## MDA-231





**Supplementary Figure 1.** Morphological aspect of MCF-7 and MDA-231 cells observed by phase contrast microscopy. MCF-7 has a cobblestone-like phenotype with strong cell-cell adhesion, whereas MDA-231 cells have an elongated fribroblast-like morphology, and pronounced cellular scattering. Basal mRNA and protein expression of mesenchymal and epithelial markers in MCF-7 and MDA-231 cells were detected using RT-PCR and western blotting. Expression of Vimentin, Snail, Slug and Twist was markedly increased in MDA-231. Tubulin  $\alpha$  and GAPDH were used as loading controls. Lysates from MDA-435 cell line served as positive control when evaluating the N-cadherin expression in Western blot analysis experiments.

## Electronic Supplementary Material (ESI) for Molecular BioSystems This journal is O The Royal Society of Chemistry 2013



**Supplementary Figure 2.** Spot volume % histograms of identified differentially expressed spots. p<0.05 \*; p<0.01 \*\*; p<0.001 \*\*\*.



**Supplementary Figure 3.** Principal component analysis (PCA) and Partial least square Discriminant analysis (PLS-DA) model. Analysis was performed using MetaboAnalyst 2.0 and Simca 13.0 on differentially expressed proteins of the 6 samples analyzed by 2-DE. PCA and PLS-DA were performed after autoscaling of data set as described in material and methods. a) 2D PCA Score plot ( $R^2$ =0.977,  $Q^2$ =0.947) showing a significant separation between the two breast cancer cell line: MCF7 (red circles) and MDA-231 (green circles). PC1 and PC2 account 93.8% and 3.9% of samples variation respectively. b) PCA Loadings plot based on the data set shows proteins that contribute to separation of score plot clustering. c) Scree Plot. Green lines represent the cumulative variance of the components. The blue line shows the proportion of variance for each principal component. PLS-DA model permutation test plots. 200 random permutations were carried out. d) MCF7 ( $R^2$ =0.489 Q<sup>2</sup>= -0.302). e) MDA-231 ( $R^2$ =0.506 Q<sup>2</sup>= -0.282). The low value of intercepts shows that the model has a statistical significance (not over-fitting). f) Variable Importance in the Projection (VIP) score of variables from the PLS-DA analysis. VIP score >1 indicates important x variables that predict y responses.



Supplementary Figure 4. Cell morphology of HBL-100, MDA-361, and MDA-435 breast cancer cell lines as examined by phase contrast microscopy. Western blot analysis of extracts from HBL-100, MDA-361, and MDA-435. Tubulin  $\alpha$  levels were used as loading control.