

DEFINING RENAL DISEASES BASED ON PROTEOM ANALYSIS

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We have utilized capillary electrophoresis coupled to high-resolution mass spectrometry (CE-MS) to examine urine samples from healthy volunteers and patients with different diseases with the aim to identify disease-specific polypeptides (biomarkers). Using this technology, typically, >1000 polypeptides can be analyzed per sample with very high reproducibility.

We will present data on samples from in total more than 1000 healthy volunteers and patients with IgA nephropathy, focal-segmental glomerulosclerosis, membranous glomerulonephritis, minimal-change disease, lupus nephritis, renal cancer, vasculitis and diabetes/diabetic nephropathy. Due to the advanced analysis technology, all data on individual patients can be compared. Comparison enabled the definition of between 10 and 100 biomarkers specific for a given renal disease, which allowed diagnosis and discrimination between different renal diseases with generally > 90% sensitivity and specificity. These results were further validated in a blinded study. Several of these biomarkers were identified using CE-MS/MS sequencing.

The high number of biomarkers defined for a specific disease suggested that this approach might enable staging and evaluation of disease progression and therapy. First results indicate that this is the case. These promising results indicate that CE-MS analysis might be a powerful tool for the evaluation of therapeutic strategies.

The “ideal biomarker”, indicative exclusively for one disease does not exist. Instead, an array of polypeptides that shows significant differences in distribution between disease and healthy can be detected using CE-MS. The combination of biomarkers to generate distinct diagnostic patterns will make the method both more robust and specific. Spurious occurrences of peptides can be eliminated without jeopardizing the overall results and diagnosis. This also indicates that for the evaluation of a pathophysiological state, not a single (or a few) biomarkers should be used, but an array of distinct biomarkers that result in a specific pattern. Combining these polypeptides to a signature or pattern results in a) increased ability to distinguish between disease and healthy and b) increased stability, since the absence or untypical amplitude of single peptides does not result in a significant change of

the typical pattern.

These results suggest that CE-MS analysis of urinary polypeptides should be capable of displaying most pathological changes in the renal system. Further, this will most likely be possible at a very early point in time, since the initial pathological changes must be reflected by indicative changes in polypeptides.

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