The influence of mesenchymal stem cell differentiation in the development of a pre-metastatic niche for breast cancer bone metastases

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Introduction: Despite advances in treatment, over 80% of patients with advanced breast cancer will develop bone metastases for which there is no cure. Chemokines secreted from bone derived cells are believed to create a favourable microenvironment for the homing and engraftment of circulating cancer cells. Mesenchymal Stem Cells (MSCs) are found predominantly in bone marrow stroma and being pluripotent, have the ability to differentiate into a number of cell types, including osteoblasts. This differentiation may affect the chemokines secreted and the predilection for metastatic development. The aim of this experiment was to investigate chemokines secreted during MSC differentiation to mature osteoblasts, their relative levels of expression, quantification of certain chemokines and their role in the migration of circulating breast cancer cells to bone.

Materials and Methods: Primary culture of MSCs, osteoblast progenitor cells (NHOst cells) and breast cancer cell lines (MDA-MB-231 and BT-474) was performed. Breast cancer cell migration in response to MSCs and NHOst cells was measured using Transwell™ inserts. Media containing β-Glycerophosphate, ascorbic acid and dexamethasone was used to induce MSC differentiation into osteoblasts. MCP-1 and VEGF were quantified using ChemiArray™ and ELISA™ at various stages of differentiation. The potential role of MCP-1 in breast cancer cell migration was investigated using a monoclonal antibody to the chemokine

Results: There was a significant increase in migration of both breast cancer cell lines in response to factors secreted by NHOst cells (5-10 fold increase) and MSCs (6-10 fold increase). MSCs were shown to secrete a range of chemokines including IL-6 & 8, TIMP 1 & 2 and MCP-1. Levels of MCP-1 secreted by differentiating MSCs increased from 319 pg/ml (Day 3) to 12,280 pg/ml (Day 21), while VEGF increased from 100 pg/ml (Day 3) to 1040 pg/ml (Day 21). MSC differentiation into osteoblasts was confirmed by the presence of calcium deposits following Von Kossa staining. A monoclonal antibody to MCP-1 resulted in inhibition of MDA-MB-231 (20% reduction) and BT-474 (30% reduction) migration in response to NHOst cells, confirming a role for this chemokine in the migratory effects seen.

Discussion: Chemokine secretion has been implicated in mediating cell-cell interactions in the metastatic cascade. The variation in both type and level of chemokines detected during differentiation may influence the development of a favourable premetastatic niche and subsequent homing and engraftment of circulating cancer cells. Further investigation of the specific mode of action of these chemokines may provide novel therapeutic targets for treatment of advanced breast cancer.