Sains Malaysiana 55(6)(2026): 1009-1020
http://doi.org/10.17576/jsm-2026-5506-06

Understanding Diabetic Retinopathy Using Multi-Omics Approaches: A Narrative Review

(Memahami Retinopati Diabetes Menggunakan Pendekatan Multi-Omik: Satu Ulasan Naratif)

ZHANQING LUO[1], MUHAMMAD ZULFIQAH SADIKAN[2,3], FAIDRUZ AZURA JAM[3], RUSDIAH RUZANNA JUSOH[3], XIAOHUI TONG[4], RONGCHUN HAN[5], MURNI NAZIRA SARIAN[1], AHMED MEDIANI[1] & HAMIZAH SHAHIRAH HAMEZAH[1,*]

[1]*Institute of Systems Biology (INBIOSIS), Universiti Kebangsaan Malaysia, 43600 UKM Bangi, Selangor, Malaysia*
[2]*Faculty of Pharmacy and Health Sciences, Universiti Kuala Lumpur Royal College of Medicine Perak, Jalan Greentown, 30450 Ipoh, Perak, Malaysia*
[3]*Faculty of Medicine, Manipal University College Malaysia (MUCM), Jalan Padang Jambu, Bukit Baru, 75150 Melaka, Malaysia*
[4]*School of Life Sciences, Anhui University of Chinese Medicine, Hefei 230012, China*
[5]*School of Pharmacy, Anhui University of Chinese Medicine, Hefei 230012, China*

Received: 30 September 2025/Accepted: 15 May 2026

ABSTRACT

Diabetic retinopathy (DR) is one of the leading causes of vision loss in diabetic patients worldwide. DR is a complex disease with diverse clinical symptoms ranging from mild nonproliferative stages to severe proliferative stages, and also with significant differences in response to treatment. This heterogeneity suggests that DR is not a single disease but rather a series of retinal pathologies involving multiple molecular mechanisms. Interventions such as anti-VEGF therapy and laser therapy have made significant progress in recent years, improving patient prognosis. However, these treatments are still unable to completely stop the disease progression and vision loss. Therefore, it is particularly urgent to further study the pathogenesis of DR. In recent years, multi-omics approaches have shown great potential in showing novel biomarkers and molecular pathways. This review focuses on integrating findings from three key omics approaches (genomics, proteomics, and metabolomics) in DR research. We discuss the role of genomics in understanding genetic susceptibility, the contributions of proteomics to elucidating inflammation, angiogenesis, and therapeutic target discovery, and the application of metabolomics in characterizing metabolic disorders and biomarker screening. Integration of multi-omics data may help elucidate complex pathological mechanisms and provide insights that support precision medicine and personalized treatment strategies for DR.

Keywords: Biomarkers; diabetic retinopathy; genomics; metabolomics; proteomics

ABSTRAK

Retinopati diabetes (DR) merupakan salah satu punca utama kehilangan penglihatan dalam kalangan pesakit diabetes di seluruh dunia. DR ialah sejenis penyakit kompleks yang disertai dengan pelbagai gejala klinikal, bermula daripada peringkat awal bukan proliferatif yang ringan hingga ke peringkat proliferatif yang teruk, serta menunjukkan perbezaan ketara dalam tindak balas terhadap rawatan. Kepelbagaian ini menunjukkan bahawa DR bukanlah satu penyakit tunggal, tetapi merupakan suatu rangkaian patologi retina yang melibatkan pelbagai mekanisme molekul. Intervensi seperti terapi anti-VEGF dan terapi laser telah mencapai kemajuan yang ketara dalam beberapa tahun kebelakangan ini, sekali gus memperbaiki prognosis pesakit. Namun begitu, rawatan ini masih belum mampu menghentikan sepenuhnya perkembangan penyakit dan kehilangan penglihatan. Oleh itu, kajian lanjut mengenai patogenesis DR adalah amat mendesak untuk dilaksanakan. Dalam beberapa tahun kebelakangan ini, pendekatan multi-omik telah menunjukkan potensi besar dalam mendedahkan penanda biologi dan laluan molekul baharu. Tinjauan ini memfokuskan dan mengintegrasikan penemuan daripada tiga lapisan omik utama (genomik, proteomik dan metabolomik) dalam penyelidikan DR. Ulasan ini membincangkan peranan genomik dalam memahami kerentanan genetik, sumbangan proteomik dalam menjelaskan keradangan, angiogenesis dan penemuan sasaran terapeutik, serta aplikasi metabolomik dalam mencirikan gangguan metabolik dan saringan penanda biologi. Dengan mengintegrasikan data multi-omik, mekanisme patologi yang kompleks dapat didedahkan dan asas saintifik dapat disediakan bagi pelaksanaan perubatan kepersisan serta rawatan yang diperibadikan untuk penyakit DR.

Kata kunci: Genomik; metabolomik; penanda biologi; proteomik; retinopati diabetes

INTRODUCTION

Diabetic retinopathy (DR) is a microvascular complication among patients with diabetes and one of the leading causes of blindness in adults worldwide (GDB 2021). Characterized by microvascular damage, inflammation, and neurodegeneration, DR exhibits significant heterogeneity in molecular mechanisms and clinical phenotypes (Brown et al. 2021; Husain et al. 2019). Generally, poor glycemic control, hypertension, long duration of diabetes, and dyslipidemia are considered key risk factors associated with DR (Al-Ghamdi 2020; Bryl et al. 2022). While multidisciplinary screening has improved (Fung et al. 2022; Lim 2024), the specific mechanisms underlying neurodegeneration and diabetic macular edema (DME) remain inadequately understood (Rossino, Dal Monte & Casini 2019; Suciu et al. 2022). Overall, DR is a complex and heterogeneous disease, and a comprehensive and in-depth understanding of its pathogenesis and personalized intervention strategies will help to improve prevention and treatment outcomes and bring better vision and quality of life to patients.

At present, numerous challenges remain in the diagnosis and treatment of DR, including the limited efficacy of nanotechnology-based drug delivery due to ocular physiological barriers (Li, Chen & Fu 2023). Additionally, traditional classification systems often fail to capture the complexity of DR. While artificial intelligence (Jagadesh et al. 2023) and supplemental therapies offer potential, such as Vitamin D (Gverović Antunica et al. 2023) and antioxidants (Alfonso-Muñoz et al. 2021), their conventional implementation requires further validation. Furthermore, ethnic and regional disparities necessitate more personalized intervention strategies, especially in resource-limited areas (Tan & Wong 2022).

Traditional single-perspective research is insufficient for deciphering the multi-layered pathogenesis of DR. Consequently, systems biology approaches integrating genomics, proteomics, and metabolomics have emerged as transformative tools (Cabrera et al. 2020; Weber et al. 2021). For example, metabolomics analysis of serum, tears, and vitreous fluid from patients with different degrees of DR can identify metabolites associated with disease progression (Lin et al. 2020; Mokhtar et al. 2023; Wen et al. 2023). This review aims to provide a comprehensive synthesis of findings from these three omics approaches, genomics, proteomics, and metabolomics, illustrating their individual and interconnected roles in DR pathology. In the future, further integration of such multi-omics data with advanced technologies, such as artificial intelligence, will provide stronger support for the precise prevention and control of DR, so as to ultimately reduce the blindness rate of DR and improve the quality of life of diabetic patients.

GENOMICS APPROACH TO UNDERSTAND DIABETIC RETINOPATHY

With advancements in molecular biology, particularly the emergence of next-generation sequencing (NGS) technologies, our understanding of DR has turned from traditional clinical observations to in-depth exploration of molecular mechanisms. The development of genomics can be broadly categorized into two phases: the 'pre-NGS era' before the advent of NGS technologies and the 'post-NGS era' following their widespread application (Rossi et al. 2022).

In the pre-NGS era, identifying Vascular Endothelial Growth Factor (VEGF) was a milestone. Hypoxia-induced VEGF secretion by glial and Müller cells may promote retinal neovascularization and vascular permeability (Miller et al. 1994; Plate et al. 1992; Stone et al. 1995). This phenomenon, resembling the characteristic lesions of proliferative diabetic retinopathy (PDR), further suggests that VEGF is a key mediator of vascular permeability and angiogenesis in the retina. A meta-analysis demonstrated VEGF levels in the vitreous or aqueous humor of PDR patients are statistically significantly higher than those in controls, which may provide a solid theoretical foundation for its use as a therapeutic target (Mason et al. 2022). Family-based studies have further uncovered genetic susceptibilities. For instance, specific pancreatic and duodenal homeobox 1 (PDX1) variants (c.443G>T, p.Arg148Leu and c.442C>G, p.Arg148Gly) are linked to familial DR in two Japanese families with Maturity-Onset Diabetes of the Young (MODY) (Tanaka et al. 2025), while dual mutations in mitochondrially encoded TRNA-Leu (UUA/G) 1 (MT-TL1, m.3243A>G, 41.76% heteroplasmy) and carboxyl ester lipase (CEL, c.1336G>A) have been identified in Chinese cohorts (Che et al. 2025). Investigations into individual gene polymorphisms highlight associations between receptor for advanced glycation end products (RAGE, rs1800625, rs1800624) and transforming growth factor beta 1 (TGF-β1, 915G>C) genes variants with non-proliferative diabetic retinopathy (NPDR) severity (Mihoubi et al. 2022; Qayyum, Afzal & Naveed 2021a, 2021b). Additionally, interactions between vitamin D receptor (VDR) polymorphisms (TT genotype of rs1544410 and the GG genotype of rs731236) and circulating Vitamin D levels contribute to the individual variability in DR occurrence (Chen et al. 2022).

Genome-wide association studies (GWAS), a pivotal tool in the post-NGS era, analyze SNPs across the genome to identify genetic loci associated with DR susceptibility. Key findings include the association of the folliculin (FLCN) gene with type 1 diabetes-related DR (Skol et al. 2020) and the identification of functional gene sets linked to oxidative stress and lipid metabolism (Sobrin et al. 2022). Furthermore, mendelian randomization and GWAS analyses suggested a genetic overlap between Alzheimer's disease and DR, implicating shared loci such as oxidative stress-responsive 1 (OARD1) and triggering receptor expressed on myeloid cells 1 (TREM1) (Ouyang et al. 2024). Furthermore, population-specific GWAS have identified unique loci like WD repeat domain 72 (WDR72) variants (rs12906891 and rs11070992) in African populations (Liu et al. 2019) and STT3 oligosaccharyltransferase complex catalytic subunit B (STT3B, rs12630354) in Japanese

cohorts (Imamura et al. 2021), highlighting the importance of genetic background. Functional validation has also confirmed the significance of transcription factor 7-like 2 (TCF7L2, rs7903146) and calcium voltage-gated channel auxiliary aubunit beta 2 (CACNB2, rs11014284) variants in DR progression (Yu & Rong 2023).

Epigenetic studies have implicated potential roles for DNA methylation and histone modifications in DR development under diabetic conditions. Under hyperglycemic conditions, dynamic DNA methylation and histone modifications (H3K9me3) upregulate Rac1 expression, which may accelerate mitochondrial damage and potentially worsen retinal degeneration (Kowluru, Radhakrishnan & Mohammad 2021; Sahajpal, Kowluru & Kowluru 2019). Targeting Rac1 and its regulatory mechanisms may offer novel approaches for DR prevention and treatment. Additionally, gene-editing technologies, such as CRISPR/Cas9, hold great promise for targeted DR therapies. For instance, CRISPR/Cas9-mediated knockout of Axl, as well as the pharmacological inhibition of Axl using the specific inhibitor R428, offer promising therapeutic avenues (Wu et al. 2021). However, challenges in data interpretation, ethical concerns, and clinical translation remain significant in this field (Bansal 2025).

PROTEOMICS APPROACH TO UNDERSTAND DIABETIC RETINOPATHY

Following the completion of the human genome sequencing, proteomics has become an important tool for biomarker discovery, focusing on the expression, function, and dynamic changes of proteins (Fahmy 2024; Gomes 2023). Mass spectrometry (MS), as the core tool in proteomics research, identifies and quantifies proteins by mass-to-charge ratio (m/z) and fragmentation patterns (Peliciari-Garcia et al. 2024). The application of MS in proteomics is primarily divided into targeted and untargeted approaches (Sobsey et al. 2020). Untargeted (discovery) proteomics comprehensively analyzes proteins without predefined targets to identify novel disease-related markers, while targeted proteomics validates preselected proteins identified in discovery studies. Through the analysis of different samples, these approaches provide unique perspectives for showing the molecular mechanisms of DR.

Vitreous fluid is one of the key samples used to study DR, as it is in direct contact with the retina and can reflect changes in the retinal microenvironment. Untargeted analysis has identified elevated inflammatory (interleukin-6 (IL-6), IL-8, tumor necrosis factor-alpha (TNF-α)) and complement proteins (C3a/C3, C5a/C5, CFB) in DR patients, which may cause retinal cell damage, accelerating disease progression (Mandava et al. 2020; Ulhaq et al. 2023; Wu et al. 2020). Future studies should clarify whether complement inhibitors can alleviate DR using animal models with conditional complement gene knockout, as current evidence is limited to observational data. The abnormal expression of VEGF in the vitreous fluid of DR patients has also received widespread attention. Significantly increased VEGF levels not only promote the formation of new blood vessels, but also may lead to vascular leakage and retinal edema (Yenihayat et al. 2019; Youngblood et al. 2019). However, approximately 30% of patients do not respond well to anti-VEGF injections (Zhang et al. 2022). The CD47/SIRPα pathway and metalloproteinase-1 (TIMP1) are implicated in anti-VEGF resistance (Feng et al. 2024), while retinal binding protein 3 (RBP3) exhibits protective effects by inhibiting VEGF receptor phosphorylation (Yokomizo et al. 2019). However, the synergistic effect between TIMP1 and VEGF remains to be investigated. The stability of RBP3 in long-term stored samples remains to be evaluated, and feasible administration routes for its therapeutic application need to be explored in future research. These findings provide important insights for developing novel anti-VEGF therapies.

Changes in the expression of ECM-related proteins in vitreous fluid also provide important clues for understanding the tissue remodeling mechanisms in DR. Upregulation of fibrinogen (fibrinogen γ-chain and β-chain) suggests that ECM remodeling may play a crucial role in the vascular abnormalities and tissue structural changes observed in DR (Sen et al. 2023). Additionally, Shahulhameed et al. (2020) reported elevated MMP9 levels in the vitreous of patients with PDR, whereas Valdivia et al. (2023) observed no significant change in MMP9 but noted an upregulation of MMP13. This discrepancy may arise from differences in sample processing protocols, such as storage temperatures and processing durations. Furthermore, upstream regulatory analysis indicated that nuclear receptors such as Peroxisome proliferator-activated receptor-α (PPAR-α), retinoid X receptor (RXR), and liver X receptor (LXR) may act as key pathological drivers in DR (Li et al. 2021). Moreover, comparing DR transgenic pig models, 266 differentially expressed proteins were identified, particularly ITGB1, COX2, and GRIFIN, which significantly increased. MYC and mTORC1 signaling pathways were also found to be closely associated with the pathological process of DR (Degroote et al. 2024). However, the density and permeability of retinal blood vessels in pigs differ from those in humans, these findings require direct validation in human DR vitreous samples.

Aqueous humor analysis provides complementary markers for DR stages. The protein composition of aqueous humor reflects pathological changes in both the anterior and posterior segments of the eye. Research has shown significant increases in inflammation factors such as IL-23 and IL-17 (Zhang et al. 2020). The concentration of immune-related protein Cystatin C (CST3) in aqueous humor is negatively correlated with the severity of DR and central retinal thickness. The activation of LXR/RXR and high expression of VEGF are also closely related to neovascularization in DR (Han et al. 2022). Since the collection of aqueous humor involves an invasive

paracentesis procedure, it is not an ideal approach for the regular screening of patients with early DR. In addition, other angiogenesis-related proteins such as placental growth factor (PlGF) (Podkowinski et al. 2020) and angiopoietin-1 (Ang-1) (Mccarthy et al. 2019) have been found to be associated with DR. Their synergistic effect may further exacerbate vascular abnormalities in the retina. Abnormal expression of metabolic-related proteins in plasma has also attracted attention. For example, the abnormal expression of apolipoprotein C2 (APOC2) (Li et al. 2019) and apolipoprotein H (APOH) (Chen et al. 2022) suggests a potential role of lipid metabolism disorders in DR.

Serum and plasma, as easily accessible sample types, provide important clues for the early diagnosis and monitoring of DR. Quantitative proteomic analysis has found that elevated TNF Alpha Induced Protein 8 (TNFAIP8) in small extracellular vesicles (sEVs), upregulated fibrinogen-α subunit (FIBA) and downregulated 1-methylhistidine (1-MH) in exosomes, all indicating an association between endothelial dysfunction and DR (Vähätupa et al. 2023; Yang, Liu & Liu 2022). The detection of sEVs and exosomal proteins requires specialized equipment and a high level of technical expertise, which may limit their widespread clinical application. Abnormal expression of metabolic-related proteins in plasma is also noteworthy. The decrease of apolipoprotein A2 and HDL-4 subclasses is closely related to microvascular complications of the disease and can be used as race-specific diabetic biomarkers (Yuan et al. 2023).

Tear, as a non-invasive sample type, has been receiving increasing attention in DR research. Studies have found that most proteins in the tear fluid of DR patients are related to oxidative stress and sEVs. High expression of IL-2/-5/-18 and TNF suggests that local inflammation plays a role in DR (Amorim et al. 2022). Proteomics also plays a key role in guiding clinical treatment decisions. A study established a blood-based proteome atlas for diabetic retinopathy and correlated it with dynamic retinal changes, offering a potent diagnostic adjunct that enhances the clinical utility of personalized management protocols (Yang et al. 2025). Meanwhile, protein expression in tears is highly sensitive to environmental factors and ocular surface conditions, which may introduce potential bias. Therefore, future studies should implement stringent inclusion criteria and standardized sample collection protocols to enhance the reproducibility of findings.

METABOLOMICS APPROACH TO UNDERSTAND DIABETIC RETINOPATHY

Metabolomics analyzes small molecules (<1500 Da) to reflect the real-time status of biological systems (Muthubharathi, Gowripriya & Balamurugan 2021). In DR research, metabolomics, with its dynamic nature and sensitivity to changes in metabolite levels, has become a key approach for exploring pathological mechanisms and discovering potential biomarkers. Liquid chromatography-mass spectrometry (LC-MS) and gas chromatography-mass spectrometry (GC-MS) enable high-sensitivity metabolite detection but requires complex sample pretreatment, while nuclear magnetic resonance (NMR) offers non-destructive analysis with lower sensitivity for low-abundance metabolites (Nagana Gowda & Raftery 2023; Si et al. 2024). Technologies like data-independent acquisition (DIA) and ion mobility spectrometry techniques further enhance analytical depth (Wang, Yin & Zhu 2019). Research strategies include untargeted analysis for global metabolic fingerprinting and targeted analysis for quantifying known metabolites in specific pathways (Bingol 2018; Hertzog et al. 2022).

Metabolic pathway dysregulation is central to DR pathogenesis. In amino acid metabolism, studies have shown that branched-chain amino acids (BCAAs), including leucine, isoleucine, and valine, exhibit elevated levels in plasma, cerebrospinal fluid, and retina samples from DR patients (Lin et al. 2019; Wang et al. 2022). Specifically, the disruption of BCAA metabolism may promote retinal glutamate neurotoxicity (Ola, Alhomida & LaNoue 2019) or activate the mTOR pathway (Gong et al. 2022), which could further aggravate the pathological process of DR. Collectively, the current evidence, while suggestive, is largely correlative. It remains unclear whether this is a primary driver of DR pathology or a secondary compensatory response.

Beyond individual metabolite changes, two metabolic pathways (arginine metabolism and tryptophan metabolism) are also closely associated with the pathophysiological mechanisms of DR. The arginine metabolism pathway also shows significant disruption in DR, with imbalances in metabolites such as arginine, citrulline, and glutamate γ-semialdehyde (Sumarriva et al. 2019). In tryptophan metabolism, particularly in the kynurenine pathway (KP), abnormalities are also observed in DR (Ancel et al. 2023). Disturbances in tryptophan metabolism could affect gut homeostasis, thereby accelerating the progression of DR (Guo et al. 2022). However, no studies have confirmed that gut-derived KP metabolites can directly cross the blood-retinal barrier to translocate into the retina. Furthermore, the specific microbial taxa that regulate KP metabolism in diabetic retinopathy (DR) remain unclear. Imbalances in tryptophan metabolism may also worsen retinal degenerative changes by influencing the blood-brain barrier and immune responses (Platten et al. 2019).

Lipid and energy metabolism disturbances are pivotal in DR pathology. Significant changes in serum and retinal triglycerides, saturated/unsaturated fatty acids, and sphingolipids indicate profound metabolic dysregulation (Fernandes Silva et al. 2024; Fort et al. 2021). Specifically, reduced acylcarnitine suggests impaired fatty acid β-oxidation and mitochondrial dysfunction (Fort et al. 2021), while altered polyunsaturated fatty acids (PUFAs) affect retinal cell viability (Li et al. 2020) and oxidative

stress repair (Saenz De Viteri et al. 2020). In glucose metabolism, elevated glycolytic products (lactate, pyruvate) in the vitreous reflect metabolic adaptation to hypoxia (Tomita et al. 2021). Furthermore, disruptions in the tricarboxylic acid (TCA) cycle (citrate, succinate (Wang et al. 2022), fumarate (Zhu et al. 2019) levels) and oxidative phosphorylation (OXPHOS) may impair photoreceptor function (Todorova et al. 2023). Notably, impaired antioxidant defense, as characterized by abnormal glutathione levels, can be partially relieved by proline supplementation, providing a potential therapeutic approach (Yam et al. 2019; Yousri et al. 2022). These metabolic alterations are not only pathophysiologically meaningful but also therapeutically relevant.

Emerging metabolomic studies have increasingly focused on identifying diagnostically useful metabolite signatures rather than individual metabolites. In plasma, amino acid metabolites such as pseudouridine, leucylleucine, and N-acetyltryptophan in plasma (Sun et al. 2021), and lipid metabolites like linoleic acid, nicotinuric acid, ornithine, phenylacetylglutamine (Zuo et al. 2021), diacylphosphatidylcholine, and lysophosphatidylcholine (Shen et al. 2022) are considered potential biomarkers for DR. A key advance in this field is that several metabolite combinations have demonstrated superior diagnostic performance over traditional clinical markers. Notably, metabolite combinations (12-HETE and 2-piperidone) demonstrate higher diagnostic accuracy than hemoglobin A1c (HbA1c) (Xuan et al. 2020). Moreover, metabolites such as lactate, pyruvate, and acetate have also been suggested as potential blood biomarkers (Hou, Wang & Pan 2021). Ocular fluid profiling has identified some potential metabolic factors, including fatty acid desaturase 2 (FADS2) (Fang et al. 2023), isocitrate, and fructose-6-phosphate (Wang et al. 2020). However, as these intraocular findings require invasive sample collection, their clinical utility as routine biomarkers remains limited. The combined analysis of azelaic acid and guanosine in tear fluid showed excellent predictive performance for non-invasive DR diagnosis (Wen et al. 2023). This highlights a promising shift towards accessible biofluids.

In urine, decreases in citric acid, ethanolamine, formic acid, and hypoxanthine levels are positively correlated with DR risk, while lower levels of 3-hydroxyisobutyrate are associated with vision loss (Quek et al. 2021). Emerging research also links gut microbial dysbiosis to DR pathology. DR patients exhibit reduced microbial diversity, with depletion of Coriobacteriaceae, Veillonellaceae, and Akkermansia, while Burkholderiaceae and other taxa increased (Ye et al. 2021; Zhou et al. 2021). These findings support microbiota-targeted interventions (He et al. 2023), such as Qilian Tablets (CQLT), which may slow DR progression by improving gut composition and glycolysis (Jia et al. 2024). Notably, consistent signatures of alanine, lactate, and glutamine across tissues reinforce these core metabolic disturbances as high-confidence therapeutic targets (Du et al. 2022).

OMICS INTEGRATION IN DIABETIC RETINOPATHY

In this review, we provide a deeper analysis of the multilevel molecular mechanisms of DR by integrating data from different 'omics'. Following the integrative framework described in previous research (Rossi et al. 2022), the Omicsnet 2.0 platform (https://www.omicsnet.ca/) was used to combine proteomics and metabolomics data, showing their interactions and functional connections. Our focus is on DR target molecules identified through proteomics and metabolomics approaches, offering a systematic multi-layered analysis of molecular changes to more comprehensively describe the pathological features of this complex disease.

First, the proteins mentioned in this study were identified and standardized using the STRING database (https://cn.string-db.org/) to obtain their Ensembl protein IDs (Supplementary Table 1). Similarly, the metabolites mentioned in this study were converted into KEGG IDs using the db2db tool in the bioDBnet database (https://biodbnet-abcc.ncifcrf.gov/db/db2db.php) (Supplementary Table 2). These identifiers were then uploaded to the OmicsNet 2.0 platform with the organism set to *Homo sapiens* (human). As of January 2025, this resulted in 28 valid protein inputs and 25 valid metabolite inputs. By matching these candidate molecules with the IntAct and KEGG databases using default settings, a protein-metabolite interaction network was constructed (Figure 1). This network is composed of seven metabolite-centered subnetworks. Notably, L-glutamate, pyruvate, and ethylsuccinic acid serve as key hubs connecting multiple protein nodes, which indicates their central importance in the network.

Finally, an integrated KEGG pathway enrichment analysis was performed. Based on the selection criteria of $P < 0.05$ and a count > 2, thirty-four significantly enriched pathways were identified (Supplementary Table 3). The top 10 most significant biological pathways were visualized using a bar chart (Figure 2). Among these pathways, the 'alanine, aspartate, and glutamate metabolism' pathway showed the highest gene enrichment and statistical significance, suggesting its central role in the studied biological processes. The 'Citrate cycle (TCA cycle)' pathway also exhibited significant results, which are closely related to its critical role in energy metabolism and biosynthesis. Notably, the three core metabolites in the PPI network are involved in the 'Glyoxylate and dicarboxylate metabolism' pathway, which aligns with related studies (Han et al. 2023). Furthermore, these metabolites were significantly enriched in the 'Butanoate metabolism' pathway, although their potential relationship with DR has not yet been verified. It must be acknowledged that the database-driven network inference in the current omics integration strategy has several inherent limitations. First, the identified protein-metabolite interactions are derived from existing knowledge in curated databases such as IntAct and KEGG, which means that well-studied molecules tend to become central hubs, while novel or poorly characterized

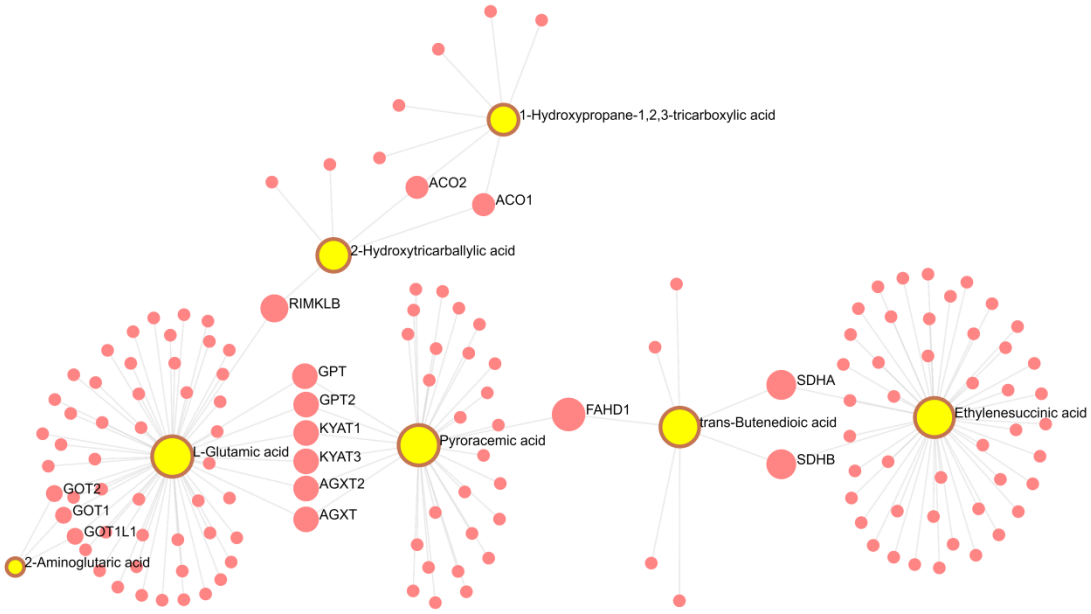

FIGURE 1. Interaction network of key metabolites and potential targets in DR. Yellow nodes represent metabolites, and red nodes represent proteins. The lines between nodes indicate molecular interactions. The size of each node is based on the degree value, where a higher degree value indicates greater connectivity and suggests a more important role in the metabolic network

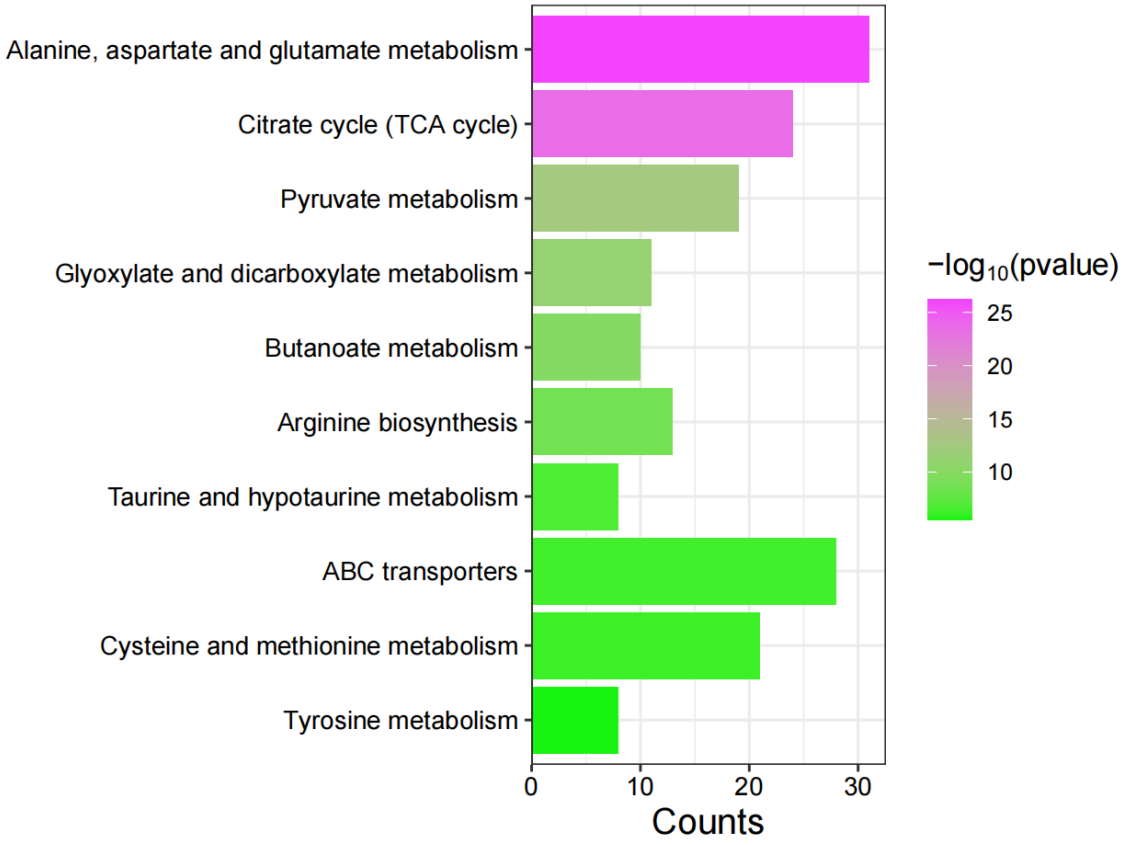

FIGURE 2. Enrichment analysis of metabolic pathways from integrated omics. The x-axis represents the number of metabolites and proteins enriched in each metabolic pathway, and the y-axis lists the names of the pathways involved in the analysis. The color gradient of the bars indicates the statistical significance, represented by -log10 (p value), with colors transitioning from green (lower significance) to pink (higher significance)

regulatory elements may be underrepresented. Second, these databases may lack tissue-specific context. Lastly, the network represents a static snapshot of molecular connections and does not capture the dynamic temporal progression of DR pathology. Despite these constraints, this integrative approach is essential for transforming fragmented data into a systemic molecular map.

CONCLUSION

As we delve deeper into the field of 'omics', it becomes clear that a single omics discipline can only provide a partial perspective on the complexity of a disease. By integrating genomics, proteomics and metabolomics, we have gained a more comprehensive perspective. It also promotes the discovery of new therapeutic targets and biomarkers. In this review, we fully utilize the advantages of multi-omics to show the complex molecular mechanisms of DR, not only going beyond simple correlation studies but also deeply exploring the potential changes that may lead to the disease. Combining multi-omics data has proven useful in understanding the many factors behind DR. Unlike diseases caused by a single factor, DR is influenced by the interaction of genetic, protein, and metabolic factors. Our analysis shows how these omics datasets interact and points out important pathways and factors that affect DR. This approach connects the molecular causes of DR to its clinical symptoms, offering a clearer understanding of the disease.

DR is a complex disease with many biological processes working together. While each omics method provides useful insights into different parts of the disease, combining them helps capture the full picture and turn these findings into practical clinical knowledge. This approach is important for understanding the large amounts of data from multi-omics research, especially in complex diseases like DR. Although there are challenges like data complexity and the lack of standardized methods, multi-omics gives us a powerful tool to study complex diseases. This approach improves the diagnosis, prognosis, and treatment of DR and helps us understand the complex networks of molecular interactions, ultimately leading to better patient outcomes.

ACKNOWLEDGMENTS

This work was supported by Universiti Kebangsaan Malaysia (DIP-2024-021) and Manipal University College Malaysia (MUCM-MRB/003/2024), and Universiti Kuala Lumpur Royal College of Medicine Perak. All supplementary data is available upon requests from the corresponding author.

REFERENCES

Al-Ghamdi, A.H. 2020. Clinical predictors of diabetic retinopathy progression; A systematic review. *Current Diabetes Reviews* 16(3): 242-247.

Alfonso-Muñoz, E.A., Burggraaf-Sánchez De Las Matas, R., Mataix Boronat, J., Molina Martín, J.C. & Desco, C. 2021. Role of oral antioxidant supplementation in the current management of diabetic retinopathy. *International Journal of Molecular Sciences* 22(8): 4020.

Amorim, M., Martins, B., Caramelo, F., Gonçalves, C., Trindade, G., Simão, J., Barreto, P., Marques, I., Leal, E.C., Carvalho, E., Reis, F., Ribeiro-Rodrigues, T., Girão, H., Rodrigues-Santos, P., Farinha, C., Ambrósio, A.F., Silva, R. & Fernandes, R. 2022. Putative biomarkers in tears for diabetic retinopathy diagnosis. *Frontiers in Medicine (Lausanne)* 9: 873483.

Ancel, P., Martin, J.C., Doukbi, E., Houssays, M., Gascon, P., Righini, M., Matonti, F., Svilar, L., Valmori, M., Tardivel, C., Venteclef, N., Julla, J.B., Gautier, J.F., Resseguier, N., Dutour, A. & Gaborit, B. 2023. Untargeted multiomics approach coupling lipidomics and metabolomics profiling reveals new insights in diabetic retinopathy. *International Journal of Molecular Sciences* 24(15): 12053.

Bansal, M. 2025. Advances in retina genetics: Progress, potential, and challenges. *Indian Journal of Ophthalmology* 73(1): 31-36.

Bingol, K. 2018. Recent advances in targeted and untargeted metabolomics by NMR and MS/NMR methods. *High Throughput* 7(2): 9.

Brown, D.M., Wykoff, C.C., Boyer, D., Heier, J.S., Clark, W.L., Emanuelli, A., Higgins, P.M., Singer, M., Weinreich, D.M., Yancopoulos, G.D., Berliner, A.J., Chu, K., Reed, K., Cheng, Y. & Vitti, R. 2021. Evaluation of intravitreal aflibercept for the treatment of severe nonproliferative diabetic retinopathy: Results from the PANORAMA randomized clinical trial. *JAMA Ophthalmology* 139(9): 946-955.

Bryl, A., Mrugacz, M., Falkowski, M. & Zorena, K. 2022. The effect of diet and lifestyle on the course of diabetic retinopathy-A review of the literature. *Nutrients* 14(6): 1252.

Cabrera, A.P., Monickaraj, F., Rangasamy, S., Hobbs, S., Mcguire, P. & Das, A. 2020. Do genomic factors play a role in diabetic retinopathy? *Journal of Clinical Medicine* 9(1): 216.

Che, X.D., Wei, Z.L., Gong, W., Qin, L., Liu, S., Jin, Y.H. & Wang, H.Y. 2025. Clinical and genetic characteristics of young-onset diabetes with concurrent mitochondrial m.3243A>G and CEL gene mutations: A case report. *World Journal of Diabetes* 16(12): 113238.

Chen, H., Wang, T., Wang, E., Li, N. & Min, H. 2022. Pursuing diabetic nephropathy through aqueous humor proteomics analysis. *Oxid. Med. Cell Longev.* 2022: 5945828.

Chen, X., Wan, Z., Geng, T., Zhu, K., Li, R., Lu, Q., Lin, X., Liu, S., Chen, L., Guo, Y., Shan, Z., Liu, L., Pan, A., Manson, J.E. & Liu, G. 2022. Vitamin D status, vitamin D receptor polymorphisms, and risk of microvascular complications among individuals with type 2 diabetes: A prospective study. *Diabetes Care* 46(2): 270-277.

Degroote, R.L., Schmalen, A., Renner, S., Wolf, E., Hauck, S.M. & Deeg, C.A. 2024. Diabetic retinopathy from the vitreous proteome perspective: The INS^{C94Y} transgenic pig model study. *Proteomics* 24(20): 2300591.

Du, X., Yang, L., Kong, L., Sun, Y., Shen, K., Cai, Y., Sun, H., Zhang, B., Guo, S. & Zhang, A. & Wang, X. 2022. Metabolomics of various samples advancing biomarker discovery and pathogenesis elucidation for diabetic retinopathy. *Frontiers in Endocrinology (Lausanne)* 13: 1037164.

Fahmy, R. 2024. Linkage of diabetic retinopathy with blood antioxidants and gut microbiota in type two diabetes mellitus saudi patients. *Journal of Ophthalmology and Advance Research* 14(1): 25-30.

Fang, J., Wang, H., Niu, T., Shi, X., Xing, X., Qu, Y., Liu, Y., Liu, X., Xiao, Y., Dou, T., Shen, Y. & Liu, K. 2023. Integration of vitreous lipidomics and metabolomics for comprehensive understanding of the pathogenesis of proliferative diabetic retinopathy. *Journal of Proteome Research* 22(7): 2293-2306.

Feng, T-T., Gao, X., Liang, A-R., Zhao, B-W., He, G-H. & Chen, S. 2024. Proteomic study of vitreous in proliferative diabetic retinopathy patients after treatment with aflibercept: A quantitative analysis based on 4D label-free technique. *International Journal of Ophtalmology* 17(4): 676-685.

Fernandes Silva, L., Hokkanen, J., Vangipurapu, J., Oravilahti, A. & Laakso, M. 2024. Metabolites as risk factors for diabetic retinopathy in patients with type 2 diabetes: A 12-year follow-up study. *The Journal of Clinical Endorinology & Metabolism* 109(1): 100-106.

Fort, P.E., Rajendiran, T.M., Soni, T., Byun, J., Shan, Y., Looker, H.C., Nelson, R.G., Kretzler, M., Michailidis, G., Roger, J.E., Gardner, T.W., Abcouwer, S.F., Pennathur, S. & Afshinnia, F. 2021. Diminished retinal complex lipid synthesis and impaired fatty acid β-oxidation associated with human diabetic retinopathy. *JCI Insight* 6(19): e152109.

Fung, T.H., Patel, B., Wilmot, E.G. & Amoaku, W.M. 2022. Diabetic retinopathy for the non-ophthalmologist. *Clin. Med. (Lond)* 22(2): 112-116.

GDB. 2021. Trends in prevalence of blindness and distance and near vision impairment over 30 years: An analysis for the Global Burden of Disease Study. *The Lancet Global Health* 9(2): e130-e143.

Gomes, K.B. 2023. Proteomic biomarkers: What they are and how type 2 diabetes mellitus has similarities with other diseases. In *Biomarkers in Diabetes,* edited by Patel, V.B. & Preedy, V.R. Cham: Springer International Publishing. pp. 311-328.

Gong, Q., Zhang, R., Wei, F., Fang, J., Zhang, J., Sun, J., Sun, Q. & Wang, H. 2022. SGLT2 inhibitor-empagliflozin treatment ameliorates diabetic retinopathy manifestations and exerts protective effects associated with augmenting branched chain amino acids catabolism and transportation in db/db mice. *Biomedicine & Pharmacotherapy* 152: 113222.

Guo, C., Jiang, D., Xu, Y., Peng, F., Zhao, S., Li, H., Jin, D., Xu, X., Xia, Z., Che, M., Lai, M., Huang, R., Wang, H., Zheng, C. & Mao, G. 2022. High-coverage serum metabolomics reveals metabolic pathway dysregulation in diabetic retinopathy: A propensity score-matched study. *Front. Mol. Biosci.* 9: 822647.

Gverović Antunica, A., Znaor, L., Ivanković, M., Puzović, V., Marković, I. & Kaštelan, S. 2023. Vitamin D and diabetic retinopathy. *International Journal of Molecular Sciences* 24(15): 12014.

Han, R., Gong, R., Liu, W. & Xu, G. 2022. Proteome changes associated with the VEGFR pathway and immune system in diabetic macular edema patients at different diabetic retinopathy stages. *Current Eye Research* 47(7): 1050-1060.

Han, X., Zhang, L., Kong, L., Tong, M., Shi, Z., Li, X.M., Zhang, T., Jiang, Q. & Biao, Y. 2023. Comprehensive metabolic profiling of diabetic retinopathy. *Experimantal Eye Research* 233: 109538.

He, S., Sun, L., Chen, J. & Ouyang, Y. 2023. Recent advances and perspectives in relation to the metabolomics-based study of diabetic retinopathy. *Metabolites* 13(9): 1007.

Hertzog, A., Selvanathan, A., Devanapalli, B., Ho, G., Bhattacharya, K. & Tolun, A.A. 2022. A narrative review of metabolomics in the era of "-omics": integration into clinical practice for inborn errors of metabolism. *Translational Pediatrics* 11(10): 1704-1716.

Hou, X-W., Wang, Y. & Pan, C-W. 2021. Metabolomics in diabetic retinopathy: A systematic review. *Investigative Ophtalmology & Visual Science* 62(10): 4.

Husain, M., Birkenfeld, A.L., Donsmark, M., Dungan, K., Eliaschewitz, F.G., Franco, D.R., Jeppesen, O.K., Lingvay, I., Mosenzon, O., Pedersen, S.D., Tack, C.J., Thomsen, M., Vilsbøll, T., Warren, M.L. & Bain, S.C. 2019. Oral semaglutide and cardiovascular outcomes in patients with type 2 diabetes. *The New England Journal of Medicine* 381(9): 841-851.

Imamura, M., Takahashi, A., Matsunami, M., Horikoshi, M., Iwata, M., Araki, S.I., Toyoda, M., Susarla, G., Ahn, J., Park, K.H., Kong, J., Moon, S., Sobrin, L., Yamauchi, T., Tobe, K., Maegawa, H., Kadowaki, T. & Maeda, S. 2021. Genome-wide association studies identify two novel loci conferring susceptibility to diabetic retinopathy in Japanese patients with type 2 diabetes. *Human Molecular Genetics* 30(8): 716-726.

Jagadesh, B.N., Karthik, M.G., Siri, D., Shareef, S.K.K., Mantena, S.V. & Vatambeti, R. 2023. Segmentation using the IC2T Model and classification of diabetic retinopathy using the rock hyrax swarm-based coordination attention mechanism. *IEEE Access* 11: 124441-124458.

Jia, J., Liu, B., Wang, X., Ji, F., Wen, F., Xu, H. & Ding, T. 2024. Metabolomics combined with intestinal microbiota reveals the mechanism of compound Qilian tablets against diabetic retinopathy. *Frontiers in Microbiology* 15: 1453436.

Kowluru, R.A., Radhakrishnan, R. & Mohammad, G. 2021. Regulation of Rac1 transcription by histone and DNA methylation in diabetic retinopathy. *Scientific Reports* 11(1): 14097.

Li, J-S., Wang, T., Zuo, J-J., Guo, C-N., Peng, F., Zhao, S-Z., Li, H-H., Hou, X-Q., Lan, Y., Wei, Y-P., Zheng, C. & Mao, G-Y. 2020. Association of n-6 PUFAs with the risk of diabetic retinopathy in diabetic patients. *Endocrine Connections* 9(12): 1191-1201.

Li, S., Chen, L. & Fu, Y. 2023. Nanotechnology-based ocular drug delivery systems: Recent advances and future prospects. *Journal of Nanobiotechnology* 21(1): 232.

Li, S., Jin, E., Shi, X., Cai, Y., Zhang, H. & Zhao, M. 2021. Proteomics of vitreous humor reveals PPARA, RXR, and LXR are possible upstream regulators of proliferative diabetic retinopathy. *Frontiers in Medicine* 8: 724695.

Li, Z-C., Chen, J-B., Zhang, H-B., Mei, W-Q., Mao, N., Ouyang, J., Qian, J-L. & Liu, Y. 2019. Plasma proteomics research in patients with diabetic retinopathy. *Jie Fang Jun Yi Xue Za Zhi* 44(1): 42-50.

Lim, A. 2024. Diabetic retinopathy: Screening, prevention, and treatment. *Cleveland Clinic Journal of Medicine* 91(12): 723.

Lin, A.L., Roman, R.J., Regan, K.A., Bolch, C.A., Chen, C.J. & Iyer, S.S.R. 2020. Eicosanoid profiles in the vitreous humor of patients with proliferative diabetic retinopathy. *International Journal of Molecular Sciences* 21(20): 7451.

Lin, H-T., Cheng, M-L., Lo, C-J., Lin, G., Lin, S-F., Yeh, J-T., Ho, H-Y., Lin, J-R. & Liu, F.C. 2019. ^{1}H nuclear magnetic resonance (NMR)-based cerebrospinal fluid and plasma metabolomic analysis in type 2 diabetic patients and risk prediction for diabetic microangiopathy. *Journal of Clinical Medicine* 8(6): 874.

Liu, C., Chen, G., Bentley, A.R., Doumatey, A., Zhou, J., Adeyemo, A., Yang, J. & Rotimi, C. 2019. Genome-wide association study for proliferative diabetic retinopathy in Africans. *npj Genomic Medicine* 4: 20.

Mandava, N., Tirado-Gonzalez, V., Geiger, M.D., Patnaik, J.L., Frazer-Abel, A., Lynch, A.M., Mandava, N., Palestine, A.G., Holers, V.M., Wagner, B.D., Sanchez-Santos, I., Meizner, D., Quiroz-Mercado, H. & Smith, J.M. 2020. Complement activation in the vitreous of patients with proliferative diabetic retinopathy. *Investigative Ophtalmology & Visual Science* 61(11): 39.

Mason, R.H., Minaker, S.A., Lahaie Luna, G., Bapat, P., Farahvash, A., Garg, A., Bhambra, N. & Muni, R.H. 2022. Changes in aqueous and vitreous inflammatory cytokine levels in proliferative diabetic retinopathy: A systematic review and meta-analysis. *Eye (Lond)*: 60(1): e100-e116.

Mccarthy, C.P., Shrestha, S., Ibrahim, N., van Kimmenade, R.R.J., Gaggin, H.K., Mukai, R., Magaret, C., Barnes, G., Rhyne, R., Garasic, J.M. & Januzzi, J.L. 2019. Performance of a clinical/proteomic panel to predict obstructive peripheral artery disease in patients with and without diabetes mellitus. *Open Heart* 6(1): e000955.

Miller, J.W., Adamis, A.P., Shima, D.T., D'Amore, P.A., Moulton, R.S., O'Reilly, M.S., Folkman, J., Dvorak, H.F., Brown, L.F., Berse, B., Yeo, T-K. & Yeo, K-T. 1994. Vascular endothelial growth factor/vascular permeability factor is temporally and spatially correlated with ocular angiogenesis in a primate model. *The American Journal of Pathology* 145(3): 574-584.

Mihoubi, E., Amroun, H., Bouldjennet, F., Azzouz, M., Touil-Boukoffa, C., Raache, R. & Attal, N. 2022. The 869C>T and 915G>C polymorphisms of *TGF-β1* and type 1 diabetic retinopathy in the Algerian Population. *Journal Francais D'ophtalmologie* 45(8): 908-914.

Mokhtar, E.R., Mahmoud, D.A., Ebrahim, G.E., Al Anany, M.G., Seliem, N. & Hassan, M.M. 2023. Serum metabolomic profiles and semaphorin-3A as biomarkers of diabetic retinopathy progression. *The Egyptian Journal of Immunology* 30(2): 83-98.

Muthubharathi, B.C., Gowripriya, T. & Balamurugan, K. 2021. Metabolomics: Small molecules that matter more. *Molecular Omics* 17(2): 210-229.

Nagana Gowda, G.A. & Raftery, D. 2023. Quantitative NMR methods in metabolomics. *Handbook of Experimental Pharmacology* 277: 143-164.

Ola, M.S., Alhomida, A.S. & LaNoue, K.F. 2019. Gabapentin attenuates oxidative stress and apoptosis in the diabetic rat retina. *Neurotoxicity Research* 36(1): 81-90.

Ouyang, F., Yuan, P., Ju, Y., Chen, W., Peng, Z. & Xu, H. 2024. Alzheimer's disease as a causal risk factor for diabetic retinopathy: A mendelian randomization study. *Front Endocrinol (Lausanne)* 15: 1340608.

Peliciari-Garcia, R.A., De Barros, C.F., Secio-Silva, A., De Barros Peruchetti, D., Romano, R.M. & Bargi-Souza, P. 2024. Multi-omics investigations in endocrine systems and their clinical implications. *Advances in Experimental Medicine and Biology* 1443: 187-209.

Plate, K.H., Breier, G., Weich, H.A. & Risau, W. 1992. Vascular endothelial growth factor is a potential tumour angiogenesis factor in human gliomas *in vivo*. *Nature* 359(6398): 845-848.

Platten, M., Nollen, E.A.A., Röhrig, U.F., Fallarino, F. & Opitz, C.A. 2019. Tryptophan metabolism as a common therapeutic target in cancer, neurodegeneration and beyond. *Nature Reviews Drug Discovery* 18(5): 379-401.

Podkowinski, D., Orlowski-Wimmer, E., Zlabinger, G., Pollreisz, A., Mursch-Edlmayr, A.S., Mariacher, S., Ring, M. & Bolz, M. 2020. Aqueous humour cytokine changes during a loading phase of intravitreal ranibizumab or dexamethasone implant in diabetic macular oedema. *Acta Ophtalmologica* 98(4): e407-e415.

Qayyum, S., Afzal, M. & Naveed, A.K. 2021a. Association analysis of 374T/A (rs1800624) receptor for advanced glycation end-products (RAGE) gene polymorphism with diabetic retinopathy in Pakistani patients. *Pakistan Journal of Medical Sciences* 37(3): 733-739.

Qayyum, S., Afzal, M. & Naveed, A.K. 2021b. Association analysis of -429T/C receptor for advanced glycation end products (RAGE) gene polymorphism with type 2 diabetic retinopathy and serum soluble RAGE levels in Pakistani patients. *Journal of the Pakistan Medical Association* 71(4): 1175-1180.

Quek, D.Q.Y., He, F., Sultana, R., Banu, R., Chee, M.L., Nusinovici, S., Thakur, S., Qian, C., Cheng, C-Y., Wong, T.Y. & Sabanayagam, C. 2021. Novel serum and urinary metabolites associated with diabetic retinopathy in three Asian cohorts. *Metabolites* 11(9): 614.

Rossi, C., Cicalini, I., Cufaro, M.C., Consalvo, A., Upadhyaya, P., Sala, G., Antonucci, I., Del Boccio, P., Stuppia, L. & De Laurenzi, V. 2022. Breast cancer in the era of integrating "Omics" approaches. *Oncogenesis* 11(1): 17.

Rossino, M.G., Dal Monte, M. & Casini, G. 2019. Relationships between neurodegeneration and vascular damage in diabetic retinopathy. *Frontiers in Neuroscience* 13: 1172.

Saenz De Viteri, M., Hernandez, M., Bilbao-Malavé, V., Fernandez-Robredo, P., González-Zamora, J., Garcia-Garcia, L., Ispizua, N., Recalde, S. & Garcia-Layana, A. 2020. A higher proportion of eicosapentaenoic acid (EPA) when combined with docosahexaenoic acid (DHA) in omega-3 dietary supplements provides higher antioxidant effects in human retinal cells. *Antioxidants* 9(9): 828.

Sahajpal, N., Kowluru, A. & Kowluru, R.A. 2019. The regulatory role of Rac1, a small molecular weight GTPase, in the development of diabetic retinopathy. *Journal of Clinical Medicine* 8(7): 965.

Sen, S., Udaya, P., Maheshwari, J.J., Kohli, P., Parida, H., Kannan, N.B., Ramasamy, K. & Dharmalingam, K.J. 2023. Comparative proteomics of proliferative diabetic retinopathy in people with type 2 diabetes highlights the role of inflammation, visual transduction, and extracellular matrix pathways. *Indian Journal of Ophtalmology* 71(8): 3069-3079.

Shahulhameed, S., Vishwakarma, S., Chhablani, J., Tyagi, M., Pappuru, R.R., Jakati, S., Chakrabarti, S. & Kaur, I. 2020. A systematic investigation on complement pathway activation in diabetic retinopathy. *Frontiers in Immunology* 11: 154.

Shen, Y., Wang, H., Xu, X., Chen, C., Zhu, S., Cheng, L., Fang, J., Liu, K. & Xu, X. 2022. Metabolomics study of treatment response to conbercept of patients with neovascular age-related macular degeneration and polypoidal choroidal vasculopathy. *Frontiers in Pharmacology* 13: 991879.

Si, Y., Huang, H., Pan, J., Luo, X., Zhang, J., Guo, Y. & Liu, D. 2024. Identification of potential biomarkers and pathways involved in high-altitude pulmonary edema using GC-MS and LC-MS metabolomic methods. *Scientific Reports* 14(1): 30978.

Skol, A.D., Jung, S.C., Sokovic, A.M., Chen, S., Fazal, S., Sosina, O., Borkar, P.P., Lin, A., Sverdlov, M., Cao, D., Swaroop, A., Bebu, I., Stranger, B.E. & Grassi, M.A. 2020. Integration of genomics and transcriptomics predicts diabetic retinopathy susceptibility genes. *Elife* 9: e59980.

Sobrin, L., Susarla, G., Stanwyck, L., Rouhana, J.M., Li, A., Pollack, S., Igo Jr., R.P., Jensen, R.A., Li, X., Ng, M.C.Y., Smith, A.V., Kuo, J.Z., Taylor, K.D., Freedman, B.I., Bowden, D.W., Penman, A., Chen, C.J., Craig, J.E., Adler, S.G., Chew, E.Y., Cotch, M.F., Yaspan, B., Mitchell, P., Wang, J.J., Klein, B.E.K., Wong, T.Y., Rotter, J.I., Burdon, K.P., Iyengar, S.K. & Segrè, A.V. 2022. Gene set enrichment analsyes identify pathways involved in genetic risk for diabetic retinopathy. *American Journal of Ophthalmology* 233: 111-123.

Sobsey, C.A., Ibrahim, S., Richard, V.R., Gaspar, V., Mitsa, G., Lacasse, V., Zahedi, R.P., Batist, G. & Borchers, C.H. 2020. Targeted and untargeted proteomics approaches in biomarker development. *Proteomics* 20(9): e1900029.

Stone, J., Itin, A., Alon, T., Pe'er, J., Gnessin, H., Chan-Ling, T. & Keshet, E. 1995. Development of retinal vasculature is mediated by hypoxia-induced vascular endothelial growth factor (VEGF) expression by neuroglia. *Journal of Neuroscience* 15(7 Pt 1): 4738-4747.

Suciu, C.I., Suciu, V.I., Cuţaş, A. & Nicoară, S.D. 2022. Interleaved optical coherence tomography: Clinical and laboratory biomarkers in patients with diabetic macular edema. *Journal of Personalized Medicine* 12(5): 765.

Sumarriva, K., Uppal, K., Ma, C., Herren, D.J., Wang, Y., Chocron, I.M., Warden, C., Mitchell, S.L., Goodwin Burgess, L., Goodale, M.P., Osborn, M.P., Ferreira, A.J., Law, J.C., Cherney, E.F., Jones, D.P. & Brantley Jr., M.A. 2019. Arginine and carnitine metabolites are altered in diabetic retinopathy. *Investigative Ophtalmology & Visual Science* 60(8): 3119-3126.

Sun, Y., Zou, H., Li, X., Xu, S. & Liu, C. 2021. Plasma metabolomics reveals metabolic profiling for diabetic retinopathy and disease progression. *Front. Endocrinol. (Lausanne)* 12: 757088.

Tan, T.E. & Wong, T.Y. 2022. Diabetic retinopathy: Looking forward to 2030. *Frontiers in Endocrinology* 13: 1077669.

Tanaka, S., Akagawa, H., Hase, M. & Iwasaki, N. 2025. Two novel pathogenic PDX1 variants in two Japanese patients with maturity-onset diabetes of the young. *Human Genome Variation* 12(1): 8.

Todorova, V., Stauffacher, M.F., Ravotto, L., Nötzli, S., Karademir, D., Ebner, L.J., Imsand, C., Merolla, L., Hauck, S.M. & Samardzija, M.J. 2023. Deficits in mitochondrial TCA cycle and OXPHOS precede rod photoreceptor degeneration during chronic HIF activation. *Molecular Neurodegeneration* 18(1): 15.

Tomita, Y., Cagnone, G., Fu, Z., Cakir, B., Kotoda, Y., Asakage, M., Wakabayashi, Y., Hellström, A., Joyal, J-S., Talukdar, S., Smith, L.E.H. & Usu, Y. 2021. Vitreous metabolomics profiling of proliferative diabetic retinopathy. *Diabetologia* 64: 70-82.

Ulhaq, Z.S., Hasan, Y.T.N., Rosyidin, N.K., Istifiani, L.A., Pamungkas, S.A. & Soraya, G.V. 2023. A systematic proteomic profiling and pathway analysis of protein biomarkers in diabetic retinopathy with subsequent validation of the IL-6 upstream regulator. *Journal of Diabetes & Metabolic Disorders* 22(1): 801-815.

Vähätupa, M., Nättinen, J., Aapola, U., Uusitalo-Järvinen, H., Uusitalo, H. & Järvinen, T.A.H. 2023. Proteomics analysis of R-Ras deficiency in oxygen induced retinopathy. *International Journal of Molecular Sciences* 24(9): 7914.

Valdivia, A.O., He, Y., Ren, X., Wen, D., Dong, L., Nazari, H. & Li, X. 2023. Probable treatment targets for diabetic retinopathy based on an integrated proteomic and genomic analysis. *Translational Vision Science & Technology* 12(2): 8.

Wang, H., Fang, J., Chen, F., Sun, Q., Xu, X., Lin, S-H. & Liu, K.J. 2020. Metabolomic profile of diabetic retinopathy: A GC-TOFMS-based approach using vitreous and aqueous humor. *Acta Diabetologica* 57: 41-51.

Wang, R., Yin, Y. & Zhu, Z.J. 2019. Advancing untargeted metabolomics using data-independent acquisition mass spectrometry technology. *Analytical and Bioanalytical Chemistry* 411(19): 4349-4357.

Wang, R., Jian, Q., Hu, G., Du, R., Xu, X. & Zhang, F. 2022. Integrated metabolomics and transcriptomics reveal metabolic patterns in retina of STZ-induced diabetic retinopathy mouse model. *Metabolites* 12(12): 1245.

Wang, Z., Tang, J., Jin, E., Zhong, Y., Zhang, L., Han, X., Liu, J., Cheng, Y., Hou, J. & Shi, X. 2022. Serum untargeted metabolomics reveal potential biomarkers of progression of diabetic retinopathy in Asians. *Frontiers in Molecular Biosciences* 9: 871291.

Weber, S.R., Zhao, Y., Ma, J., Gates, C., Da Veiga Leprevost, F., Basrur, V., Nesvizhskii, A. I., Gardner, T.W. & Sundstrom, J.M. 2021. A validated analysis pipeline for mass spectrometry-based vitreous proteomics: New insights into proliferative diabetic retinopathy. *Clinical Proteomics* 18(1): 28.

Wen, X., Ng, T.K., Liu, Q., Wu, Z., Zhang, G. & Zhang, M. 2023. Azelaic acid and guanosine in tears improve discrimination of proliferative from non-proliferative diabetic retinopathy in type-2 diabetes patients: A tear metabolomics study. *Heliyon* 9(5): e16109.

Wu, F., Lamy, R., Ma, D., Laotaweerungsawat, S., Chen, Y., Zhao, T., Ma, W., Zhang, F., Psaras, C. & Stewart, J.M. 2020. Correlation of aqueous, vitreous, and plasma cytokine levels in patients with proliferative diabetic retinopathy. *Investigative Ophtalmology & Visual Science* 61(2): 26.

Wu, W., Xia, X., Tang, L., Yao, F., Xu, H. & Lei, H. 2021. Normal vitreous promotes angiogenesis via activation of Axl. *The Faseb Journal* 35(1): e21152.

Xuan, Q., Ouyang, Y., Wang, Y., Wu, L., Li, H., Luo, Y., Zhao, X., Feng, D., Qin, W., Hu, C., Zhou, L., Liu, X., Zou, H., Cai, C., Wu, J., Jia, W. & Xu, G. 2020. Multiplatform metabolomics reveals novel serum metabolite biomarkers in diabetic retinopathy subjects. *Advanced Science* 7(22): 2001714.

Yam, M., Engel, A.L., Wang, Y., Zhu, S., Hauer, A., Zhang, R., Lohner, D., Huang, J., Dinterman, M. & Zhao, C. 2019. Proline mediates metabolic communication between retinal pigment epithelial cells and the retina. *Journal of Biological Chemistry* 294(26): 10278-10289.

Yang, J., Liu, D. & Liu, Z. 2022. Integration of metabolomics and proteomics in exploring the endothelial dysfunction mechanism induced by serum exosomes from diabetic retinopathy and diabetic nephropathy patients. *Frontiers in Endocrinology* 13: 830466.

Yang, S., Xin, Z., Xiong, R., Zhu, Z., Li, H., Chen, Y., Zhong, Z., Du, L., Zhu, L.Z., Shang, X., Huang, W., Zhang, L., Chen, S., He, C., Tan, S., He, M., Congdon, N., Jonas, J.B., Tham, Y.C., Cheng, C.Y. & Wang, W. 2025. Proteome atlas for mechanistic discovery and risk prediction of diabetic retinopathy. *Nature Communications* 16(1): 9636.

Ye, P., Zhang, X., Xu, Y., Xu, J., Song, X. & Yao, K. 2021. Alterations of the gut microbiome and metabolome in patients with proliferative diabetic retinopathy. *Frontiers in Microbiology* 12: 667632.

Yenihayat, F., Özkan, B., Kasap, M., Karabaş, V.L., Güzel, N., Akpınar, G. & Pirhan, D. 2019. Vitreous IL-8 and VEGF levels in diabetic macular edema with or without subretinal fluid. *International Ophtalmology* 39: 821-828.

Yokomizo, H., Maeda, Y., Park, K., Clermont, A.C., Hernandez, S.L., Fickweiler, W., Li, Q., Wang, C-H., Paniagua, S.M. & Simao, F.J. 2019. Retinol binding protein 3 is increased in the retina of patients with diabetes resistant to diabetic retinopathy. *Science Translational Medicine* 11(499): 6627.

Youngblood, H., Robinson, R., Sharma, A. & Sharma, S. 2019. Proteomic biomarkers of retinal inflammation in diabetic retinopathy. *International Journal of Molecular Sciences* 20(19): 4755.

Yousri, N.A., Suhre, K., Yassin, E., Al-Shakaki, A., Robay, A., Elshafei, M., Chidiac, O., Hunt, S.C., Crystal, R.G. & Fakhro, K.A. 2022. Metabolic and metabo-clinical signatures of type 2 diabetes, obesity, retinopathy, and dyslipidemia. *Diabetes* 71(2): 184-205.

Yu, X. & Rong, S. 2023. Genome-wide associations and confirmatory meta-analyses in diabetic retinopathy. *Genes (Basel)* 14(3): 653.

Yuan, L., Li-Gao, R., Verhoeven, A., Van Eyk, H.J., Bizino, M.B., Rensen, P.C.N., Giera, M., Jazet, I.M., Lamb, H.J., Rabelink, T.J. & van den Berg, B.M. 2023. Altered high-density lipoprotein composition is associated with risk for complications in type 2 diabetes mellitus in South Asian descendants: A cross-sectional, case-control study on lipoprotein subclass profiling. *Diabetes, Obesity and Metabolism* 25(8): 2374-2387.

Zhang, H., Liang, L., Huang, R., Wu, P. & He, L. 2020. Comparison of inflammatory cytokines levels in the aqueous humor with diabetic retinopathy. *International Ophtalmology* 40: 2763-2769.

Zhang, J., Zhang, J., Zhang, C., Zhang, J., Gu, L., Luo, D. & Qiu, Q. 2022. Diabetic macular edema: Current understanding, molecular mechanisms and therapeutic implications. *Cells* 11(21): 3362.

Zhou, Z., Zheng, Z., Xiong, X., Chen, X., Peng, J., Yao, H., Pu, J., Chen, Q. & Zheng, M. 2021. Gut microbiota composition and fecal metabolic profiling in patients with diabetic retinopathy. *Frontiers in Cell and Developmental Biology* 9: 732204.

Zhu, X.R., Yang, F.Y., Lu, J., Zhang, H.R., Sun, R., Zhou, J.B. & Yang, J.K. 2019. Plasma metabolomic profiling of proliferative diabetic retinopathy. *Nutr. Metab. (Lond)* 16: 37.

Zuo, J., Lan, Y., Hu, H., Hou, X., Li, J., Wang, T., Zhang, H., Zhang, N., Guo, C., Peng, F., Zhao, S., Wei, Y., Jia, C., Zheng, C. & Mao, G. 2021. Metabolomics-based multidimensional network biomarkers for diabetic retinopathy identification in patients with type 2 diabetes mellitus. *BMJ Open Diabetes Research & Care* 9(1): e001443.

*Corresponding author; email: hamizahshahirah@ukm.edu.my