Comparative qualitative proteomic analysis of conditioned media from breast cancer cell lines

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ABSTRACT

Introduction: One of the best ways to diagnose breast cancer early or to predict therapeutic response is to use serum or tissue biomarkers. Unfortunately, for breast cancer, we do not have effective biomarkers for either early detection or prediction of therapeutic response. Therefore, we are examining the tissue culture supernatants of breast cancer cell lines.

Methods: A proteomic approach, a two-dimensional liquid chromatography-mass spectrometry (2D-LC-MS/MS) strategy was utilized to identify and compare levels of extracellular and membrane-bound proteins. Seventy-five secreted human breast cancer cell lines were cultured in serum-free media. Proteins from the conditioned media were digested overnight at 37°C using Sequencing grade modified trypsin. Digested peptides were loaded onto a SCX column (LC-MS/MS) to identify the peptides present. Comparative analysis of the protein identified in the normal, non-invasive and metastatic cell lines will provide candidate tumor markers for further investigation.

Results: 581 proteins were successfully identified over 400 proteins, using a 2-D linear ion-trap mass spectrometer (LTQ). These proteins, from conditioned media of the breast cancer cell line MDA-MB-468. Two independent samples of the cell lines were processed and over 250 secreted and/or membrane-bound proteins were identified in total. Among the identified proteins were proteases, receptors, protease inhibitors, cytokines and cadherin development (cell growth, differentiation, metastasis), are linked to early onset breast cancer (E- breast cancer: a review.

Objectives: (1) to validate if identified proteins can be used as breast cancer biomarker.

SUMMARY & CONCLUSION

(1) Identified proteins are candidate molecules which can be examined in more detail for their ability to act as circulating breast cancer biomarkers.

REFERENCES


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Figure 2: Classification of proteins by cellular localization from MDA-MB-468 and the emerged of proteins identified by MS between sample #1 and #2. (A) Sample #1: A total of 437 proteins were identified with >40% being classified as extracellular and membrane-bound. (B) Table illustrating the breakdown of the cellular localization for sample #1. (C) Venn diagram: 193 proteins (~10%) are common between the two samples.