Quantitative Mass Spectrometry Based Breast Cancer Biomarkers Discovery in a Mouse Model, Jian Li, MCB seminar, spring 2010

During 2002 to 2006, 123.8 per 100,000 women were diagnosed for cancer of the breast in the United States, and 24.5 per 100,000 were died of the breast cancer. Early diagnosis significantly increases the 5-year relative survival rate of the breast cancer patients. Biomarkers, genes differentially expressed in normal and cancerous tissues, have great implication in cancer diagnosis and treatment. The microarray and proteomics are powerful tools in detecting the differential gene expression at transcriptional and translational levels. However, the difficulties laying in the refinement of hundreds of the candidates generated surrender their feasibilities. Only very few biomarkers are in clinical application. Paulovich et al reported a quantitative mass spectrometry based method to identify the biomarkers for breast cancer in a mouse model. Specifically, the researchers generated a mouse model for breast cancer and the mammary tissue from cancerous and normal mouse were subjected to LC-MS/MS (Liquid chromatography-Mass spectrometry/Mass spectrometry). Based on the observation frequency in the MS spectra, they identified the up-regulation of 341 proteins (false discovery rate < 0.10) in the tumor tissue, among which only 24 proteins have commercially available antibodies for western blotting. Instead of generating antibodies for each of the candidates, the authors explored the selected reaction monitoring (SRM) mass spectrometry to quantify the biomarker candidates in the tumor tissues and in the plasma. In semi-quantitative SRM, a unique peptide was chosen to represent each of the candidates (60 proteins were randomly chosen from the 341 candidates). The signature peptides are selectively monitored by m/z filtration in a triple quadrupole mass spectrometer. 48 out of 60 proteins were confirmed to be more abundant in the tumor tissues, one was reported to have no change, and eleven were not detected. Furthermore, the researchers did quantitative SRM with heavy isotope labeled synthetic signature peptide. Based on the relative intensity calculated from endogenous peptides against synthetic peptides, they confirmed the elevated expression levels of eleven proteins among the fifteen proteins tested. Because of the low abundance of most cancer biomarker (in ng/ml range) in the plasma, the researcher incorporated an antibody based immuno-enrichment step into the quantitative SRM and successfully identified fibulin-2 as a circulating biomarker for the breast cancer. In a later study, the researchers confirmed the precision and high reproducibility of the quantitative SRM across different laboratories and instrumental platforms. The results from these studies proved that quantitative mass spectrometry is promising in the discovery of the breast cancer biomarkers, as well as other human diseases.
References


