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A matter of perspective—Cutting-edge technology-driven urine proteome in COVID-19

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ABSTRACT

In a recent issue of Nature Communications, we highlighted in-depth urine proteomic research in which significant immunosuppression was revealed in early SARS-CoV-2- infected patients 1. The application of urine in mapping the landscape of molecular changes closely associated with human diseases has been widely accepted. Herein, we take a systematic review of the published article from the perspective of both methodology and clinical significance.

In a recent issue of *Nature Communications*, we highlighted in-depth urine proteomic research in which significant immunosuppression was revealed in early SARS-CoV-2-infected patients.[1] The application of urine in mapping the landscape of molecular changes closely associated with human diseases has been widely accepted. Herein, we take a systematic review of the published article from the perspective of both methodology and clinical significance.

Sophisticated cutting-edge mass spectrometry-based urine proteomic technology

Urine has been recognized as an ideal source for biomarker identification largely owing to its sensitivity to homeostasis. It may reflect alterations for both systemic and primary renal diseases at an early stage.[2–4] One of the most crucial points in successful urine proteomic evaluation depends on qualitative and quantitative accuracy of the approach. According to different quantitative strategies, proteomic technology can be divided into two types of acquisition: data-dependent acquisition (DDA) and data-independent acquisition (DIA). While DDA only allows limited peptide ions with stronger signals to enter the secondary mass spectrum for fragmentation analysis, DIA divides the entire scan range of the mass spectrum into several windows and performs high-speed and circular analysis of all of the ions in each window. With data availability greatly improved, DIA is widely accepted as being superior to DDA in terms of the number of identified proteins, repeatability, and quantitative accuracy.[5,6] However, traditional DIA may make the spectrogram highly complex, which can bring great challenges to data analysis. Thus, certain disputes regarding its quantitative reliability remain.

In response to this problem, we applied the most advanced 4D-DIA (dia-PASEF) approach to reveal the urine proteomic pattern of the clinical cohorts. It is well-known that four-dimensional (4D) proteomics demonstrates enhanced scanning speed and detection sensitivity with the addition of ion mobility separation to the traditional 3D proteomics (retention time, m/z, and intensity). By innovatively combining the advantages of 4D proteomics and DIA technology, the 4D-DIA approach has attracted increasing attention in recent years. It not only overcomes the original disadvantages of DIA, but it also achieves improved protein identification ability, detection sensitivity, and data integrity.[7] A new-generation technique, 4D-DIA is a comprehensive upgrade of DIA strategy, and it has broad application prospects in future proteomic research.

Notably, a recent third-party report comprehensively evaluated our data quality from multiple perspectives, including but not limited to the width of chromatographic peak, the extraction range of XIC, and the number of data points per peak (DPPP).[8] All of the evidence pointed to the favorable quantitative stability, data integrity, and coverage of our approach. The spectral library containing 6,346 proteins was established with high reliability under the strictest triple FDR quality control (PSM, peptide, and protein) at 1%. While the previous urine proteome was

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usually maintained at a coverage of 1000–2000 proteins, the current study achieves up to 3,500 proteins, which represents the highest level, to the best of our knowledge.

Clinical Insights into COVID-19 from the immunosuppressive urine proteomic pattern

According to the proteomic pattern of COVID-19 patients, the total number of down-regulated proteins, mainly involved in immune response and tight junction formation, was 10 times greater than that of up-regulated proteins. Distinct from other respiratory viruses, increasing evidence has revealed that SARS-CoV-2 is extremely "tricky", which may subvert any traditional cognition of virologists and immunologists.[9,10] It is noteworthy that another study published on the same day in *Nature Communications* reported a similar immunosuppressive pattern of SARS-CoV-2.[11] Specifically, massive innate immune pathways like toll-like receptors and interleukin and chemokine signaling demonstrate an attenuated response to the virus. Other studies found that COVID-induced organ dysfunction is predominantly mediated by immunosuppression and endothelial activation, rather than by significant inflammatory cytokine elevation.[12,13] When cytokines are not highly produced, the efficiency of immunomodulatory therapy can be very limited and should be re-evaluated. Supporting evidence can be found in a recent clinical study, in which it was reported that there was no difference in the primary outcomes between tocilizumab (targeting interleukin-6) and placebo in COVID-19 patients.[14]

Considering all of the factors, including results from our ongoing research, other peer-reviewed publications, and also valuable successful experiences from our cooperative clinicians on the front line, it is advisable to enhance immunity at the early stage of COVID-19 infection. Early intervention can be very helpful in repairing the damaged tissues and improving metabolic functions. Alternative effective attempts may contain appropriate nutritional supplementation and close observation of immune functions. With the ultimate goal of winning the fight against the pandemic, the current study provides important insights into developing more precise therapeutic intervention strategies at different stages of disease progression.

Declaration of competing interest

The authors declare no conflict of interests.

References

1. Tian W, Nan Z, Jin R, et al. Immune suppression in the early stage of COVID-19 disease. *Nat Commun*. 2020;11:5859.
2. Rodriguez-Suarez E, Siwy J, Zurbig P, Mischak H. Urine as a source for clinical proteome analysis: from discovery to clinical application. *Bba-Proteins Proteom*. 2014; 1844:884–898.
3. Gao Y. *Urine Promising Biomarker Source for Early Disease Detection*. Springer Nature Singapore Pte Ltd.; 2019.
4. Shimura T, Dayde D, Wang H, et al. Novel urinary protein biomarker panel for early diagnosis of gastric cancer. *Br J Canc*. 2020;123:1656–1664.
5. Bilbao A, Varesio E, Luban J, et al. Processing strategies and software solutions for data-independent acquisition in mass spectrometry. *Proteomics*. 2015;15:964–980.
6. Muntel J, Xuan Y, Berger S, et al. Advancing urinary protein biomarker discovery by data-independent acquisition on a quadrupole-orbitrap mass spectrometer. *J Proteome Res*. 2015;14:4752–4762.
7. Meier F, Brunner A, Frank M, et al. diaPASEF: parallel accumulation-serial fragmentation combined with data-independent acquisition. *Nat Methods*. 2020;17: 1229–1236.
8. Omicsolution. *DIA*技术转化重磅成果, 黄超兰、高福团队新冠尿液研究实现超高深度单针定量. 2020.
9. Standl F, Jockel KH, Brune B, Schmidt B, Stang A. Comparing SARS-CoV-2 with SARS-CoV and influenza pandemics. *The Lancet. Infect Dis*. 2020.
10. Mann R, Perisetti A, Gajendran M, Gandhi Z, Umapathy C, Goyal H. Clinical characteristics, diagnosis, and treatment of major coronavirus outbreaks. *Front Med*. 2020;7:581521.
11. Mick E, Kamm J, Pisco A, et al. Upper airway gene expression reveals suppressed immune responses to SARS-CoV-2 compared with other respiratory viruses. *Nat Commun*. 2020;11:5854.
12. Su H, Yang M, Wan C, et al. Renal histopathological analysis of 26 postmortem findings of patients with COVID-19 in China. *Kidney Int*. 2020;98:219–227.
13. Leisman DE, Deutschman CS, Legrand M. Facing COVID-19 in the ICU: vascular dysfunction, thrombosis, and dysregulated inflammation. *Intensive Care Med*. 2020; 46:1105–1108.
14. Chen LYC, Hoiland RL, Stukas S, Wellington CL, Sekhon MS. Confronting the controversy: interleukin-6 and the COVID-19 cytokine storm syndrome. *Eur Respir J*. 2020;56.