Heliyon 10 (2024) e37156

Contents lists available at ScienceDirect

Heliyon

journal homepage: www.cell.com/heliyon

Research article

Integrated gene-metabolite association network analysis reveals key metabolic pathways in gastric adenocarcinoma

Botao Xu [a,1], Yuying Shi [b,c,d,1], Chuang Yuan [e], Zhe Wang [f], Qitao Chen [b,c], Cheng Wang [b,c,**], Jie Chai [a,*]

[a] Department of Gastrointestinal Surgery, Shandong Cancer Hospital and Institute, Shandong First Medical University and Shandong Academy of Medical Science, Jinan, China
[b] Department of Biostatistics, School of Public Health, Cheeloo College of Medicine, Shandong University, Jinan, 250012, China
[c] National Institute of Health Data Science of China, Shandong University, Jinan, 250000, China
[d] National Science Library (Chengdu), Chinese Academy of Sciences, Chengdu, 610299, China
[e] Department of Biochemistry and Biophysics, School of Basic Medical Sciences, Peking University, Beijing, 100191, China
[f] Department of Radiation Oncology, Shandong Cancer Hospital and Institute, Shandong First Medical University and Shandong Academy of Medical Science, Jinan, Shandong, China

ARTICLE INFO

Keywords:
Gastric adenocarcinoma
Metabolomics
Biomarker
Topological analysis
Metabolic pathway

ABSTRACT

Gastric adenocarcinoma is one of the most death cause cancers worldwide. Metabolomics is an effective approach for investigating the occurrence and progression of cancer and detecting prognostic biomarkers by studying the profiles of small bioactive molecules. To fully decipher the functional roles of the disrupted metabolites that modulate the cellular mechanism of gastric cancer, integrated gene-metabolite association network methods are critical to map the associations between metabolites and genes. In this study, we constructed a knowledge-based gene-metabolite association network of gastric cancer using the dysregulated metabolites and genes between gastric cancer patients and control group. The topological pathway analysis and gene-protein-metabolite-disease association analysis revealed four key gene-metabolite pathways which include eleven metabolites associated with modulated genes. The integrated gene-metabolite association network enables mechanistic investigation and provides a comprehensive overview regarding the investigation of molecular mechanisms of gastric cancer, which facilitates the in-depth understanding of metabolic biomarker roles in gastric cancer.

1. Introduction

Gastric cancer (GSC) is the fifth most common cancer and the third leading cause of cancer-related deaths worldwide [1–3]. Gastric adenocarcinoma is the most prevalent type of GSC [1,4]. It is often detected when patients present with obvious symptoms, leading to a diagnosis at an advanced stage, which severely impacts treatment and prognosis [5,6]. Therefore, early detection using effective

* Corresponding author. Department of Gastrointestinal Surgery, Shandong Cancer Hospital and Institute, Shandong First Medical University and Shandong Academy of Medical Science, Jinan, China.
** Corresponding author. Department of Biostatistics, School of Public Health, Cheeloo College of Medicine, Shandong University, Jinan, 250012, China.
E-mail addresses: chengwangsdu@outlook.com (C. Wang), jchai@sdfmu.edu.cn (J. Chai).
[1] These authors contributed equally to this work.

https://doi.org/10.1016/j.heliyon.2024.e37156
Received 5 March 2024; Received in revised form 22 July 2024; Accepted 28 August 2024
Available online 30 August 2024

screening methods is crucial to improving diagnosis rates and reducing mortality.

Metabolomics is a comprehensive method for qualitative and quantitative profiling of all endogenous small molecules in biological samples such as tissues, urine, and plasma [7–9]. Cancer metabolomics studies focus on identifying cancer specific metabolites from tumor tissues that can serve as potential biomarkers for clinical applications [10,11]. Metabolic differences between tumor cells and surrounding cells help understand the mechanisms underlying tumor growth, invasion, and metastasis [11–13]. Research on metabolic biomarkers for GSC has grown rapidly over the decades with advanced analytical platforms. Several gastric cancer biomarkers, such as carcinoembryonic antigen (CEA), are used for diagnosis, clinical staging, assessment of treatment response and screening for recurrence after successful treatment [14–17].

The discovery of new biomarkers, such as the expression levels of various proteins and genes in body fluid samples, has created new opportunities for diagnosing and monitoring patients with GSC [18]. These findings may provide valuable targets for the early diagnosis and personalized treatment of GSC. Functional analysis based on identified metabolites is critical for understanding the molecular mechanisms of gastric cancer [19]. Gu et al. identified several important metabolites of gastric cancer through metabolomics analysis and performed metabolic pathway analysis [20]. They revealed multiple significantly disrupted metabolic pathways, including oxidative stress, choline phosphorylation, amino acid metabolism, the Krebs cycle, and glycolysis. Three metabolic pathways are consistently disrupted during GSC development and progression: taurine and hypotaurine metabolism, glutamine and glutamate metabolism, and alanine, aspartate, and glutamate metabolism [21]. These alterations may be due to abnormal energy supply for tumor cell proliferation and growth.

Metabolomics approaches have been widely studied in gastric cancer, but the deep exploration of upstream pathways and functions of gastric cancer metabolites is still relatively limited [19,22]. Specifically, most metabolomics studies focus on the enriched metabolic pathways, while some key modulated metabolites may be ignored. In this work, we conducted an integrated gene-metabolite association network approach using the differentially expressed metabolites of gastric adenocarcinoma and its paraneoplastic tissues. Hundreds of metabolic pathways were enriched using disrupted metabolites. By using topological pathway analysis and knowledge-based networks, the gene-protein-metabolite-disease interaction network was constructed to identify core regulated metabolites and genes. The identification of specific metabolic biomarkers and pathways provides potential targets for early detection and diagnosis of gastric cancer. These targets can be further explored for developing drugs that specifically disrupt cancer metabolism, potentially leading to more effective treatments. In addition, this integrated gene-metabolite association pipeline provides molecular insights into the mechanisms of gastric cancer and helps discover new potential metabolic biomarkers.

2. Methods

2.1. Dataset

Twelve metabolomics studies on GSC were included. We extracted the disrupted metabolites that showed statistical differences between GSC patients and control group. All metabolites were measured using nuclear magnetic resonance (NMR) spectroscopy or mass spectrometry (MS). The details of the twelve metabolomics studies and metabolites are listed in Table 1.

2.2. Pathway enrichment analysis

Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway enrichment analysis was applied to understand the functions and pathways related to differential metabolites. Using the KEGG Compound Database (https://www.genome.jp/kegg/compound/) and MetaboAnalyst, we performed pathway enrichment analysis based on several libraries containing approximately hundreds of metabolites [23,24]. The identified metabolites were matched to the KEGG pathway database. We used Fisher's exact test to verify the *P*-value of the calculated pathway enrichment and Holm-Bonferroni adjustment for multiple test corrections. The Benjamini-Hochberg method was employed to control the false discovery rate (FDR) and reduce false positives. To provide more information on the number of metabolites, we listed the total number of metabolites in the pathway and the matched number of metabolites. The results of the enriched pathways were ranked by *P*-values, and pathways with $P < 0.01$ were selected for topological analysis. The top 25 pathways with significant analysis were visualized using bar plots and scatter plots.

2.3. Topological analysis

The most significant metabolic pathways were identified based on *P*-value and FDR. We then used the relative-betweenness Centrality method for topological analysis through the GenomeNet Database (https://www.genome.jp/). Betweenness centrality (BC) describes the importance of a node by the number of shortest paths passing through it. In other words, all shortest paths between any two nodes in the network were calculated. Nodes with more of these paths passing through them were considered to have high betweenness. Using the KEGG Pathway Database, we manually mapped the relevant metabolites and removed unnecessary pathways. We then converted this pathway information into a directed graph where each metabolite was uniquely identified by its chemical name. We calculated BC based on the number of connections each metabolite had and further exploring their molecular and cellular functions.

Table 1
Differential metabolites in serum, urine, and tissue collected on different analytical platforms.

Author and year	Platform	Sample size	TNM stage	Grade	Sample (species)	Group	Disrupted metabolites
Jingping Gu et al. (2020) [21]	NMR	Model mice (n = 52) CON mice (n = 32)			Mice tissue	GS vs CON	lactate, glutamate, glutamine, aspartate, creatine, choline, PC, GPC, taurine, AMP, Pyruvate, myo-Inositol.
						LGD vs GS	glutamate, glutamine, creatine, choline, PC, GPC, taurine, AMP, NAD^+.
						HGD vs LGD	lactate, acetate, GPC, glycine, creatine, choline, PC, taurine, AMP.
						GSC vs HGD	leucine, isoleucine, valine, oxybutyrate, lactate, lysine, aspartate, GPC, glycine, myo-Inositol, PC, AMP.
	GC–MS NMR				Human tissue, serum, urine, gastric content	20 trials (summarized from review article)	lactate, valine, glutamine, fumarate, pyrimidine, nucleotides, guanosine, myo-inositol, acetaldehyde, ketones, acetone, β-hydroxybutyrate
Naresh Doni Jayavelu et al. (2014) [27]	^{1}H NMR LC GC MS CE				Human serum, urine, tissue	8 trials (summarized from review article)	lactic acid, serine, proline, malic acid, fatty acids, sarcosine, azelaic acid, glutamate, urate, creatinine, threonate, 3-hydroxypropionic acid, pyruvic acids, valine, isoleucine, serine, phosphoserine, 1-acyl-lysophosphatidylcholines, polyunsaturated fatty acid, lactic acid, butanedioic acid
Hyuk Nam Kwon et al. (2020) [28]	NMR	GSC:n = 103 (67 male, 36 female, mean age:53.7 ± 10.1 years) CON:n = 100 (70 male, 30 female, mean age 54.5 ± 10.5 years)	Stage I (69 patients) Stage II (10 patients) Stage III (15 patients) Stage IV(9 patients)	Differentiated (n = 55), Undifferentiated (n = 48)	Human urine	GSC (stage1,2,3,4) vs CON	alanine, citrate, creatine, creatinine, glycerol, ippurate, phenylalanine, taurine,3-hydroxybutyrate
Huijuan Wang et al. (2016) [29]	^{1}H NMR	GSC: n = 125 (91 male, 34 female, median age:60, range: 28–86 years) CON: n = 54 (39 male, 15 female, median age:61, range: 28–80 years)	Stage I (30 patients) Stage II (46 patients) Stage III (37 patients) Stage IV (12 patients)	Poorly differentiated (n = 74), Moderately differentiated (n = 46), Well-differentiated (n = 0), Not applicable (n = 5)	Human tissue	GSC (stage1,2,3,4) vs CON	isoleucine, lactate, glutamate, glutathione, 4-hydroxyphenylactate, tyrosine, phenyacetylglutamine, hypoxanthine, citrulline, valine, acetoacetate, methylamine
Younes Aftabi et al. (2021) [30]	NMR LC GC CE				Serum, urine, tissue, breath, fecal	18 trials (summarized from review article)	acylcarnitines, amino acids and biogenic amines, sphingolipids, glycerophospholipids, hexoses, tryptophan, phenylalanine, kynurenine, 2-Hydroxybutyrate, pyroglutamate, glutamate, asparagine, azelaic acid, ornithine, urate, g-tocopherol, organic acids, fatty acids, 3-Hydroxypropionic acid, pyruvic acid, L-alanine, glucuronoic lactone steroids, heptanedioic acid, L-valine, acetamide, L-isoleucine, serine.
V. Tugnoli et al. (2006) [31]	HR-MAS	GSC: n = 5 (2 male, 3 female, mean age 70.6 ± 12.9 years, range: 59–86		Well differentiated (n = 3), Poorly differentiated	Human tissue	GSC vs CON	glucose, imidazole, oleic acid, valine, threonine, lactate, lidocaine, chlorohydrate, β-alanine, lysine, arginine, glutamate, glutamine, proline,

(*continued on next page*)

Table 1 (*continued*)

Author and year	Platform	Sample size	TNM stage	Grade	Sample (species)	Group	Disrupted metabolites
		years) CON: n = 11 (7 male, 4 female, mean age: 49.3 ± 17.3 years, range: 30–86 years)		and Undifferentiated (n = 1)			methionine, asparagine, creatine, tyrosine, phenylalanine, ethanolamine, phosphorylethanolamine, glycerophosphorylethanolamine, glycerophosphorylcholine, phosphorylcholine, β-glucose, taurine, myoinositol, scylloinositol, α-glucose, glycine, PEG, glycerol, UDP, uracil, UMP, formate.
Shivanand Pudakalakatti et al. (2022) [32]	NMR LC-MS	Villin-PPARD mouse model (n = 5) WT littermates (n = 5)			Mice tissue	Villin-PPARD mouse model vs WT littermates	glucose, imidazole, IMP, uracil, phenylalanine, glycine, isocitrate, UDP-glucose, AMP, inosine, palmitic acid, linoleic acid, steric acid, oleic acid.
Angela W Chan et al. (2016) [33]	H NMR	GSC: n = 43 (28 male, 15 female, mean age: 65.2 ± 12.0 years) BN: n = 40 (198 male, 21 female, mean age: 63.1 ± 9.0 years) HE: n = 40 (23 male, 17 female, mean age: 63.2 ± 8.8 years)	Stage I (6 patients) Stage II (11 patients) Stage III (10 patients) Stage IV (14 patients) Unknown (2 patients)	Well differentiation (n = 3), Moderate differentiation (n = 8), Moderate to poor differentiation (n = 5), Poor differentiation (n = 29), Not reported (n = 3)	Human urine	GSC vs HE	2-hydroxyisobutyrate, 3-indoxyl-sulfate, alanine
Gokula Krishnan Ramachandran et al. (2016) [34]	NMR	13 gastric cancer cell lines			Human gastric cancer cell	GSC vs CON	choline, choline related compounds, creatine, 2-hydroxy-glutarates, glutamine, methylamine, 2-oxoglutarate
Jinping Gu et al. (2016) [20]	NMR	Model mice (n = 52) CON mice (n = 32)		GS (n = 11), LGD (n = 15), HGD(n = 15), GSC(n = 11)	Mice serum	Model (GS, LGD, HGD, GSC) vs CON	seine, tyrosine, phenylalanine, glycine, lysine, histidine, asparagine, hydroxybutyrate, glycerol, arginine, glutamine, threonine, alanine, LDL/VLDL, lactate, xanthine, phosphocholine, α-acid glycoprotein, polyunsaturated fatty acid
Chi-Woong Mun et al. (2004) [35]	NMR	Noncancerous (n = 22) Cancerous (n = 13)			Human tissue	Noncancerous vs Cancerous	lipids, alanine, N-acetyl neuraminic acid, glutathione, lactate, choline

GSC represents the gastric cancer group, CON is the normal population and represents the control group, GS represents the gastritis group, LGD represents the low-grade gastric dysplasia group, HGD represents the high-grade gastric dysplasia group, BN represents the benign gastric disease group, HE represents the healthy group.

2.4. Network analysis

To create knowledge-based networks, metabolites and genes were mapped to the interaction network to generate subnetworks containing these seeds and their direct neighbors (first-order subnetworks). This process often results in one large subnetwork along with several smaller ones. We extracted genes associated with gastric adenocarcinoma from the Disgenet database [25]. Chemical and human gene associations were extracted from the Search Tool for Interacting Chemicals (STITCH), which explores known and predicted interactions between chemicals and proteins, using only highly confident interactions [26]. Most associations in STITCH are based on the PubMed database, including reactions from similar chemical structures and molecular activities. Since reaction direction was not considered in this study, the metabolic response network plot was undirected, and edge weights were not specified. Node characteristics included size and color. We used degree centrality to measure node importance. Degree centrality is terms as the node degree, meaning the number of edges it has. The higher the degree, the more central the node, which indicates a higher correlation between the metabolites and gastric adenocarcinoma.

3. Results and discussion

3.1. Study design

The overview of this study design is shown in Fig. 1. We first summarized the differentially expressed metabolites associated with gastric adenocarcinoma from twelve metabolomics studies on the disease. Table 1 provides examples of differential metabolites in serum, urine, and tissue based on NMR and MS. We then performed functional analyses, including pathway enrichment analysis, topological analysis, and gene-protein-metabolite-disease interaction network analysis. The molecular mechanisms of key metabolites and metabolic pathways were explored through literature cross-referencing. We mapped differentially expressed metabolites onto relevant metabolic pathways. Based on the significance level of the *P*-value adjusted by FDR, significant metabolic pathways were selected to identify the top differentially expressed metabolites with biological significance between different groups. Topological analysis of these significant metabolic pathways was conducted to discover potential interconnections between metabolites. To investigate the pathogenesis of gastric adenocarcinoma from a genetic regulation perspective, we linked the related genes of gastric adenocarcinoma and significant metabolites to construct a gene-metabolite interaction network. The genes most related to the metabolic pathway of gastric adenocarcinoma were identified to explain the development of gastric adenocarcinoma at both genetic and metabolic levels.

3.2. Metabolic profiling

Through the analysis of analytical platforms and methods, we identified and summarized 80 disrupted metabolites specific to gastric adenocarcinoma. These metabolites are listed in Table S1. The ten most confident differential metabolites are glutamate, glutamine, AMP, choline, aspartate, isoleucine, lactate, valine, citrate, and fumarate. There are five crucial disrupted metabolic processes in gastric adenocarcinoma: amino acid metabolism, carbohydrate metabolism, fatty acid metabolism, energy metabolism, and quaternary ammonium metabolism. Branched-chain amino acids (leucine, isoleucine, and valine) and lysine were significantly upregulated. Glutamate, glutamine, aspartate, isoleucine, and valine were involved in amino acid metabolism. Lactate, which belongs to carbohydrate metabolism, gradually increased during gastric adenocarcinoma. Fatty acid metabolism showed a roughly increased level of myo-inositol. Adenosine 5′-monophosphate was markedly increased in energy metabolism. Choline was significantly increased in quaternary ammonium metabolism.

3.3. Metabolic pathway enrichment analysis

KEGG pathway enrichment analysis was used to identify key pathways in the development of gastric adenocarcinoma. The significant KEGG pathways and core metabolite sets were analyzed, and the results are shown in Fig. 2. There are ten pathways with an adjusted *P*-value <0.01 and FDR <0.01, including aminoacyl-tRNA biosynthesis, arginine biosynthesis, alanine, aspartate, and glutamate metabolism, glyoxylate and dicarboxylate metabolism, valine, leucine, and isoleucine biosynthesis, pantothenate and CoA biosynthesis, the citrate cycle, glycine, serine, and threonine metabolism, D-glutamine and D-glutamate metabolism, and butanoate metabolism. The details of the enrichment analysis are shown in Table S2. As shown in Fig. 2, there are seven pathways with $-\log(p) \geq 1.5$ and impact >0.3, including arginine biosynthesis, alanine, aspartate, and glutamate metabolism, glyoxylate, and dicarboxylate metabolism, D-glutamine and D-glutamate metabolism, phenylalanine metabolism, phenylalanine, tyrosine and tryptophan biosynthesis, and the synthesis and degradation of ketone bodies.

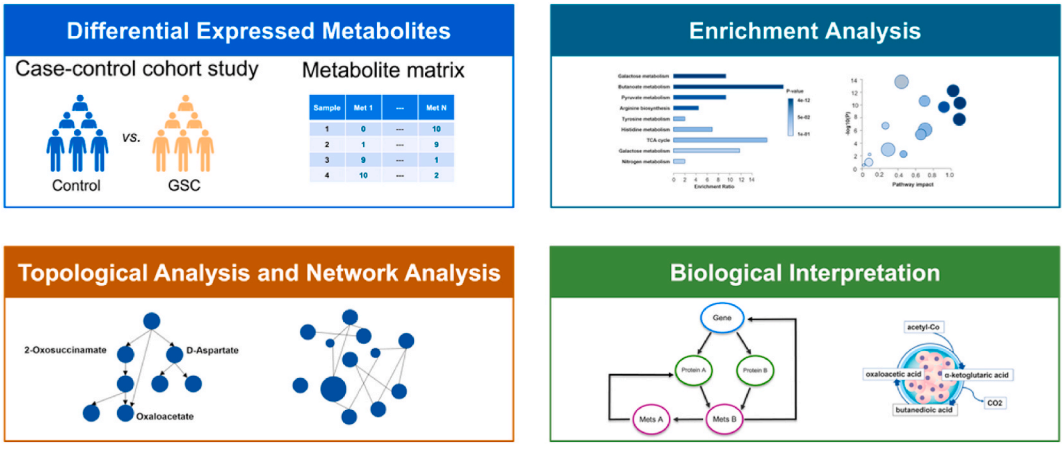

Fig. 1. The workflow of the integrated gene-metabolite association network for mechanistic investigation of gastric adenocarcinoma.

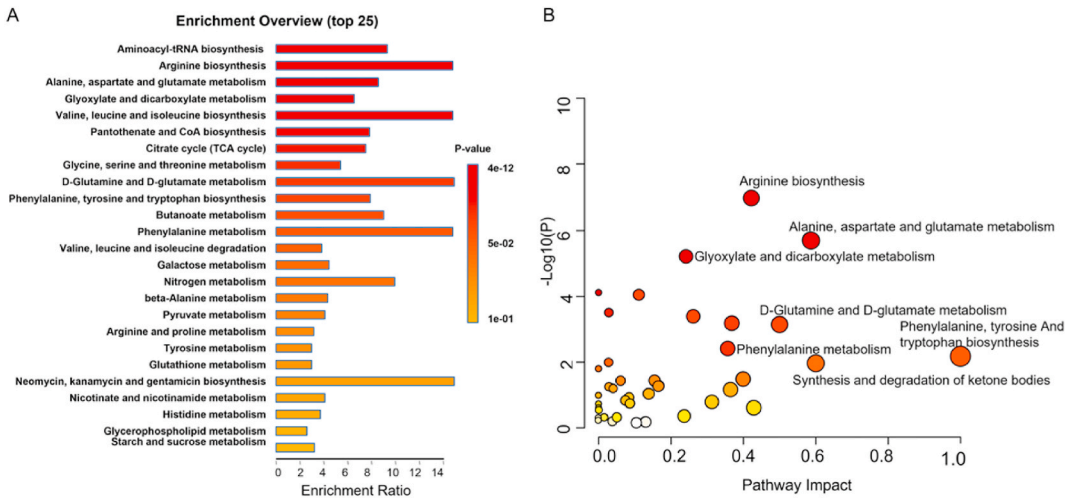

Fig. 2. The metabolic pathway enrichment analysis. Panel (a) shows the pathway enrichment overview (top 25 pathways). The horizontal axis represents the enrichment ratio, and the vertical axis represents the name of the pathway. The color of the histogram represents the *P*-value, representing the significant degree of enrichment. The darker the color, the smaller the Q value and the higher the degree of enrichment. Panel (b) shows all matched pathways according to the *P*-values from the pathway enrichment analysis and pathway impact values from the pathway topological analysis. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

3.4. Topological analysis

Four metabolic pathways have been validated to be associated with gastric adenocarcinoma: arginine biosynthesis (Pathway Impact, PI: 0.42, *P*-value: 1.04E-7), alanine, aspartate, and glutamate metabolism (PI: 0.58, *P*-value: 2.00E-6), D-glutamine and D-glutamate metabolism (PI: 0.5, *P*-value: 0.7E-3), and phenylalanine metabolism (PI: 0.36, *P*-value:0.0038). Differentially expressed metabolites were mapped to these pathways as shown in Fig. 3.

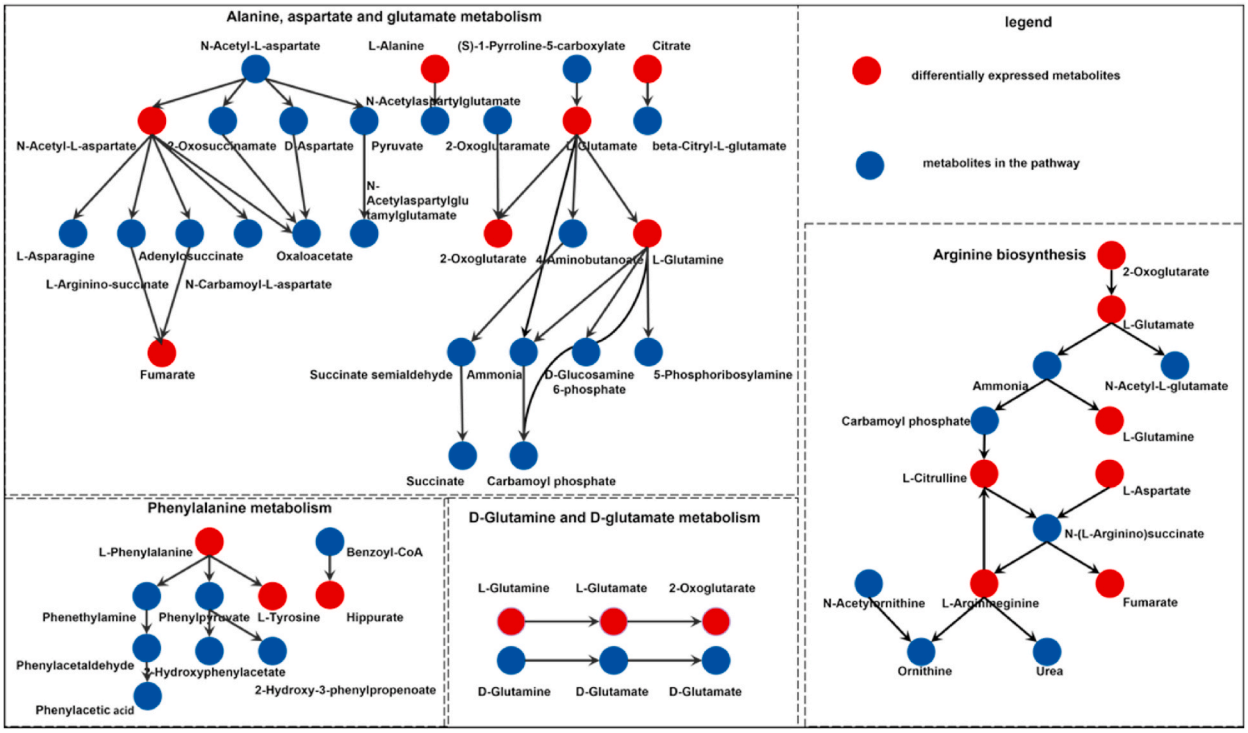

Fig. 3. Topological analysis of four enrichment pathways related to gastric adenocarcinoma. It shows the metabolic pathway of alanine, aspartate and glutamate metabolism, D-glutamine and D-glutamate metabolism, Phenylalanine metabolism, and Arginine biosynthesis.

We identified that alanine, aspartate, and glutamate were significantly different in gastric adenocarcinoma tissues compared to normal tissues. In a study by Yuan et al. using a multi-omics approach, the metabolic profile of gastric adenocarcinoma showed that the metabolism of alanine, aspartate, and glutamate (AAG) was significantly related to the occurrence and development of gastric adenocarcinoma [36]. The metabolomic analysis highlighted the co-expression relationship between AAG metabolism, glycolysis/gluconeogenesis metabolism (G/G), and HER2 levels in gastric adenocarcinoma [36]. This finding could contribute to the development of more targeted therapies for gastric adenocarcinoma.

3.5. Gene-metabolite association network analysis

The results of the genes associated with gastric adenocarcinoma from the Disgenet database are shown in Table S3. We used 80 differentially expressed metabolites and 675 gastric adenocarcinoma-related genes to create a gene-metabolic network. A gene-metabolite interaction network with all the differentially expressed metabolites was then constructed, as shown in Fig. 4. Based on node degree, the top eleven metabolites associated with genes are L-glutamic acid, glutathione, citric acid, oxoglutaric acid, succinic acid, L-aspartic acid, adenine, glycine, L-glutamine, sucrose, and taurine. The top four genes are DECR1, CAT, GLUL, and IDH2. These metabolomic changes provide new insights for selecting effective diagnostic markers and targeted therapy for gastric adenocarcinoma. Additionally, these genes offer clues for screening and treating gastric adenocarcinoma at the transcriptional level. The metabolite that interacted most with genes was L-glutamic acid.

The metabolites that interacted most with genes were L-glutamic acid. Fig. 5 shows a gene-metabolite interaction network for the four top enriched pathways: alanine, aspartate, and glutamate metabolism; D-glutamine and D-glutamate metabolism; arginine biosynthesis; and phenylalanine metabolism. As shown in Fig. 5, L-glutamic acid and oxoglutaric acid are produced by three metabolic

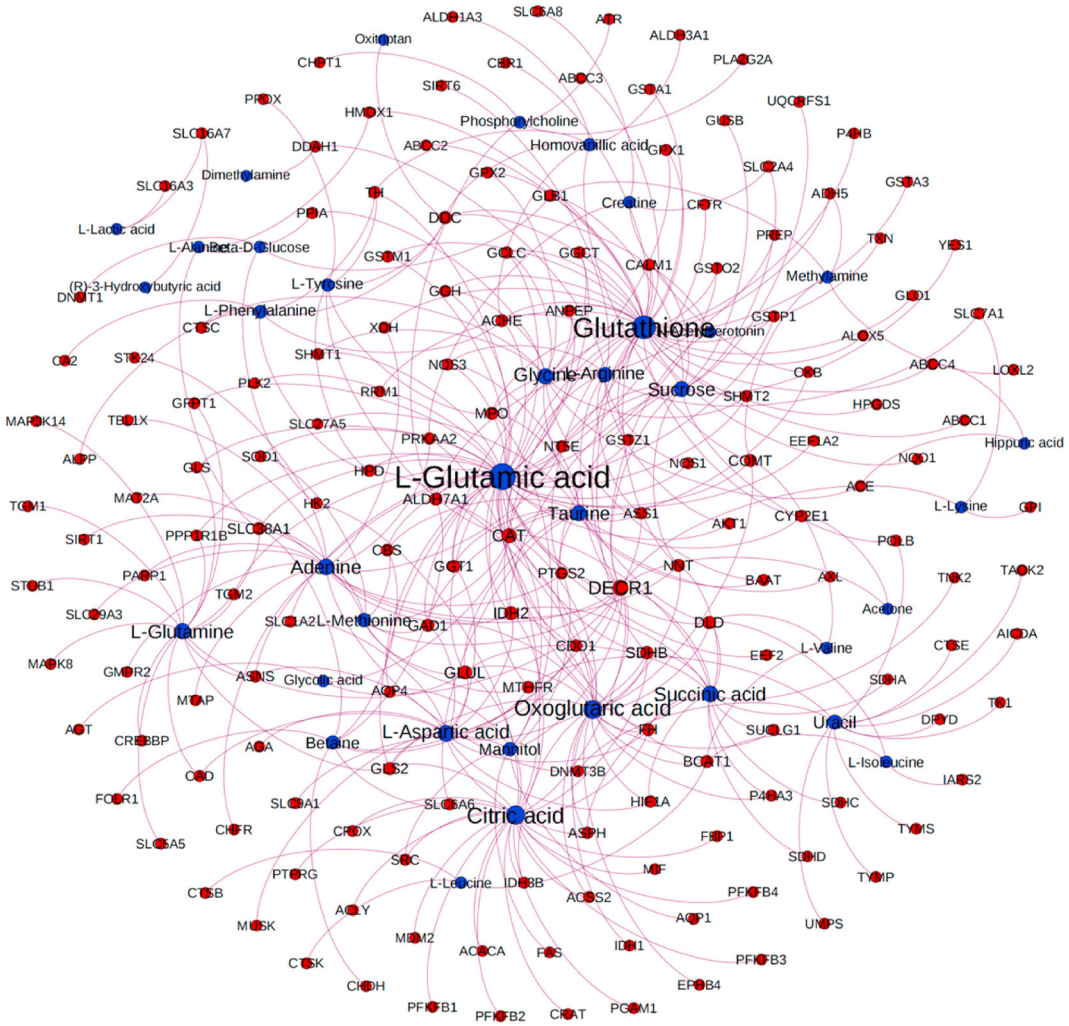

Fig. 4. Integrated network analysis of genes and metabolites associated with gastric adenocarcinoma. Red labels indicate genes and blue labels indicate metabolites. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

pathways. The genes ACE and FH are also closely related to these pathways. This highlights the potential of related signaling pathways as therapeutic targets for gastric adenocarcinoma. Angiotensin-converting enzyme (ACE) is a type I cell surface zinc metallopeptidase responsible for catalyzing the conversion of Ang I to Ang II [37]. There is increasing evidence that ACE is also involved in the pathological process of carcinogenesis [38]. ACE is differentially expressed in several malignancies and affects tumor cell proliferation [38], migration, angiogenesis, and metastatic behavior [39,40]. A recent study discovered that ACE mRNA and protein levels are significantly upregulated in gastric adenocarcinoma [41]. Rocken et al. demonstrated that ACE influences the progression and metastatic behavior of gastric adenocarcinoma, but not its incidence [42]. Experimental studies have shown that the local renin-angiotensin system can affect tumor biology in various ways: (a) increasing neoangiogenesis mediated by vascular endothelial growth factor and microvascular density in solid tumors, which is essential for tumor growth [38,43]; (b) promoting tumor cell proliferation [44]; and (c) remodeling the mesenchymal stroma, which forms the scaffold for tumor cells [45]. Recent studies indicate that combination therapies including ACE inhibitors may be effective in cancer treatment [44]. Many ACE inhibitors are readily available, affordable, well-tolerated, and may reduce the of side effects of other chemotherapeutic agents [46].

The FH gene encodes both cytosolic and mitochondrial variants, which differ in their N-terminal peptide sequences [47]. The mitochondrial FH protein is part of the tricarboxylic acid (TCA) cycle, catalyzing the reversible hydration of fumarate to malate [48]. FH-deficient cells respond to mitochondrial damage compensatory metabolic changes [49]. Typically, as observed in mitochondrial diseases, FH-deficient cells increase their glycolytic rate, shunting glucose to lactate production instead of oxidizing it in the mitochondria [50] and other branches of glycolysis [51]. This glycolytic shift is supported by the transcriptional reprogramming of glycolytic enzymes and the inhibition of pyruvate dehydrogenase (PDH), which prevents glucose from entering the mitochondria [52]. As glucose entry into mitochondria is reduced, glutamine replaces glucose as the main carbon source for the truncated TCA cycle [53]. Sporadic deletion of FH has been reported in several tumors, including paragangliomas [53,54], adrenocortical carcinomas [55], neuroblastomas [56], gliomas, osteosarcomas and Ewing's sarcoma [57]. Its transcriptional downregulation has been found in sporadic clear cell carcinoma and colorectal cancer [58], and there is evidence of FH mutations in breast, bladder, and testicular cancers [59]. These findings imply a critical role for FH deletion in human cancers [49]. However, how its deletion promotes tumorigenesis remains controversial, and its role in gastric carcinogenesis and progression requires further investigation.

4. Discussion

In this study, we identified key metabolic pathways associated with gastric adenocarcinoma using topological and gene-metabolite association network analyses. Three metabolomics studies served as independent validation cohorts to determine whether the dysregulated metabolites and metabolic pathways were consistent with external findings. The results are provided in Table S6. Among the differentially expressed metabolites, 84.3 % showed a consistent trend with the summarized metabolites, and 90.3 % of the key metabolic pathways were validated. These findings demonstrate the validity of these dysregulated metabolic pathways in gastric cancer [60–62].

For the metabolism of aromatic amino acids, both phenylalanine and tryptophan which are essential amino acids, have been validated as being linked to gastric cancer. According to a study by Deng K. et al., high levels of aromatic amino acids in gastric juice are associated with stomach cancer and are necessary for the formation of the non-essential amino acid tyrosine [63,64]. They also found elevated levels of tyrosine, phenylalanine, and tryptophan in gastric fluid samples during the early stages of gastric carcinogenesis, supporting the discovery of elevated levels of aromatic amino acids in gastric contents [64]. An essential indicator of metabolic reprogramming in gastric adenocarcinoma is abnormal arginine metabolism. This reflects changes in the pathophysiology

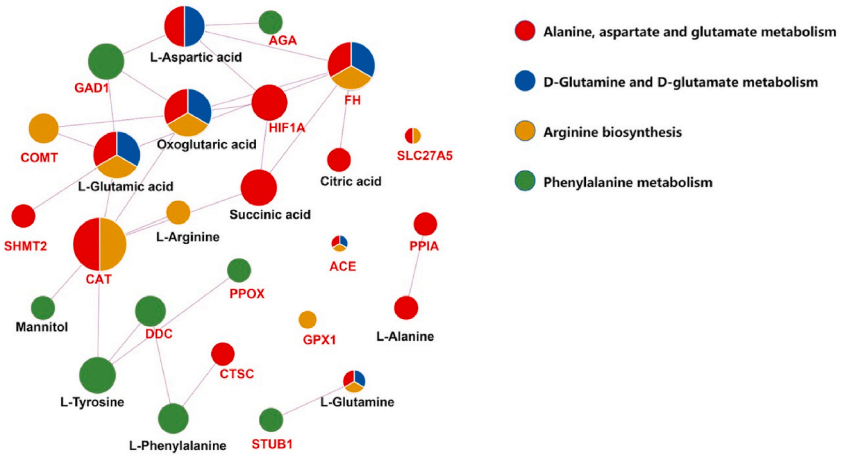

Fig. 5. A gene-metabolite interaction network of four key metabolic pathways. The color of the circle represents the different signalling pathways. The size represents the degree to which a metabolite or gene is associated with gastric adenocarcinoma. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

and subtype of the disease, as well as interactions among enzymes and intermediates in the metabolic pathways [65].

Glutamine plays a crucial role in cancer cell metabolism and is essential for tumor growth, development, and treatment response [66–69]. Glutamine enters the cell via the amino acid transporter ASCT2/SLC1A5 and is converted to glutamate in the mitochondria through a deamination reaction catalyzed by glutaminase (GLS) [70,71]. Glutamate is then converted to the TCA cycle intermediate α-ketoglutarate (α-KG) by glutamate dehydrogenase (GDH)/alanine or aspartate aminotransferases (TAs) [72]. The increased presence of glutamine and glutamate in metabolic data underscores the importance of this pathway in cancer metabolism [70,71]. The absorption of glutamine by the tumor microenvironment differs significantly from that of healthy tissues [73,74]. Glutamine metabolism in the tumor microenvironment has been shown to enhance tumor growth and reduce the immune system's ability to fight tumors [73]. Many cancer cells exhibit an oncogene-dependent addiction to glutamine, promoting proliferative signaling [75,76]. For example, the influx of glutamine via SLC1A5 is closely linked to the efflux of molecules via the SLC7A5/LAT1 transport protein, which also facilitates leucine entry into cells and promotes mTORC1-mediated cell growth [77]. Additionally, signaling molecules like Akt, Ras, and AMPK activate glycolytic enzymes and induce lactate production (Warburg effect), forcing cancer cells to rely on glutamine metabolism to meet the increased energy demands [76]. The proto-oncogene c-Myc promotes glutamine catabolism by transcriptionally activating GLS and SLC1A5 genes [76–78]. Glutamine-mediated protein glycosylation, including that of growth factor receptors, transports proteins to the cell surface and activates them [78].

Aspartate is another crucial metabolite to consider. Aspartate β-hydroxylase (ASPH) has been identified as a cell surface protein associated with the malignant transformation of tumor cells [79,80]. ASPH is a key target for controlling tumor cell migration and invasion [81,82]. Increased expression of ASPH has been observed at both transcriptional and translational levels in various transformed cell lines and human cancer tissues, including hepatocellular carcinoma, pancreatic cancer, colon cancer, prostate cancer, lung cancer, breast cancer, ovarian and cervical cancer, cholangiocarcinoma, neuroblastoma, and gastric adenocarcinoma [83]. ASPH levels have also been linked to cell motility and invasion in in vitro studies. The Wnt/β-catenin [84,85] and insulin/insulin-like growth factor 1 (IGF1)/insulin receptor substrate 1 (IRS1) signaling pathways [86], via extracellular signal-regulated kinase (ERK)/mitogen-activated protein kinase (MAPK) and phosphatidylinositol 3-kinase/protein kinase B [66], upregulate ASPH gene expression. ASPH has been proposed as a common link between the Wnt/β-catenin and insulin/IGF1/IRS1 signaling pathways [87]. This study deploys an integrated multi-omics approach and comprehensive network analysis that provides valuable insights into the metabolic alterations in gastric cancer. The identified metabolic and genomic biomarkers and pathways show significant potential for improving early diagnosis, personalizing treatment, and developing new therapeutic strategies for gastric cancer.

4.1. Limitations of study

Though this study presents key metabolic pathways in gastric adenocarcinoma based on metabolomics data, several limitations should be noted. The metabolomics data were pooled from multiple observational cohort studies, which limits the ability to establish causality between the observed metabolic alterations and the pathophysiology of gastric adenocarcinoma. Additionally, the study results were derived from a specific cohort of patients, which may not represent all demographic groups affected by gastric adenocarcinoma. Genetic diversity and environmental factors, which vary widely across populations, can influence the disease's metabolic pathways, potentially limiting the applicability of our findings to other groups. This study utilized metabolomics data and a topological approach to infer the gene-metabolite association network. To gain a more comprehensive understanding of the disease mechanisms and the metabolic diversity of gastric adenocarcinoma, additional data such as transcriptomics and proteomics should be incorporated. This integrated approach could help tailor personalized therapeutic strategies for gastric adenocarcinoma.

5. Conclusions

In this study, the gene-metabolite interaction network analysis provided insights into the transcriptional regulation mechanisms of these metabolic pathways associated with gastric adenocarcinoma. These findings highlight several potential biomarkers for early detection, diagnosis, and monitoring, as well as targets for personalized therapeutic strategies. This integrated analysis offers significant mechanistic insights into the metabolic and genetic disruptions in gastric adenocarcinoma, paving the way for improved clinical interventions and outcomes.

Fundings

This research was funded by the Key Research and Development Project of the Shandong Province of China (2019JZZY011008 to J. C.), Shandong Natural Science Foundation (ZR2021MH108 to J.C., ZR2022QB152 to C.W.), and the Young Scholars Program of Shandong University (21320082164070 to C.W.).

Ethics approval

Not applicable.

Consent to participate

Not applicable.

Consent for publication

Not applicable.

Data availability statement

All data generated or analyzed during this study are included in this article and its supplementary information files. The source code and data are also available with request to the corresponding author (C.W.).

CRediT authorship contribution statement

Botao Xu: Writing – original draft, Validation, Resources, Investigation, Data curation. **Yuying Shi:** Writing – review & editing, Visualization, Software, Methodology, Formal analysis. **Chuang Yuan:** Investigation, Data curation. **Zhe Wang:** Investigation, Validation, Writing – review & editing. **Qitao Chen:** Investigation, Validation, Writing – review & editing. **Cheng Wang:** Supervision, Conceptualization, Funding acquisition, Investigation, Project administration. **Jie Chai:** Project administration, Funding acquisition, Conceptualization, Writing – review & editing.

Declaration of competing interest

The authors have no relevant financial or non-financial interests to disclose.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.heliyon.2024.e37156.

References

[1] P. Rawla, A. Barsouk, Epidemiology of gastric cancer: global trends, risk factors and prevention 14 (2019) 26–38, https://doi.org/10.5114/pg.2018.80001. Pg.
[2] E.C. Smyth, M. Nilsson, H.I. Grabsch, N.C. van Grieken, F. Lordick, Gastric cancer, Lancet 396 (2020) 635–648, https://doi.org/10.1016/S0140-6736(20)31288-5.
[3] J. Machlowska, J. Baj, M. Sitarz, R. Maciejewski, R. Sitarz, Gastric cancer: epidemiology, risk factors, classification, genomic characteristics and treatment strategies, IJMS 21 (2020) 4012, https://doi.org/10.3390/ijms21114012.
[4] H. Zali, M. Rezaei-Tavirani, M. Azodi, Gastric cancer: prevention, risk factors and treatment, Gastroenterol Hepatol Bed Bench 4 (2011) 175–185.
[5] N. Health Commission of the PRC, National Health Commission of the People's Republic of China, National guidelines for diagnosis and treatment of gastric cancer 2022 in China (English version), Chin. J. Cancer Res. 34 (2022) 207–237, https://doi.org/10.21147/j.issn.1000-9604.2022.03.04.
[6] M. Banks, D. Graham, M. Jansen, T. Gotoda, S. Coda, M. Di Pietro, N. Uedo, P. Bhandari, D.M. Pritchard, E.J. Kuipers, M. Rodriguez-Justo, M.R. Novelli, K. Ragunath, N. Shepherd, M. Dinis-Ribeiro, British Society of Gastroenterology guidelines on the diagnosis and management of patients at risk of gastric adenocarcinoma, Gut 68 (2019) 1545–1575, https://doi.org/10.1136/gutjnl-2018-318126.
[7] E. Holmes, I.D. Wilson, J.K. Nicholson, Metabolic phenotyping in health and disease, Cell 134 (2008) 714–717, https://doi.org/10.1016/j.cell.2008.08.026.
[8] L.D. Roberts, A.L. Souza, R.E. Gerszten, C.B. Clish, Targeted metabolomics, Curr. Protoc. Mol. Biol. 98 (2012), https://doi.org/10.1002/0471142727.mb3002s98.
[9] S. Qiu, Y. Cai, H. Yao, C. Lin, Y. Xie, S. Tang, A. Zhang, Small molecule metabolites: discovery of biomarkers and therapeutic targets, Sig Transduct Target Ther 8 (2023) 132, https://doi.org/10.1038/s41392-023-01399-3.
[10] R. Beger, A review of applications of metabolomics in cancer, Metabolites 3 (2013) 552–574, https://doi.org/10.3390/metabo3030552.
[11] W. Wang, Z. Rong, G. Wang, Y. Hou, F. Yang, M. Qiu, Cancer metabolites: promising biomarkers for cancer liquid biopsy, Biomark. Res. 11 (2023) 66, https://doi.org/10.1186/s40364-023-00507-3.
[12] T. Han, D. Kang, D. Ji, X. Wang, W. Zhan, M. Fu, H.-B. Xin, J.-B. Wang, How does cancer cell metabolism affect tumor migration and invasion? Cell Adhes. Migrat. 7 (2013) 395–403, https://doi.org/10.4161/cam.26345.
[13] D.R. Schmidt, R. Patel, D.G. Kirsch, C.A. Lewis, M.G. Vander Heiden, J.W. Locasale, Metabolomics in cancer research and emerging applications in clinical oncology, CA A Cancer J. Clin. 71 (2021) 333–358, https://doi.org/10.3322/caac.21670.
[14] T.O. Tobore, On the need for the development of a cancer early detection, diagnostic, prognosis, and treatment response system, Future Science OA 6 (2020) FSO439, https://doi.org/10.2144/fsoa-2019-0028.
[15] W. Jelski, B. Mroczko, Molecular and circulating biomarkers of gastric cancer, IJMS 23 (2022) 7588, https://doi.org/10.3390/ijms23147588.
[16] T. Jiang, L. Mei, X. Yang, T. Sun, Z. Wang, Y. Ji, Biomarkers of gastric cancer: current advancement, Heliyon 8 (2022) e10899, https://doi.org/10.1016/j.heliyon.2022.e10899.
[17] M. Ahmed, A.M. Semreen, W. El-Huneidi, Y. Bustanji, E. Abu-Gharbieh, M.A.Y. Alqudah, A. Alhusban, M. Shara, A.Y. Abuhelwa, N.C. Soares, M.H. Semreen, K. H. Alzoubi, Preclinical and clinical applications of metabolomics and proteomics in glioblastoma research, IJMS 24 (2022) 348, https://doi.org/10.3390/ijms24010348.
[18] T. Matsuoka, M. Yashiro, Biomarkers of gastric cancer: current topics and future perspective, WJG 24 (2018) 2818–2832, https://doi.org/10.3748/wjg.v24.i26.2818.
[19] S. Huang, Y. Guo, Z. Li, Y. Zhang, T. Zhou, W. You, K. Pan, W. Li, A systematic review of metabolomic profiling of gastric cancer and esophageal cancer, Cancer Biol. Med. 17 (2020) 181–198, https://doi.org/10.20892/j.issn.2095-3941.2019.0348.
[20] J. Gu, X. Hu, W. Shao, T. Ji, W. Yang, H. Zhuo, Z. Jin, H. Huang, J. Chen, C. Huang, D. Lin, Metabolomic analysis reveals altered metabolic pathways in a rat model of gastric carcinogenesis, Oncotarget 7 (2016) 60053–60073, https://doi.org/10.18632/oncotarget.11049.
[21] J. Gu, C. Huang, X. Hu, J. Xia, W. Shao, D. Lin, Nuclear magnetic resonance-based tissue metabolomic analysis clarifies molecular mechanisms of gastric carcinogenesis, Cancer Sci. 111 (2020) 3195–3209, https://doi.org/10.1111/cas.14443.
[22] S. Xiao, L. Zhou, Gastric cancer: Metabolic and metabolomics perspectives (review), Int. J. Oncol. 51 (2017) 5–17, https://doi.org/10.3892/ijo.2017.4000.

[23] Z. Pang, J. Chong, G. Zhou, D.A. de Lima Morais, L. Chang, M. Barrette, C. Gauthier, P.-É. Jacques, S. Li, J. Xia, MetaboAnalyst 5.0: narrowing the gap between raw spectra and functional insights, Nucleic Acids Res. 49 (2021) W388–W396, https://doi.org/10.1093/nar/gkab382.
[24] Z. Pang, G. Zhou, J. Ewald, L. Chang, O. Hacariz, N. Basu, J. Xia, Using MetaboAnalyst 5.0 for LC–HRMS spectra processing, multi-omics integration and covariate adjustment of global metabolomics data, Nat. Protoc. 17 (2022) 1735–1761, https://doi.org/10.1038/s41596-022-00710-w.
[25] J. Piñero, À. Bravo, N. Queralt-Rosinach, A. Gutiérrez-Sacristán, J. Deu-Pons, E. Centeno, J. García-García, F. Sanz, L.I. Furlong, DisGeNET: a comprehensive platform integrating information on human disease-associated genes and variants, Nucleic Acids Res. 45 (2017) D833–D839, https://doi.org/10.1093/nar/gkw943.
[26] D. Szklarczyk, A. Santos, C. von Mering, L.J. Jensen, P. Bork, M. Kuhn, Stitch 5: augmenting protein–chemical interaction networks with tissue and affinity data, Nucleic Acids Res. 44 (2016) D380–D384, https://doi.org/10.1093/nar/gkv1277.
[27] N.D. Jayavelu, Metabolomic studies of human gastric cancer, Review, WJG 20 (2014) 8092, https://doi.org/10.3748/wjg.v20.i25.8092.
[28] H.N. Kwon, H. Lee, J.W. Park, Y.-H. Kim, S. Park, J.J. Kim, Screening for early gastric cancer using a noninvasive urine metabolomics approach, Cancers 12 (2020) 2904, https://doi.org/10.3390/cancers12102904.
[29] H. Wang, H. Zhang, P. Deng, C. Liu, D. Li, H. Jie, H. Zhang, Z. Zhou, Y.-L. Zhao, Tissue metabolic profiling of human gastric cancer assessed by 1H NMR, BMC Cancer 16 (2016) 371, https://doi.org/10.1186/s12885-016-2356-4.
[30] Y. Aftabi, J. Soleymani, A. Jouyban, Efficacy of analytical technologies in metabolomics studies of the gastrointestinal cancers, Crit. Rev. Anal. Chem. 52 (2022) 1593–1605, https://doi.org/10.1080/10408347.2021.1901646.
[31] V. Tugnoli, A. Mucci, L. Schenetti, V. Righi, C. Calabrese, A. Fabbri, G. Di Febo, M. Tosi, Ex vivo HR-MAS Magnetic Resonance Spectroscopy of human gastric adenocarcinomas: a comparison with healthy gastric mucosa, Oncol. Rep. (2006), https://doi.org/10.3892/or.16.3.543.
[32] S. Pudakalakatti, M. Titus, J.S. Enriquez, S. Ramachandran, N.M. Zacharias, I. Shureiqi, Y. Liu, J.C. Yao, X. Zuo, P.K. Bhattacharya, Identifying the metabolic signatures of PPARD-overexpressing gastric tumors, IJMS 23 (2022) 1645, https://doi.org/10.3390/ijms23031645.
[33] A.W. Chan, P. Mercier, D. Schiller, R. Bailey, S. Robbins, D.T. Eurich, M.B. Sawyer, D. Broadhurst, 1H-NMR urinary metabolomic profiling for diagnosis of gastric cancer, Br. J. Cancer 114 (2016) 59–62, https://doi.org/10.1038/bjc.2015.414.
[34] G.K. Ramachandran, W.P. Yong, C.H. Yeow, Identification of gastric cancer biomarkers using 1H nuclear magnetic resonance spectrometry, PLoS One 11 (2016) e0162222, https://doi.org/10.1371/journal.pone.0162222.
[35] C.-W. Mun, J.-Y. Cho, W.-J. Shin, K.-S. Choi, C.-K. Eun, S.S. Cha, J. Lee, Y. Yang, S.-H. Nam, J. Kim, S.Y. Lee, Ex vivo proton MR spectroscopy (1H-MRS) for evaluation of human gastric carcinoma, Magn. Reson. Imag. 22 (2004) 861–870, https://doi.org/10.1016/j.mri.2004.01.045.
[36] Q. Yuan, D. Deng, C. Pan, J. Ren, T. Wei, Z. Wu, B. Zhang, S. Li, P. Yin, D. Shang, Integration of transcriptomics, proteomics, and metabolomics data to reveal HER2-associated metabolic heterogeneity in gastric cancer with response to immunotherapy and neoadjuvant chemotherapy, Front. Immunol. 13 (2022) 951137, https://doi.org/10.3389/fimmu.2022.951137.
[37] H. Yoshiji, S. Kuriyama, M. Kawata, J. Yoshii, Y. Ikenaka, R. Noguchi, T. Nakatani, H. Tsujinoue, H. Fukui, The angiotensin-I-converting enzyme inhibitor perindopril suppresses tumor growth and angiogenesis: possible role of the vascular endothelial growth factor, Clin. Cancer Res. 7 (2001) 1073–1078.
[38] H. Abali, I.H. Güllü, H. Engin, I.C. Haznedaroğlu, M. Erman, G. Tekuzman, Old antihypertensives as novel antineoplastics: angiotensin-I-converting enzyme inhibitors and angiotensin II type 1 receptor antagonists, Med. Hypotheses 59 (2002) 344–348, https://doi.org/10.1016/S0306-9877(02)00185-8.
[39] B. Bauvois, Transmembrane proteases in cell growth and invasion: new contributors to angiogenesis? Oncogene 23 (2004) 317–329, https://doi.org/10.1038/sj.onc.1207124.
[40] H. Yoshiji, S. Kuriyama, H. Fukui, Perindopril: possible use in cancer therapy, Anti Cancer Drugs 13 (2002) 221–228, https://doi.org/10.1097/00001813-200203000-00003.
[41] S. Carl-McGrath, U. Lendeckel, M. Ebert, A.-B. Wolter, A. Roessner, C. Röcken, The ectopeptidases CD10, CD13, CD26, and CD143 are upregulated in gastric cancer, Int. J. Oncol. 25 (2004) 1223–1232.
[42] C. Röcken, U. Lendeckel, J. Dierkes, S. Westphal, S. Carl-McGrath, B. Peters, S. Krüger, P. Malfertheiner, A. Roessner, M.P.A. Ebert, The number of lymph node metastases in gastric cancer correlates with the angiotensin I–converting enzyme gene insertion/deletion polymorphism, Clin. Cancer Res. 11 (2005) 2526–2530, https://doi.org/10.1158/1078-0432.CCR-04-1922.
[43] M. Fujita, I. Hayashi, S. Yamashina, M. Itoman, M. Majima, Blockade of angiotensin AT1a receptor signaling reduces tumor growth, angiogenesis, and metastasis, Biochem. Biophys. Res. Commun. 294 (2002) 441–447, https://doi.org/10.1016/S0006-291X(02)00496-5.
[44] M. Yasumaru, S. Tsuji, M. Tsujii, T. Irie, M. Komori, A. Kimura, T. Nishida, Y. Kakiuchi, N. Kawai, H. Murata, M. Horimoto, Y. Sasaki, N. Hayashi, S. Kawano, M. Hori, Inhibition of angiotensin II activity enhanced the antitumor effect of cyclooxygenase-2 inhibitors via insulin-like growth factor I receptor pathway, Cancer Res. 63 (2003) 6726–6734.
[45] T. Suehiro, T. Morita, M. Inoue, Y. Kumon, Y. Ikeda, K. Hashimoto, Increased amount of the angiotensin-converting enzyme (ACE) mRNA originating from the ACE allele with deletion, Hum. Genet. 115 (2004), https://doi.org/10.1007/s00439-004-1136-4.
[46] C.A. Haiman, S.O. Henderson, P. Bretsky, L.N. Kolonel, B.E. Henderson, Genetic variation in angiotensin I-converting enzyme (ACE) and breast cancer risk: the multiethnic cohort, Cancer Res. 63 (2003) 6984–6987.
[47] S. Picaud, K.L. Kavanagh, W.W. Yue, W.H. Lee, S. Muller-Knapp, O. Gileadi, J. Sacchettini, U. Oppermann, Structural basis of fumarate hydratase deficiency, J of Inher Metab Disea 34 (2011) 671–676, https://doi.org/10.1007/s10545-011-9294-8.
[48] C. Frezza, Mitochondrial metabolites: undercover signalling molecules, Interface Focus 7 (2017) 20160100, https://doi.org/10.1098/rsfs.2016.0100.
[49] C. Schmidt, M. Sciacovelli, C. Frezza, Fumarate hydratase in cancer: a multifaceted tumour suppressor, Semin. Cell Dev. Biol. 98 (2020) 15–25, https://doi.org/10.1016/j.semcdb.2019.05.002.
[50] C. Frezza, L. Zheng, O. Folger, K.N. Rajagopalan, E.D. MacKenzie, L. Jerby, M. Micaroni, B. Chaneton, J. Adam, A. Hedley, G. Kalna, I.P.M. Tomlinson, P. J. Pollard, D.G. Watson, R.J. Deberardinis, T. Shlomi, E. Ruppin, E. Gottlieb, Haem oxygenase is synthetically lethal with the tumour suppressor fumarate hydratase, Nature 477 (2011) 225–228, https://doi.org/10.1038/nature10363.
[51] Y. Yang, A.N. Lane, C.J. Ricketts, C. Sourbier, M.-H. Wei, B. Shuch, L. Pike, M. Wu, T.A. Rouault, L.G. Boros, T.W.-M. Fan, W.M. Linehan, Metabolic reprogramming for producing energy and reducing power in fumarate hydratase null cells from hereditary leiomyomatosis renal cell carcinoma, PLoS One 8 (2013) e72179, https://doi.org/10.1371/journal.pone.0072179.
[52] E. Gonçalves, M. Sciacovelli, A.S.H. Costa, M.G.B. Tran, T.I. Johnson, D. Machado, C. Frezza, J. Saez-Rodriguez, Post-translational regulation of metabolism in fumarate hydratase deficient cancer cells, Metab. Eng. 45 (2018) 149–157, https://doi.org/10.1016/j.ymben.2017.11.011.
[53] G.R. Clark, M. Sciacovelli, E. Gaude, D.M. Walsh, G. Kirby, M.A. Simpson, R.C. Trembath, J.N. Berg, E.R. Woodward, E. Kinning, P.J. Morrison, C. Frezza, E. R. Maher, Germline FH mutations presenting with pheochromocytoma, The Journal of Clinical Endocrinology & Metabolism 99 (2014) E2046–E2050, https://doi.org/10.1210/jc.2014-1659.
[54] L.J. Castro-Vega, A. Buffet, A.A. De Cubas, A. Cascón, M. Menara, E. Khalifa, L. Amar, S. Azriel, I. Bourdeau, O. Chabre, M. Currás-Freixes, V. Franco-Vidal, M. Guillaud-Bataille, C. Simian, A. Morin, R. Letón, Á. Gómez-Graña, P.J. Pollard, P. Rustin, M. Robledo, J. Favier, A.-P. Gimenez-Roqueplo, Germline mutations in FH confer predisposition to malignant pheochromocytomas and paragangliomas, Hum. Mol. Genet. 23 (2014) 2440–2446, https://doi.org/10.1093/hmg/ddt639.
[55] A. Fieuw, C. Kumps, A. Schramm, F. Pattyn, B. Menten, F. Antonacci, P. Sudmant, J.H. Schulte, N. Van Roy, S. Vergult, P.G. Buckley, A. De Paepe, R. Noguera, R. Versteeg, R. Stallings, A. Eggert, J. Vandesompele, K. De Preter, F. Speleman, Identification of a novel recurrent 1q42.2-1qter deletion in high risk MYCN single copy 11q deleted neuroblastomas, Int. J. Cancer 130 (2012) 2599–2606, https://doi.org/10.1002/ijc.26317.
[56] A. Nakazawa, C. Haga, M. Ohira, H. Okita, T. Kamijo, A. Nakagawara, Correlation between the international neuroblastoma pathology classification and genomic signature in neuroblastoma, Cancer Sci. 106 (2015) 766–771, https://doi.org/10.1111/cas.12665.
[57] J. Zhang, M.F. Walsh, G. Wu, M.N. Edmonson, T.A. Gruber, J. Easton, D. Hedges, X. Ma, X. Zhou, D.A. Yergeau, M.R. Wilkinson, B. Vadodaria, X. Chen, R. B. McGee, S. Hines-Dowell, R. Nuccio, E. Quinn, S.A. Shurtleff, M. Rusch, A. Patel, J.B. Becksfort, S. Wang, M.S. Weaver, L. Ding, E.R. Mardis, R.K. Wilson,

A. Gajjar, D.W. Ellison, A.S. Pappo, C.-H. Pui, K.E. Nichols, J.R. Downing, Germline mutations in predisposition genes in pediatric cancer, N. Engl. J. Med. 373 (2015) 2336–2346, https://doi.org/10.1056/NEJMoa1508054.
[58] J. Hu, J.W. Locasale, J.H. Bielas, J. O'Sullivan, K. Sheahan, L.C. Cantley, M.G.V. Heiden, D. Vitkup, Heterogeneity of tumor-induced gene expression changes in the human metabolic network, Nat. Biotechnol. 31 (2013) 522–529, https://doi.org/10.1038/nbt.2530.
[59] C. Frezza, P.J. Pollard, E. Gottlieb, Inborn and acquired metabolic defects in cancer, J Mol Med 89 (2011) 213–220, https://doi.org/10.1007/s00109-011-0728-4.
[60] Z. Xu, Y. Huang, C. Hu, L. Du, Y.-A. Du, Y. Zhang, J. Qin, W. Liu, R. Wang, S. Yang, J. Wu, J. Cao, J. Zhang, G.-P. Chen, H. Lv, P. Zhao, W. He, X. Wang, M. Xu, P. Wang, C. Hong, L.-T. Yang, J. Xu, J. Chen, Q. Wei, R. Zhang, L. Yuan, K. Qian, X. Cheng, Efficient plasma metabolic fingerprinting as a novel tool for diagnosis and prognosis of gastric cancer: a large-scale, multicentre study, Gut 72 (2023) 2051–2067, https://doi.org/10.1136/gutjnl-2023-330045.
[61] J. Yu, J. Zhao, T. Yang, R. Feng, L. Liu, Metabolomics reveals novel serum metabolic signatures in gastric cancer by a mass spectrometry platform, J. Proteome Res. 22 (2023) 706–717, https://doi.org/10.1021/acs.jproteome.2c00295.
[62] Y. Chen, B. Wang, Y. Zhao, X. Shao, M. Wang, F. Ma, L. Yang, M. Nie, P. Jin, K. Yao, H. Song, S. Lou, H. Wang, T. Yang, Y. Tian, P. Han, Z. Hu, Metabolomic machine learning predictor for diagnosis and prognosis of gastric cancer, Nat. Commun. 15 (2024) 1657, https://doi.org/10.1038/s41467-024-46043-y.
[63] K. Deng, S. Lin, L. Zhou, Q. Geng, Y. Li, M. Xu, R. Na, Three aromatic amino acids in gastric juice as potential biomarkers for gastric malignancies, Anal. Chim. Acta 694 (2011) 100–107, https://doi.org/10.1016/j.aca.2011.03.053.
[64] K. Deng, S. Lin, L. Zhou, Y. Li, M. Chen, Y. Wang, Y. Li, High levels of aromatic amino acids in gastric juice during the early stages of gastric cancer progression, PLoS One 7 (2012) e49434, https://doi.org/10.1371/journal.pone.0049434.
[65] I. Bednarz-Misa, M.G. Fleszar, P. Fortuna, Ł. Lewandowski, M. Mierzchała-Pasierb, D. Diakowska, M. Krzystek-Korpacka, Altered L-arginine metabolic pathways in gastric cancer: potential therapeutic targets and biomarkers, Biomolecules 11 (2021) 1086, https://doi.org/10.3390/biom11081086.
[66] C.T. Hensley, A.T. Wasti, R.J. DeBerardinis, Glutamine and cancer: cell biology, physiology, and clinical opportunities, J. Clin. Invest. 123 (2013) 3678–3684, https://doi.org/10.1172/JCI69600.
[67] T.-L. Nguyen, R.V. Durán, Glutamine Metabolism in Cancer Therapy, CDR, 2018, https://doi.org/10.20517/cdr.2018.08.
[68] L. Yuan, X. Sheng, A.K. Willson, D.R. Roque, J.E. Stine, H. Guo, H.M. Jones, C. Zhou, V.L. Bae-Jump, Glutamine promotes ovarian cancer cell proliferation through the mTOR/S6 pathway, Endocr. Relat. Cancer 22 (2015) 577–591, https://doi.org/10.1530/ERC-15-0192.
[69] G.W. Kim, D.H. Lee, Y.H. Jeon, J. Yoo, S.Y. Kim, S.W. Lee, H.Y. Cho, S.H. Kwon, Glutamine synthetase as a therapeutic target for cancer treatment, IJMS 22 (2021) 1701, https://doi.org/10.3390/ijms22041701.
[70] N. Abbassi-Ghadi, S. Kumar, J. Huang, R. Goldin, Z. Takats, G.B. Hanna, Metabolomic profiling of oesophago-gastric cancer: a systematic review, Eur. J. Cancer 49 (2013) 3625–3637, https://doi.org/10.1016/j.ejca.2013.07.004.
[71] Y. Tian, W. Du, S. Cao, Y. Wu, N. Dong, Y. Wang, Y. Xu, Systematic analyses of glutamine and glutamate metabolisms across different cancer types, Chin. J. Cancer 36 (2017) 88, https://doi.org/10.1186/s40880-017-0255-y.
[72] L. Jin, G.N. Alesi, S. Kang, Glutaminolysis as a target for cancer therapy, Oncogene 35 (2016) 3619–3625, https://doi.org/10.1038/onc.2015.447.
[73] M.D. Claiborne, R. Leone, Differential glutamine metabolism in the tumor microenvironment - studies in diversity and heterogeneity: a mini-review, Front. Oncol. 12 (2022) 1011191, https://doi.org/10.3389/fonc.2022.1011191.
[74] L. Zhu, X. Zhu, Y. Wu, Effects of glucose metabolism, lipid metabolism, and glutamine metabolism on tumor microenvironment and clinical implications, Biomolecules 12 (2022) 580, https://doi.org/10.3390/biom12040580.
[75] M.H. Kim, H. Kim, Oncogenes and tumor suppressors regulate glutamine metabolism in cancer cells, J Cancer Prev 18 (2013) 221–226, https://doi.org/10.15430/JCP.2013.18.3.221.
[76] L. Chen, H. Cui, Targeting glutamine induces apoptosis: a cancer therapy approach, IJMS 16 (2015) 22830–22855, https://doi.org/10.3390/ijms160922830.
[77] D. Xiao, L. Zeng, K. Yao, X. Kong, G. Wu, Y. Yin, The glutamine-alpha-ketoglutarate (AKG) metabolism and its nutritional implications, Amino Acids 48 (2016) 2067–2080, https://doi.org/10.1007/s00726-016-2254-8.
[78] M.J. Lukey, K.F. Wilson, R.A. Cerione, Therapeutic strategies impacting cancer cell glutamine metabolism, Future Med. Chem. 5 (2013) 1685–1700, https://doi.org/10.4155/fmc.13.130.
[79] N. Ince, S.M. de la Monte, J.R. Wands, Overexpression of human aspartyl (asparaginyl) beta-hydroxylase is associated with malignant transformation, Cancer Res. 60 (2000) 1261–1266.
[80] L. Lavaissiere, S. Jia, M. Nishiyama, S. de la Monte, A.M. Stern, J.R. Wands, P.A. Friedman, Overexpression of human aspartyl(asparaginyl)beta-hydroxylase in hepatocellular carcinoma and cholangiocarcinoma, J. Clin. Invest. 98 (1996) 1313–1323, https://doi.org/10.1172/JCI118918.
[81] L.-M. Sturla, M. Tong, N. Hebda, J. Gao, J.-M. Thomas, M. Olsen, S.M. de la Monte, Aspartate-β-hydroxylase (ASPH): a potential therapeutic target in human malignant gliomas, Heliyon 2 (2016) e00203, https://doi.org/10.1016/j.heliyon.2016.e00203.
[82] K. Nagaoka, X. Bai, K. Ogawa, X. Dong, S. Zhang, Y. Zhou, R.I. Carlson, Z.-G. Jiang, S. Fuller, M.S. Lebowitz, H. Ghanbari, J.R. Wands, Anti-tumor activity of antibody drug conjugate targeting aspartate-β-hydroxylase in pancreatic ductal adenocarcinoma, Cancer Lett. 449 (2019) 87–98, https://doi.org/10.1016/j.canlet.2019.02.006.
[83] H. Yang, K. Song, T. Xue, X.-P. Xue, T. Huyan, W. Wang, H. Wang, The distribution and expression profiles of human Aspartyl/Asparaginyl beta-hydroxylase in tumor cell lines and human tissues, Oncol. Rep. 24 (2010) 1257–1264, https://doi.org/10.3892/or_00000980.
[84] Y. Tomimaru, H. Koga, H. Yano, S. de la Monte, J.R. Wands, M. Kim, Upregulation of T-cell factor-4 isoform-responsive target genes in hepatocellular carcinoma, Liver Int. 33 (2013) 1100–1112, https://doi.org/10.1111/liv.12188.
[85] S.M. de la Monte, S. Tamaki, M.C. Cantarini, N. Ince, M. Wiedmann, J.J. Carter, S.A. Lahousse, S. Califano, T. Maeda, T. Ueno, A. D'Errico, F. Trevisani, J. R. Wands, Aspartyl-(asparaginyl)-β-hydroxylase regulates hepatocellular carcinoma invasiveness, J. Hepatol. 44 (2006) 971–983, https://doi.org/10.1016/j.jhep.2006.01.038.
[86] M. Kanwal, M. Smahel, M. Olsen, J. Smahelova, R. Tachezy, Aspartate β-hydroxylase as a target for cancer therapy, J. Exp. Clin. Cancer Res. 39 (2020) 163, https://doi.org/10.1186/s13046-020-01669-w.
[87] W. Chung, M. Kim, S. de la Monte, L. Longato, R. Carlson, B.L. Slagle, X. Dong, J.R. Wands, Activation of signal transduction pathways during hepatic oncogenesis, Cancer Lett. 370 (2016) 1–9, https://doi.org/10.1016/j.canlet.2015.09.016.