



**Karolinska
Institutet**

Department of Medical Biochemistry and Biophysics

PROTEOME STUDIES, WITH EMPHASIS ON THE KIDNEY GLOMERULUS

AKADEMISK AVHANDLING

som för avläggande av medicine doktorexamen vid Karolinska
Institutet offentligen försvaras i Hörsal Samuelsson,
Tomtebodavägen 6, Karolinska Institutet, Solna

Fredagen den 11 maj 2012 kl 9.30

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Stockholm 2012

ABSTRACT

Proteomics aims to link genes to protein expression and function. Proteomics is a very versatile and technology-dependent discipline and the development of mass spectrometry-based proteomics has advanced the understanding of the complex and dynamic nature of proteomes. Proteomics does not have a “one-size-fits-all” strategy to answer all biological questions. New technical approaches are needed to overcome the hurdles, e.g. issues related to complexity and dynamic ranges of the samples, high throughput and post-translational modifications, in the field of proteomics. Due to recent technological development, proteomics has become complementary to genomics and transcriptomics in systems biology and can provide unique contributions.

In this thesis a novel microfluidic compact disc based system was tested in use of high throughput sample preparation of digested peptide samples prior to matrix assisted laser desorption/ionization mass spectrometry (Paper I). A novel microfluidic electrocapture technology able to immobilize charged biomolecules in an electric field along a hydrodynamic flow was further developed to establish its ability to separate peptides of protein digests (Paper III). The same technology was later also used in analysis of glomerular membrane proteins as a part of a two-dimensional fractionation system in combination with liquid chromatography mass spectrometry (Paper IV).

The kidney glomerulus functions as a specialized blood filter unit that forms the primary urine. The filtration barrier is composed of a three-layer barrier, which consists of endothelial cells, the glomerular basement membrane and the slit diaphragm between epithelial podocyte foot processes. The glomeruli are sensitive to damage and glomerular diseases underlie the majority of diseases leading to chronic kidney disease and renal failure. Even though the molecular composition of the glomerulus is quite well known, the pathomechanisms of glomerular diseases are still poorly unknown. The mouse glomerular proteome was now studied with two-dimensional gel electrophoresis in combination with mass spectrometry (Paper II). Finally a meta-analysis of currently available proteomic and transcriptomic glomerular expression data in normal and disease states was performed to explore the usability of expression profiling in the diagnosis, prognosis and prediction of glomerular diseases (Paper V).

ISBN 978-91-7457-696-2