Towards obtaining molecular signatures of glomerulonephritis by MALDI-MSI: Preliminary evidence

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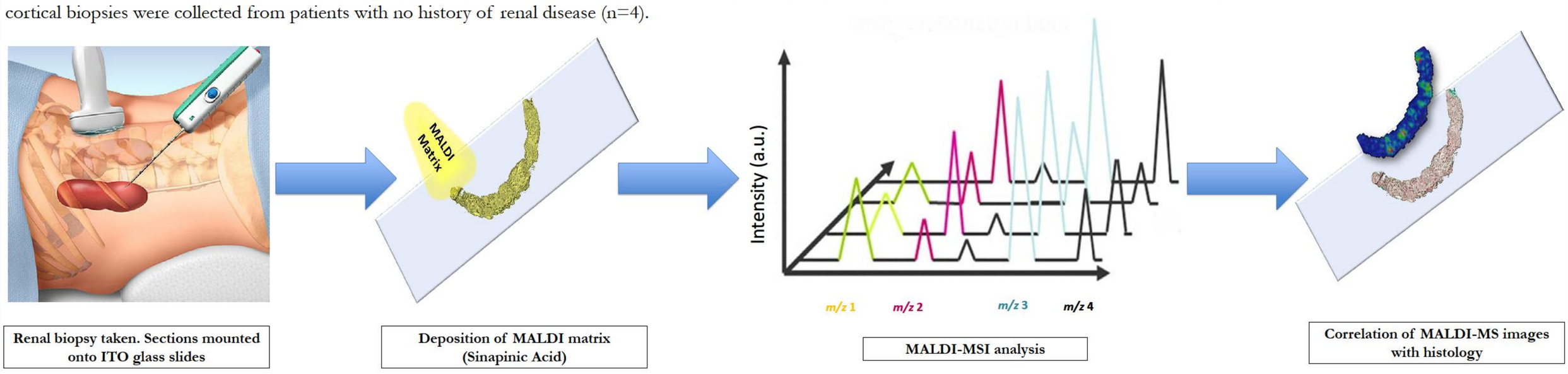
Introduction

Nephropathology is still founded upon traditional approaches, with morphological analysis, along with direct immunofluorescence (IF) and electron microscopy (EM), representing the gold standard for the analysis of renal biopsies[1]. However, in particular cases of glomerulonephritis (GNs), the difficult to interpret analysis and inter-observer variability can lead to an indeterminate or even misdiagnosis of a patient[2]. This results in the need for multiple invasive biopsies to be taken from the same patient. Therefore, there is a strong need for an alternative approach that improves the diagnostic success rate and is less invasive.

Matrix-assisted laser desorption/ionization (MALDI) mass spectrometry imaging (MSI) is a unique proteomic technology that explores the spatial distribution of biomolecules directly *in situ*, thus integrating molecular and morphological information. The feasibility of this technology for the analysis of frozen renal biopsies that have been routinely collected from the clinic has previously been explored[3]. Here, the study aims applies the same technology in order to establish molecular signatures of GNs that could potentially assist in the differential diagnosis of ambiguous cases of GNs.

Materials and methods

Sample collection - Renal biopsies with focal segmental glomerulosclerosis (FSGS, n=6), IgA Nephropathy (IgAN, n=6) and membranous glomerulonephritis (MGN, n=7) were selected. Normal cortical biopsies were collected from patients with no history of renal disease (n=4).



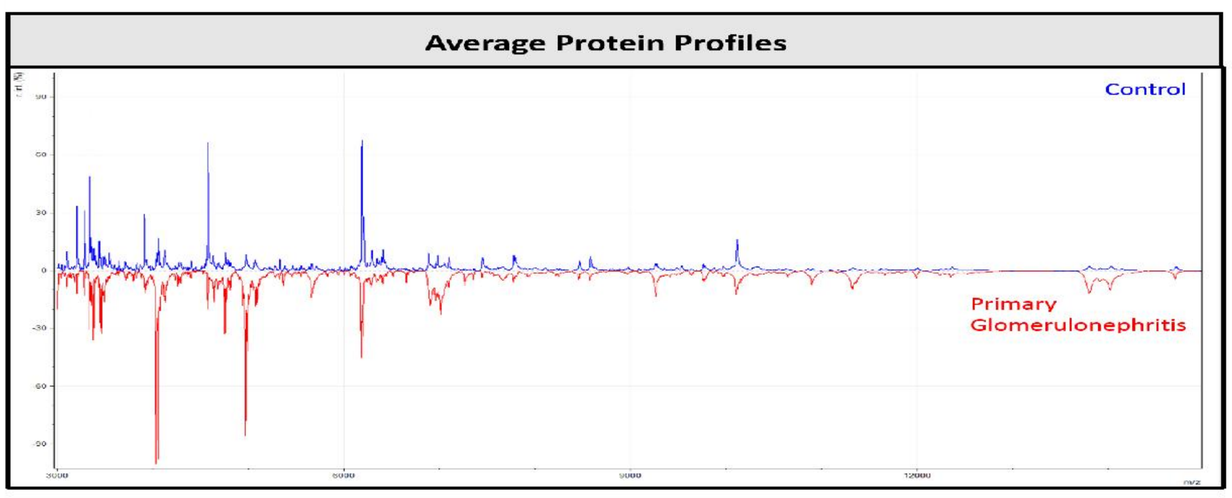
Renal biopsy taken. Sections mounted onto ITO glass slides

Deposition of MALDI matrix (Sinapinic Acid)

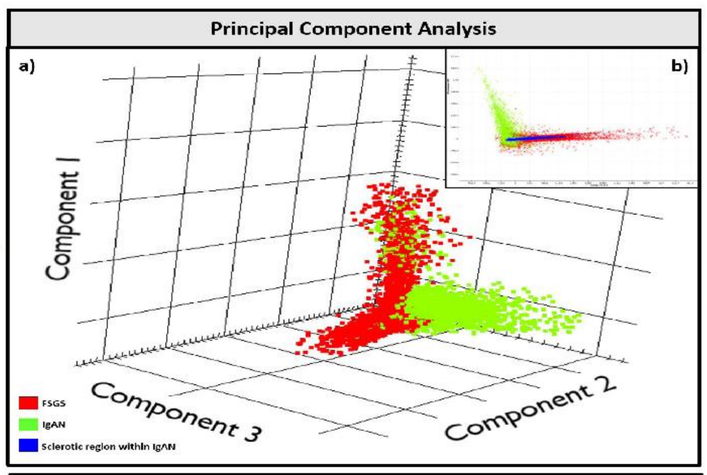
MALDI-MSI analysis

Correlation of MALDI-MS images with histology

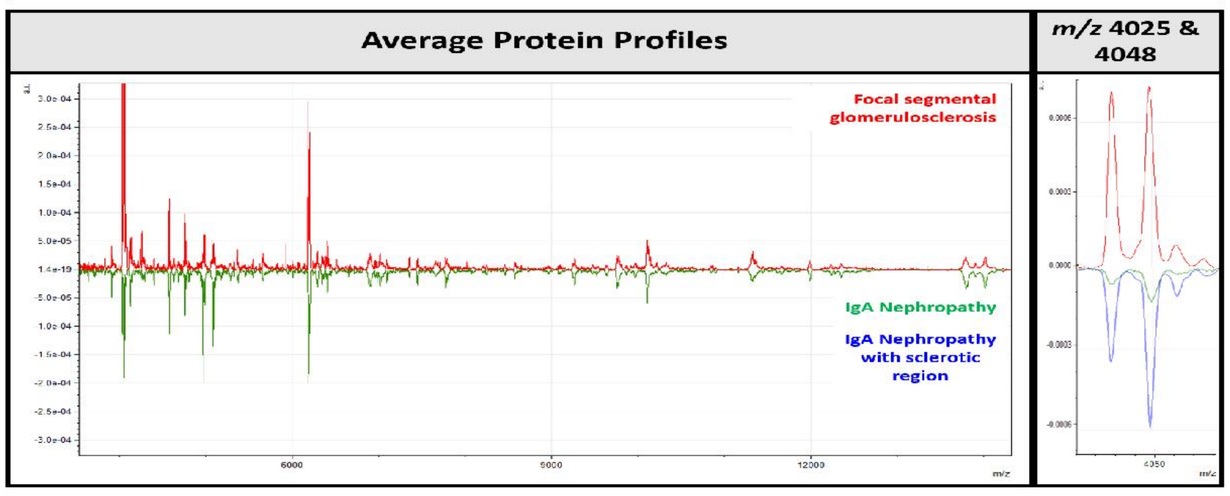
Results



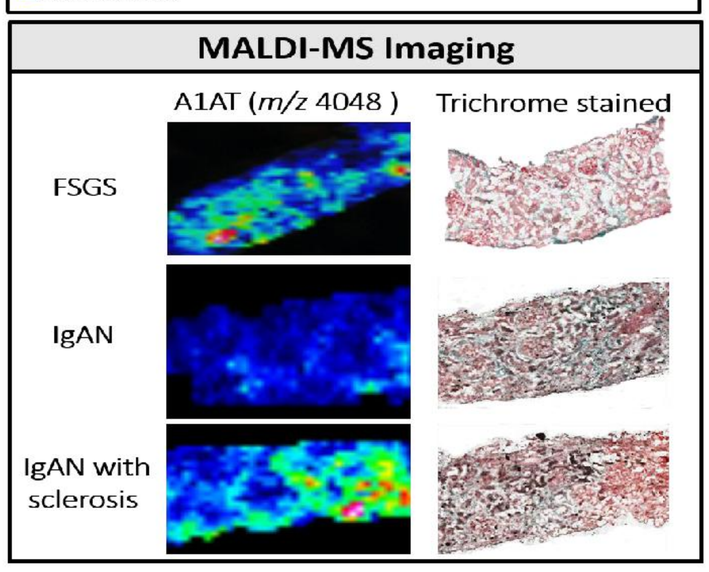
Average proteomic profiles obtained from control renal cortical tissue and from patients histologically diagnosed with primary glomerulonephritis (FSGS, IgAN or MGN)



A) Distribution of spectra obtained from FSGS and IgAN tissue, highlighting the proteomic differences present.. B) With the addition of spectra virtually micro-dissected from sclerotic lesions within IgAN



(Left) Average proteomic profiles obtained from FSGS and IgAN. (Right)Up-regulation of two signals in FSGS cases and sclerotic lesions in IgAN (AUC > 0.8)



(Left) The molecular distribution of a protein, identified as α-1 antitrypsin (m/z 4048), in FSGS, IgAN and IgAN with sclerosis. (Right) Corresponding trichrome stained images from the same section used for MALDI-MSI analysis

Conclusions

The application of MALDI-MSI represents an innovative approach in the analysis of renal biopsies. In this study, we have highlighted the potential of this technology to generate molecular signatures of glomerulonephritis. More specifically, different types of GNs present altered protein profiles, with the specific up and down regulation of proteins associated with individual types of GNs. Here, a protein, identified as α-1 antitrypsin was shown to be correlated with sclerotic lesions of bioptic tissue and up-regulated in cases of FSGS and sclerotic lesions in IgAN.

This work highlights the potential of MALDI-MSI to assist in the diagnosis of renal biopsies, especially in ambiguous and difficult cases. Furthermore, there is the possibility of detect diagnostically significant biomarkers of GNs that could be translated into immunohistochemical tests.

Acknowledgements

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