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Data Article

Datasets from label-free quantitative proteomic analysis of human glomeruli with sclerotic lesions

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ABSTRACT

Human glomeruli with intermediate (i-GS) and advanced (GS) sclerotic lesions as well as the normal control (Nor) were captured from laser microdissection, digested by trypsin and subjected to shotgun LC-MS/MS analysis (LTQ-Orbitrap XL). The label-free quantification was performed using the Normalized Spectral Index (SI_N) to assess the relative molar concentration of each protein identified in a sample. All the experimental data are shown in this article. The data is associated to the research article submitted to Journal of Proteomics [1].

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Specifications Table

Subject area	Chemistry, Biology
More specific subject area	Label-free quantitative proteomics
Type of data	a. *Excel datasheets with peptide and protein identifications in each sample.* b. *Excel datasheet with Normalized Spectral Index (SI_N) of the proteins identified in each sample.* c. *Venn diagrams for peptide/protein identifications in each group.* d. *Graphs for comparison of SI_N values between the sample groups.*
How data was acquired	MS/MS data acquired from LTQ-Orbitrap XL (Thermo Scientific) combined with nanoscale C18 reversed phase liquid chromatography (DiNa-A, KYA technologies). Peptide/protein identification results exported from Mascot.
Data format	.xlsx (peptide/protein identifications and SI_N values) .PDF (Venn diagrams and comparison of SI_N values)
Experimental factors	No sample pretreatment applied
Experimental features	Human glomerular sections were collected by laser microdissection and digested by trypsin. The peptide sample was purified with StageTips C18 before analyzed by LC-MS/MS in triplicates.
Data source location	Niigata City, Japan
Data accessibility	All the experimental data are available in this article.

Value of the data

- In-depth proteomic profiles provide comprehensive protein composition of human sclerotic glomeruli.
- SI_N-based label-free quantitative datasets are useful to explore the key biological events which would be critically involved in the progression of human glomerulosclerosis.
- Detailed information on peptide-spectrum matches and ion intensities for spectral queries enables further bioinformatic analysis.

1. Data, experimental design, materials and methods

We aimed to characterize human glomeruli with intermediate and advanced sclerotic lesions by label-free quantitative proteomic approach in combination with laser microdissection. Macroscopically normal kidney tissues were obtained from patients who underwent nephrectomy due to urological cancers. Sclerotic glomeruli, which were excluded from specific renal diseases and assumed to be aging-related, were divided into two groups, intermediate (i-GS) and advanced (GS) sclerosis, as well as the normal control (Nor). Glomerular sections (10 μm–thick, fixed by methyl–Carnoy) were collected by laser microdissection (50 sections/sample, 3 samples/group from 3 patients). The detailed information on patients and specimens are given in the associated article submitted to Journal of Proteomics [1].

Each glomerular sample was directly digested with 15 μl of activated trypsin solution (20 ng/μl) at 37 °C overnight. After trypsin digestion, 1 μl of 50% trifluoacetic acid (TFA) was added to the peptide mixture to quench the trypsin activity. Peptides were eluted and purified using StageTips C18 (Thermo Scientific) according to the manual instructions [2]. Briefly, the C18 tips were firstly activated with solvent A (80% acetonitrile, 5% formic acid) and re-equilibrated with solvent B (5% formic acid) before sample loading. Then the peptides were eluted twice with 20 μl of solvent A. Finally, the peptide eluate was dehydrated in a Speedvac for dryness and stored at −30 °C until LC-MS/MS analysis. We estimated that around 1 μg of peptides could be extracted from one sample according to BCA assay. As the peptide yield is very limited, we did not perform BCA assay for the samples which were analyzed by LC-MS/MS. All peptides extracted from each sample were directly analyzed in triplicate.

Each peptide sample was solubilized in the sample solution (2% acetonitrile, 0.1% formic acid) and measured in triplicates by LTQ-Orbitrap XL (Thermo Scientific) combined with nanoscale C18 reversed