Proteomics-based systems biology analyses of bladder cancer unravel a functional structure with prognostic value

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Introduction
Bladder cancer is the fourth most common tumor type among males (1). It divides into low-grade non-muscle invasive cancers and high-grade muscle-invasive cancers (MIBC). Invasive bladder tumors progress rapidly to become metastatic and they present high mortality (2). Traditionally, bladder cancer classification is based in histological features, although a molecular classification of MIBC has been proposed recently (3,4).

Material and methods
Tissue samples from MIBC were obtained from 58 patients. Proteome was analyzed applying a high-throughput proteomics approach to routinely archived formalin-fixed, paraffin-embedded tumor tissue. Tryptic digests were analyzed by mass spectrometry for protein identification using a Q-Exactive mass spectrometer. Subgroups were defined by random forest and Significance Analysis of Microarrays. Functional structure was developed using probabilistic graphical models with local minimum Bayesian Information Criterion and Gene Ontology Analysis. Data analysis was done using MeV, BRB Array Tools, R and Cytoscape software suites, as well as Uniprot and DAVID webtools.

Result 1
Using proteomics data and random-forest, two different molecular groups with differential prognosis were identified. Gene ontology analyses showed that proteins showing differential expression between both groups are mainly related with cellular adhesion. These results were confirmed using a Significance Analysis of Microarrays. Some of these proteins have been previously related with bladder cancer diagnosis and prognosis (5-8).

Result 2
Probabilistic graphical models showed that wide protein expression assessment allows building a functional network structure, with nodes showing a defined biological activity. Eighteen nodes with a majority function were defined and evaluated between both groups.

Result 3
Focal adhesion, metabolism, RNA and splicing nodes activities were different between both groups. In addition, focal adhesion node activity had prognosis value in MIBC.

Conclusions
• Protein data analysis through random forest showed MIBC groups with different prognosis.
• Results provided independently by Significance Analysis of Microarrays and random forest analyses support that different molecular subgroups exist in MIBC.
• We were able to establish different functional nodes with diagnostic and prognosis value in MIBC.
• These differential biological processes may represent new therapeutic opportunities for bladder cancer treatment.

Bibliography