

Spatial metabolomics: A new tool for unravelling the metabolic disorders and heterogeneity in diabetic kidney disease (Review)

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Received September 22, 2025; Accepted January 13, 2026

DOI: 10.3892/ijmm.2026.5747

Abstract. Diabetic kidney disease (DKD) is a microvascular complication of diabetes, characterized by region-specific metabolic reprogramming that disrupts kidney function and markedly impairs patient prognosis. By enabling *in situ* visualization and analysis of metabolite distribution within kidney tissue, spatial metabolomics offers a unique advantage in detecting spatial heterogeneity in metabolic alterations, which is inaccessible through conventional metabolomics. This approach not only enhances the understanding of DKD pathophysiology but also provides a solid foundation for the development of precision nephrology strategies informed by spatial metabolite data. The present review discusses the fundamental workflows and spatial resolution capabilities of spatial metabolomics, summarizing the key metabolites involved in regional metabolic disruptions in multiple DKD animal models. Moreover, it highlights notable metabolites, including glucose, succinate, phosphatidylserine, lysophosphatidylglycerol, phosphatidylglycerol, sphingomyelin, phosphatidylcholine, phosphatidylethanolamine, taurine, glutamate, L-carnitine, choline, adenosine monophosphate and guanosine monophosphate. The continued advancement of imaging technologies and data analysis methodologies is

expected to further refine the spatial resolution and precision of spatial metabolomics, thereby facilitating its broader application in clinical practice.

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1. Introduction

Diabetic kidney disease (DKD) is defined as kidney damage resulting from diabetes. In total, ~40% of patients with diabetes will eventually develop DKD (1,2). Moreover, with disease progression, patients with DKD experience a decline in renal function, ultimately culminating in kidney failure and end-stage kidney disease (3). Under the pathological condition of DKD, metabolic dysregulation can trigger oxidative stress (4-7), inflammatory cascades (8), tubulointerstitial injury (9,10) and glomerular endothelial dysfunction via fenestration alterations (11). Therefore, there is an urgent requirement to fully understand the pathological changes in different regions of DKD and explore effective diagnostic and therapeutic approaches.

Metabolomics is an omics technology that enables the comprehensive analysis of metabolites in organisms (12). Classic metabolomics methods focus on identifying and quantifying metabolites, providing a global metabolic pathway profile that captures systemic alterations under physiological or pathological conditions (13,14). This enables the identification

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Key words: diabetic kidney disease, spatial metabolomics, metabolic disorders, heterogeneity, mass spectrometry imaging

of metabolic disparities across diverse sample cohorts (15,16). However, data obtained by these methods lack information on the spatial distribution of metabolites within tissues. This limitation has driven the development of spatial metabolomics.

Spatial metabolomics is a technology developed over the past two decades, and its application scope has been continuously expanded in the last 5 years with the advancement of technical methodologies (17,18). This technique integrates metabolomics with spatial biology, offering a novel approach to explore the heterogeneity of metabolites within tissue microenvironments under disease conditions. Recent studies on cancer types such as hepatocellular carcinoma have reported that the spatial distribution patterns of immune cells can more accurately predict diseases than gene expression levels alone (19-22). This highlights the importance of spatial location information in disease research. Currently, spatial metabolomics has demonstrated great potential in several fields, including neuroscience (23,24), microbiology (25), plant science (26), drug development (27) and clinical applications (28). Notably, the application of spatial metabolomics in kidney research is still in its developmental stage, with most existing studies concentrating on the heterogeneity between the renal cortex and medulla (29-31).

The present review discusses the fundamental principles and advantages of spatial metabolomics technology, summarizing recent advancements in its application to metabolic disorders in DKD. The focus is on characteristics associated with carbohydrate metabolism, lipid metabolism, amino acid metabolism and nucleic acid metabolism within cortical and medullary regions. Finally, the review outlines potential future research directions, aiming to provide valuable insights and contributions to researchers in the fields of DKD and related pathophysiology.

2. Basic principles of spatial metabolomics technology

Spatial metabolomics represents a cutting-edge technology that integrates mass spectrometry imaging (MSI) with the principles of metabolomics. The fundamental working principle involves employing specific ionization methods to ionize metabolites present in tissue sections of samples (32). Currently, three primary ionization techniques are widely utilized in spatial metabolomics: Matrix-assisted laser desorption/ionization (MALDI)-MSI, desorption electrospray ionization (DESI)-MSI and secondary ion (SIMS)-MSI (33-38). These ionized metabolites are then sent to a mass spectrometer for mass analysis, where they are analyzed to obtain information on their mass-to-charge ratio. At the same time, the spatial distribution images of metabolites in the tissue sections are reconstructed by combining the mass spectrometry data with the spatial coordinate information of the tissue samples (32,39,40). To enable co-localization analysis, previous studies spatially aligned consecutive kidney sections stained with hematoxylin and eosin with MSI data (41,42) (Fig. 1).

3. Comparison of major spatial metabolomics technologies

The core advantage of spatial metabolomics is its capability to resolve spatial information, enabling *in situ* localization of

metabolites within biological tissues. This is particularly valuable in kidney research, where the average cellular diameter is $\sim 10\ \mu\text{m}$ (43). MALDI-MSI boasts high sensitivity and the capability to detect low-abundance metabolites, thus occupying a dominant position in the field of spatial metabolomics. Using MALDI-MSI with a spatial resolution of $20\ \mu\text{m}$, the overall metabolic heterogeneity distribution from the cortex to the medulla in DKD kidney sections can be clearly visualized, revealing metabolic gradient changes in different functional regions under disease conditions (31). By contrast, employing a higher resolution of $5\ \mu\text{m}$ with MALDI-MSI and isotope labeling enables *in situ*, cell-type-specific dynamic metabolic measurements, uncovering cellular metabolism within the tissue structure (44). A key advantage of DESI-MSI is its compatibility with fresh tissues, facilitating rapid detection of clinical samples. Operating at room temperature and atmospheric pressure, DESI-MSI requires no complex sample pretreatment (45,46). With a typical spatial resolution of $50\text{-}200\ \mu\text{m}$ (47) and a moderate detectable molecular weight range, it is more suitable for imaging large tissue regions. This facilitates the investigation of the pervasive metabolic landscape in advanced DKD, a condition often characterized by extensive interstitial fibrosis. SIMS-MSI operates under high-vacuum conditions and has stringent temperature requirements. While SIMS-MSI achieves an ultra-high spatial resolution of $<1\ \mu\text{m}$, it has the narrowest detectable molecular weight range among all the discussed techniques. Nevertheless, it provides the highest spatial resolution among the technologies addressed here, making it ideally suited for single-cell metabolomics research (35,37). Therefore, the selection of an appropriate spatial metabolomics technique for DKD research necessitates a trade-off between its spatial resolution and metabolite coverage. Future studies may integrate multiple technologies to obtain more comprehensive metabolic information, thereby enhancing our in-depth understanding of disease mechanisms.

4. Functional specialization and metabolic disturbances in DKD

The kidney is a vital excretory and regulatory organ, whose functions rely on the sophisticated structure and division of labor between the renal cortex and medulla (48,49). The renal cortex, composed primarily of abundant renal corpuscles and proximal tubules, is principally responsible for blood filtration, reabsorption and secretion of substances (50,51). By contrast, the renal medulla, through the loops of Henle and collecting ducts, maintains internal environmental homeostasis by regulating water-electrolyte balance and urine concentration (52,53). Owing to these functional distinctions between the two regions, there are notable differences in their demand for and utilization of metabolites (54).

In the onset and progression of DKD, the dysregulation of regional metabolic functions is particularly critical, and metabolomics research provides an essential technological approach for systematically elucidating its molecular mechanisms (55,56). Classical metabolomics, through the synergistic application of non-targeted metabolic profiling and targeted metabolite quantification strategies, has systematically constructed a comprehensive molecular map of the metabolic

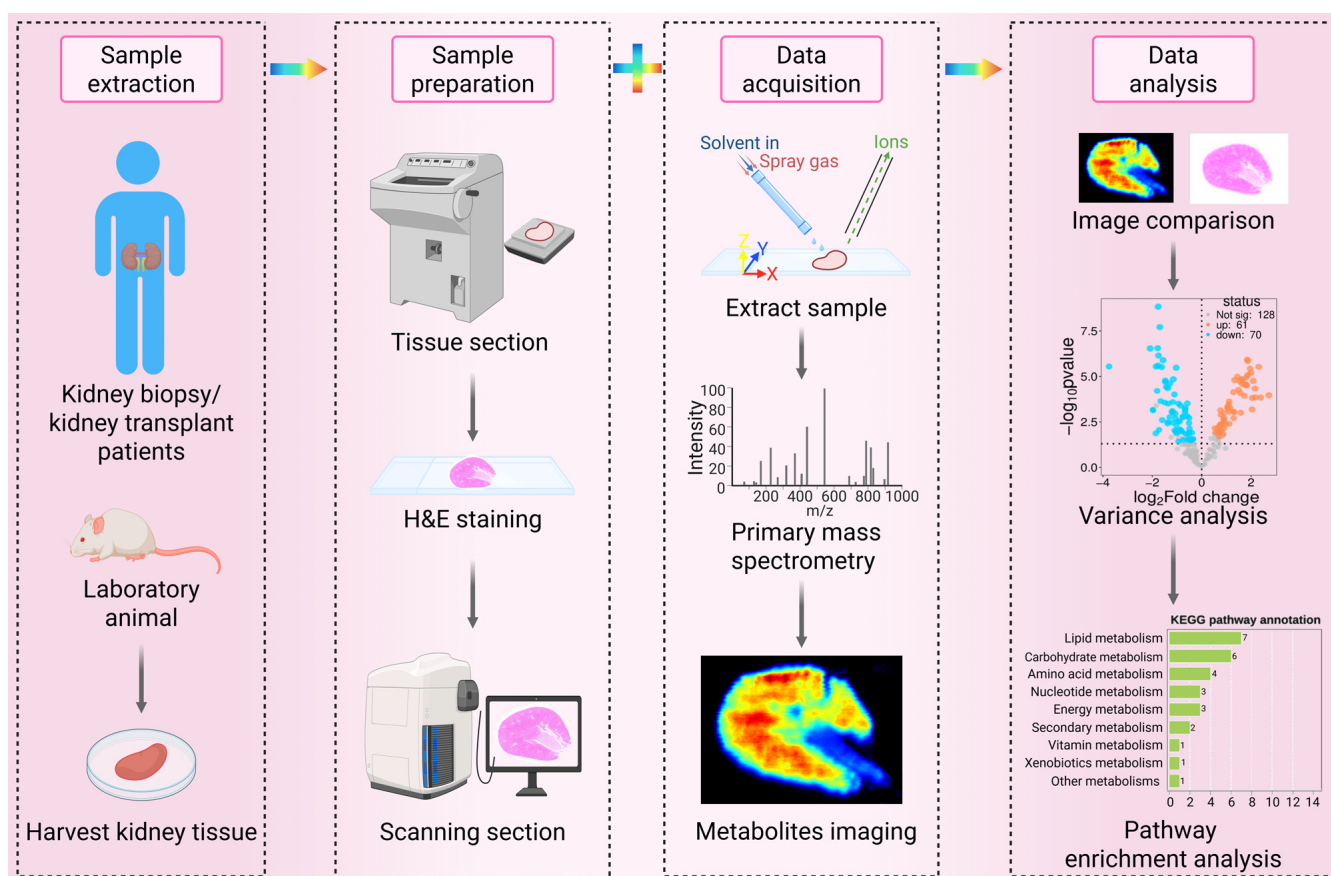


Figure 1. Schematic workflow of spatial metabolomics analysis for kidney samples. After kidney tissue is obtained, the workflow encompasses sample preparation, high-resolution mass spectrometry imaging, multimodal data collection, and integration of variance analysis and pathway enrichment analysis. This results in the construction of a spatial metabolic map that captures metabolic heterogeneity between the renal cortex and medulla, offering a visual reference for mechanistic investigations into metabolic diseases such as diabetic kidney disease. H&E, hematoxylin and eosin; KEGG, Kyoto Encyclopedia of Genes and Genomes.

network of the kidney (Fig. 2), revealing ATP depletion driven by mitochondrial dysfunction, imbalance of tricarboxylic acid (TCA) cycle intermediates and inhibition of fatty acid oxidation (FAO), which ultimately leads to apoptosis, autophagy, oxidative stress and lipid droplet accumulation (57,58). Therefore, the integration of spatial metabolomics has enabled unprecedented precision in regional metabolite localization (59,60).

5. Spatial metabolomics reveals region-specific disturbances in glucose metabolism and the TCA cycle in DKD

A hyperglycemic environment can induce an imbalance in energy metabolism homeostasis, characterized by mitochondrial oxidative phosphorylation dysfunction and metabolic reprogramming (61,62). Notably, the renal medulla mainly relies on glucose for energy (63,64). Previous research reported that the levels of hexoses in the cortical tissue increased when male C57BL/6J-Ins2Akita mice reached 17 weeks of age (65). When DKD rats were 24 and 28 weeks old, respectively, the kidney glucose levels exhibited a marked global increase, with particularly pronounced elevations observed in the cortex region (31,66). By contrast, rats subjected to prolonged feeding exhibited notably reduced glucose levels in the renal medulla region (66). However, it should be noted that these findings reflect the complexity of metabolic heterogeneity across

different regions and time points. The discrepancies in glucose levels between the renal cortex and medulla have led to varied interpretations of the pathophysiological mechanisms of DKD, highlighting the necessity of considering tissue specificity and its potential impacts on overall disease progression when investigating metabolic alterations.

The distribution of TCA cycle intermediates also exhibits spatial heterogeneity. Clinical studies have reported that, compared with healthy controls, succinic acid and malic acid are markedly reduced in kidney biopsies of patients with type 1 diabetes across several kidney regions, suggesting subclinical mitochondrial dysfunction (67). These spatial distribution abnormalities are highly congruent with the global findings of classical metabolomics. In individuals with DKD, enzymes involved in glycolytic, sorbitol, methylglyoxal and mitochondrial pathways have been reported to exhibit reduced expression and activity. Moreover, pyruvate kinase M2 demonstrates marked decreases in both its transcriptional expression and enzymatic activity (68). From the perspective of metabolic pathways, the accumulation of TCA cycle intermediates induces a burst of mitochondrial reactive oxygen species, driving the progression of DKD (69,70). Table I consolidates information on spatially resolved metabolic alterations in carbohydrate and TCA cycle intermediates.

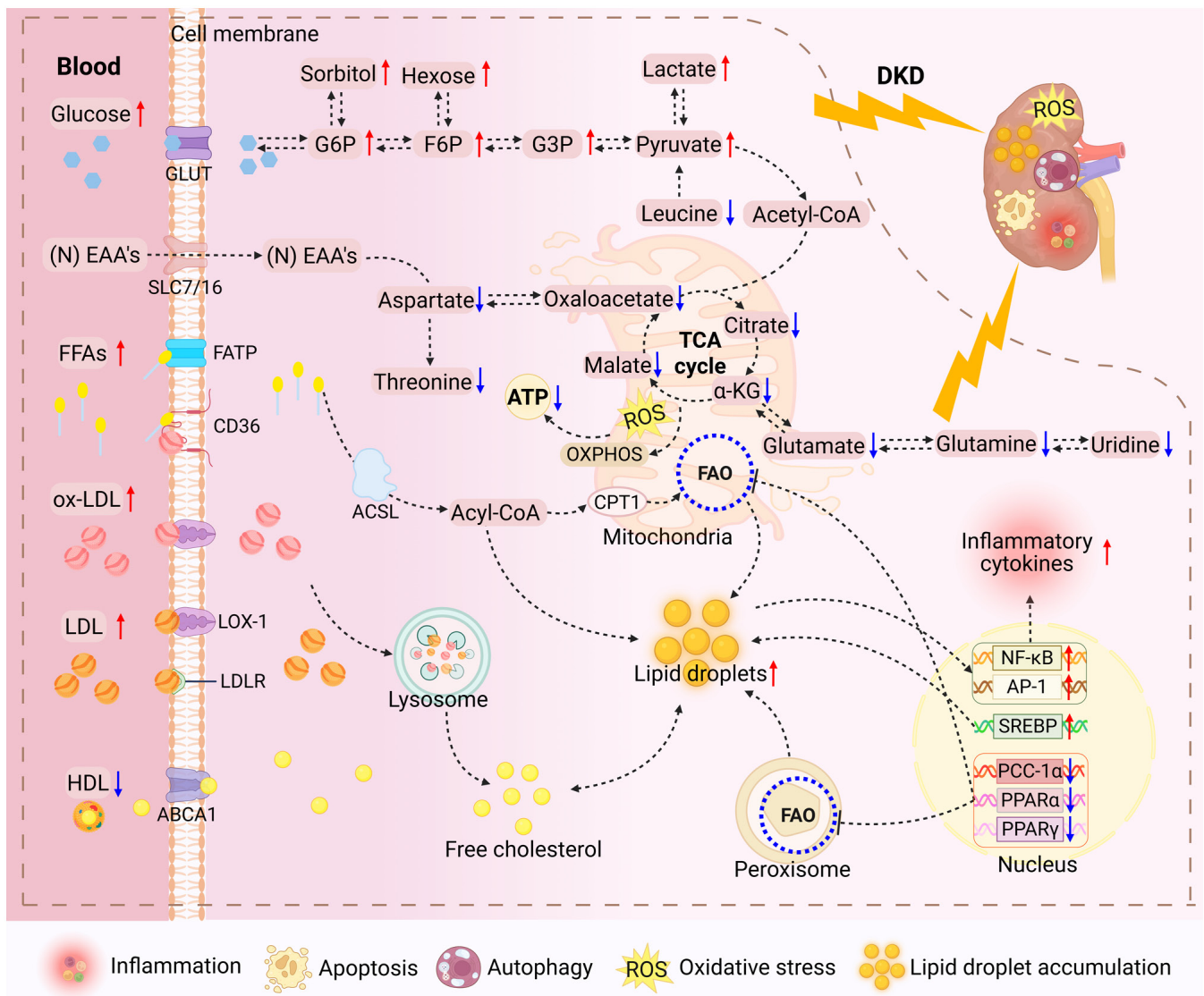


Figure 2. Global metabolic disorder network of DKD revealed by classical metabolomics. The key metabolic alterations and their pathogenic relevance to DKD progression are shown. Intracellular glucose influx is increased, leading to cytoplasmic glucose accumulation that activates the polyol pathway and disrupts glycolytic intermediate balance, further exacerbating lactate buildup. These changes drive a metabolic shift toward anaerobic glycolysis. FFAs are taken up through CD36 and FATP receptors, resulting in the accumulation of oxidized low-density lipoprotein. The functions of cholesterol reverse transport proteins ABCA1/LDLR are impaired, leading to lipid droplet accumulation and oxidative stress. PGC-1 α and PPAR regulate FAO, while SREBPs can affect cholesterol synthesis. Mitochondrial oxidative phosphorylation dysfunction and peroxisome dysfunction further worsen lipotoxicity. The reduction of various amino acids is closely related to the decrease of intermediates in the TCA cycle. Red arrows indicate an increase; blue arrows indicate a decrease. DKD, diabetic kidney disease; GLUT, glucose transporter; G6P, glucose-6-phosphate; F6P, fructose-6-phosphate; G3P, glyceraldehyde-3-phosphate; (N)EAA's, (non)essential amino acids; FFAs, free fatty acids; FATP, fatty acid transport protein; CD36, cluster of differentiation 36; ox-LDL, oxidized low-density lipoprotein; LDL, low-density lipoprotein; LOX-1, lectin-like oxidized low-density lipoprotein receptor-1; LDLR, low-density lipoprotein receptor; HDL, high-density lipoprotein; ABCA1, ATP-binding cassette transporter A1; TCA cycle, tricarboxylic acid cycle; ATP, adenosine triphosphate; ROS, reactive oxygen species; OXPHOS, oxidative phosphorylation; FAO, fatty acid oxidation; ACSL, acyl-CoA synthetase long-chain family; CPT1, carnitine palmitoyltransferase 1; α -KG, α -ketoglutarate; NF- κ B, nuclear factor κ -light-chain-enhancer of activated B cells; AP-1, activator protein-1; SREBP, sterol regulatory element-binding protein; PGC-1 α , peroxisome proliferator-activated receptor γ coactivator 1- α ; PPAR α , peroxisome proliferator-activated receptor α ; PPAR γ , peroxisome proliferator-activated receptor γ .

6. Lipid metabolism in DKD

Classical metabolomics studies have systematically characterized the systemic lipid metabolic dysregulation in DKD, elucidating imbalances in cholesterol esterification, phospholipid remodeling and triglyceride (TG) accumulation (71-73). Moreover, spatial metabolomics has revealed notable regional variations in phospholipids and TGs in DKD kidneys (66,74). Phospholipids are key components of cell membranes

and are associated with the pathogenesis of DKD (75). Lysophosphatidic acid (LPA), lysophosphatidylcholine (LPC), lysophosphatidylethanolamine (LPE) and lysophosphatidylethanolamine (GPE) are all lipid metabolites derived from phospholipids. In DKD model mice, the levels of LPA, LPC, LPE and GPE in the cortex were reported to be markedly elevated (65,76,77). Moreover, changes in the phosphatidylcholine (PC)/phosphatidylethanolamine (PE) ratio have been reported to have an impact on cellular processes associated

Table I. Spatial metabolomics reveals dysregulation of glycolysis and tricarboxylic acid cycle intermediates in DKD.

Experimental model	Substructure		Overall structure	Applied method	Spatial resolution, μm	(Refs.)
	Cortex	Medulla				
F1 C57BL/6J-Ins2Akita male mice	Hexose $\uparrow$, threonic acid $\uparrow$			DESI-MSI	200	(65)
HFD and STZ-treated DKD rats	Glucose $\uparrow$, sorbitol $\uparrow$	Glucose $\downarrow$, sorbitol $\uparrow$, succinate $\uparrow$, glucose 6-phosphate $\uparrow$, glyceraldehyde 3-phosphate $\uparrow$	Citric acid $\downarrow$, malate $\downarrow$	AFADESI-MSI, MALDI-MSI	20	(31)
HFD/STZ-induced diabetic rats	Glucose $\uparrow$	SGLTs $\downarrow$	Citrate $\downarrow$	AFADESI-MSI	20	(66)
HFD/STZ-induced db/db mice	Glucose $\uparrow$, succinate $\uparrow$	SGLTs $\uparrow$, succinate $\uparrow$	Malate $\downarrow$	AFADESI-MSI	20	(66)
8 participants with T1D and preserved kidney function, and 5 HC			Succinic acid $\downarrow$, malic acid $\downarrow$	MALDI-MSI	20	(67)

HC, healthy control; DKD, diabetic kidney disease; HFD, high-fat diet; STZ, streptozotocin; T1D, type 1 diabetes; MSI, mass spectrometry imaging; MALDI-MSI, matrix-assisted laser desorption/ionization-MSI; DESI, desorption electrospray ionization-MSI; AFADESI-MSI, airflow-assisted desorption electrospray ionization-MSI; SGLT, sodium-glucose transporter.

with health and disease (77). In cardiovascular disease patients at risk of insulin resistance, changes in the PC/PE ratio affect cellular processes related to cardiovascular health and disease progression by regulating inflammatory responses and the physiological state of cardiomyocytes (78). Notably, in db/db mice, the levels of PC/PE ratio in the cortex and medulla showed opposite changes, indicating that metabolic responses in different kidney regions during disease progression are both complex and region-specific (66). TGs are subject to rapid turnover and re-arrangement of fatty acids (FAs) (79). A recent spatial multi-omics study of long-term DKD reported a marked accumulation of TG in the medullary region (74). Furthermore, in DKD rats at 24 weeks of age, renal cortical oleic acid levels were elevated, whilst polyunsaturated FAs, including linoleic acid, were markedly reduced (31).

Spatial metabolomics has advanced the understanding of lipid changes in the kidneys of DKD to the level of proximal tubular cells. Under physiological conditions, the kidney tubules predominantly utilize FAs (64,80,81), and this utilization requires carnitine as a carrier, facilitating the transport of long-chain fatty acids into the mitochondria via the carnitine shuttle system (82). Spatial metabolomics and multi-omics analyses of human kidney samples reported that the detection of acylcarnitines in cortical samples was insufficiently sensitive (83). This observation is consistent with the known downregulation of FAO in chronic kidney disease, indicating impaired FAO in proximal tubular cells of patients with kidney disease (84). Detailed lipid changes are presented in Table II.

It should be noted that the inherent differences in sample size, animal model selection and experimental methodologies among the aforementioned studies may impose significant constraints on the reproducibility and generalizability of the results. Although spatial metabolomics has successfully

revealed the regional heterogeneity of lipid distribution in the kidney, existing research is mostly confined to single-species models (e.g., db/db mice and DKD rats). Moreover, the sample size used for spatial metabolomics analysis is only up to 6 animals per group, coupled with the lack of dynamic tracking data across different disease stages. This may not only induce biases in interpreting the causal relationship between lipid metabolic disorders and the pathological progression of DKD, but may also hinder the accurate reflection of the clinical metabolic characteristics of human DKD. Therefore, future studies should prioritize optimizing experimental designs by expanding sample sizes, diversifying animal models and conducting dynamic monitoring across multiple disease stages, so as to more comprehensively elucidate the metabolic response mechanisms of distinct renal regions during DKD progression.

7. Amino acid metabolism in DKD

The dysregulation of amino acid metabolism in DKD is not merely a consequence of renal dysfunction, but an active driver of pathogenesis (85,86). Spatial metabolomics techniques have revealed changes in amino acids in specific regions of the kidney. In db/db mice, glutamate, spermine, taurine and spermidine were notably reduced in both the cortex and medulla, whilst histamine, putrescine and indoxyl sulfate markedly accumulated in the cortex (66,87,88). Notably, spermidine has been reported to exhibit multiple pharmacological effects, including anti-aging, anti-oxidation, anti-inflammation and cardiovascular protection (89-92). Sulfate is a protein-binding gut-derived uremic toxin that has been reported to be closely associated with podocyte loss in DKD (93). Our previous research demonstrated that

Table II. Identification of lipid metabolism disorders in DKD utilizing spatial metabolomics.

Experimental model	Substructure			Overall structure	Applied method	Spatial resolution, μm	(Refs.)
	Cortex	Medulla					
C57BLKS db/db mouse model of type 2 DN F1 C57BL/6J-Ins2Akita male mice	LPA $\uparrow$, LPC $\uparrow$, NeuGe-GM3 $\uparrow$, Amadori-PE (16:0/20:4) $\uparrow$			MALDI-IMS	10	(76)	
	GPE $\uparrow$, PI $\downarrow$, PS $\downarrow$, PG $\downarrow$, PE $\downarrow$, Cer $\downarrow$, unsaturated FA $\uparrow$			DESI-MSI	200	(65)	
	LPE $\uparrow$	PS (O-16:0/16:0) $\uparrow$	PE (18:0/20:4) $\uparrow$	DESI-MSI	50	(77)	
	FA (18:1) $\downarrow$	L-palmitoyl-carnitine $\downarrow$, propionyl-carnitine $\downarrow$	L-Carnitine $\downarrow$, FA $\uparrow$	AFADESI-MSI	100	(87)	
db/db mice	Docosahexaenoic acid $\downarrow$	PI [18:0/18:2 (9Z, 12Z)] $\downarrow$		MALDI MSI		(88)	
HFD and STZ-treated DKD rats	Oleic acid $\uparrow$, linolenic acid $\downarrow$, linoleic acid $\downarrow$, arachidonic acid $\downarrow$, eicosapentaenoic acid $\downarrow$, docosahexaenoic acid $\downarrow$, acetyl-carnitine $\downarrow$	DAG (18:1/18:1) $\uparrow$, DAG (18:1/18:2) $\uparrow$, PC $\uparrow$, PE (36:4) $\uparrow$, LysoPC (16:0) $\uparrow$, LysoPG (18:1) $\uparrow$, LysoPG (20:4) $\uparrow$	LysoPG (22:6) $\downarrow$, PE (38:6) $\downarrow$, PE (38:4) $\downarrow$, SM (d18:1/16:0) $\uparrow$, phosphorylethanolamine $\downarrow$, long-chain acyl-carnitines $\uparrow$	AFADESI-MSI, MALDI-MSI	20	(31)	
	PC/PE $\uparrow$, LysoPG $\uparrow$, PI $\downarrow$, PS metabolites $\downarrow$, PG metabolites $\downarrow$, SM (d18:1/16:0) $\uparrow$	PC/PE $\uparrow$, PS $\downarrow$, PG $\downarrow$, PI $\downarrow$, SM (d18:1/16:0) $\uparrow$	PC/PE $\uparrow$, PA $\downarrow$, choline $\downarrow$, L-carnitine $\downarrow$, stearoyl-carnitine $\uparrow$	AFADESI-MSI	20	(66)	
HFD/STZ-induced db/db mice	PC/PE $\downarrow$, PS $\uparrow$, PG $\downarrow$, LysoPG $\uparrow$, PI $\uparrow$, SM (d18:1/16:0) $\uparrow$	PC/PE $\uparrow$, PS $\uparrow$, PG $\downarrow$, PI $\uparrow$, SM (d18:1/16:0) $\uparrow$	PC/PE $\uparrow$, PA $\uparrow$, choline $\downarrow$, betaine $\uparrow$, palmitoyl-carnitine $\uparrow$, linoleyl-carnitine $\uparrow$, octadecenoyl-carnitine $\uparrow$	AFADESI-MSI	20	(66)	
Long-standing DKD (diabetes duration >10 years)		Glycerophospholipids $\uparrow$ TG $\uparrow$, PE $\uparrow$, sphingolipids $\uparrow$	Tridecatrienoic acid $\uparrow$, octadecanoic acid $\uparrow$, hydroxyoctadecanoic acid $\uparrow$		50	(74)	
DKD, diabetic kidney disease; HFD, high-fat diet; STZ, streptozotocin; MSI, mass spectrometry imaging; MALDI-MSI, matrix-assisted laser desorption/ionization-MSI; DESI-MSI, desorption electrospray ionization-MSI; AFADESI-MSI, airflow-assisted desorption electrospray ionization-MSI; LPA, lysophosphatidic acid; LPC, lysophosphatidylcholine; NeuGe-GM3, N-glycolylneuraminic acid ganglioside GM3; PE, phosphatidylethanolamine; GPE, glycerophosphatidylethanolamine; PI, phosphatidylinositol; PS, phosphatidylserine; PG, phosphatidylglycerol; Cer, ceramide; FA, fatty acid; LPE, lysophosphatidylethanolamine; DAG, diacylglycerol; LysoPG, lysophosphatidylglycerol; LysoPC, lysophosphatidylcholine; PC, phosphatidylcholine; PA, phosphatidic acid; SM, sphingomyelin; TG, triglyceride.							

DKD, diabetic kidney disease; HFD, high-fat diet; STZ, streptozotocin; MSI, mass spectrometry imaging; MALDI-MSI, matrix-assisted laser desorption/ionization-MSI; DESI-MSI, desorption electrospray ionization-MSI; AFADESI-MSI, airflow-assisted desorption electrospray ionization-MSI; LPA, lysophosphatidic acid; LPC, lysophosphatidylcholine; NeuGc-GM3, N-glycolylneuraminic acid ganglioside GM3; PE, phosphatidylethanolamine; GPE, glycerophosphatidylethanolamine; PI, phosphatidylinositol; PS, phosphatidylserine; PG, phosphatidylglycerol; Cer, ceramide; FA, fatty acid; LPE, lysophosphatidylethanolamine; DAG, diacylglycerol; LysoPG, lysophosphatidylglycerol; LysoPC, lysophosphatidylcholine; PC, phosphatidylcholine; PA, phosphatidylcholine; TG, triglyceride.

Table III. Cortex-medulla heterogeneity in amino acid and related metabolism of DKD revealed by spatial metabolomics.

Experimental model	Substructure		Applied method	Spatial resolution, μm	(Refs.)
	Cortex	Medulla			
HFD and STZ-induced DKD rats	Glutamine↓, aspartate↓, threonine↓, leucine/isoleucine↓	Glutamine↓, aspartate↓, threonine↓, leucine/isoleucine↓	AFADESI-MSI MALDI-MSI	20	(31)
db/db mice	Histamine↑, putrescine↑, taurine↓, spermidine↓, spermine↓, glutathione disulfide↑, L-cysteine↑, 5-L-glutamy-aurine↑	Glutathione↓, taurine↓, spermidine↓, spermine↓, glutathione disulfide↑, L-cysteine↑, 5-L-glutamy-aurine↑	AFADESI-MSI	100	(87)
Obese ZDF male rats	Tyrosinamide↑, L-threonine↓	Tyrosinamide↑, L-threonine↓	MALDI-MSI	20	(95)
HFD/STZ-induced diabetic rats		Glutamate↓, aspartate↓	AFADESI-MSI	20	(66)
HFD/STZ-induced db/db mice	Glutamate↓	Glutamate↓	AFADESI-MSI	20	(66)
db/db mice	Indoxyl sulfate↑		MALDI-MSI	50	(88)

DKD, diabetic kidney disease; HFD, high-fat diet; STZ, streptozotocin; MSI, mass spectrometry imaging; AFADESI-MSI, airflow-assisted desorption electrospray ionization-MSI; MALDI-MSI, matrix-assisted laser desorption/ionization-MSI; ZDF, Zucker Diabetic Fatty.

Tangshen formula can effectively decrease the concentration of sulfate during the treatment of DKD (94). Furthermore, compared with that in db/db mice, glutamate was reported to be similarly reduced in the cortex and medulla of high-fat diet and STZ-induced DKD rats (31,66). Additionally, glutamine, aspartate, threonine and leucine/isoleucine were reported to be markedly reduced (31,66,95). The heterogeneity of amino acid and related metabolism in the cortex and medulla of DKD revealed by spatial metabolomics is summarized in Table III.

8. Nucleic acid metabolism in DKD

Classic metabolomic studies have demonstrated that after inducing diabetes in rats via injection of alloxan, the levels of purine metabolites in their bodies are significantly elevated, including uric acid (the end product of purine metabolism) and metabolic intermediates such as xanthine, hypoxanthine, adenosine monophosphate (AMP) and inosine (96-98). Notably, a large-scale clinical metabolomics study, involving 4,503 healthy controls and 1,875 patients with DKD, reported that the levels of uracil in patients with DKD were markedly reduced (99). These phenomena suggest unique nucleic acid metabolic disturbances. However, to clarify their local *in situ* distribution and pathological significance within the kidney, spatial metabolomics is required.

Spatially resolved analyses have elucidated compartment-specific nucleotide dysregulation, offering mechanistic insights into DKD pathology (31,100). In db/db mice, the level of AMP, a key intermediate in purine nucleotide metabolism, was reported to be notably elevated across all renal regions (66). Moreover, in DKD rats, the levels of AMP and

guanosine monophosphate (GMP) were reported to be markedly increased in the cortex, whilst they showed a decreasing trend in the medulla (31). Mammalian target of rapamycin (mTOR) integrates growth factor and insulin signaling pathways to orchestrate critical cellular processes, including cell proliferation, motility, survival, protein synthesis, autophagy and transcriptional regulation (101). In DKD, abnormal activation of the mTOR pathway can mediate metabolic reprogramming, with adenine reported to drive kidney pathological progression through this pathway. Spatial metabolomics studies reveal that in normal kidneys, adenine is primarily localized in the glomeruli and vascular regions (102,103). However, in the pathology of DKD, adenine diffusely increases throughout the entire tissue section, particularly in the regions of sclerotic vessels, mildly sclerotic glomeruli, atrophic renal tubules and areas of interstitial inflammation (102). The importance of adenine in rescue and targeted therapies has been elucidated using multimodal omics approaches (104). As the most abundant modified nucleoside in several RNA species, pseudouridine has been recognized as a new biomarker demonstrating better performance than creatinine in chronic kidney disease stratification (105). Previous studies have reported that diabetic mice exhibit relatively high levels of pseudouridine in the cortex tissue (65). Table IV summarizes spatial metabolomics research revealing dysregulation of nucleotide and purine metabolism in kidneys of organisms with DKD.

9. Discussion and future perspectives

In-depth analyses of metabolic remodeling and pathological mechanisms in the renal microenvironment is

Table IV. Spatial metabolomics reveals nucleotide and purine metabolism dysregulation in DKD.

Experimental model	Substructure		Applied method	Spatial resolution, μm	(Refs.)
	Cortex	Medulla			
F1 C57BL/6J-Ins2Akita male mice	Pseudouridine $\uparrow$		DESI-MSI	200	(65)
HFD and STZ-treated DKD rats	AMP $\uparrow$, GMP $\uparrow$, ADP $\uparrow$, ATP $\uparrow$, inosine $\downarrow$, hypoxanthine $\downarrow$, xanthine $\downarrow$, uric acid $\downarrow$, uridine $\downarrow$	AMP $\downarrow$, GMP $\downarrow$, ADP $\uparrow$, ATP $\uparrow$	AFADESI-MSI, MALDI-MSI	20	(31)
Diabetic patients (T1D, T2D)	Endogenous adenine $\uparrow$	Endogenous adenine $\uparrow$	MALDI-MSI		(99)
HFD/STZ-induced diabetic rats		AMP $\downarrow$	AFADESI-MSI	20	(66)
HFD/STZ-induced db/db mice	AMP $\uparrow$, GMP $\uparrow$		AFADESI-MSI	20	(66)

DKD, diabetic kidney disease; HFD, high-fat diet; STZ, streptozotocin; T1D, type 1 diabetes; T2D, type 2 diabetes; MSI, mass spectrometry imaging; DESI-MSI, desorption electrospray ionization-MSI; AFADESI-MSI, airflow-assisted desorption electrospray ionization-MSI; MALDI-MSI, matrix-assisted laser desorption/ionization-MSI; AMP, adenosine monophosphate; GMP, guanosine monophosphate; ADP, adenosine diphosphate; ATP, adenosine triphosphate.

the theoretical cornerstone for developing new targeted intervention strategies for DKD. The present review systematically summarizes the cutting-edge advancements in spatial metabolomics in this field. Its core contribution lies in overcoming the limitations of traditional global tissue analysis, revealing the fundamental spatial heterogeneity of metabolic regulation along the cortical-medullary axis and within specific pathological structures. Synthesizing existing research findings, the metabolite profiles in the renal cortex and medulla exhibit marked region-specific dysregulation across multiple DKD animal models. In the cortex region, the levels of glucose, AMP, GMP, PS(36:2), PS(36:1), LysoPG(18:1) and SM(d18:1/16:0) are significantly upregulated; concurrently, the levels of taurine, glutamate, L-carnitine, PG(32:0), PC/PE and choline are notably downregulated. In the medulla region, the levels of succinate, PS(36:1), SM(d18:1/16:0) and PC/PE show significant elevation; meanwhile, the levels of taurine, glutamate, L-carnitine and choline also display a decreasing trend. Fig. 3 offers an intuitive visual overview and acts as a reference for identifying regional or global metabolic targets in subsequent targeted interventions. Metabolites exhibit similar or opposite distribution and regional accumulation in different functional areas of the kidney. These are key spatial details that traditional tissue homogenate-based metabolomics cannot capture. Conventional methods provide overall averages, masking contrasting trends across anatomical regions. This obscures the true picture of metabolic disorders.

The core value of spatial metabolomics lies in addressing the ‘where’ and ‘how’ questions that conventional techniques cannot resolve, thereby revealing the molecular mechanisms and causal relationships driving pathological processes. It

is known that metabolic disorders disrupt cellular energy homeostasis; for example, in the renal hyperglycemic environment, the pentose phosphate, polyol and hexosamine pathways are activated, leading to the local accumulation of toxic metabolites, which in turn activate key signaling pathways and ultimately trigger core pathological processes such as inflammation, oxidative stress and fibrosis (106,107). However, classical metabolomics cannot clearly illustrate in which region of the kidney these metabolic disorders and signaling pathway activations occur, nor how the spatial distribution of metabolites drives pathological processes. This is where spatial metabolomics demonstrates its unique value, and the research on endogenous adenine metabolism is a prime example. The accumulation of adenine in specific regions inhibits 5'-AMP-activated protein kinase whilst activating the mTORC1-S6K signaling axis, thereby translating metabolic dysregulation into typical pathological phenotypes such as cellular hypertrophy and tissue fibrosis (108-111). By integrating spatial metabolomics and spatial proteomics, research has reported that metformin acts in the cortical-medullary outer zone, upregulating nephrosis 2 and inhibiting IL-17 signaling (112). This specifically reverses purine metabolic imbalance, thereby blocking glomerulosclerosis and interstitial fibrosis (112). This intuitively shows how the three-dimensional spatial coupling of metabolism, transcription and proteins precisely remodels the pathological microenvironment. Nevertheless, notably, knowledge gaps remain regarding the regional dynamics of metabolites and the precise mechanisms by which they promote renal pathology at the molecular and cellular levels. The key direction for future research should shift from current descriptive associations to systematic functional validation and mechanistic exploration of these spatially anchored metabolites.

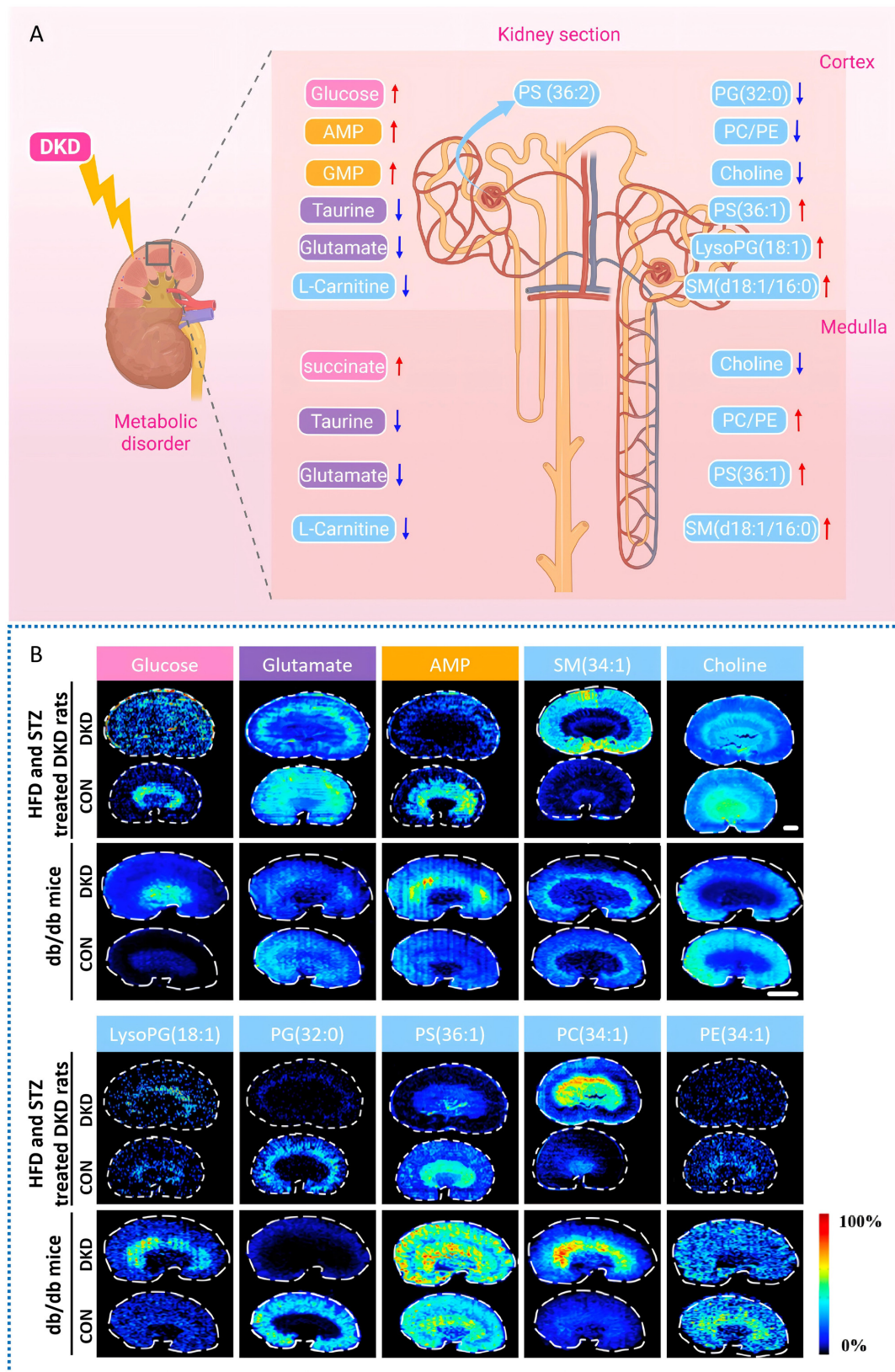


Figure 3. Spatial metabolic heterogeneity is observed in the cortex and medulla of DKD animal models. (A) Schematic diagram of regional distribution of metabolites with consistent change trends across two or more DKD animal models. Through regional demarcation and arrow notation (↑ denotes elevation, ↓ denotes reduction), the region-specific dysregulation of metabolites implicated in glucose metabolism (e.g., glucose), lipid metabolism (e.g., PS, SM), amino acid metabolism (e.g., taurine, glutamic acid), and nucleotide metabolism (e.g., AMP) are explicitly illustrated. (B) Spatial distribution images of representative metabolites obtained via spatial metabolomics. Adapted from Zhang *et al* (66), <https://doi.org/10.3390/metabol13030324>, under the terms of the CC BY 4.0 license: Glucose and glutamate in this figure are adapted from Fig. 8 of this study, AMP from Fig. 9, choline from Fig. 10, and SM(34:1), LysoPG(18:1), PG(32:0), PS(36:1), PC(34:1) and PE(34:1) from Figs. 6 and 7. The images compare the control CON and DKD groups across two models (HFD and STZ treated rats, db/db mice), with the color scale (from 0 to 100%) indicating metabolite abundance. Scale bar, 3 mm. CON, control; DKD, diabetic kidney disease; HFD, high-fat diet; STZ, streptozotocin; PG, phosphatidylglycerol; LysoPG, lysophosphatidylglycerol; PS, phosphatidylserine; PC/PE, the ratio of phosphatidylcholine to phosphatidylethanolamine; SM(34:1), SM(d18:1/16:0); SM, sphingomyelin; AMP, adenosine monophosphate; GMP, guanosine monophosphate.

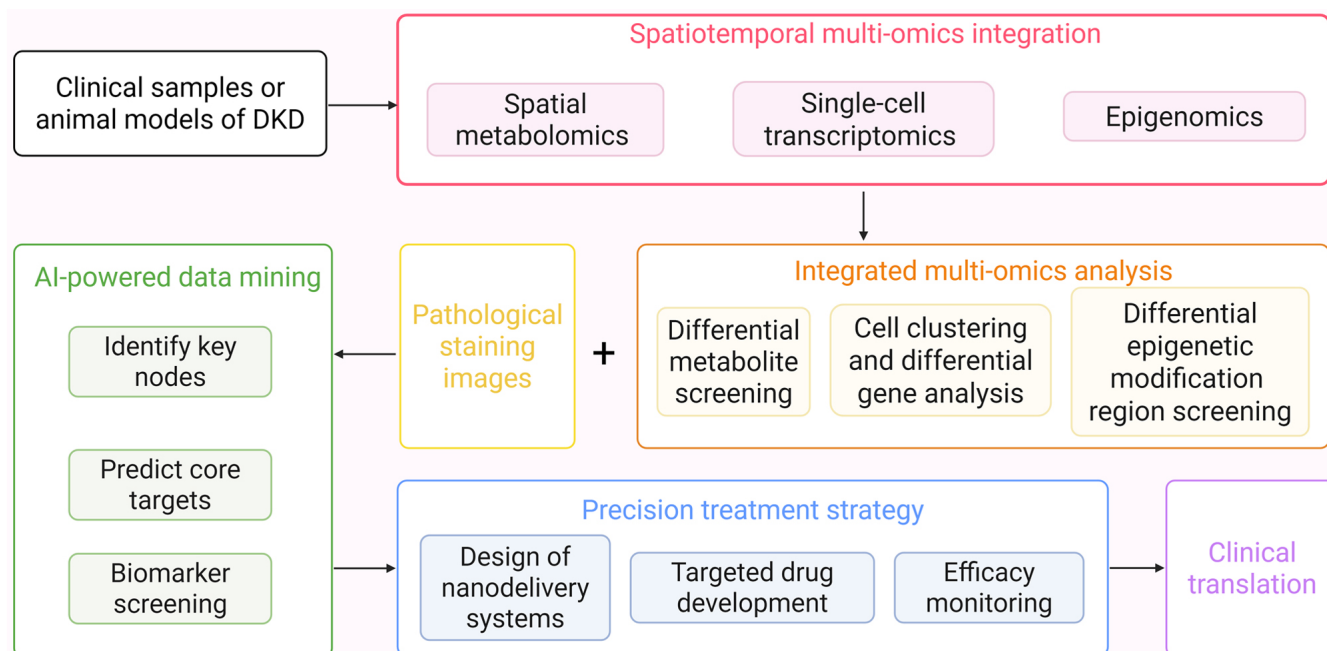


Figure 4. Spatial metabolomics-driven precision medicine research for DKD. This framework illustrates the multi-omics integration strategy of spatial metabolomics with single-cell transcriptomics, epigenomics and pathological imaging. Leveraging AI-powered data mining technologies, a ‘metabolite-gene-cell’ spatial interaction network is constructed to systematically decipher the heterogeneous pathological mechanisms of DKD. This network further generates three major clinical translation outcomes: Novel spatial biomarkers, precise molecular typing systems and efficient therapeutic targets. Ultimately, these findings support the development of precision treatment strategies and promote the advancement of precision medicine in DKD diagnosis and treatment through therapeutic efficacy monitoring. DKD, diabetic kidney disease; AI, artificial intelligence.

As a pivotal methodology for investigating the spatial distribution of metabolites *in vivo*, spatial metabolomics is an indispensable component of spatial multi-omics research. This technology not only provides a novel spatial perspective for understanding DKD but also deepens the comprehension of disease-specific metabolic processes. However, its application in the field of DKD still faces several pressing challenges that need to be addressed, for example: i) There are limitations in technical sensitivity and resolution. Low-abundance metabolites are prone to loss in imaging (113). Most existing studies only localize metabolites to large anatomical regions, such as the cortex or medulla, and they struggle to accurately depict the true distribution of these metabolites within fine functional units, such as glomeruli and specific tubular segments. If the resolution is enhanced, the acquisition speed may consequently be reduced (114). ii) The identification and annotation of metabolites often lack sufficient precision. When annotating metabolites, certain metabolites, such as isomers, cannot be annotated with absolute certainty (17,115,116). iii) Data analysis and interpretation present challenges. Crucial challenges exist in data and signal processing, data comparability and the need for optimization tailored to each tissue type (117). These challenges indicate that engineers and researchers need to address the shortcomings to provide more accurate spatial metabolic maps for the mechanistic analysis of complex diseases such as DKD.

Moreover, spatial metabolomics can directly present actual biochemical activities and functional metabolic outcomes, offering new opportunities for DKD research: Firstly, applying integrated spatial multi-omics to establish causal

mechanisms. As life science research progresses, single-omics approaches are increasingly inadequate for comprehensively deciphering complex biological processes. The integration of spatial metabolomics with transcriptomics and proteomics has become more prevalent (118). By integrating data from spatial metabolomics and spatial transcriptomics, researchers can associate changes in metabolite levels with alterations in gene expression, enabling a deeper exploration of the molecular mechanisms underlying metabolic regulation (119). Secondly, targeted strategies can address regional metabolic heterogeneity. This moves beyond conventional treatment, enabling precision medicine. Spatial metabolomics technology can assess changes in the distribution of metabolites after drug intervention, effectively evaluate DKD drugs, clarify the mechanism of drug treatment and monitor treatment responses, thereby helping to further optimize treatment regimens (31). By combining nanotechnology with drug targeted delivery systems, precise treatment can be implemented for specific regions of metabolic heterogeneity (120). The combined application of these technologies could provide new possibilities for formulating personalized DKD treatment plans and achieving accurate prognostic assessment for patients with DKD (Fig. 4).

10. Conclusion

The present review systematically examines the regional metabolic characteristics of DKD, and it highlights the unique value of spatial metabolomics in elucidating the pathological mechanisms of DKD. With continuous improvements in spatial resolution and quantitative analytical techniques, this technology is expected to serve a critical role in the precision

diagnosis and treatment of DKD. Future research may delve deeper into the cellular level, focusing on elucidating the more refined spatial localization and functional associations of specific metabolites. With the continuous advancement of multi-center data accumulation and clinical trial validation, region-specific metabolites or key enzymes are expected to be translated into reliable diagnostic biomarkers and intervenable therapeutic targets. Such initiatives will enable the tailored development of individualized therapies, ultimately boosting early diagnostic efficiency and optimizing long-term outcomes for patients with DKD.

Acknowledgements

Not applicable.

Funding

The present study was supported by the National Natural Science Foundation of China (grant no. 82374224), the Beijing Natural Science Foundation (grant no. 7252270), the Noncommunicable Chronic Diseases-National Science and Technology Major Project (grant no. 2023ZD0509306), National High Level Hospital Clinical Research Funding (grant no. 2024-NHLHCRF-PY I-07) and National High Level Hospital Clinical Research Funding (grant no. 2024-NHLHCRF-JBGS-ZH-01).

Availability of data and materials

Not applicable.

Authors' contributions

HL made substantial contributions to writing the original draft, reviewing and editing the manuscript, acquiring resources, conducting investigation, developing the conceptualization and performing visualization. TZ participated in reviewing and editing the manuscript, carrying out investigation, securing funding acquisition and conceptualization. DF contributed to reviewing and editing the manuscript, as well as conceptualization. YL and BZ both took part in reviewing and editing the manuscript, and visualization. WC and GM were involved in visualization and investigation. JR made contributions to writing the original draft and visualization. SD participated in investigation and visualization. All authors have read and approved the final manuscript. Data authentication is not applicable.

Ethics approval and consent to participate

Not applicable.

Patient consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

References

1. GBD 2021 Diabetes Collaborators: Global, regional, and national burden of diabetes from 1990 to 2021, with projections of prevalence to 2050: A systematic analysis for the Global Burden of Disease Study 2021. *Lancet* 402: 203-234, 2023.
2. Joumaa JP, Raffoul A, Sarkis C, Chatrich E, Zaidan S, Attieh P, Harb F, Azar S and Ghadieh HE: Mechanisms, biomarkers, and treatment approaches for diabetic kidney disease: Current insights and future perspectives. *J Clin Med* 14: 727, 2025.
3. Johansen KL, Gilbertson DT, Li SL, Li S, Liu J, Roetker NS, Ku E, Schulman IH, Greer RC, Chan K, *et al*: US renal data system 2023 annual data report: Epidemiology of kidney disease in the United States. *Am J Kidney Dis* 83: A8-A13, 2024.
4. Vartak T, Godson C and Brennan E: Therapeutic potential of pro-resolving mediators in diabetic kidney disease. *Adv Drug Deliv Rev* 178: 113965, 2021.
5. Cleveland KH and Schnellmann RG: Pharmacological targeting of mitochondria in diabetic kidney disease. *Pharmacol Rev* 75: 250-262, 2023.
6. Tuttle KR, Agarwal R, Alpers CE, Bakris GL, Brosius FC, Kolkhof P and Uribarri J: Molecular mechanisms and therapeutic targets for diabetic kidney disease. *Kidney Int* 102: 248-260, 2022.
7. Sharma V, Khokhar M, Panigrahi P, Gadwal A, Setia P and Purohit P: Advancements, Challenges, and clinical implications of integration of metabolomics technologies in diabetic nephropathy. *Clin Chim Acta* 561: 119842, 2024.
8. Rayego-Mateos S, Rodrigues-Diez RR, Fernandez-Fernandez B, Mora-Fernández C, Marchant V, Donate-Correa J, Navarro-González JF, Ortiz A and Ruiz-Ortega M: Targeting inflammation to treat diabetic kidney disease: The road to 2030. *Kidney Int* 103: 282-296, 2023.
9. Xu F, Jiang H, Li X, Pan J, Li H, Wang L, Zhang P, Chen J, Qiu S, Xie Y, *et al*: Discovery of PRDM16-Mediated TRPA1 induction as the mechanism for low Tubulo-interstitial fibrosis in diabetic kidney disease. *Adv Sci (Weinh)* 11: e2306704, 2024.
10. Liu D, Chen X, He W, Lu M, Li M, Zhang S, Xie J, Zhang Y and Wang W: Update on the pathogenesis, diagnosis, and treatment of diabetic tubulopathy. *Integrat Med Nephrol Androl* 11: e23-00029, 2024.
11. Empitu MA, Rinastiti P and Kadariswantiningsih IN: Targeting endothelin signaling in podocyte injury and diabetic nephropathy-diabetic kidney disease. *J Nephrol* 38: 49-60, 2025.
12. Muthubharathi BC, Gowripriya T and Balamurugan K: Metabolomics: Small molecules that matter more. *Mol Omics* 17: 210-229, 2021.
13. Zhou M, Sun W, Gao Y, Jiang B, Sun T, Xu R, Zhang X, Wang Q, Xuan Q and Ma S: Metabolomic profiling reveals interindividual metabolic variability and its association with cardiovascular-kidney-metabolic syndrome risk. *Cardiovasc Diabetol* 24: 315, 2025.
14. Barovic M, Hahn JJ, Heinrich A, Adhikari T, Schwarz P, Mirtschink P, Funk A, Kabisch S, Pfeiffer AFH, Blüher M, *et al*: Proteomic and metabolomic signatures in prediabetes progressing to diabetes or reversing to normoglycemia within 1 year. *Diabetes Care* 48: 405-415, 2025.
15. Pereira PR, Carrageta DF, Oliveira PF, Rodrigues A, Alves MG and Monteiro MP: Metabolomics as a tool for the early diagnosis and prognosis of diabetic kidney disease. *Med Res Rev* 42: 1518-1544, 2022.
16. Md Dom ZI, Moon S, Satake E, Hirohama D, Palmer ND, Lampert H, Ficociello LH, Abedini A, Fernandez K, Liang X, *et al*: Urinary Complement proteome strongly linked to diabetic kidney disease progression. *Nat Commun* 16: 7291, 2025.
17. Alexandrov T: Spatial metabolomics and imaging mass spectrometry in the age of artificial intelligence. *Annu Rev Biomed Data Sci* 3: 61-87, 2020.
18. Sharma K, Hansen J, Susztak K, Eberlin L, Anderton CR, Alexandrov T and Iyengar R: Spatial metabolomics and multiomics integration for breakthroughs in precision medicine for kidney disease. *Nat Rev Nephrol*: Oct 9, 2025 doi: 10.1038/s41581-025-01007-3 (Epub ahead of print).
19. Najumudeen AK and Vande voorde J: Spatial metabolomics to unravel cellular metabolism. *Nat Rev Genet* 26: 228, 2025.
20. Allam M and Coskun AF: Combining spatial metabolomics and proteomics profiling of single cells. *Nat Rev Immunol* 24: 701, 2024.

21. Sun N, Krauss T, Seeliger C, Kunzke T, Stöckl B, Feuchtinger A, Zhang C, Voss A, Heisz S, Prokopchuk O, *et al.*: Inter-organ cross-talk in human cancer cachexia revealed by spatial metabolomics. *Metabolism* 161: 156034, 2024.
22. Jia G, He P, Dai T, Goh D, Wang J, Sun M, Wee F, Li F, Lim JCT, Hao S, *et al.*: Spatial immune scoring system predicts hepatocellular carcinoma recurrence. *Nature* 640: 1031-1041, 2025.
23. Jiang X, Li T, Zhou Y, Wang X, Dan Z, Huang J and He J: A new direction in metabolomics: Analysis of the central nervous system based on spatially resolved metabolomics. *TrAC Trends Analytical Chemist* 165: 117103, 2023.
24. Miller A, York EM, Stopka SA, Martínez-François JR, Hossain MA, Baquer G, Regan MS, Agar NYR and Yellen G: Spatially resolved metabolomics and isotope tracing reveal dynamic metabolic responses of dentate granule neurons with acute stimulation. *Nat Metab* 5: 1820-1835, 2023.
25. Dean DA, Klechka L, Hossain E, Parab AR, Eaton K, Hinsdale M and McCall LI: Spatial metabolomics reveals localized impact of influenza virus infection on the lung tissue metabolome. *mSystems* 7: e0035322, 2022.
26. Yu X, Liu Z and Sun X: Single-cell and spatial multi-omics in the plant sciences: Technical advances, applications, and perspectives. *Plant Commun* 4: 100508, 2023.
27. Wang X, Zhang J, Zheng K, Du Q, Wang G, Huang J, Zhou Y, Li Y, Jin H and He J: Discovering metabolic vulnerability using spatially resolved metabolomics for antitumor small molecule-drug conjugates development as a precise cancer therapy strategy. *J Pharm Anal* 13: 776-787, 2023.
28. Bag S, Oetjen J, Shaikh S, Chaudhary A, Arun P and Mukherjee G: Impact of spatial metabolomics on immune-microenvironment in oral cancer prognosis: A clinical report. *Mol Cell Biochem* 479: 41-49, 2024.
29. He T, Lin K, Xiong L, Zhang W, Zhang H, Duan C, Li X and Zhang J: Disorder of phospholipid metabolism in the renal cortex and medulla contributes to acute tubular necrosis in mice after cantharidin exposure using integrative lipidomics and spatial metabolomics. *J Pharm Anal* 15: 101210, 2025.
30. Qiu S, Wang Z, Wang X, Guo S, Cai Y, Xie D, Hu Z, Wang S, Yang Q and Zhang A: Spatial metabolomics identifies riboflavin metabolism as a therapeutic target of Huangqi Guizhi Wuwu decoction in diabetic nephropathy. *Biomed Chromatogr* 39: e70239, 2025.
31. Wang Z, Fu W, Huo M, He B, Liu Y, Tian L, Li W, Zhou Z, Wang B, Xia J, *et al.*: Spatial-resolved metabolomics reveals tissue-specific metabolic reprogramming in diabetic nephropathy by using mass spectrometry imaging. *Acta Pharm Sin B* 11: 3665-3677, 2021.
32. Min X, Zhao Y, Yu M, Zhang W, Jiang X, Guo K, Wang X, Huang J, Li T, Sun L and He J: Spatially resolved metabolomics: From metabolite mapping to function visualizing. *Clin Transl Med* 14: e70031, 2024.
33. Tuck M, Grélard F, Blanc L and Desbenoit N: MALDI-MSI towards multimodal imaging: Challenges and perspectives. *Front Chem* 10: 904688, 2022.
34. Kumar BS: Desorption electrospray ionization mass spectrometry imaging (DESI-MSI) in disease diagnosis: An overview. *Anal Methods* 15: 3768-3784, 2023.
35. Yang S, Wang Z, Liu Y, Zhang X, Zhang H, Wang Z, Zhou Z and Abliz Z: Dual mass spectrometry imaging and spatial metabolomics to investigate the metabolism and nephrotoxicity of nitidine chloride. *J Pharm Anal* 14: 100944, 2024.
36. Song X, Zang Q, Li C, Zhou T and Zare RN: Immuno-desorption electrospray ionization mass spectrometry imaging identifies functional macromolecules by using Microdroplet-cleavable mass tags. *Angew Chem Int Ed Engl* 62: e202216969, 2023.
37. Lockyer NP, Aoyagi S, Fletcher JS, Gilmore I, van der heide P, Moore KL, Tyler BJ and Weng LT: Secondary ion mass spectrometry. *Nat Rev Methods Primers* 4: 32, 2024.
38. Coello Y, Jones AD, Gunaratne TC and Dantus M: Atmospheric pressure femtosecond laser imaging mass spectrometry. *Anal Chem* 82: 2753-2758, 2010.
39. Chen H, Durand S, Bawa O, Bourgin M, Montégut L, Lambertucci F, Motiño O, Li S, Nogueira-Recalde U, Anagnostopoulos G, *et al.*: Biomarker identification in liver cancers using desorption electrospray ionization mass spectrometry (DESI-MS) imaging: An approach for spatially resolved metabolomics. *Methods Mol Biol* 2769: 199-209, 2024.
40. He MJ, Pu W, Wang X, Zhong X, Zhao D, Zeng Z, Cai W, Liu J, Huang J, Tang D and Dai Y: Spatial metabolomics on liver cirrhosis to hepatocellular carcinoma progression. *Cancer Cell Int* 22: 366, 2022.
41. Lakkimsetty SS, Weber A, Bemis KA, Stehl V, Bronsert P, Föll MC and Vitek O: MSReg: An R package for unsupervised coregistration of mass spectrometry and H&E images. *Bioinformatics* 40: btae624, 2024.
42. Zickuhr GM, Um IH, Laird A, Harrison DJ and Dickson AL: DESI-MSI-guided exploration of metabolic-phenotypic relationships reveals a correlation between PI 38:3 and proliferating cells in clear cell renal cell carcinoma via single-section co-registration of multimodal imaging. *Anal Bioanal Chem* 416: 4015-4028, 2024.
43. Wang G, Heijs B, Kostidis S, Rietjens RGJ, Koning M, Yuan L, Tiemeier GL, Mahfouz A, Dumas SJ, Giera M, *et al.*: Spatial dynamic metabolomics identifies metabolic cell fate trajectories in human kidney differentiation. *Cell Stem Cell* 29: 1580-1593, e7, 2022.
44. Wang G, Heijs B, Kostidis S, Mahfouz A, Rietjens RGJ, Bijkerk R, Koudijs A, van der Pluijm LAK, van den Berg CW, Dumas SJ, *et al.*: Analyzing cell-type-specific dynamics of metabolism in kidney repair. *Nat Metab* 4: 1109-1118, 2022.
45. Lin J, Lin H, Li C, Liao N, Zheng Y, Yu X, Sun Y and Wu L: Unveiling characteristic metabolic accumulation over enzymatic-catalyzed process of Tieguanyin oolong tea manufacturing by DESI-MSI and multiple-omics. *Food Res Int* 181: 114136, 2024.
46. Banerjee S, Wong AC, Yan X, Wu B, Zhao H, Tibshirani RJ, Zare RN and Brooks JD: Early detection of unilateral ureteral obstruction by desorption electrospray ionization mass spectrometry. *Sci Rep* 9: 11007, 2019.
47. Qi K, Wu L, Liu C and Pan Y: Recent advances of ambient mass spectrometry imaging and its applications in lipid and metabolite analysis. *Metabolites* 11: 780, 2021.
48. Blanc T, Goudin N, Zaidan M, Traore MG, Bienaime F, Turinsky L, Garbay S, Nguyen C, Burtin M, Friedlander G, *et al.*: Three-dimensional architecture of nephrons in the normal and cystic kidney. *Kidney Int* 99: 632-645, 2021.
49. Li H, Li D and Humphreys BD: Chromatin conformation and histone modification profiling across human kidney anatomic regions. *Sci Data* 11: 797, 2024.
50. Zhang SY and Mahler GJ: A glomerulus and proximal tubule microphysiological system simulating renal filtration, reabsorption, secretion, and toxicity. *Lab Chip* 23: 272-284, 2023.
51. Fan G, Jiang C, Huang Z, Tian M, Pan H, Cao Y, Lei T, Luo Q and Yuan J: 3D autofluorescence imaging of hydronephrosis and renal anatomical structure using cryo-micro-optical sectioning tomography. *Theranostics* 13: 4885-4904, 2023.
52. Hinze C, Karaikos N, Boltengagen A, Walentin K, Redo K, Himmerkus N, Bleich M, Potter SS, Potter AS, Eckardt KU, *et al.*: Kidney Single-cell transcriptomes predict spatial corticomedullary gene expression and tissue osmolality gradients. *J Am Soc Nephrol* 32: 291-306, 2021.
53. Gao G, Sumrall ES, Pitchiaya S, Bitzer M, Alberti S and Walter NG: Biomolecular condensates in kidney physiology and disease. *Nat Rev Nephrol* 19: 756-770, 2023.
54. Ding Y, Zhao F, Hu J, Zhao Z, Shi B and Li S: A conjoint analysis of renal structure and omics characteristics reveal new insight to yak high-altitude hypoxia adaptation. *Genomics* 116: 110857, 2024.
55. Gurung RL, Zheng H, Tan JLI, Liu S, Chan C, Ang K, Tan C, Liu JJ, Subramaniam T, Sum CF and Lim SC: Integrative metabolomic and proteomic analysis of diabetic kidney disease progression with younger-onset type 2 diabetes. *Diabetes Obes Metab* 27: 7454-7463, 2025.
56. Jiang X, Liu X, Qu X, Zhu P, Wo F, Xu X, Jin J, He Q and Wu J: Integration of metabolomics and peptidomics reveals distinct molecular landscape of human diabetic kidney disease. *Theranostics* 13: 3188-3203, 2023.
57. Fan X, Yang M, Lang Y, Lu S, Kong Z, Gao Y, Shen N, Zhang D and Lv Z: Mitochondrial metabolic reprogramming in diabetic kidney disease. *Cell Death Dis* 15: 442, 2024.
58. Li S and Susztak K: Mitochondrial dysfunction has a central role in diabetic kidney disease. *Nat Rev Nephrol* 21: 77-78, 2025.
59. Poorna R, Chen WW, Qiu P and Cicerone MT: Toward Gene-correlated spatially resolved metabolomics with fingerprint coherent Raman imaging. *J Phys Chem B* 127: 5576-5587, 2023.
60. Alexandrov T: Spatial metabolomics: From a niche field towards a driver of innovation. *Nat Metabolism* 5: 1443-1445, 2023.
61. Zhang J, Wu T, Li C and Du J: A glycopolymerosome strategy for 'drug-free' treatment of diabetic nephropathy. *J Control Release* 372: 347-361, 2024.

62. Luo A, Wang R, Gong J, Wang S, Yun C, Chen Z, Jiang Y, Liu X, Dai H, Liu H and Zheng Y: Syntaxin 17 translocation mediated mitophagy switching drives hyperglycemia-induced vascular injury. *Adv Sci (Weinh)* 12: e2414960, 2025.
63. Rebelos E, Mari A, Oikonen V, Iida H, Nuutila P and Ferrannini E: Evaluation of renal glucose uptake with [¹⁸F] FDG-PET: Methodological advancements and metabolic outcomes. *Metabolism* 141: 155382, 2023.
64. Liu X, Du H, Sun Y and Shao L: Role of abnormal energy metabolism in the progression of chronic kidney disease and drug intervention. *Ren Fail* 44: 790-805, 2022.
65. Zhang G, Zhang J, DeHoog RJ, Pennathur S, Anderton CR, Venkatachalam MA, Alexandrov T, Eberlin LS and Sharma K: DESI-MSI and METASPACE indicates lipid abnormalities and altered mitochondrial membrane components in diabetic renal proximal tubules. *Metabolomics* 16: 11, 2020.
66. Zhang X, Liu Y, Yang S, Gao X, Wang S, Wang Z, Zhang C, Zhou Z, Chen Y, Wang Z and Abliz Z: Comparison of local metabolic changes in diabetic rodent kidneys using mass spectrometry imaging. *Metabolites* 13: 324, 2023.
67. Zhang G, Liu L, Tamayo IM, De Leon NGP, Vigers TB, Tommerdahl KL, Nelson RG, Ladd PE, Alexandrov T, Birznieks C, *et al*: 406-P: Spatial metabolomics of human kidney tissues reveal impaired tricarboxylic acid (TCA) cycle turnover in type 1 Diabetes (T1D). *Diabetes* 72: 406, 2023.
68. Qi W, Keenan HA, Li Q, Ishikado A, Kannt A, Sadowski T, Yorek MA, Wu IH, Lockhart S, Coppey LJ, *et al*: Pyruvate kinase M2 activation may protect against the progression of diabetic glomerular pathology and mitochondrial dysfunction. *Nat Med* 23: 753-762, 2017.
69. Hasegawa S and Inagi R: Harnessing metabolomics to describe the pathophysiology underlying progression in diabetic kidney disease. *Curr Diab Rep* 21: 21, 2021.
70. Murphy DP, Wolfson J, Reule S, Johansen KL, Ishani A and Drawz PE: A cohort study of sodium-glucose cotransporter-2 inhibitors after acute kidney injury among Veterans with diabetic kidney disease. *Kidney Int* 106: 126-135, 2024.
71. Wang Y, Liu T, Wu Y, Wang L, Ding S, Hou B, Zhao H, Liu W and Li P: Lipid homeostasis in diabetic kidney disease. *Int J Biol Sci* 20: 3710-3724, 2024.
72. Mitrofanova A, Merscher S and Fornoni A: Kidney lipid dysmetabolism and lipid droplet accumulation in chronic kidney disease. *Nat Rev Nephrol* 19: 629-645, 2023.
73. Iizuka K: Commentary: Comprehensive lipidome profiling of the kidney in early-stage diabetic nephropathy. *Front Endocrinol (Lausanne)* 13: 1015305, 2022.
74. Zhang YR, Piao HL and Chen D: Identification of spatial specific lipid metabolic signatures in long-standing diabetic kidney disease. *Metabolites* 14: 641, 2024.
75. Hao Y, Fan Y, Feng J, Zhu Z, Luo Z, Hu H, Li W, Yang H and Ding G: ALCAT1-mediated abnormal cardiolipin remodelling promotes mitochondrial injury in podocytes in diabetic kidney disease. *Cell Commun Signal* 22: 26, 2024.
76. Grove KJ, Voziyan PA, Spraggins JM, Wang S, Pauksakon P, Harris RC, Hudson BG and Caprioli RM: Diabetic nephropathy induces alterations in the glomerular and tubule lipid profiles. *J Lipid Res* 55: 1375-1385, 2014.
77. McCrimmon A, Corbin S, Shrestha B, Roman G, Dhungana S and Stadler K: Redox phospholipidomics analysis reveals specific oxidized phospholipids and regions in the diabetic mouse kidney. *Redox Biol* 58: 102520, 2022.
78. Vianello E, Ambrogi F, Kalousová M, Badalyan J, Dozio E, Tacchini L, Schmitz G, Zima T, Tsongalis GJ and Corsi-Romanelli MM: Circulating perturbation of phosphatidylcholine (PC) and phosphatidylethanolamine (PE) is associated to cardiac remodeling and NLRP3 inflammasome in cardiovascular patients with insulin resistance risk. *Exp Mol Pathol* 137: 104895, 2024.
79. Wunderling K, Zurkovic J, Zink F, Kuerschner L and Thiele C: Triglyceride cycling enables modification of stored fatty acids. *Nat Metab* 5: 699-709, 2023.
80. Darshi M, Tumova J, Saliba A, Kim J, Baek J, Pennathur S and Sharma K: Crabtree effect in kidney proximal tubule cells via late-stage glycolytic intermediates. *Iscience* 26: 106462, 2023.
81. Hall AM: Protein handling in kidney tubules. *Nat Rev Nephrol* 21: 241-252, 2025.
82. Panov AV, Mayorov VI, Dikalova AE and Dikalov SI: Long-Chain and Medium-Chain fatty acids in energy metabolism of murine kidney mitochondria. *Int J Mol Sci* 24: 379, 2023.
83. Li H, Li D, Ledru N, Xuanyuan Q, Wu H, Asthana A, Byers LN, Tullius SG, Orlando G, Waikar SS and Humphreys BD: Transcriptomic, epigenomic, and spatial metabolomic cell profiling redefines regional human kidney anatomy. *Cell Metab* 36: 1105-1125.e10, 2024.
84. Kang HM, Ahn SH, Choi P, Ko YA, Han SH, Chinga F, Park AS, Tao J, Sharma K, Pullman J, *et al*: Defective fatty acid oxidation in renal tubular epithelial cells has a key role in kidney fibrosis development. *Nat Med* 21: 37-46, 2015.
85. Li C, Gao L, Lv C, Li Z, Fan S, Liu X, Rong X, Huang Y and Liu J: Active role of amino acid metabolism in early diagnosis and treatment of diabetic kidney disease. *Front Nutr* 10: 1239838, 2023.
86. Liu L, Xu J, Zhang Z, Ren D, Wu Y, Wang D, Zhang Y, Zhao S, Chen Q and Wang T: Metabolic homeostasis of amino acids and diabetic kidney disease. *Nutrients* 15: 184, 2023.
87. Linnan B, Yanzhe W, Ling Z, Yuyuan L, Sijia C, Xinmiao X, Fengqin L and Xiaoxia W: In situ metabolomics of metabolic reprogramming involved in a mouse model of type 2 diabetic kidney disease. *Front Physiol* 12: 779683, 2021.
88. Han J, Li P, Sun H, Zheng Y, Liu C, Chen X, Guan S, Yin F and Wang X: Integrated metabolomics and mass spectrometry imaging analysis reveal the efficacy and mechanism of Huangkui capsule on type 2 diabetic nephropathy. *Phytomedicine* 138: 156397, 2025.
89. Castoldi F, Kroemer G and Pietrocola F: Spermidine rejuvenates T lymphocytes and restores anticancer immunosurveillance in aged mice. *Oncoimmunology* 11: 2146855, 2022.
90. Zou D, Zhao Z, Li L, Min Y, Zhang D, Ji A, Jiang C, Wei X and Wu X: A comprehensive review of spermidine: Safety, health effects, absorption and metabolism, food materials evaluation, physical and chemical processing, and bioprocessing. *Compr Rev Food Sci Food Saf* 21: 2820-2842, 2022.
91. Aihara S, Torisu K, Uchida Y, Imazu N, Nakano T and Kitazono T: Spermidine from arginine metabolism activates Nrf2 and inhibits kidney fibrosis. *Commun Biol* 6: 676, 2023.
92. Li X, Zhou X, Liu X, Li X, Jiang X, Shi B and Wang S: Spermidine protects against acute kidney injury by modulating macrophage NLRP3 inflammasome activation and mitochondrial respiration in an eIF5A hypusination-related pathway. *Mol Med* 28: 103, 2022.
93. Jia M, Lin L, Xun K, Li D, Wu W, Sun S, Qiu H and Jin D: Indoxyl sulfate aggravates podocyte damage through the TGF- β 1/Smad/ROS signalling pathway. *Kidney Blood Press Res* 49: 385-396, 2024.
94. Zhao T, Zhang H, Yin X, Zhao H, Ma L, Yan M, Peng L, Wang Q, Dong X and Li P: Tangshen formula modulates gut Microbiota and reduces gut-derived toxins in diabetic nephropathy rats. *Biomed Pharmacother* 129: 110325, 2020.
95. Hejazi L, Sharma S, Ruiz A, Zhang G, Tucci FC and Sharma K: Spatial metabolomics analysis by MSI-DeepPath identifies key pathways in ZDF diabetic kidney disease model. *Diabetes* 72 (Suppl 1): 400-P, 2023.
96. Varadaiah YGC, Sivanesan S, Nayak SB and Thirumalarao KR: Purine metabolites can indicate diabetes progression. *Arch Physiol Biochem* 128: 87-91, 2022.
97. Zubaidi SN, Wong PL, Qadi WSM, Dawoud EAD, Hamezah HS, Baharum SN, Jam FA, Abas F, Moreno A and Mediani A: Deciphering the mechanism of Annona muricata leaf extract in alloxan-nicotinamide-induced diabetic rat model with 1H-NMR-based metabolomics approach. *J Pharm Biomed Anal* 260: 116806, 2025.
98. Shen XL, Liu H, Xiang H, Qin XM, Du GH and Tian JS: Combining biochemical with (1)H NMR-based metabolomics approach unravels the antidiabetic activity of genipin and its possible mechanism. *J Pharm Biomed Anal* 129: 80-89, 2016.
99. Yuan Y, Huang L, Yu L, Yan X, Chen S, Bi C, He J, Zhao Y, Yang L, Ning L, *et al*: Clinical metabolomics characteristics of diabetic kidney disease: A meta-analysis of 1875 cases with diabetic kidney disease and 4503 controls. *Diabetes Metab Res Rev* 40: e3789, 2024.
100. Jung I, Nam S, Lee DY, Park SY, Yu JH, Seo JA, Lee DH and Kim NH: Association of succinate and adenosine nucleotide metabolic pathways with diabetic kidney disease in patients with type 2 diabetes mellitus. *Diabetes Metab J* 48: 1126-1134, 2024.
101. Mohandes S, Doke T, Hu H, Mukhi D, Dhillion P and Susztak K: Molecular pathways that drive diabetic kidney disease. *J Clin Invest* 133: e165654, 2023.
102. Sharma K, Zhang GS, Hansen J, Bjornstad P, Lee HJ, Menon R, Hejazi L, Liu JJ, Franzone A, Looker HC, *et al*: Endogenous adenine mediates kidney injury in diabetic models and predicts diabetic kidney disease in patients. *J Clin Invest* 133: e170341, 2023.

103. Ragi N and Sharma K: Deliverables from metabolomics in kidney disease: Adenine, new insights, and implication for clinical Decision-making. *Am J Nephrol* 55: 421-438, 2024.
104. Drexler Y and Fornoni A: Adenine crosses the biomarker bridge: From 'omics to treatment in diabetic kidney disease. *J Clin Invest* 133: e174015, 2023.
105. Hocher B and Adamski J: Metabolomics for clinical use and research in chronic kidney disease. *Nat Rev Nephrol* 13: 269-284, 2017.
106. Efiiong EE, Maedler K, Effa E, Osuagwu UL, Peters E, Ikebiuro JO, Soremekun C, Ihediwa U, Niu J, Fuchs M, *et al*: Decoding diabetic kidney disease: A comprehensive review of interconnected pathways, molecular mediators, and therapeutic insights. *Diabetol Metab Syndr* 17: 192, 2025.
107. Sinha SK and Nicholas SB: Pathomechanisms of diabetic kidney disease. *J Clin Med* 12: 7349, 2023.
108. Huynh C, Ryu J, Lee J, Inoki A and Inoki K: Nutrient-sensing mTORC1 and AMPK pathways in chronic kidney diseases. *Nat Rev Nephrol* 19: 102-122, 2023.
109. Zhang J, Fuhrer T, Ye H, Kwan B, Montemayor D, Tumova J, Darshi M, Afshinnia F, Scialla JJ, Anderson A, *et al*: High-throughput metabolomics and diabetic kidney disease progression: Evidence from the chronic renal insufficiency (CRIC) study. *Am J Nephrol* 53: 215-225, 2022.
110. Hong YA and Nangaku M: Endogenous adenine as a key player in diabetic kidney disease progression: An integrated multiomics approach. *Kidney Int* 105: 918-920, 2024.
111. Demko J, Saha B, Takagi E, Manis A, Richman P and Pearce D: Renal tubule mTORC2 deletion increases gluconeogenesis and urinary glucose excretion. *Physiology* 38: 5735039, 2023.
112. Qiu S, Xie D, Guo S, Wang Z, Wang X, Cai Y, Lin C, Yao H, Guan Y, Zhao Q, *et al*: Spatially segregated multiomics decodes metformin-mediated function-specific metabolic characteristics in diabetic kidney disease. *Life Metabolism* 4: loaf019, 2025.
113. Wu Q, Chu JL, Rubakhin SS, Gillette MU and Sweedler JV: Dopamine-modified TiO₂ monolith-assisted LDI MS imaging for simultaneous localization of small metabolites and lipids in mouse brain tissue with enhanced detection selectivity and sensitivity. *Chem Sci* 8: 3926-3938, 2017.
114. Spraggins JM, Rizzo DG, Moore JL, Noto MJ, Skaar EP and Caprioli RM: Next-generation technologies for spatial proteomics: Integrating ultra-high speed MALDI-TOF and high mass resolution MALDI FTICR imaging mass spectrometry for protein analysis. *Proteomics* 16: 1678-1689, 2016.
115. Neumann EK, Migas LG, Allen JL, Caprioli RM, Van de Plas R and Spraggins JM: Spatial metabolomics of the human kidney using MALDI trapped ion mobility imaging mass spectrometry. *Anal Chem* 92: 13084-13091, 2020.
116. Fu T, Oetjen J, Chapelle M, Verdu A, Szesny M, Chaumot A, Degli-Esposti D, Geffard O, Clément Y, Salvador A and Aycirix S: In situ isobaric lipid mapping by MALDI-ion mobility separation-mass spectrometry imaging. *J Mass Spectrom* 55: e4531, 2020.
117. Spatial Omics DataBase (SODB): Increasing accessibility to spatial omics data. *Nat Methods* 20: 359-360, 2023.
118. Vandergrift GW, Veličković M, Day LZ, Gorman BL, Williams SM, Shrestha B and Anderton CR: Untargeted spatial metabolomics and spatial proteomics on the same tissue section. *Anal Chem* 97: 392-400, 2024.
119. Abedini A, Levinsohn J, Klötzer KA, Dumoulin B, Ma Z, Frederick J, Dhillon P, Balzer MS, Shrestha R, Liu H, *et al*: Single-cell multi-omic and spatial profiling of human kidneys implicates the fibrotic microenvironment in kidney disease progression. *Nat Genet* 56: 1712-1724, 2024.
120. Chuang AE, Chen YL, Chiu HJ, Nguyen HT and Liu CH: Nasal administration of polysaccharides-based nanocarrier combining hemoglobin and diferuloylmethane for managing diabetic kidney disease. *Int J Biol Macromol* 282: 136534, 2024.



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