Mass spectrometry-based proteomics to investigate biomarkers for non-small cell lung cancer

Saime Sürmen[1], Mustafa Gani Sürmen[2], Arslan Ali[3], Çağrı Çakıcı[4], İlhan Yaylım[1] and Nesrin Emekli[4]

[1]Department of Molecular Medicine, Aziz Sancar Institute of Experimental Medicine, Istanbul University, Turkey
[2]Department of Molecular Medicine, Hamidiye Institute of Health Sciences, University of Health Sciences, Turkey
[3]Dr. Panjwani Center for Molecular Medicine and Drug Research, International Center for Chemical and Biological Sciences, University of Karachi, Karachi 75270, Pakistan
[4]Department of Medical Biochemistry, Faculty of Medicine, Istanbul Medipol University, Turkey

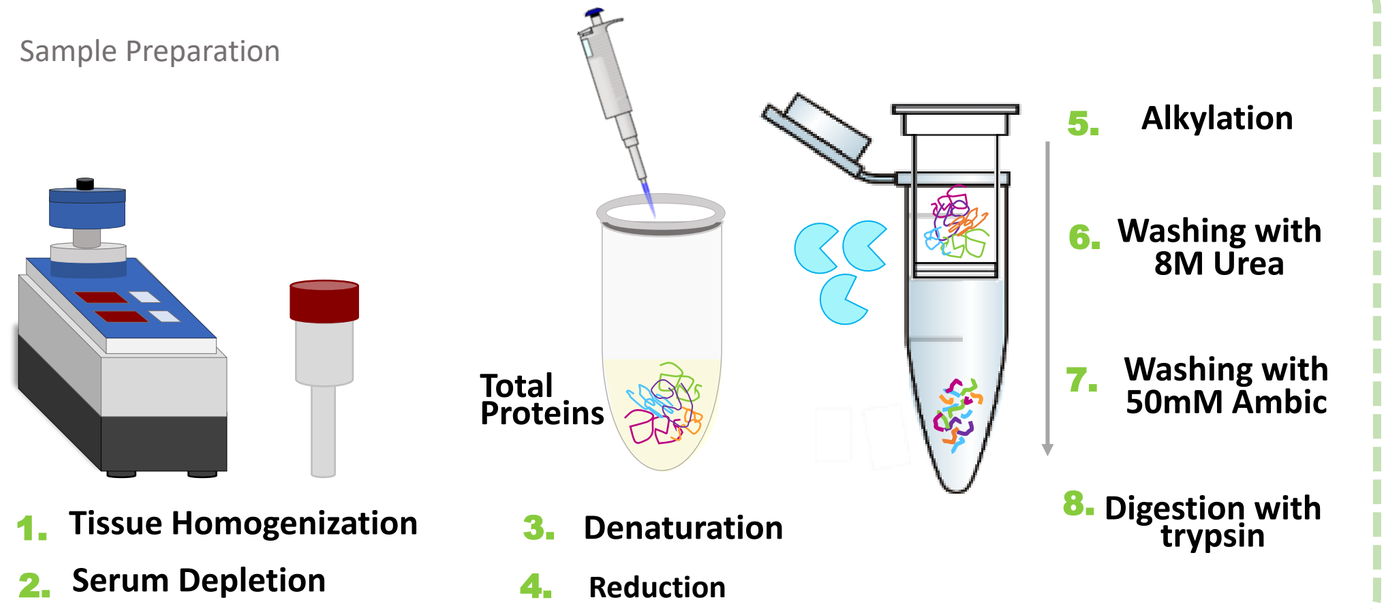
Background & Aim:

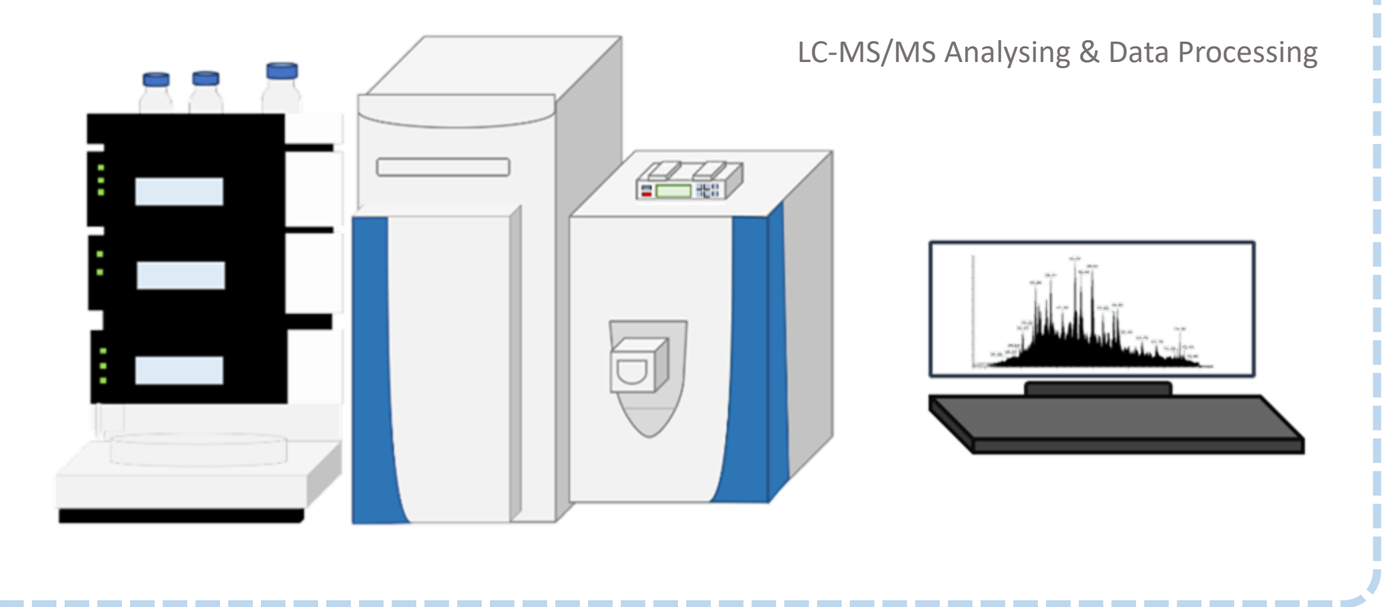
Lung cancer is a heterogeneous disease that includes various histological types. Large cell carcinoma is defined as non-small cell lung cancer (NSCLC) originating from the epithelial cells of the lung and indicating no apparent histological evidence of differentiation. To date, there is still very little knowledge about available that reveal the molecular features of LCC. On the other hand, proteomic studies using constantly updated methods and technologies provide important clues about how proteins in tumor cells change and interact.

Mass spectrometers, especially powerful tools in this field, are promising for a better understanding of proteins associated with cancer processes. Here, we aimed to reveal dysregulated proteins in lung tissue and serum samples from LCC patients by MS-based proteomic approaches.

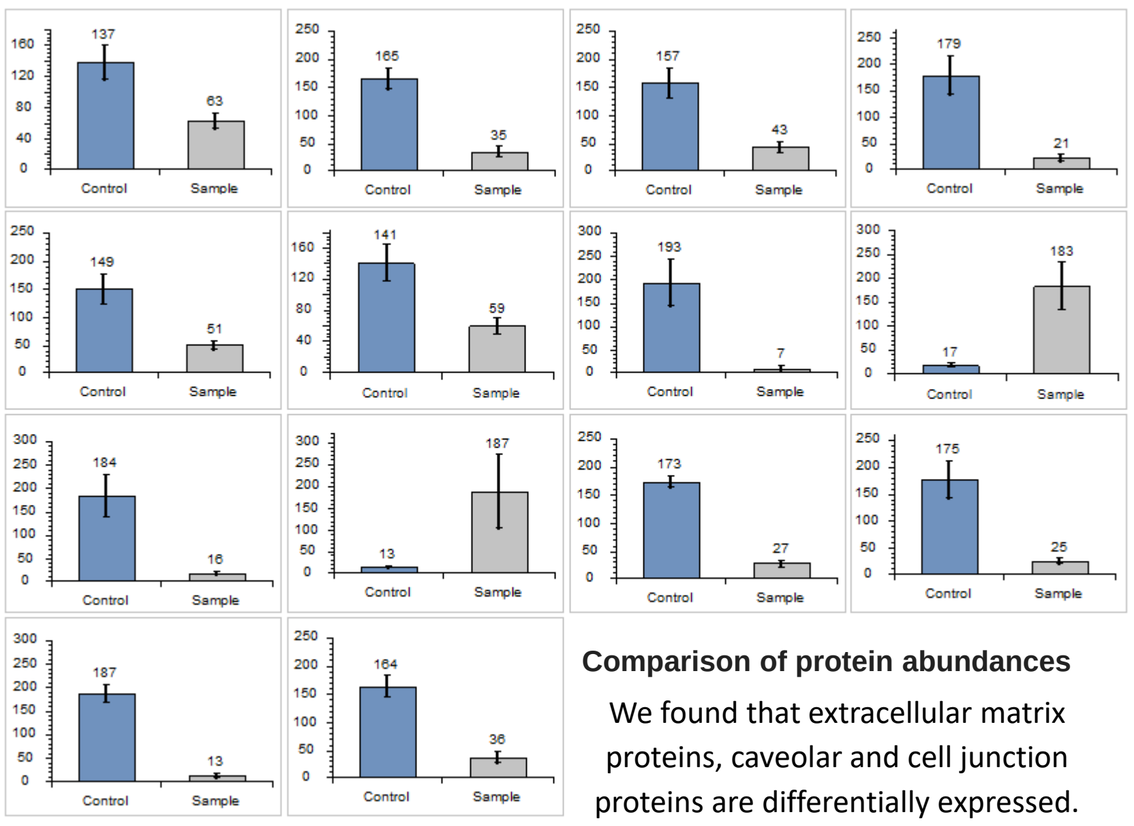
Material and Methods:

Tissue samples were homogenized in the buffer containing protease inhibitor cocktail, DTT and SDS. After that, digestion method was applied. Briefly, this method involves steps of alkylation, washing with urea, washing with ammonium bicarbonate and digestion with trypsin. MS analyzes were performed with the UltiMate 3000 RSLC nano system and Q Exactive Plus Mass spectrometer which was equipped with an ESI (electrospray ionization) source. Peptide and protein identifications were made using Proteome Discoverer software. The label-free LC-MS/MS method was used for quantification.





Results



Comparison of protein abundances

We found that extracellular matrix proteins, caveolar and cell junction proteins are differentially expressed.

Protein accession numbers: P12111, O15230, Q969G5, Q03135, P0301, P11047, O95810, P24821, P22105-4, P01033, P56199, Q6NZI2-1, A6NMZ7, P55268

Pathway enrichment analysis of differentially expressed proteins

a.

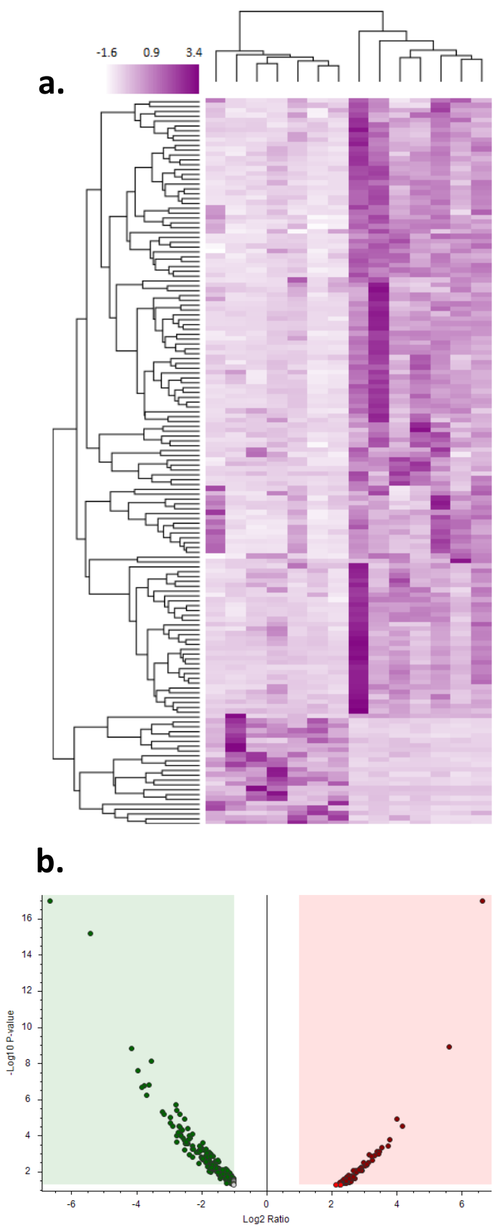
#term ID	term description	observed count	background count	FDR
HSA-1474244	Extracellular matrix organization	31	298	8.66e-16
HSA-3000178	ECM proteoglycans	15	75	9.18e-11
HSA-1474228	Degradation of the extracellular matrix	17	139	1.97e-09
HSA-3000157	Laminin interactions	10	30	5.95e-09
HSA-1500931	Cell-Cell communication	14	127	2.44e-07
HSA-216083	Integrin cell surface interactions	12	83	2.44e-07
HSA-445355	Smooth Muscle Contraction	8	32	2.09e-06
HSA-109582	Hemostasis	26	601	5.76e-06
HSA-2022090	Assembly of collagen fibrils and other multimeric structures	9	60	1.07e-05
HSA-8874081	MET activates PTK2 signaling	7	29	1.36e-05

b.

#term ID	term description	observed count	background count	FDR
HSA-8953854	Metabolism of RNA	26	652	0.00033
HSA-69278	Cell Cycle, Mitotic	20	483	0.0022
HSA-5696397	Gap-filling DNA repair synthesis and ligation in GG-NER	5	25	0.0086
HSA-174411	Polymerase switching on the C-strand of the telomere	4	14	0.0127
HSA-6781827	Transcription-Coupled Nucleotide Excision Repair (TC-NER)	7	78	0.0127
HSA-69091	Polymerase switching	4	14	0.0127
HSA-69190	DNA strand elongation	5	31	0.0127
HSA-73933	Resolution of Abasic Sites (AP sites)	5	36	0.0127
HSA-74160	Gene expression (Transcription)	35	1366	0.0127
HSA-8868773	rRNA processing in the nucleus and cytosol	10	189	0.0127

a. Downregulated proteins b. Upregulated proteins

Differentially expressed proteins were significantly enriched according to the Reactome pathway analysis.



Significantly changed proteins in lung tissues
a. Heatmap b. Volcano-plot

Conclusion

Our results address changes in proteins that explain the invasion and metastasis. Consequently, quantitative proteomic analyzes imply that these proteins may have an important role as potential cancer biomarkers for LCC.

References

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Keywords: Lung cancer, large carcinoma cell, quantitative proteomics, mass spectrometry

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