Vascular Pharmacology 112 (2019) 17–23

Contents lists available at ScienceDirect

Vascular Pharmacology

journal homepage: www.elsevier.com/locate/vph

Review

Role of amino acid metabolism in angiogenesis

Roxana E. Oberkersch, Massimo M. Santoro*

Department of Biology, University of Padua, Italy

ABSTRACT

The role of endothelial metabolism represents a crucial element governing the formation and the differentiation of blood vessels, termed angiogenesis. Besides glycolysis and fatty acid oxidation, endothelial cells rely on specific amino acids to proliferate, migrate, and survive. In this review we focus on the metabolism of those amino acids and the intermediates that hold an established function within angiogenesis and endothelial pathophysiology. We also discuss recent work which provides a rationale for specific amino acid-restricted diets and its beneficial effects on vascular tissues, including extending the life span and preventing the development of a variety of diseases.

1. Introduction

The vascular system is a multi-branched network lined by endothelial cells (ECs) which delivers oxygen and nutrients to the tissues in the body [1]. The ability to expand this network is a process called angiogenesis. This process is vital in many physiological settings and is also implicated in the pathogenesis of several disorders including: diabetic retinopathy, ischemic heart disease, rheumatoid arthritis, psoriasis, and tumor growth [2]. A better understanding of the molecular mechanisms involved in angiogenesis is crucial in the fight against such diseases. The metabolism of ECs has been recognized as a driving force of angiogenesis [3]. Changes in endothelial metabolism are able to induce or inhibit different steps within the angiogenic process. In addition, the adoption of an angiogenic phenotype may reprogram EC metabolism [4]. A growing amount of evidence indicates that glycolysis, fatty acid oxidation (FAO) and glutamine metabolism are all essential during blood vessel formation. ECs *per se* are highly glycolytic and produce up to 85% of their ATP through aerobic glycolysis [5]. It has been hypothesized that the use of glycolysis as the main source of ATP holds several advantages. High glycolytic flux can produce faster ATP with respect to oxidative metabolism, which represents an advantage for ECs in a hypoxic microenvironment [6]. The anaerobic metabolism of ECs might save oxygen towards becoming perivascular mural cells [7]. Glycolysis is also a source of intermediate metabolites, which are crucial in many biosynthetic pathways: nucleotide biosynthesis (through ribose-5-phosphate in the pentose phosphate pathway (PPP)) [8], hexosamine biosynthesis [9], and serine-glycine biosynthesis [10]. During sprouting angiogenesis, additional energy and biomass are needed, therefore proliferating and migrating ECs will double their glycolytic flux compared to quiescent ECs [11] by increasing the expression of a glucose transporter (GLUT1) [12], while glycolytic enzymes lactate dehydrogenase-A (LDH-A) [13], and 6-phosphofructo-2-kinase/fructose-2,6-phisphosphatase (PFKFB3) [14].

Although glycolysis is essential during the sprouting of ECs it is not

Abbreviations: AA, amino acid; AAR, amino acid response; Acetyl-CoA, acetyl coenzyme A; α-KG, alpha-ketoglutarate; ARG, arginase; ASNS, asparagine synthetase; ATF4, activating transcription factor; ATP, adenosine triphosphate; bFGF, basic fibroblast growth factor; CAT, cystathionine aminotransferase; CBS, cystathionine-β-synthase; CGL or CSE, cystathionine-γ-lyase; CPT1a, carnitine palmytoyltransferase 1a; CPT2, carnitine palmytoyltransferase 2; CSA, cysteine sulfinic acid; EAA, essential amino acid; EC, endothelial cell; endoMT, endothelial-to-mesenchymal transition; eNOS, endotelial NOS; ER, endoplasmic reticulum; FA, fatty acid; FAO, fatty acid oxidation; GLS, glutaminase; GLUT1, glucose transporter 1; GSAL, L-glutamate-γ-semialdehyde; GSALDH, GSAL dehydrogenase; GSH, reduced glutathione; GSSG, oxidized glutathione; H_2S, hydrogen sulphide; HCEC, human corneal endothelial cells; HUVEC, human umbilical vein endothelial cells; IGF-1, insulin-like growth factor 1; iNOS, inducible NOS; LCFA, long-chain fatty acid; LDH-A, lactate dehydrogenase-A; 3-MST, 3-mercaptopyruvate sulfurtransferase; NEAA, non-essential amino acid; NO, nitric oxide; NOS, nitric oxide synthase; OAT, ornithine δ-amino acid transferase; ODC, ornithine decarboxylase; P5C, 1-pyrroline-5-carboxylate; P5CR, pyrroline-5-carboxylate reductase; P5CS, P5C synthase; PFKFB3, 6-phosphofructo-2-kinase/fructose-2,6-phisphosphatase; 3PG, 3-phosphoglycerate; PHGDH, phosphoglycerate dehydrogenase; 3PHP, 3-phosphohydroxypyruvate; PPP, pentose phosphate pathway; PRODH, proline dehydrogenase; PSAT1, phosphoserine aminotransferase 1; PSPH, phosphoserine phosphatase; ROS, reactive oxygen species; SAA, sulfur amino acid; SAH, S-adenosylhomocysteine; SAM, S-adenosylmethionine; SHMT, serinehydroxymethyltransferase; SSP, serine synthesis pathway; TCA, tricarboxylic acid cycle; TGFβ, transforming growth factor beta; TSP, transsulfuration pathway; UPR, unfolded protein response; VEGF, vascular endothelial growth factor; VEGFR, VEGF receptor

* Corresponding author.
E-mail address: massimo.santoro@unipd.it (M.M. Santoro).

https://doi.org/10.1016/j.vph.2018.11.001
Received 19 June 2018; Accepted 2 November 2018
Available online 10 November 2018

the only metabolic determinant of the angiogenic process. Indeed, fatty acid (FA) metabolism plays important role within ECs [15]. FAs can be imported, oxidized, synthesized, and exported by ECs. FAO contributes to approximately 5% to the total cellular ATP production in ECs and ^{13}C-palmitate tracing experiments have revealed that FAO predominantly produce acetyl coenzyme A (acetyl-CoA), an anaplerotic substrate [16]. EC-specific deletion of carnitine palmytoyltransferase 1a (CPT1a), long-chain fatty acid (LCFA) importer localized in the outer mitochondrial membrane, impairs *de novo* nucleotide synthesis that alters EC proliferation, which does not cause energy depletion or disturb redox homeostasis [17]. The phenotype of CPT1a-silenced ECs is rescued by acetate (metabolized to acetyl-CoA, thereby substituting for the depleted FAO-derived acetyl-CoA) or nucleoside mix. Endothelial FAO is a critical regulator of endothelial-to-mesenchymal transition (endoMT) [18]. Finkel and colleagues demonstrated that the inhibition of FAO through EC-specific deletion of carnitine palmytoyltransferase 2 (CPT2), the inner mitochondrial transporter of LCFAs, markedly reduces the stability of SMAD7, a potent inhibitor of transforming growth factor beta (TGF-β) signaling. Although, these data support that inhibition of CPT2 it also induces the endoMT, the implication of FAO in pathological angiogenesis is still needed to be proved.

In recent years, it has been shown that amino acid (AA) metabolism also plays an important role in regulating and maintaining vascular function. These functions include vascular tone, coagulation and fibrinolysis, cell growth and differentiation, redox homeostasis, plus immune and inflammatory responses. Each AA has specific characteristics which are defined by the side chain, and based on the polarity of their side chains they can then be classified as hydrophobic, polar, or charged AAs. AAs had traditionally been classified as nutritionally essential (EAA, indispensable) or non-essential (NEAA, dispensable) depending on its synthesized *de novo* or not in an organism. Although well-defined metabolic pathways exist to allow cells to synthesize NEAAs *de novo*, highly proliferative cells require an exogenous supply of some of these AAs for optimal growth. Various reasons can underlie these needs. First, the *de novo* synthesis pathways may not always be able to provide adequate amounts of each AA to meet the demands of growing cells. Second, the increase of oxidative stress or different cellular stimuli can reprogram the metabolism and follow a shift *de novo* AA synthesis. And, conversely endothelial specific activation pathways that allow ECs to synthesize NEAAs can make them less dependent on exogenous supplies, but may also lead to a selective dependency on these endogenous alterations. NEAAs normally contribute for different biosynthetic purposes, including energy production, provision of carbon and nitrogen, and antioxidant defence. Under conditions of stress and catabolic states, NEEAs may become essential when the capacity of endogenous AA synthesis is exceeded. The proposal of this review is to provide a comprehensive overview of recent discoveries on endothelial AA metabolism and its role in the angiogenic processes.

2. Glutamine and asparagine

Glutamine is the most abundant free AA in blood plasma with physiological levels around 0.65 mM. Glutamine constitutes the major transporter of nitrogen from sites of glutamine synthesis (skeletal muscle, liver, and lung) to sites of utilization, which includes endothelium [19]. Several membrane transporters which differ in transport modes ensure glutamine homeostasis by coordinating its absorption, reabsorption, and delivery to tissues [20]. In ECs, around 90% of glutamine is transported by the sodium-dependent system ASC [21]. The uptake of glutamine in ECs is highly regulated. Indeed, in lung microvascular ECs, glucocorticoids and endotoxin inhibit glutamine uptake at low concentrations of glutamine. While at high glutamine concentrations (> 1 mM), endotoxin and glucocorticoids, exhibit no effect on glutamine consumption [21]. On the other hand, in human umbilical vein endothelial cells (HUVEC), endotoxin and cytokines do not significantly alter glutamine uptake, but direct activation of protein kinase C inhibits glutamine transportation [22]. Recently, Zhang et al. demonstrated that corneal endothelium uses glutamine to produce ATP to support Na^+-K^+ ATPase pump activity more than it supports biosynthesis [23].

Earlier studies demonstrated that the transamination of glutamine rather than oxidative dehydrogenation is the predominant pathway for the degradation of glutamine-derived glutamate in different ECs [24]. Glutaminase (GLS) is a phosphate-dependent enzyme that converts glutamine into glutamate and ammonia. Two isozymes of glutaminase have been identified in mammalian cells: kidney-type glutaminase encoded by the GLS gene and liver-type glutaminase encoded by GLS2 gene. Each gene produces two different variants. The *GLS* gene expresses the kidney-type glutaminase (KGA) and the C-terminal truncated glutaminase C (GAC) isoforms (both usually referred to as GLS1). The *GLS2* gene also expresses a longer isoform called LGA long or GAB, and a shorter one (LGA short). The N-termini of the GLS variants begin with a 16-residue sequence predicted to localize the protein to the mitochondria; and indeed, there is experimental consensus that GLS is localized to this organelle [25]. In contrast, there are no experimental data about the GLS2 subcellular localization in EC or other cells [26]. Both isozymes possess discrete tissue distribution, structural properties, enzyme kinetics, and molecular regulation. It has been recently reported that glutamine catabolism is essential for EC proliferation through its role in the replenishment of the Krebs cycle (Fig. 1). Without glutamine catabolism there is near complete loss of tricarboxylic acid (TCA) cycle intermediates and with no compensation from glucose-derived anaplerosis. Mechanistically, the addition of exogenous alpha-ketoglutarate (α-KG) replenishes TCA intermediates and rescues cellular growth [27]. Concomitantly, it was also reported that depriving ECs of glutamine or inhibiting glutaminase 1 (GLS1) caused vessel sprouting defects due to impaired proliferation and migration. In this study, the combination of the TCA cycle replenishment plus an asparagine supplementation were able to restore angiogenesis upon glutamine deprivation. Glutamine provided the nitrogen needed for the asparagine synthesis to sustain cellular homeostasis. While ECs can take up asparagine, silencing asparagine synthetase (ASNS, which converts glutamine-derived nitrogen and aspartate to asparagine) impaired EC sprouting even with the presence of glutamine [28].

The role of GLS2 in angiogenesis is less studied. It has been shown that EC-specific deletion of GLS2 produced an increase of reactive oxygen species (ROS) due to a drop of glutathione (GSH)/oxidized glutathione(GSSG) ratio [29]. Generation of genetic models with conditional alleles of *GLS2* will help in the characterization of GLS2 function in angiogenesis. Although glutamine catabolism is critical for TCA cycle, ATP production, biosynthesis of NEAA, and the control of GSH levels in ECs, whether GLS1 and GLS2 play a different role in angiogenesis is still unclear and need to be investigated further.

Although glutamine is more abundant than asparagine (blood asparagine concentration is ranging from 0.05 to 0.1 mM), Ras-transformed cancer cells can survive glutamine deprivation-mediated apoptosis, by using exogenous asparagine. This anti-apoptotic function depends on the ability of ASNS to maintain glutamine-dependent biosynthesis of asparagines [30,31]. ASNS converts aspartate and glutamine to asparagine and glutamate in an ATP-dependent reaction and is present in most mammalian organs, but varies widely in basal expression (Fig. 1) [32]. In agreement with Zhang et al. [31], glutamine starvation in ECs induces the expression of ASNS through activating transcription factor (ATF) 4 (ATF4) activation and EC-specific deletion of ASNS impairing EC sprouting [28]. Although asparagine presents a crucial role in endothelial homeostasis and its levels are strictly controlled by ASNS, the exact molecular mechanism by which asparagine regulates angiogenesis remains unexplored. These findings reveal a novel link between endothelial glutamine and asparagine metabolism in vessel sprouting.

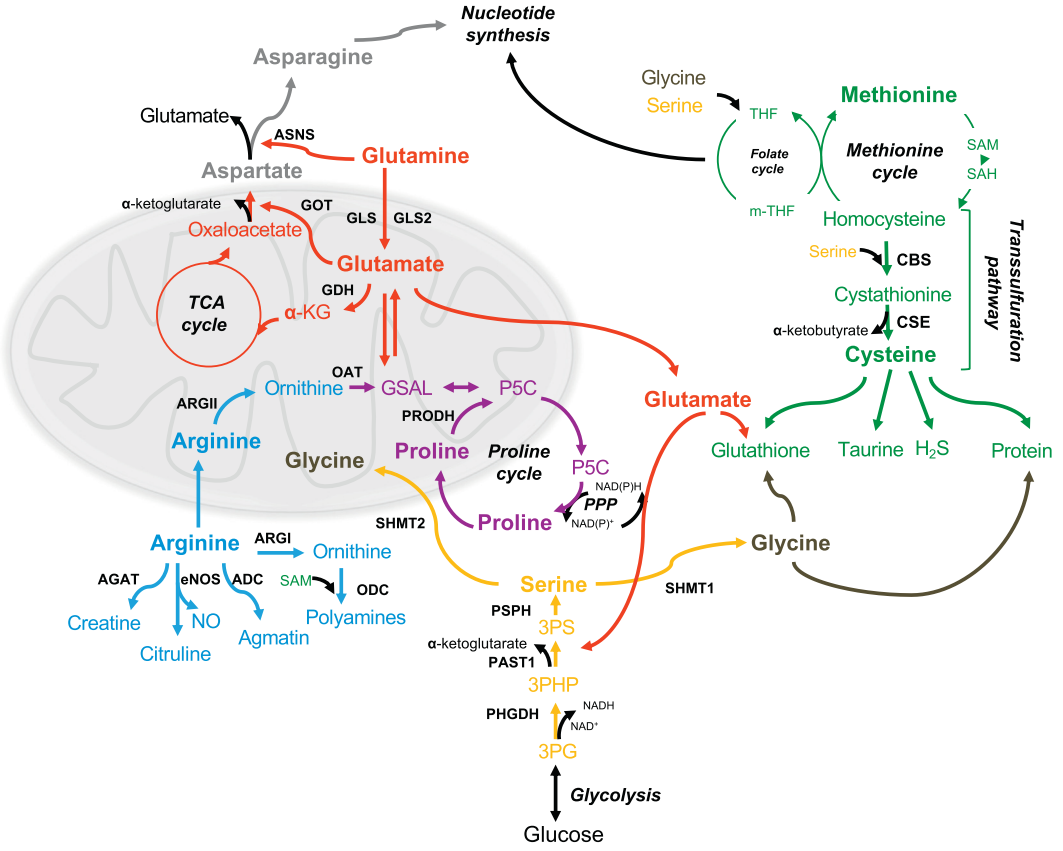

Fig. 1. Schematic overview of amino acid metabolism in endothelial cells. The connection between those NEAA and the enzymes that play an important role in ECs have been highlighted and discussed in this review. Metabolic pathways are in italic font. Enzymes are indicated in bold capital letters. The metabolism of glutamine, asparagine, arginine, serine, glycine, proline and sulfur amino acids are indicated in red, grey, blue, yellow, brown, violet, and green fonts, respectively. Abbreviations used: ADC, arginine decarboxylase; AGAT, arginine-glycine-amidinotransferase; α-KG, alpha-ketoglutarate; ARGI/II, arginase; ASNS, asparagine synthetase; CBS, cystathionine-β-synthase; CSE, cystathionine-γ-lyase; eNOS, endothelial nitric oxide synthase; GLS, glutaminase; GLS2, glutaminase 2; GSAL, L-glutamate-γ-semialdehyde; GSALDH, GSAL dehydrogenase; m-THF, 5-methyltetrahydrofolate; NAD + (P), oxidized nicotinamide adenine dinucleotide (phosphate); NADH(P), reduced nicotinamide adenine dinucleotide (phosphate); NO, nitric oxide; OAT, ornithine δ-amino acid transferase; ODC, ornithine decarboxylase; P5C, 1-pyrroline-5-carboxylate; P5CS, P5C synthase; PAST1; phosphoserine aminotransferase 1; PHGDH, phosphoglycerate dehydrogenase; PPP, pentose phosphate pathway; PRODH, proline dehydrogenase; PSPH, phosphoserine phosphatase; SAH, S-adenosylhomocysteine; SAM, S-adenosylmethionine; SHMT, hydroxymethyltransferase; TCA cycle, tricarboxylic acid cycle; THF, tetrahydrofolate.

3. Arginine

Arginine is a semi-essential amino acid with physiological levels of 0.1 mM. Arginine has been involved in numerous cellular metabolic and signaling pathways through the different products of its catabolism. Indeed, arginine is not only involved in protein synthesis, it is also a single substrate for endothelial nitric oxide synthase (eNOS) to produce the important vasoprotective molecule nitric oxide (NO) (Fig. 1) [33].

Extensive cancer research of arginine metabolic pathways led to the establishment of arginine-depriving enzymes as a potential anti-cancer strategy against arginine auxotrophic tumors [34,35]. But we must ask, what is the role of arginine in angiogenesis? Answering this question is not easy considering that arginine is metabolized into several products: NO, urea, creatine, polyamides, proline, glutamate, and agmatine which all have an impact on vessel formation in their own specific ways (Fig. 1) [36].

NO is a signaling molecule that regulates many vascular functions, such as vascular tone, blood pressure, neurotransmission, immune response, and oxidation-sensitive mechanisms [37]. Regarding angiogenesis, conflicting data suggests that NO can inhibit or stimulate angiogenesis depending upon the NO level and the angiogenic model used [38]. NO can also affect angiogenesis directly or through secondary messengers. Much of the data regarding NO's role in angiogenesis has been derived from *in vitro* experiments using NO donors and nonspecific NOS inhibitors. These experiments have shown that NO promotes new vessel growth through mediated NO upregulation of angiogenic factors, such as vascular endothelial growth factor (VEGF) [39], basic fibroblast growth factor (bFGF) [40], and matrix metalloproteinases during angiogenesis. *In vivo*, mice with genetic ablation of eNOS gene demonstrated a reduction in capillary growth in implanted collagen plugs, impaired angiogenic responses to murine hindlimb ischemia, and abnormal retinal development, and in all cases the phenotype could not be rescued by the administration of VEGF [41,42]. NO also affects the proliferation of endothelial and smooth muscle cells, through the cyclic guanosine monophosphate signaling pathway [43]. These findings show the crucial role of NO in angiogenesis.

Arginine is also converted into ornithine by arginase (ARG), after that, ornithine decarboxylase (ODC) converts ornithine into putrescine, a polyamine precursor [44]. Polyamines promote angiogenesis by increasing translation, protein synthesis, and gene expression [45]. Interestingly, experiments using an irreversible inhibitor of ODC showed an inhibition of B16 melanoma-induced angiogenesis in the chick embryo chorioallantoic membrane assay [46]. It has been shown that arginine promotes wound healing and functions as a secretagogue, stimulating the release of insulin-like growth factor 1 (IGF-1), insulin, and prolactin [47]. Taken together, the above-mentioned findings show that arginine plays a crucial role regulating the angiogenic process through different products of its catabolism.

4. Serine

Serine can be taken up into the cells using different transporters or can be synthesized by the cells. The serine synthesis pathway (SSP) is one of many side branches of glycolysis. Starting with the glycolytic intermediate, 3-phosphoglycerate (3PG), PHGDH catalyses the NAD^+-dependent conversion of 3PG into 3-phosphohydroxypyruvate (3PHP), which is then converted into phosphoserine by phosphoserine aminotransferase 1 (PSAT1). This is a transamination reaction that uses glutamate-derived nitrogen and produces α-KG. Serine is then generated from phosphoserine through phosphoserine phosphatase (PSPH) (Fig. 1) [10]. Serine is required for a number of biosynthetic and signaling pathways, including the synthesis of other AAs and the production of phospholipids such as sphingolipids and phosphatidylserine. Serine is also a major donor of one-carbon units to the folate cycle and serine-derived one-carbon units are used for the *de novo* synthesis of adenosine, guanosine, thymidylate, and for remethylation of homocysteine to support the methionine cycle (Fig. 1). Serine is also a precursor for the synthesis of glycine and cysteine through the transsulfuration pathway (TSP), which is essential for hydrogen sulphide (H_2S) and GSH production (Fig. 1). Serine metabolism function has been recently addressed in angiogenesis. Neonatal mouse deficiency for PHGDH in ECs have shown to suffer lethal vascular defects due to the reduction of heme synthesis which is required to maintain mitochondrial respiration and homeostasis [48]. The SSP is vital for ECs, as PHGDH loss induces apoptosis, even without external serine deprivation. In addition, Gopalakrishnan and colleagues have shown that serine elicits a substantial antihypertensive effect in the NO-compromised state [49]. It is conceivable to believe that ECs are dependent on serine being able to use part of their glucose to synthetize serine by PHGDH. Indeed, PHGDH silencing also importantly affect ROS levels in tumor ECs supporting a role for this metabolic branch in regulating endothelial redox homeostasis [50].

5. Glycine

Glycine can be generated by serine *via* serinehydroxymethyltransferase (SHMT) and as such feeds one-carbon metabolism, which is essential for nucleotide synthesis and the protection from oxidative stress (Fig. 1) [51]. The mitochondrial SHMT (SHMT2) appears to be ubiquitous and responsible for the bulk of glycine synthesis in most cell types, whereas the cytosolic SHMT (SHMT1) occurs primarily in the liver and kidneys and is less active in catalyzing the conversion of serine to glycine compared to SHMT2 [52]. Whether glycine, the simplest NEAA, which participates in the synthesis of proteins and many physiologically important functions, directly promotes angiogenesis is unclear. Amin et al., investigated the effects of glycine on angiogenesis during embryogenesis showing that exogenous glycine inhibited angiogenesis by > 50% in chorioallantoic membrane assay [53]. It has been found that dietary glycine is a potent anti-angiogenic agent that can reduce wound healing and tumor growth through the reduction of inducible NOS (iNOS) expression [54]. Glycine blunts VEGF-mediated EC proliferation supporting a negative role for the glycine receptor in angiogenesis [55]. On the contrary, it has been shown that glycine promote angiogenesis both *in vitro* and *in vivo* [56]. Here, the authors demonstrated that VEGF stimulation increased intracellular glycine levels in ECs by activating the glycine transporter 1. How glycine directly affect angiogenesis is still unclear although the increase in mitochondrial function have been shown after glycine treatment of ECs *in vitro*. In addition, glycine might play an important role in endothelial redox homeostasis, in regards to is being a precursor of GSH in ECs and other tissues (Fig. 1).

6. Proline

Proline, a unique secondary AA, is required for protein biosynthesis but it has a critical role in cellular bioenergetics, osmoregulation, stress protection, cellular signaling processes such us apoptosis and cancer cell metabolism [57–59]. Proline is synthesized from ornithine or glutamate, with both precursors leading to L-glutamate-γ-semialdehyde (GSAL), an intermediate that spontaneously cyclizes to 1-pyrroline-5-carboxylate (P5C). The formation of GSAL from ornithine is catalyzed by ornithine δ-amino acid transferase (OAT), while the route to GSAL from glutamate requires the bifunctional enzyme P5C synthase (P5CS). The final step for both proline biosynthetic routes is the reduction of P5C to proline catalyzed by NAD(P)H-dependent P5CR. In humans, P5CR is known as PYCR, with isoforms PYCR1 and PYCR2 in the mitochondrion and isoform PYCRL in the cytosol. On the other hand, proline is broken down by proline dehydrogenase (PRODH) and GSAL dehydrogenase (GSALDH) to P5C in the mitochondria. Considering that P5C is not only the precursor of proline but also as its immediate degradative product constitutes a catalytic cycle transferring reducing the potential of it into mitochondrial and the cycling of proline-P5C participates in a metabolic interlock with the PPP (Fig. 1).

In ECs, elevated expression of ARGI or ARGII increased the production of both proline and glutamate from arginine. It has been reported that the production of proline is greater than glutamate from arginine. Considering that proline is a major AA for the synthesis of collagen and the generation of extracellular matrix, this increase in proline synthesis from arginine may play an important role in vascular remodeling [60]. It is clear that the proline metabolic axis can serve as a scaffold upon which a variety of regulatory mechanisms are integrated. It is unclear how proline metabolism may contribute to vessel growth.

7. Sulfur amino acids: methionine, cysteine, homocysteine and taurine

Sulfur amino acids (SAA) are amino acids which contain sulfhydryl (-SH) and include methionine, cysteine, homocysteine, and taurine. Some of them play a crucial role in chronic cardiovascular disease and diabetes [61,62]. Among the SAAs, methionine and cysteine are considered as the primary SAAs and are the only proteogenic. Methionine is an indispensable AA in mammals since it cannot be synthesized in amounts sufficient to maintain the normal growth of mammals. Whereas, cysteine is a semi-essential AA since it can be produced through TSP from methionine and serine (Fig. 1) [63]. While dietary sources are required only for methionine, both methionine and cysteine are common constituents in most dietary proteins. Nevertheless, the deficiency and excess of methionine and cysteine in the diet affect the normal growth of tissues and blood vessels [64]. Both methionine and cysteine contain thiol groups making them extremely sensitive to almost all forms of reactive oxygen species, which thus makes them critical in controlling redox reactions in cells [65].

Methionine serves as a major component for protein synthesis. In addition to this, methionine contributes to various metabolic processes, including being a major methyl group donor as well as a precursor of sulfur compounds, including cysteine, taurine, and glutathione (Fig. 1). Through transmethylation reactions, its methyl group serves as a critical intermediate acting both as the methyl group donor for nucleic acid intermediates (transmethylation reactions), but, also as methyl acceptor from 5-methylTHF and folate (remethylationreactions) [51]. During the donation of its methyl group to multiple different methyl acceptor (transmethylation), methionine is converted to S-adenosylmethionine (SAM) and S-adenosylhomocysteine (SAH) and then to homocysteine. Homocysteine can be either reconverted in methionine (remethylation) or metabolize to cysteine *via* TSP. Here, homocysteine is irreversibly catabolized into cysteine in a series of reactions catalyzed by the transsulphuration enzymes, cystathionine-β-synthase (CBS) and cystathionine-γ-lyase (CGL or CSE) (Fig. 1).

Cysteine is a critical component of all proteins and essential to regulate protein secondary structure and conformation. Cysteine is present in the active site of many enzymes and can be regulated by

redox reaction on its thiol group [62]. In addition to forming protein structures, cysteine is catabolized and used to form important metabolic redox cofactors, including GSH, H_2S, taurine, and sulfate. While methionine is considered the metabolic precursor for cysteine, only the sulfur atom from methionine is transferred to cysteines while the rest become α-KG; instead, the carbon skeleton of cysteine is donated by serine. Cysteine is not a precursor for methionine because of the irreversibility of the TSP reactions catalyzed by CBS and CSE.

Blockade of methionine and cysteine metabolism in ECs influences oxidative stress through the involvement of metabolic intermediates causing endothelial damage or inflammation [66]. Methionine is clearly accepted as a dietary indispensable AA, and cysteine as a dietary dispensable AA, which can be replaced entirely with dietary methionine in human adults. Recent work indicates that SAA restriction triggers angiogenesis *via* the GCN2/ATF4-mediated induction [64]. Mice kept in a diet with limited amounts of methionine and no cysteine, showed an increased vascular density in skeletal muscle. SAA restriction diet also renders these mice more prone to neovascularization in an experimental hindlimb ischemia model. Mechanistically, SAA restriction activate ATF4 that in turn is able to promote VEGF transcription and TSP activation leading to production of H_2S [64,67]. It has also been reported that methionine restriction in diet alleviated oxidative stress by reducing mitochondrial ROS production *in vivo* [68,69]. Accordingly, it has been shown that the supplementation of methionine mitigates the ROS-induced damage by increasing the levels of GSH [70]. Interestingly, cysteine supplementation to HUVEC cells can promote angiogenesis *in vitro* in a NO-dependent manner [71]. Cysteine is an indispensable AA for human corneal endothelial cells (HCECs) that have high aerobic metabolic activity. By supporting high demands for antioxidant power, cysteine protects HCECs from oxidative stress, and support their growth and survival [72].

Although not proteogenic, homocysteine and taurine are considered non-diet SAA. They are important AA intermediates of the methionine salvage pathway and cysteine catabolism that can exert independent functions [73]. Homocysteine is present in the plasma of normal adults at approximately 10 μM. Elevation of plasma levels, called hyperhomocysteinemia, is known to be an independent risk factor for cardiovascular disease in adults [61,74], as well as ischemic and hemorrhagic stroke in newborns and children [75–77]. Homocysteine plays an important role in angiogenesis but its function in ECs is still not clear. It has been found that homocysteine can be a potent inducer of the master angiogenic regulator, VEGF [78,79]. Homocysteine activates the endoplasmic reticulum (ER)-stress response pathway known as the unfolded protein response (UPR), that in turn induces the ER stress-responsive genes, GRP78, GADD153, and ATF4 [80,81]. The UPR represents a set of signaling cascades by which the ER communicates to the protein translation machinery and to the nucleus in order to balance the folding capacity of the ER with the protein processing demand [82]. The significance of the stress-induced ATF3 and ATF4 gene expression as metabolic sensor in angiogenesis were later on discovered to show that expression of VEGF can also be increased by other thiol-containing reductive compounds *via* activation of the same ATF4-dependent gene transcription pathway [83]. Several works have proposed that homocysteine inhibit angiogenesis *in vitro* and *in vivo* by blocking angiogenic receptors such as VEGFR and angiopoietins and at the same time perturbing cytoskeletal remodeling [84–89]. Clearly more work needs to be performed to identify the role of homocysteine on VEGF signaling and angiogenesis.

Taurine, a non-canonical AA, is abundant in many tissues. It is mostly found in free form within human plasma where its concentration ranges from 5 to 30 mM. Intracellular pool of taurine is around 50-200 μM. This non proteinogenic SAA is synthesized from cysteine that is then converted into cysteine sulfinic acid (CSA) by a dioxygenase. CSA is then decarboxylate by a sulfinoalanine decarboxylase to form hypotaurine. Hypotaurine is enzymatically oxidized to yield taurine by hypotaurine dehydrogenase. Alternatively, hypotaurine can also be generated by cysteamine *via* another dioxygenase. Mammalian taurine synthesis occurs in the pancreas *via* the cysteine sulfinic acid pathway. Despite the fact that the health effects of taurine are largely unknown, in recent years taurine has become a popular supplement and ingredient in commercially available energy drinks. The role of taurine in angiogenesis is unclear [90,91]. This molecule has been found to be particularly effective in the prevention of cardiovascular and cerebrovascular diseases [92]. A report suggests that extracellular taurine increased angiogenesis *in vitro* and *in vivo* by unknown mechanisms [93]. Taurine can provide protective action against endothelial dysfunction induced by a combination of hyperglycemia and/or oxidized low density lipoproteins [94,95]. Taurine treatment has been found to be beneficial for young male type 1 diabetes [96]. On the other hand, intracellular taurine through its antioxidant capacity can block angiogenesis in a tumor context [97]. The molecular mechanism (s) involved in the vascular effects of taurine is largely unknown and requires further investigations.

Although it is not a sulfur amino acid *per se*, an important product of SAA catabolism is H_2S. The gaseous signal transducer H_2S is a novel crucial player in angiogenesis [98]. H_2S is synthesized from cysteine *via* three enzymes: CBS, CSE, and 3-mercaptopyruvate sulfurtransferase (3-MST). The first two are cytosolic and vitamin B6-dependent and, then, directly use cysteine to form H_2S. The synthesis of H_2S by 3-MST (which is also located in the mitochondria) occurs in two steps: first cystathionine aminotransferase (CAT) converts cysteine into 3-mercaptopyruvate using α-KG as substrate, and second 3-MST uses 3-mercaptopyruvate to form H_2S and pyruvate. All three H_2S-producing enzymes are expressed by vascular cells [99–101]. H_2S, released by the desulfurization of cysteine has numerous important endothelial functions, including the regulation of cell cycle, antioxidant response, and metabolic reprogramming [98]. Little is currently known regarding the functional regulation of these enzymes in ECs. Reactive oxygen species and laminar shear flow have been shown to enhance the expression of CSE and 3-MST, respectively. It has also been proposed that H_2S can mitigate vascular aging *via* NAD-dependent deacetylase sirtuin-1 [102,103]. Indeed, recent work shows that H_2S and NAD-dependent deacetylase sirtuin-1 functions as a critical axis regulating angiogenesis with the potential to mitigate or reverse oxidative stress-induced and aging-related changes in vascular health using pharmacological agents [104].

Interestingly, SAA restriction promotes expression of the TSP enzyme CSE, resulting in an increased hydrogen sulfide production and protection from hepatic ischemia reperfusion injury [105]. SAA starvation can also trigger H_2S production which in turn reroutes endothelial metabolism towards glucose uptake, glycolysis, and PPP while inhibiting mitochondrial oxidative phosphorylation [64]. In this context, it would be interesting to test therapeutic strategies aim to support H_2S production (or release) for providing beneficial effects in age-related vascular diseases.

8. Conclusions and perspectives

The metabolism of AAs controls many aspects of normal angiogenesis, including energetic status, proliferation, cell signaling, gene expression, and redox balance. Important studies have also demonstrated the role of some AAs in pathological angiogenesis. Indeed, it has been reported that an increase of homocysteine is a risk factor for diabetic retinopathy [106]. Also, tumor angiogenesis is enhanced by arginine through polyamide production [107], and amino acid deprivation [108]. Altogether, these findings make the therapeutic targeting of the amino acid metabolism a promising perspective in pathological angiogenesis, such as cancer, retinopathies, and atherosclerosis [109].

In the last two decades, untargeted metabolite profiling approaches and metabolic flux analysis allowed researchers to deeply characterize energetic and carbon metabolism in quiescent and proliferative ECs. We assume that in the years to come the combination of genetics,

metabolomics, and chemical biology will open to new discoveries on how endothelial amino acid metabolism govern angiogenesis. While increasing knowledge on the endothelial metabolism had offered new treatment possibilities of vascular related diseases in the past, these novel studies on endothelial amino acid metabolism in physiological and pathological angiogenesis will rapidly emphasize the tight regulation of endothelial metabolism based on microenvironmental condition for the future.

Conflict of interest

The authors declare no conflict of interest.

Acknowledgements

We thank Ellen Jane Corcoran for editorial and language assistance. This work is supported by the ERC-CoG647057 and AIRC Grant 20119 to MMS.

References

[1] W. Risau, Mechanisms of angiogenesis, Nature 386 (1997) 671–674.
[2] E.A. Logsdon, S.D. Finley, A.S. Popel, F. MacGabhann, A systems biology view of blood vessel growth and remodelling, J. Cell. Mol. Med. 18 (2014) 1491–1508.
[3] K. Rohlenova, K. Veys, I. Miranda-Santos, K. De Bock, P. Carmeliet, Endothelial cell metabolism in health and disease, Trends Cell Biol. 28 (2017) 224–236.
[4] L. Treps, L. Conradi, U. Harjes, P. Carmeliet, Manipulating angiogenesis by targeting endothelial metabolism: hitting the engine rather than the drivers — a new perspective ? Pharmacol. Rev. 68 (2016) 872–887.
[5] O. Culic, M.L. Gruwel, J. Schrader, Energy turnover of vascular endothelial cells, Am. J. Phys. 273 (1997) C205–C213.
[6] A. Zecchin, J. Kalucka, C. Dubois, P. Carmeliet, How endothelial cells adapt their metabolism to form vessels in tumors, Front. Immunol. 8 (2017) 1–8.
[7] M. Quintero, S.L. Colombo, A. Godfrey, S. Moncada, Mitochondria as signaling organelles in the vascular endothelium, Proc. Natl. Acad. Sci. 103 (2006) 5379–5384.
[8] E. Cheung, V. Olin-Sandoval, N. Grüning, The return of metabolism: biochemistry and physiology of the pentose phosphate pathway, Biol. Rev. Camb. Philos. Soc. 90 (2015) 927–963.
[9] D.C. Love, J.A. Hanover, The hexosamine signaling pathway: deciphering the “O-GlcNAc code”, Sci. Signal. (312) (2005) 1–14.
[10] M. Yang, K.H. Vousden, Serine and one-carbon metabolism in cancer, Nat. Rev. Cancer 16 (2016) 650–662.
[11] L.A. Teuwen, V. Geldhof, P. Carmeliet, How glucose, glutamine and fatty acid metabolism shape blood and lymph vessel development, Dev. Biol. (2017) 1–13.
[12] W. Yeh, C. Lin, W. Fu, Enhancement of glucose transporter expression of brain endothelial cells by vascular endothelial growth factor derived from glioma exposed to hypoxia, Mol. Pharmacol. 73 (2008) 170–177.
[13] G. Parra-Bonilla, D.F. Alvarez, M. Alexeyev, T. Stevens, Critical role for lactate dehydrogenase A in aerobic glycolysis that sustains pulmonary microvascular endothelial cell proliferation, Am. J. Physiol. Lung Cell Mol. Physiol. 299 (2010) 513–522.
[14] K. De Bock, M. Georgiadou, S. Schoors, A. Kuchnio, B.W. Wong, A.R. Cantelmo, et al., Role of PFKFB3-driven glycolysis in vessel sprouting, Cell 154 (2013) 651–663.
[15] A. Krützfeldt, R. Spahr, S. Mertens, B. Siegmund, H.M. Piper, Metabolism of exogenous substrates by coronary endothelial cells in culture, J. Mol. Cell. Cardiol. 22 (1990) 1393–1404.
[16] F. Patella, Z.T. Schug, E. Persi, L.J. Neilson, Z. Erami, D. Avanzato, et al., Proteomics-based metabolic modeling reveals that fatty acid oxidation (FAO) controls endothelial cell (EC) permeability, Mol. Cell. Proteomics 14 (2015) 621–634.
[17] S. Schoors, U. Bruning, R. Missiaen, K.C.S. Queiroz, Fatty acid carbon is essential for dNTP synthesis in endothelial cells, Nature 520 (2015) 192–197.
[18] J. Xiong, H. Kawagishi, Y. Yan, J. Liu, Q.S. Wells, L.R. Edmunds, et al., A metabolic basis for endothelial-to-mesenchymal transition, Mol. Cell 69 (2018) 689–698.e7.
[19] N.P. Curthoys, Regulation of glutaminase activity and glutamine metabolism, Annu. Rev. Nutr. 15 (1995) 133–159.
[20] G.E. Mann, D.L. Yudilevich, L. Sobrevia, Regulation of amino acid and glucose transporters in endothelial and smooth muscle cells, Physiol. Rev. 83 (2003) 183–252.
[21] M. Pan, M. Wasa, U. Ryan, W. Souba, Inhibition of pulmonary microvascular endothelial glutamine transport by glucocorticoids and endotoxin, J. Parenter. Enter. Nutr. 19 (1995) 477–481.
[22] M. Pan, M. Wasa, W. Souba, Protein kinase C activation inhibits glutamate transport by endothelial cells, J. Surg. Res. 58 (1995) 630–635.
[23] W. Zhang, H. Li, D.G. Ogando, S. Li, M. Feng, F.W. Price, et al., Glutaminolysis is essential for energy production and ion transport in human corneal endothelium, EBioMedicine 16 (2017) 292–301.
[24] G. Wu, T.E. Haynes, H. Li, C.J. Meininger, Glutamine metabolism in endothelial cells: ornithine synthesis from glutamine via pyrroline-5-carboxylate synthase, Comp. Biochem. Physiol. 126 (2000) 115–123.
[25] A. Cassago, A.P.S. Ferreira, I.M. Ferreira, C. Fornezari, E.R.M. Gomes, K.S. Greene, et al., Mitochondrial localization and structure-based phosphate activation mechanism of Glutaminase C with implications for cancer metabolism, Proc. Natl. Acad. Sci. 109 (2012) 1092–1097.
[26] W.P. Katt, M.J. Lukey, R.A. Cerione, A tale of two glutaminases: homologous enzymes with distinct roles in tumorigenesis, Future Med. Chem. 9 (2017) 223–243.
[27] B. Kim, J. Li, C. Jang, Z. Arany, Glutamine fuels proliferation but not migration of endothelial cells, EMBO J. 36 (2017) 2321–2333.
[28] H. Huang, S. Vandekeere, J. Kalucka, L. Bierhansl, A. Zecchin, U. Brüning, et al., Role of glutamine and interlinked asparagine metabolism in vessel formation, EMBO J. 36 (2017) 2334–2352.
[29] S. Suzuki, T. Tanaka, M.V. Poyurovsky, H. Nagano, T. Mayama, S. Ohkubo, et al., Phosphate-activated glutaminase (GLS2), a p53-inducible regulator of glutamine metabolism and reactive oxygen species, Proc. Natl. Acad. Sci. 107 (2010) 7461–7466.
[30] N.N. Pavlova, S. Hui, J.M. Ghergurovich, J. Fan, A.M. Intlekofer, R.M. White, et al., As Extracellular Glutamine Levels Decline, Asparagine Becomes an Essential Amino Acid, Cell Metab. 27 (2018) 428–438.e5.
[31] J. Zhang, J. Fan, S. Venneti, J.R. Cross, T. Takagi, H. Djaballah, et al., Asparagine plays a critical role in regulating cellular adaptation to glutamine depletion, Mol. Cell 56 (2015) 205–218.
[32] C.L. Lomelino, J.T. Andring, R. McKenna, M.S. Kilberg, Asparagine synthetase: function, structure, and role in disease, J. Biol. Chem. 292 (2017) 19952–19958.
[33] S.M. Morris, Recent advances in arginine metabolism: roles and regulation of the arginases, Br. J. Pharmacol. 157 (2009) 922–930.
[34] M.M. Phillips, M.T. Sheaff, P.W. Szlosarek, Targeting arginine-dependent cancers with arginine-degrading enzymes: opportunities and challenges, Cancer Res. Treat. 45 (2013) 251–262.
[35] M.D. Patil, J. Bhaumik, S. Babykutty, U.C. Banerjee, D. Fukumura, Arginine dependence of tumor cells: targeting a chink in cancer's armor, Oncogene 35 (2016) 4957–4972.
[36] S.M. Morris, Arginine metabolism in vascular biology and disease, Vasc. Med. 10 (2005) 83–87.
[37] D. Tousoulis, A.-M. Kampoli, C. Tentolouris, N. Papageorgiou, C. Stefanadis, The role of nitric oxide on endothelial function, Curr. Vasc. Pharmacol. 10 (2012) 4–18.
[38] J.P. Cooke, NO and angiogenesis, Atheroscler. Suppl. 4 (2003) 53–60.
[39] H. Kimura, H. Esumi, Reciprocal regulation between nitric oxide and vascular endothelial growth factor in angiogenesis, Acta Biochim. Pol. 50 (2003) 49–59.
[40] P. Voisine, J. Li, C. Bianchi, T.A. Khan, M. Ruel, S.H. Xu, et al., Effects of L-arginine on fibroblast growth factor 2-induced angiogenesis in a model of endothelial dysfunction, Circulation 112 (2005) 202–208.
[41] D. Fukumura, T. Gohongi, A. Kadambi, Y. Izumi, J. Ang, C.O. Yun, et al., Predominant role of endothelial nitric oxide synthase in vascular endothelial growth factor-induced angiogenesis and vascular permeability, Proc. Natl. Acad. Sci. U. S. A. 98 (2001) 2604–2609.
[42] J.M. Ha, S.H. Baek, Y.H. Kim, S.Y. Jin, H.S. Lee, S.J. Kim, et al., Regulation of retinal angiogenesis by phospholipase C-β3 signaling pathway, Exp. Mol. Med. 48 (2016) e240.
[43] C. Napoli, G. Paolisso, A. Casamassimi, M. Al-Omran, M. Barbieri, L. Sommese, et al., Effects of nitric oxide on cell proliferation: novel insights, J. Am. Coll. Cardiol. 62 (2013) 89–95.
[44] K. Soda, The mechanisms by which polyamines accelerate tumor spread, J. Exp. Clin. Cancer Res. 30 (2011) 95–104.
[45] A.E. Pegg, Mammalian polyamine metabolism and function, IUBMB Life 61 (2009) 880–894.
[46] M. Takigawa, M. Takigawa, M. Enomoto, M. Enomoto, Y. Nishida, Y. Nishida, et al., Tumor angiogensis and polyamines: alpha-difluoromethylornithine, an irreversible inhibitor of ornithine decarboxylase, inhibits B16 melanoma-induced angiogensis in ovo and the proliferation of vascular endothelial cells in vitro, Cancer Res. 50 (1990) 4131–4138.
[47] D.S. Lind, Arginine and cancer, J. Nutr. 134 (2004) 2837S–2841S.
[48] S. Vandekeere, C. Dubois, J. Kalucka, M.R. Sullivan, M. García-Caballero, J. Goveia, et al., Serine synthesis via PHGDH is essential for heme production in endothelial cells, Cell Metab. 28 (2018) 1–15.
[49] R.C. Mishra, S. Tripathy, D. Quest, K.M. Desai, J. Akhtar, I.D. Dattani, et al., L-Serine lowers while glycine increases blood pressure in chronic L-NAME-treated and spontaneously hypertensive rats, J. Hypertens. 26 (2008) 2339–2348.
[50] A.R. Cantelmo, L.C. Conradi, A. Brajic, J. Goveia, J. Kalucka, A. Pircher, et al., Inhibition of the Glycolytic Activator PFKFB3 in Endothelium Induces Tumor Vessel Normalization, Impairs Metastasis, and Improves Chemotherapy, Cancer Cell 30 (2016) 968–985.
[51] J.W. Locasale, Serine, glycine and one-carbon units: Cancer metabolism in full circle, Nat. Rev. Cancer 13 (2013) 572–583.
[52] W. Wang, Z. Wu, Z. Dai, Y. Yang, J. Wang, G. Wu, Glycine metabolism in animals and humans: Implications for nutrition and health, Amino Acids 45 (2013) 463–477.
[53] J. Li, W.R. Chao, M.W. Dewhirst, Z.A. Haroon, Dietary glycine inhibits angiogenesis during wound healing and tumor growth, Cancer Res. Ther. 2 (2003) 173–178.
[54] M.L. Rose, J. Madren, H. Bunzendahl, R.G. Thurman, Dietary glycine inhibits the growth of B16 melanoma tumors in mice, Carcinogenesis 20 (1999) 793–798.

[55] H. Bruns, D. Kazanavicius, D. Schultze, Saeedi M. Al, K. Yamanaka, K. Strupas, et al., Glycine inhibits angiogenesis in colorectal cancer: role of endothelial cells, Amino Acids 48 (2016) 2549–2558.
[56] D. Guo, C.E. Murdoch, H. Xu, H. Shi, D.D. Duan, A. Ahmed, et al., Vascular endothelial growth factor signaling requires glycine to promote angiogenesis, Sci. Rep. 7 (2017) 1–10.
[57] X. Liang, M.B. Dickman, D.F. Becker, Proline biosynthesis is required for endoplasmic reticulum stress tolerance in Saccharomyces cerevisiae, J. Biol. Chem. 289 (2014) 27794–27806.
[58] N. Krishnan, M.B. Dickman, D.F. Becker, Proline modulates the intracellular redox environment and protects mammalian cells against oxidative stress, Free Radic. Biol. Med. 44 (2008) 671–681.
[59] J.M. Phang, Proline metabolism in cell regulation and cancer biology: recent advances and hypotheses, Antioxid. Redox Signal. 5 (2017) 1–45.
[60] H. Li, C.J. Meininger, J.R. Hawker, T.E. Haynes, D. Kepka-Lenhart, S.K. Mistry, et al., Regulatory role of arginase I and II in nitric oxide, polyamine, and proline syntheses in endothelial cells, Am. J. Physiol. Endocrinol. Metab. 280 (2001) E75–E82.
[61] H. Refsum, M. Uealand, Plasma homocysteine and cardiovascular disease, Annu. Rev. Med. 49 (1998) 31–62.
[62] R.N. Carter, N.M. Morton, Cysteine and hydrogen sulphide in the regulation of metabolism: Insights from genetics and pharmacology, J. Pathol. 238 (2016) 321–332.
[63] M.H. Stipanuk, I. Ueki, Dealing with methionine/homocysteine sulfur: cysteine metabolism to tauriene and inorgani sulfur, J. Inherit. Metab. Dis. 34 (2011) 17–32.
[64] A. Longchamp, T. Mirabella, A. Arduini, M.R. MacArthur, A. Das, J.H. Treviño-Villarreal, et al., Amino acid restriction triggers angiogenesis via GCN2/ATF4 regulation of VEGF and H2S production, Cell 173 (2018) 117–129.e14.
[65] P. Bin, R. Huang, X. Zhou, Oxidation resistance of the sulfur amino acids: Methionine and cysteine, Biomed. Res. Int. 2017 (2017) 1–6.
[66] S.V. Vijaya Lakshmi, S.M. Naushad, Y. Rupasree, D. Seshagiri Rao, V.K. Kutala, Interactions of 5'-UTR thymidylate synthase polymorphism with 677C → T methylene tetrahydrofolate reductase and 66A → G methyltetrahydrofolate homocysteine methyl-transferase reductase polymorphisms determine susceptibility to coronary artery disease, J. Atheroscler. Thromb. 18 (2011) 56–64.
[67] C. Szabõ, Hydrogen sulphide and its therapeutic potential, Nat. Rev. Drug Discov. 6 (2007) 917–935.
[68] S. Maddineni, S. Nichenametla, R. Sinha, R.P. Wilson, J.P. Richie, Methionine restriction affects oxidative stress and glutathione-related redox pathways in the rat, Exp. Biol. Med. 238 (2013) 392–399.
[69] P. Caro, J. Gomez, I. Sanchez, A. Naudi, V. Ayala, M. López-Torres, et al., Forty percent Methionine Restriction Decreases Mitochondrial Oxygen Radical Production and Leak at complex I during Forward Electron Flow and Lowers Oxidative damage to Proteins and Mitochondrial DNA in Rat Kidney and Brain Mitochondria, Rejuvenation Res. 12 (2009) 421–434.
[70] Vesco A.P. Del, E. Gasparino, D.O. Grieser, V. Zancanela, F.R.S. Gasparin, J. Constantin, et al., Effects of methionine supplementation on the redox state of acute heat stress – exposed quails 1, J. Anim. Sci. 92 (2014) 806–815.
[71] C. Coletta, A. Papapetropoulos, K. Erdelyi, G. Olah, K. Modis, P. Panopoulos, et al., Hydrogen sulfide and nitric oxide are mutually dependent in the regulation of angiogenesis and endothelium-dependent vasorelaxation, Proc. Natl. Acad. Sci. 109 (2012) 9161–9166.
[72] N. Okumura, R. Inoue, K. Kakutani, M. Nakahara, S. Kinoshita, J. Hamuro, et al., Corneal endothelial cells have an absolute requirement for cysteine for survival, Cornea 0 (2017) 1–7.
[73] J. Selhub, J. Mayer, Homocysteine metabolism, Annu. Rev. Nutr. 19 (1999) 217–246.
[74] K. McCully, Homocysteine and vascular disease, Nat. Med. 2 (1996) 386–389.
[75] M. Hogeveen, H.J. Blom, M. Van Amerongen, B. Boogmans, I.M. Van Beynum, M. Van De Bor, Hyperhomocysteinemia as risk factor for ischemic and hemorrhagic stroke in newborn infants, J. Pediatr. 141 (2002) 429–431.
[76] I. Van Beynum, J. Smeitink, M. Den Heijer, M. Poele Pothoff, H. Blom, Hyperhomocysteinemia. A risk factor for ischemia stroke in children, Circulation 99 (1999) 2070–2072.
[77] J. Loscalzo, Homocysteine-mediated thrombosis and angiostasis in vascular pathobiology, J. Clin. Invest. 119 (2009) 3203–3205.
[78] Y. Cai, C. Zhang, T. Nawa, T. Aso, M. Tanaka, S. Oshiro, et al., Homocysteine-responsive ATF3 gene expression in human vascular endothelial cells: activation of c-Jun NH(2)-terminal kinase and promoter response element, Blood 96 (2000) 2140–2148.
[79] N. Roybal, S. Yang, C. Sun, D. Hurtado, D. Vander Jagt, T. Townes, et al., Homocysteine increases the expression of VEGF by a mechanism involving endoplasmic reticulum stress and transcription factor ATF4, JBC 01 (2004) 1–39.
[80] Outinen P A, S.K. Sood, S.I. Pfeifer, S. Pamidi, T.J. Podor, J. Li, et al., Homocysteine-induced endoplasmic reticulum stress and growth arrest leads to specific changes in gene expression in human vascular endothelial cells, Blood 94 (1999) 959–967.
[81] G.H. Werstuck, S.R. Lentz, S. Dayal, G.S. Hossain, S.K. Sood, Y.Y. Shi, et al., Homocysteine-induced endoplasmic reticulum stress causes dysregulation of the cholesterol and triglyceride biosynthetic pathways, J. Clin. Invest. 107 (2001) 1263–1273.
[82] L. Ellgaard, A. Helenius, Quality control in the endoplasmic reticulum, Nat. Rev. Mol. Cell Biol. 4 (2003) 181–191.
[83] F. Binet, P. Sapieha, ER stress and angiogenesis, Cell Metab. 22 (2015) 560–575.
[84] J. Duan, T. Murohara, H. Ikeda, A. Katoh, S. Shintani, K. Sasaki, et al., Hypercholesterolemia Inhibits Angiogenesis in Response to Hindlimb Ischemia, Nitric Oxide Depend. Mech. 102 (2000) (Iii-370-Iii-376).
[85] A.T. Jacovina, A.B. Deora, Q. Ling, M.J. Broekman, D. Almeida, C.B. Greenberg, et al., Homocysteine inhibits neoangiogenesis in mice through blockade of annexin A2-dependent fibrinolysis, J. Clin. Invest. 119 (2009) 3384–3394.
[86] Y. Nagai, H. Tasaki, H. Takatsu, Nihei S. Ichi, K. Yamashita, T. Toyokawa, et al., Homocysteine inhibits angiogenesis in vitro and in vivo, Biochem. Biophys. Res. Commun. 281 (2001) 726–731.
[87] A.M. Oosterbaan, E.A.P. Steegers, N.T.C. Ursem, The effects of homocysteine and folic acid on angiogenesis and VEGF expression during chicken vascular development, Microvasc. Res. 83 (2012) 98–104.
[88] D. Zhang, Y. Chen, X. Xie, J. Liu, Q. Wang, W. Kong, et al., Homocysteine activates vascular smooth muscle cells by DNA demethylation of platelet-derived growth factor in endothelial cells, J. Mol. Cell. Cardiol. 53 (2012) 487–496.
[89] L. Pan, G. Yu, J. Huang, X. Zheng, Y. Xu, Homocysteine inhibits angiogenesis through cytoskeleton remodeling, Biosci. Rep. 37 (2017) 1–10.
[90] W. Abebe, M.S. Mozaffari, Role of taurine in the vasculature: an overview of experimental and human studies, Am. J. Cardiovasc. Dis. 1 (2011) 293–311.
[91] R. Huxtable, Physiological actions of taurine, Physiol. Rev. 72 (1992) 101–163.
[92] O.P. Wójcik, K.L. Koenig, A. Zeleniuch-Jacquotte, M. Costa, Y. Chen, The potential protective effects of taurine on coronary heart disease, Atherosclerosis 208 (2010) 19–25.
[93] Y.Y. Baek, D.H. Cho, J. Choe, H. Lee, D. Jeoung, K.S. Ha, et al., Extracellular taurine induces angiogenesis by activating ERK-, Akt-, and FAK-dependent signal pathways, Eur. J. Pharmacol. 674 (2012) 188–199.
[94] G. Ulrich-Merzenich, H. Zeitler, H. Vetter, R.R. Bhonde, Protective effects of taurine on endothelial cells impaired by high glucose and oxidized low density lipoproteins, Eur. J. Nutr. 46 (2007) 431–438.
[95] Q. di Wu, J.H. Wang, F. Fennessy, H.P. Redmond, D. Bouchier-Hayes, Taurine prevents high-glucose-induced human vascular endothelial cell apoptosis, Am. J. Physiol. Cell Physiol. 277 (1999) C1229–C1238.
[96] M.A. Moloney, R.G. Casey, D.H. O'Donnell, P. Fitzgerald, C. Thompson, D.J. Bouchier-Hayes, Two weeks taurine supplementation reverses endothelial dysfunction in young male type 1 diabetics, Diabetes Vasc. Dis. Res. 7 (2010) 300–310.
[97] N. Finnegan, H.P. Redmond, Bouchier-hayes DJ. Taurine attenuates recombinant interleukin-2 -activated, lymphocyte-mediated endothelial cell injury, Am. Cancer Soc. (1998) 186–199.
[98] N.L. Kanagy, C. Szabo, A. Papapetropoulos, Vascular biology of hydrogen sulfide, Am. J. Physiol. Cell Physiol. 312 (2017) C537–C549.
[99] A. Papapetropoulos, A. Pyriochou, Z. Altaany, G. Yang, A. Marazioti, Z. Zhou, et al., Hydrogen sulfide is an endogenous stimulator of angiogenesis, Proc. Natl. Acad. Sci. 106 (2009) 21972–21977.
[100] S. Saha, P.K. Chakraborty, X. Xiong, S.K.D. Dwivedi, S.B. Mustafi, N.R. Leigh, et al., Cystathionine β-synthase regulates endothelial function via protein S-sulfhydration, FASEB J. 30 (2016) 441–456.
[101] N. Shibuya, Y. Mikami, Y. Kimura, N. Nagahara, H. Kimura, Vascular endothelium expresses 3-mercaptopyruvate sulfurtransferase and produces hydrogen sulfide, J. Biochem. 146 (2009) 623–626.
[102] Z.-S. Jiang, Hydrogen sulfide prevents H2O2-induced senescence in human umbilical vein endothelial cells through SIRT1 activation, Mol. Med. Rep. 7 (2013) 1865–1870.
[103] T.V. Arumugam, B.K. Kennedy, H2S to Mitigate Vascular Aging: a SIRT1 connection, Cell 173 (2018) 8–10.
[104] A. Das, G.X. Huang, M.S. Bonkowski, A. Longchamp, C. Li, M.B. Schultz, et al., Impairment of an Endothelial NAD + -H2S Signaling Network is a Reversible Cause of Vascular Aging, Cell 173 (2018) 74–89.e20.
[105] C. Hine, E. Harputlugil, Y. Zhang, C. Ruckenstuhl, B.C. Lee, L. Brace, et al., Endogenous hydrogen sulfide production is essential for dietary restriction benefits, Cell 160 (2015) 132–144.
[106] S. Neugebauer, T. Baba, K. Kurokawa, A. Haushofer, W.M. Halbmayer, Defective homocysteine metabolism as a risk factor for diabetic retinopathy, Lancet 349 (1997) 473–474.
[107] L.E. Davel, M.A. Jasnis, E. De la Torre, T. Gotoh, M. Diament, G. Magenta, et al., Arginine metabolic pathways involved in the modulation of tumor-induced angiogenesis by macrophages, FEBS Lett. 532 (2002) 216–220.
[108] Y. Wang, Y. Ning, G.N. Alam, B.M. Jankowski, Z. Dong, J.E. Nör, et al., Amino Acid Deprivation Promotes Tumor Angiogenesis through the GCN2/ATF4 Pathway, Neoplasia 15 (2013) 989–997.
[109] M. Wang, R.J. Kaufman, Protein misfolding in the endoplasmic reticulum as a conduit to human disease, Nature 529 (2016) 326–335.