# CANCER DEGRADOMICS: ADAM17 REGULATES TGF-BETA SIGNALING THROUGH THE CLEAVAGE OF VASORIN

***J. Malapeira [(1)], C.W. Esselens [(1)], J.J. Bech-Serra [(2)], N. Colomé [(2)], J. Arribas [(1)], F. Canals [(2)].***

[(1)] Medical Oncology Research Program. Vall Hebron Institute of Oncology (VHIO). Vall Hebron University Hospital., [(2)] Proteomics Laboratory. Vall Hebron Institute of Oncology (VHIO). Vall Hebron University Hospital..

Metalloproteases play a complex role in tumor progression. Proteomic approaches to identify the array of substrates of a metalloprotease (degradome), can help to unveil its role in tumor growth and metastasis. Here we describe a proteomic screening using a SILAC-based liquid chromatography-mass spectrometry (LC-MS) analysis, to identify new substrates of the metalloprotease ADAM17, released to the conditioned medium of invasive mammary tumor MCF7 cells.

MCF7 mammary cancer cells were differentially labelled by SILAC, and then grown in the absence or presence of a protease inhibitor specific for the ADAM17 metalloprotease. Glycoproteins released to the conditioned media of each of the two cell cultures were purified by lectin affinity chromatography. Samples of the two conditions were pooled and run on a 1D SDS-PAGE gel. The gel lane was then cut into 20 fractions, and each one was digested and analyzed by reverse phase LC-MS/MS on an Bruker ion trap mass spectrometer.

Around 3200 different peptides, corresponding to ca. 750 individual proteins, were identified and quantified. A number of known substrates of ADAM17 were identified in the analysis, such us ALCAM and Desmoglein 2. In addition several new candidate substrates of the protease were identified, including the transmembrane protein vasorin (VASN).

Further characterization validates the ADAM17 dependent cleavage of VASN, and shows that only the secreted fragment released is able to inhibit TGFbeta signaling. Furthermore, through the cleavage of VASN the metalloprotease is able to control TGF-beta mediated epithelial to mesenchymal transition.

SILAC proteomic screening for specific substrates of ADAM17 metalloprotease in mammary cancer cells allowed the identification of potential new substrates of the protease.

The TGF-beta trap protein VASN was identified and further validated as a substrate of ADAM17, unveiling a regulatory mechanism of the TGF-beta pathway through the specific proteolysis of VASN by ADAM17.