defining HER2 status on CTC ($k = 0.71$ and $p = 0.0002$).

Conclusion: Our study suggests that a subset of pts with HER2 negative primary tumors develops HER2 positive CTC during disease progression. Clinical evaluation of anti-HER2 compounds in this particular subset of pts is warranted.

		CTC[a]		
		HER2 +	HER2 −	Tot
Primary tumor[b]	HER2 +	7	5	12
	HER2 −	8	20	28
	Tot	15	25	40

[a]HER2 status evaluated by IF.
[b]HER2 status evaluated by FISH and immunohistochemistry.