Journal of Clinical and Translational Hepatology **2023** vol. 11(3) | 675–681
DOI: 10.14218/JCTH.2022.00217

Review Article

Role of Hydrogen Sulfide and Hypoxia in Hepatic Angiogenesis of Portal Hypertension

Huaxiang Yang[1], Mingjie Tan[1], Zhuqing Gao[1], Shanshan Wang[1,2], Lingna Lyu[1] and Huiguo Ding[1*]

[1]Department of Gastroenterology and Hepatology, Beijing You'an Hospital Affiliated to Capital Medical University, Beijing, China; [2]Cell Biology Laboratory, Beijing Institute of Hepatology, Beijing, China

Received: 7 May 2022 | **Revised:** 23 August 2022 | **Accepted:** 21 September 2022 | **Published online:** 4 January 2023

Abstract

The pathogenesis of portal hypertension remains unclear, and is believed to involve dysfunction of liver sinusoidal endothelial cells (LSEC), activation of hepatic stellate cells (HSC), dysregulation of endogenous hydrogen sulfide (H_2S) synthesis, and hypoxia-induced angiogenic responses. H_2S, a novel gas transmitter, plays an important role in various pathophysiological processes, especially in hepatic angiogenesis. Inhibition of endogenous H_2S synthase by pharmaceutical agents or gene silencing may enhance the angiogenic response of endothelial cells. Hypoxia-inducible factor-1 (HIF-1) is the main transcription factor of hypoxia, which induces hepatic angiogenesis through up-regulation of vascular endothelial growth factor (VEGF) in HSC and LSEC. H_2S has also been shown to be involved in the regulation of VEGF-mediated angiogenesis. Therefore, H_2S and HIF-1 may be potential therapeutic targets for portal hypertension. The effects of H_2S donors or prodrugs on the hemodynamics of portal hypertension and the mechanism of H_2S-induced angiogenesis are promising areas for future research.

Citation of this article: Yang H, Tan M, Gao Z, Wang S, Lyu L, Ding H. Role of Hydrogen Sulfide and Hypoxia in Hepatic Angiogenesis of Portal Hypertension. J Clin Transl Hepatol 2023;11(3)675–681. doi: 10.14218/JCTH.2022.00217.

Introduction

Portal hypertension is defined as a portal pressure gradient (PPG) >6 mmHg or hepatic venous pressure gradient (HVPG) >5 mmHg.[1,2] The condition is often accompanied by gastroesophageal varices, ascites, and splenomegaly. The pathogenesis of portal hypertension involves multifaceted cellular and molecular mechanisms, including dysfunction of liver sinusoidal endothelial cells (LSEC), activation of hepatic stellate cells (HSC), dysregulation of endogenous hydrogen sulfide (H_2S) synthesis, and hypoxia-induced angiogenic responses.[3,4] In portal hypertension, hepatic sinusoidal vascular remodeling leads to impaired oxygen supply to liver parenchymal cells, resulting in the formation of hepatic hypoxic microenvironment (HHME). The HHME in turn leads to pathological angiogenesis as well as other adaptive changes in the liver.[5] Hypoxia-inducible factor-1 (HIF-1) is the main transcription factor of hypoxia response and the main regulator of oxygen homeostasis.[6] The interaction between HIF-1 and proangiogenic factors is an essential pathophysiological event in the process of angiogenesis induced by hypoxic conditions.[7,8]

H_2S is a new member of the gas transmitter family.[9] Recent studies have demonstrated the involvement of endogenous H_2S in regulating angiogenic responses.[9,10] Importantly, H_2S upregulates the expression of vascular endothelial growth factor (VEGF) and participates in the regulation of VEGF-mediated angiogenic signaling pathway.[11] HIF-1 and H_2S are both potential mediators of the angiogenic response in HHME, coregulating the development of portal hypertension. Therefore, this review focuses on the pathophysiological roles as well as mechanisms of H_2S and HIF-1 in regulating hepatic angiogenesis, providing potential therapeutic targets for the treatment of portal hypertension.

Adaptive changes in HHME

Tissue oxygen tension is a key factor in maintaining cell viability. The physiological gradient of oxygen tension in the hepatic lobules has a profound effect on the function of hepatic parenchymal cells. The unique dual blood supply system of the liver produces an oxygen partial pressure (pO2) in different liver zones, with a pO2 of 60–65 mmHg in the periportal region and 30–35 mmHg in the perivenous region.[12] Thus, periportal hepatocytes and perivenous hepatocytes differ in the expression of many enzymes involved in glucose metabolism, including insulin receptors, glucagon receptors, phosphoglycerate kinase, and pyruvate kinase.[13] Periportal hepatocytes are primarily responsible for oxidative metabolism, gluconeogenesis, and synthesis of urea and bile, while perivenous hepatocytes are the primary sites of glucose up-

Keywords: Hydrogen sulfide; Hypoxia; Hypoxia-inducible factor; Angiogenesis; Portal hypertension.

Abbreviations: 3-MST, 3-Mercaptopyruvate sulfotransferase; CBS, cystathionine beta synthase; CCl_4, carbon tetrachloride; cGMP, cyclic guanosine monophosphate; CSE, cystathionine gamma lyase; eIF2α, eukaryotic translation initiation factor 2α; eNOS, endothelial nitric oxide synthase; FGF-2, fibroblast growth factor 2; HHME, hepatic hypoxic microenvironment; HIF-1, hypoxia-inducible factor-1; H_2S, hydrogen sulfide; HSC, hepatic stellate cells; HVPG, hepatic venous pressure gradient; iNOS, inducible nitric oxide synthase; LSEC, liver sinusoidal endothelial cells; mTOR, mammalian target of rapamycin; NaHS, sodium hydrosulfide; NO, nitric oxide; PDGF, platelet-derived growth factor; PHD, prolyl hydroxylase; PI3K, phosphatidylinositol 3-kinase; PKG, protein kinase G; pO, oxygen partial pressure; Sp1, transcription factor specific protein 1; TNF-α, tumor necrosis factor-α; VEGF, vascular endothelial growth factor; VEGFR, vascular endothelial growth factor receptor; VHL, von Hippel-Lindau protein.

***Correspondence to:** Huiguo Ding, Director of Department of Gastroenterology and Hepatology, Beijing You'an Hospital affiliated to Capital Medical University, No.8 Xitoutiao, You An Men Wai Street, Fengtai District, Beijing 100069, China. ORCID: https://orcid.org/0000-0002-8716-4926. Tel: +86-10-83997155, Fax: +86-10-63295525, E-mail: dinghuiguo@ccmu.edu.cn

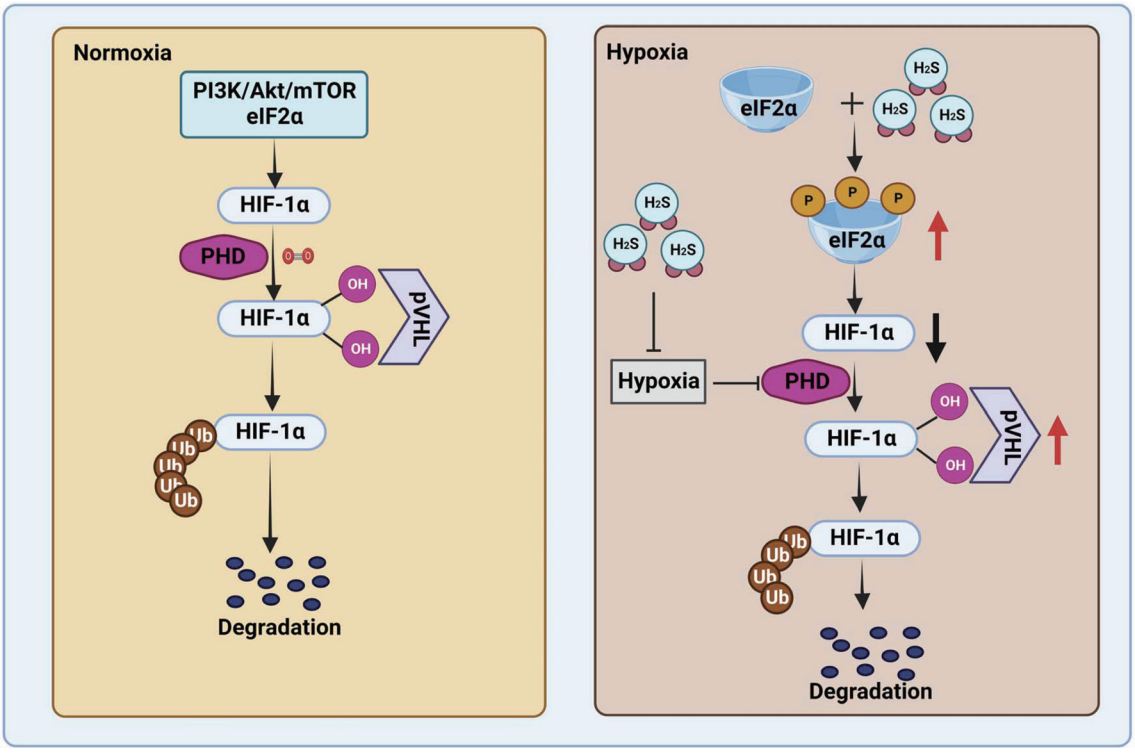

Fig. 1. Pathways of HIF-1α synthesis/degradation under normoxic and hypoxic conditions. Translation of HIF-1α protein under normoxic conditions is mainly dependent on the activation of PI3K/Akt/mTOR signaling pathway. HIF-1α is hydroxylated by PHD under normoxic conditions and subsequently binds to VHL protein to form a complex. The complex in turn recruits ubiquitin ligases to target HIF-1α for proteasomal degradation. Under hypoxic conditions, H_2S induces phosphorylation of eIF2α, thereby inhibiting HIF-1α translation. In addition, H_2S reverses hypoxia-induced inhibition of PHD activity and thus promotes the degradation of HIF-1α. HIF-1α, hypoxia-inducible factor 1α; PHD, prolyl hydroxylase; VHL, von Hippel-Lindau; H_2S, hydrogen sulfide; eIF2α, eukaryotic translation initiation factor 2α.

take, glutamine formation and metabolism.[14] Exposure of hepatocytes to different levels of oxygen tension affects their ability to respond to hypoxic microenvironments. Primary rat hepatocytes cultured under conditions approximating periportal oxygen tension (pO_2 of 60–65 mmHg) were able to survive briefly in a hypoxic environment, whereas hepatocytes cultured under conditions approximating perivenous oxygen tension (pO_2 of 30–35 mmHg) were intolerant to hypoxic stimulation and underwent rapid apoptosis.[12,15] This suggests that under hypoxic conditions such as increased hepatic metabolic demand, or tissue ischemia, perivenous hepatocytes may suffer greater injury when oxygen tension drops below a threshold level.

HIF-1, an oxygen-sensitive transcription factor, is the core factor mediating the hypoxic response.[16] HIF-1 is a heterodimer consisting of the hypoxia-inducible and oxygen-sensitive HIF-1α subunits and the constitutively expressed HIF-1β subunits.[17] HIF-1 heterodimers bind to hypoxia responsive elements in target genes, thereby enhancing transcription of target genes. Under normoxic conditions, it is difficult to detect HIF-1α protein in normal cells because of its rapid degradation. The two specific proline residues within the oxygen-dependent degradation domain in HIF-1α are hydroxylated by prolyl hydroxylase (PHD) under normoxic conditions.[18] Hydroxylated HIF-1α then binds to von Hippel-Lindau protein (VHL). This complex in turn recruits ubiquitin ligases to target HIF-1α for proteasomal degradation.[18] Translation of HIF-1α protein under normoxic conditions is dependent on activation of the PI3K-Akt-mTOR pathway and mitogen-activated protein kinase pathway.[19] Phosphorylation of eukaryotic translation initiation factor 2α (eIF2α) and inhibition of mTOR activity under hypoxic conditions are believed to be responsible for the inhibition of HIF-1α protein expression (Fig. 1).[19,20]

Interaction between hypoxia and H_2S

H_2S is believed to be the third gaseous signaling molecule which is widely expressed in mammalian cells and tissues.[21] Three enzymes are known to be involved in the production of endogenous H_2S, namely cystathionine beta synthase (CBS), cystathionine gamma lyase (CSE), and 3-Mercaptopyruvate sulfotransferase (3-MST).[9] An increasing body of evidence has revealed the cross-talk between HIF-1 and H_2S. In a study, knockdown of *CSE* was shown to affect H_2S production, which impaired the stability of HIF-1α, suggesting that endogenous H_2S is important for the regulation of HIF-1 signaling pathway.[22] However, another study found no effect of H_2S on HIF-1 level in EB8 cells under hypoxic conditions.[23] Moreover, treatment of cells with 1 mM NaHS, an exogenous H_2S donor, inhibited mitochondrial oxygen consumption, thereby enhancing oxygen levels in hypoxic cells. Under hypoxic conditions, H_2S inhibited the stability of HIF-1α protein.[23] The authors concluded that H_2S promoted the degradation of HIF-1α under hypoxic conditions due to NaHS-induced inhibition of mitochondrial oxygen consumption. Notably, the study used 1 mM NaHS, a high concentration of H_2S that is clearly outside the physiological range of endogenous H_2S (10–100 μM) and was potentially toxic to cells.

In a study using 10–100 μM NaHS, H_2S significantly reduced HIF-1α protein levels under hypoxic conditions.[24] Fur-

ther, addition of the translation inhibitor actidione blocked the effect of NaHS on HIF-1α protein levels, indicating that H_2S mediated inhibition of HIF-1α. The findings demonstrated that the key mechanism of H_2S-induced HIF-1α downregulation was inhibition of HIF-1α translation rather than through the ubiquitin-proteasome degradation pathway.[24] eIF2α was shown to be a key regulatory molecule for initiation of translation in eukaryotic cells. Phosphorylation of eIF2α was shown to prevent the formation of translational initiation complex and thus inhibit protein translation.[25] H_2S-induced downregulation of HIF-1α was partially reversed in *eIF2α* knockdown cells under hypoxic conditions.[24] The findings suggest that inhibition of HIF-1α translation under hypoxic conditions may be related to H_2S-induced eIF2α phosphorylation (Fig. 1).

Although the accumulation of HIF-1α in cells is largely dependent on VHL, new evidence suggests that VHL-independent degradation pathways may play an equally important role in regulating HIF-1α.[26,27] Cysteine synthase-1 was homologous to CBS, which had a negative regulatory effect on EGL-9 family hypoxia-inducible factor-1, thereby enhancing the stability of HIF-1.[27] Thus, H_2S may lead to the accumulation of HIF-1 under hypoxic conditions by promoting the interaction between EGL-9 family hypoxia-inducible factor (HIF) and cysteine synthase-1.[27] Although this pathway appears to be less dependent on intracellular oxygen levels, further studies are required to elucidate the potential involvement of this pathway in the regulation of HIF-1 levels by H_2S under hypoxic conditions.

Molecular mechanisms of angiogenesis in HHME

Studies have demonstrated the occurrence of pathological angiogenesis throughout the progression of portal hypertension.[28] Furthermore, hepatic angiogenesis and the concomitant vascular remodeling play a significant role in the development of portal hypertension and its associated complications.[29]

Role of hypoxia/HIF in hepatic angiogenesis

Hypoxia is one of the most potent stimuli known to drive angiogenesis.[30] The HIF-mediated hypoxic response results in enhanced transcriptional activity of a range of cell surface receptors and target genes, which enhances the sensitivity of endothelial cells to angiogenic factors and accelerates the process of liver cirrhosis.[31] The process of liver cirrhosis was shown to be accompanied by liver parenchymal hypoxia, which induced hepatic angiogenesis through up-regulation of VEGF in HSC and LSEC via HIF-1α.[32] VEGF expression induced sustained capillarization in LSEC, leading to the disappearance of fenestrations on the surface of the hepatic sinusoids. This undermined oxygen exchange between LSEC and hepatic parenchymal cells, resulting in a local hypoxic microenvironment.[33] Remodeling of the hepatic vascular architecture may increase intrahepatic vascular resistance, which leads to cirrhotic portal hypertension. Studies have shown that inhibition of HIF-1α expression reduces the synthesis and secretion of VEGF, thereby suppressing hepatic angiogenesis.[6] Apparently, HIF-1α plays an essential role in hypoxia-induced pathological angiogenesis in the liver by regulating VEGF expression. In addition, under hypoxic conditions, HIF-1α was shown to induce overexpression of angiopoietin-1, a critical factor in the regulation of angiogenesis.[34] Angiopoietin-1 then bound to its receptor Tie-2 and recruited mural cells to wrap around LSEC, thereby promoting the progression of liver fibrosis. That was confirmed by another study in which angiopoietin-1 and its specific receptor Tie-2 were significantly upregulated in liver tissue of rats with CCl_4-induced liver fibrosis.[35] In addition to HIF-1α, HIF-2α also plays a role in hypoxia-induced angiogenesis. Knockdown of *HIF-2α* gene was shown to up-regulate the expression of VEGFR1, which prevented VEGF from interacting with VEGFR2 and thus negatively regulated hepatic angiogenesis.[36] The above findings indicate that hypoxia and pathological angiogenesis may play a synergistic role in the progression of liver cirrhosis.[31]

H_2S-related signaling pathways in angiogenesis

Recent studies have confirmed the role of endogenous H_2S in the activation of mitogen-activated protein kinase cascade.[37] Elevated levels of ERK1/2 and p38 phosphorylation were observed after stimulation of endothelial cells with VEGF, which may be blocked by *CSE* silencing. The ATP-sensitive potassium channel is the main mediator of the H_2S effect and is located upstream of p38.[37] Direct inhibition of endogenous synthesis of H_2S with pharmacological agents or *CSE* gene silencing resulted in reduced intracellular cyclic guanosine monophosphate (cGMP) levels, whereas overexpression of *CSE* upregulated cGMP by direct inhibition of phosphodiesterase activity by H_2S.[38] In addition, H_2S production activated the PI3K/Akt signaling pathway, leading to endothelial nitric oxide synthase (eNOS) phosphorylation and eNOS activation.[39] Activation of the PI3K/Akt pathway may result from inhibition of lipid phosphatase and tensin homologue expression by H_2S.[40] Vasodilator stimulated phosphoprotein phosphorylation at Ser239 was upregulated in endothelial cells treated with L-cysteine, demonstrating that endogenous H_2S activated the cGMP/PKG pathway.[38] The accumulation of cGMP in turn activated PKG, which stimulated the angiogenic effect of endothelial cells.[38] In addition, the pro-angiogenic effect of the 3-MP/3-MST/H_2S pathway was shown to be significantly associated with the activation of Akt. In a study, knockdown of *3-MST* resulted in reduced levels of Akt and vasodilator-stimulated phosphoprotein phosphorylation.[41] Based on the above evidence, H_2S is considered as an essential gaseous signal molecule involved in the regulation of angiogenesis.

Interaction between H_2S and angiogenic factors

As previously described, CSE inhibitors or *CSE* gene silencing inhibited VEGF-mediated angiogenic effects.[38] This finding further suggests that treatment of endothelial cells with VEGF promotes the synthesis of H_2S. Although the underlying mechanism of this effect has not been fully elucidated, it is believed to be mediated through calcium-dependent activation of CSE.[38] However, in another study, adenovirus-mediated triple gene transfection of *CBS*, *CSE*, and *3-MST* upregulated VEGF expression and downregulated the expression of anti-angiogenic factors.[42] The above evidence indicates that H_2S is a downstream effector molecule of VEGF signaling, but may also be present upstream of VEGF signaling. Further studies are required for an in-depth characterization of the regulatory relationship between H_2S and VEGF.

The binding of VEGF to VEGFR2 causes its homodimerization, leading to phosphorylation of a series of tyrosine residues. In a recent study, a disulfide bond existing between Cys1045 and Cys1024 of VEGFR2 was found to alter the active conformation of VEGFR2 and inhibit its activity.[43] Nucleophilic attack on disulfide bonds by H_2S led to reduction of the disulfide bond and enhanced VEGFR2 tyrosine kinase activity.[43] In another study, knockdown of *CBS* in endothelial cells inhibited VEGF signaling by reducing the transcription of VEGFR2 and NRP-1.[44] That study further revealed that the

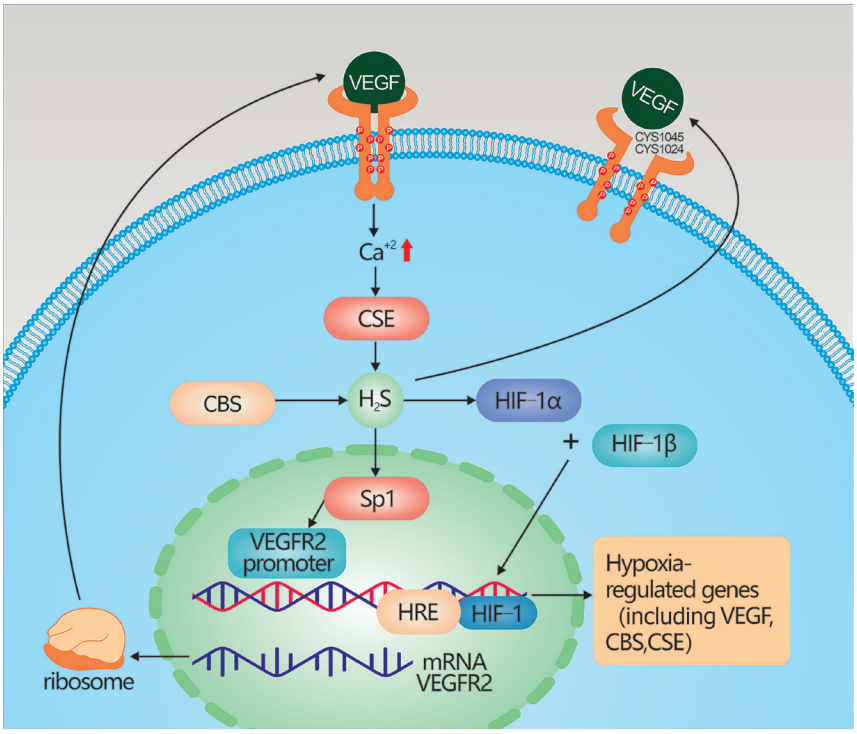

Fig. 2. Proposed interactions among H_2S, VEGF, and HIF-1. The binding of VEGF to VEGFR2 may activate CSE through a calcium-dependent pathway, which in turn promotes endogenous H_2S production. Nucleophilic attack by H_2S on the disulfide bond between Cys 1045-1024 leads to reduction of the disulfide bond and enhances VEGFR2 tyrosine kinase activity. CBS-derived and CSE-derived H_2S enhances the stability and transcriptional activity of Sp1, which further promotes the transcription of VEGFR2. H_2S results in increased HIF-1α levels, DNA binding, and transcriptional activity. VEGFR2, vascular endothelial growth factor receptor 2; VEGF, vascular endothelial growth factor; CSE, cystathionine-γ lyase; CBS, cystathionine-β synthase; HIF, hypoxia-inducible factor; HRE, hypoxia response element; Sp1, specificity protein 1; H_2S, hydrogen sulfide.

downregulation of VEGFR2 transcriptional activity was mediated by reduced stability of transcription factor specific protein 1 (Sp1).[44] Exogenous supplementation of H_2S donors in *CBS*-knockdown endothelial cells restored Sp1 levels and its binding to the VEGFR2 promoter, thereby reactivating the VEGF signaling pathway. A schematic illustration of the hypothetical interactions among H_2S, VEGF, and HIF-1 is shown in Figure 2.

Similar to VEGF, fibroblast growth factor 2 (FGF-2) is a pro-angiogenic and pro-fibrotic factor that inhibits endothelial cell apoptosis.[45] However, in contrast to VEGF, FGF-2 is insensitive to hypoxia.[45] HIF and VEGF expression occurred simultaneously and were co-localized to the same region; however, FGF-2 expression was observed much later and did not coincide with the distribution of hypoxic regions, which suggested that angiogenesis was mainly mediated by VEGF in response to hypoxia, while FGF-2 may contribute to maintain advanced angiogenesis.[46] Furthermore, H_2S inhibitors did not attenuate the FGF-mediated angiogenic response, suggesting that the pro-angiogenic effect of FGF was independent of H_2S.[47] Although H_2S may not be required for FGF signaling, H_2S can up-regulate FGF levels *in vivo*. In a recent study, both FGF and VEGF expression levels were elevated following hindlimb ischemia in wild-type mice, whereas this response was reduced in *CSE*-knockout mice.[48] Further studies are required to elucidate the potential role of H_2S in angiogenic signaling of other growth factors.

Vasodilatory effects of H_2S in portal hypertension

Studies have shown that the exogenous H_2S donor NaHS has vasodilatory effects similar to nitric oxide (NO).[49] Perfusion with NaHS attenuated norepinephrine-induced hepatic vasoconstriction in normal and cirrhotic liver.[49] Notably, although in normal liver the vasodilatory effects were reproduced by cysteine supplementation, the effects of cysteine in cirrhotic liver gradually diminished with the progressive loss of hepatic parenchymal cells.[50] H_2S acts as a vasodilator in the portal circulation, and perfusion of cirrhotic livers with exogenous H_2S donors was shown to compensate for defective NO production in a rat model of portal hypertension.[49]

In addition to causing endothelial dysfunction, homocysteine has been shown to cause contraction of HSC. This homocysteine-induced contraction of HSC is reversed by H_2S.[21] HSC contraction is associated with regulation of liver sinusoidal blood flow and intrahepatic resistance, and it is hypothesized that H_2S may be involved in regulating HSC contraction together with LSEC-derived paracrine cytokines.[21] Briefly, reduced levels of H_2S in the portal microcirculation cause an increase in intrahepatic resistance, not only because it is part of endothelial dysfunction, but also because it facilitates the activation and contraction of HSC. Another postulated mechanism of the vasodilatory effect of H_2S in liver involves its interaction with NO. However, perfusion of rat livers with nonselective eNOS and inducible nitric oxide synthase (iNOS) inhibitors failed to reverse the vasodilatory effects of H_2S on norepinephrine-induced vasoconstriction.[49] Inhibition of CSE with propargylglycin did not induce further constriction of liver sinusoidal microvessels using high flow rate liver perfusion. Interestingly, in patients with cirrhosis exhibiting splanchnic hyperdynamic circulation, NO levels

Table 1. Anti-angiogenesis therapeutic drugs in portal hypertension

Drugs	Experimental model	Site of angiogenesis	Target	Ref
Sorafenib	NASH; PBC; CCl4-induced cirrhosis	Intrahepatic angiogenesis	Raf/MEK/ERK signaling pathway; VEGFR; PDGFR	63–66
Sunitinib	CCl4-induced cirrhosis; human HSC; primary human LSEC	Intrahepatic angiogenesis	VEGFR-1/2/3; PDGFR-α/β; FGFR	67,68
Brivanib	NASH; PBC	Intrahepatic angiogenesis	VEGFR; FGFR	69,70
Simvastatin	CCl4-induced cirrhosis; human HSC	Intrahepatic angiogenesis	KLF2	71,72
Largazole	CCl4-induced cirrhosis; human HSC	Intrahepatic angiogenesis	VEGFR2; TGF-β	73
Rapamycin and imatinib	Partial portal vein ligation	Extrahepatic angiogenesis	VEGF; VEGFR2; PDGF; PDGFR-β	74
Bosentan	PBC	Extrahepatic angiogenesis	Endothelin receptors; iNOS; COX-2	75
Pioglitazone	PBC	Extrahepatic angiogenesis	NF-κB; VEGF; PDGF	76
Thalidomide	PBC	Extrahepatic angiogenesis	TNF-α-VEGF-NOS-NO pathway	77
Curcumin	PBC	Extrahepatic angiogenesis	VEGF; COX-2	78

NASH, nonalcoholic steatohepatitis; PBC, primary biliary cirrhosis; CCl4, carbon tetrachloride; HSC, hepatic stellate cells; LSEC, liver sinusoidal endothelial cells; VEGFR, vascular endothelial growth factor receptor; PDGFR, platelet-derived growth factor receptor; FGFR, fibroblast growth factor receptor; KLF2, Krüppel-like factor 2; TGF-β, transforming growth factor beta; VEGF, vascular endothelial growth factor; PDGF, platelet-derived growth factor; SSTR2, somatostatin subtype receptor 2; TNF-α, tumor necrosis factor alpha; Ang1, angiopoietin 1; eNOS, endothelial nitric oxide synthase; iNOS, inducible nitric oxide synthase; NO, nitric oxide; COX-2, cyclooxygenase-2; NF-κB, factor kappa-light-chain-enhancer of activated B cells.

were elevated by dysregulated eNOS production compared to healthy controls, while circulating H_2S levels appeared to be reduced.[51]

Hypoxia-induced angiogenesis in portal hypertension

HHME in patients with cirrhotic portal hypertension leads to liver sinusoidal capillarization, increased intrahepatic vascular resistance, and formation of intrahepatic shunts.[52,53] In addition, vasoconstrictors (e.g., endothelin-1) were upregulated in response to hypoxia, thereby increasing microcirculatory resistance in the liver and exacerbating hepatocyte hypoxia.[53] In hypoxic cirrhotic tissue, regenerative nodules were surrounded by a dense vascular plexus consisting of many microvessels that originated from pre-existing intrahepatic vascular branches. That progressed with the fibrous repair process, bypassing the liver parenchyma, and eventually leading to intrahepatic shunts.[29] Hyperdynamic splanchnic circulation is an important feature of portal hypertension.[54] Increased blood flow to the visceral organs flowing into the portal vein leads to increased portal blood flow, which causes portal hypertension. Previously, this phenomenon was believed to be associated with vasoconstriction and diastolic dysfunction, and the formation of collateral circulation was considered to be a mechanical consequence of elevated blood pressure.[55] However, recent studies have shown that angiogenesis may contribute to the maintenance of hyperdynamic splanchnic circulation and the development of collateral circulation, which is closely associated with VEGF- and PDGF-induced neovascularization and remodeling.[56]

Non-cirrhotic portal hypertension is a group of heterogeneous hepatic vascular diseases characterized by portal hypertension in the absence of cirrhosis.[57] Damage to the liver sinusoidal endothelium facilitates the entry of erythrocytes into the space of Dissé and the formation of perisinusoidal fibrosis. This hypoxic state induces excessive release of pro-angiogenic factors such as VEGF.[48] In addition, chronic hypoxia in the centrilobular areas induced nodular regenerative hyperplasia.[58] More evidence is required to fully elucidate the role of hypoxia and H_2S in regulating angiogenesis of non-cirrhotic portal hypertension.

Therapeutic implications of anti-angiogenesis agents for portal hypertension

Currently, the pharmacological effects of most anti-angiogenesis drugs have been investigated in experiments involving animal models as shown in Table 1. However, clinical evidence that supports the use of anti-angiogenesis drugs to treat portal hypertension is limited. Coriat *et al*.[59] first evaluated the effects of sorafenib on portal vein and systemic hemodynamics in seven patients with hepatocellular carcinoma and cirrhosis. Five of the patients were assessed as Child-Pugh class A, and two as Child-Pugh class B. Sorafenib (400 mg) was administered twice daily for 1 month. The results showed that the blood flow of portal vein decreased by 36%, but there was no significant change in the blood flow in the azygos vein and abdominal aorta. Another study explored the effects of sorafenib on HVPG and systemic hemodynamics in 13 patients with liver cirrhosis and hepatocellular carcinoma. Ten were Child-Pugh class A and three were Child-Pugh class B. The study also assessed the expression of genes related to liver fibrosis, angiogenesis, and inflammation. All patients received sorafenib (400 mg) twice a day for two weeks. In four of the 11 patients with clinically significant portal hypertension, HVPG was decreased by more than 20% from baseline. The levels of VEGF, PDGF, placental growth factor, and TNF-α were also downregulated.[60]

The main disadvantage of tyrosine kinase inhibitors is hepatotoxicity.[61] Poor liver function limits their application in patients with hepatocellular carcinoma complicated with cirrhotic portal hypertension. Selective delivery of drugs to target cells (especially HSC) by targeted drug delivery systems may be a promising direction to solve this problem.[62] Additional preclinical and clinical evidence on targeting HIF or H_2S agents in portal hypertension is required to support the clinical use of these drugs.

Summary and perspective

Angiogenesis is an essential pathophysiological event in the formation of the portal collateral circulation and the development of portal hypertension. The pathogenesis of portal hypertension is believed to involve H_2S- and hypoxia-induced hepatic angiogenesis. Endogenous H_2S plays a critical role in the regulation of vascular endothelial homeostasis, which may promote angiogenesis and induce vasodilation. H_2S upregulates the expression of VEGF in the HHME and participates in the regulation of VEGF-mediated angiogenesis. Therefore, H_2S and HIF are potential therapeutic targets for portal hypertension. The effects of H_2S donors or prodrugs on the hemodynamics of portal hypertension and the mechanism of H_2S-induced angiogenesis are promising areas for future research.

Acknowledgments

We appreciate Dr. Zijin Liu (Department of Gastroenterology and Hepatology, Beijing You'an Hospital Affiliated to Capital Medical University) for his assistance in preparing figures.

Funding

This work was supported by grants from the National Natural Science Foundation (81970525) and Sino-German Cooperation Group (GZ1517).

Conflict of interest

The authors have no conflict of interests related to this publication.

Author contributions

Study concept and design (HY, HD), acquisition of data (MT, ZG), analysis and interpretation of data (HY, MT, ZG), drafting of the manuscript (HY), critical revision of the manuscript for important intellectual content (SW, HD), administrative, technical, or material support (HD), and study supervision (SW, LL, HD). All authors have made a significant contribution to this study and have approved the final manuscript.

References

[1] Veldhuijzen van Zanten D, Buganza E, Abraldes JG. The Role of Hepatic Venous Pressure Gradient in the Management of Cirrhosis. Clin Liver Dis 2021;25(2):327–343. doi:10.1016/j.cld.2021.01.002, PMID:33838853.
[2] de Franchis R, Bosch J, Garcia-Tsao G, Reiberger T, Ripoll C, Baveno VII Faculty. Baveno VII - Renewing consensus in portal hypertension. J Hepatol 2022;76(4):959–974. doi:10.1016/j.jhep.2021.12.022, PMID:35120736.
[3] Gracia-Sancho J, Marrone G, Fernández-Iglesias A. Hepatic microcirculation and mechanisms of portal hypertension. Nat Rev Gastroenterol Hepatol 2019;16(4):221–234. doi:10.1038/s41575-018-0097-3, PMID:30568278.
[4] Mauro E, Gadano A. What's new in portal hypertension? Liver Int 2020; 40(Suppl 1):122–127. doi:10.1111/liv.14366, PMID:32077610.
[5] Waseem N, Chen PH. Hypoxic Hepatitis: A Review and Clinical Update. J Clin Transl Hepatol 2016;4(3):263–268. doi:10.14218/JCTH.2016.00022, PMID:27777895.
[6] Manuelli V, Pecorari C, Filomeni G, Zito E. Regulation of redox signaling in HIF-1-dependent tumor angiogenesis. FEBS J 2022;289(18):5413–5425. doi:10.1111/febs.16110, PMID:34228878.
[7] Lee SH, Golinska M, Griffiths JR. HIF-1-Independent Mechanisms Regulating Metabolic Adaptation in Hypoxic Cancer Cells. Cells 2021;10(9):2371. doi:10.3390/cells10092371, PMID:34572020.
[8] Albanese A, Daly LA, Mennerich D, Kietzmann T, Sée V. The Role of Hypoxia-Inducible Factor Post-Translational Modifications in Regulating Its Localisation, Stability, and Activity. Int J Mol Sci 2020;22(1):E268. doi:10.3390/ijms22010268, PMID:33383924.
[9] Ding H, Chang J, He F, Gai S, Yang P. Hydrogen Sulfide: An Emerging Precision Strategy for Gas Therapy. Adv Healthc Mater 2022;11(4):e2101984. doi:10.1002/adhm.202101984, PMID:34788499.
[10] Ngowi EE, Afzal A, Sarfraz M, Khattak S, Zaman SU, Khan NH, *et al*. Role of hydrogen sulfide donors in cancer development and progression. Int J Biol Sci 2021;17(1):73–88. doi:10.7150/ijbs.47850, PMID:33390834.
[11] Katsouda A, Bibli SI, Pyriochou A, Szabo C, Papapetropoulos A. Regulation and role of endogenously produced hydrogen sulfide in angiogenesis. Pharmacol Res 2016;113(Pt A):175–185. doi:10.1016/j.phrs.2016.08.026, PMID:27569706.
[12] Kietzmann T. Liver Zonation in Health and Disease: Hypoxia and Hypoxia-Inducible Transcription Factors as Concert Masters. Int J Mol Sci 2019;20(9):E2347. doi:10.3390/ijms20092347, PMID:31083568.
[13] Kling S, Lang B, Hammer HS, Naboulsi W, Sprenger H, Frenzel F, *et al*. Characterization of hepatic zonation in mice by mass-spectrometric and antibody-based proteomics approaches. Biol Chem 2022;403(3):331–343. doi:10.1515/hsz-2021-0314, PMID:34599868.
[14] Gonzalez FJ, Xie C, Jiang C. The role of hypoxia-inducible factors in metabolic diseases. Nat Rev Endocrinol 2018;15(1):21–32. doi:10.1038/s41574-018-0096-z, PMID:30275460.
[15] Wilson GK, Tennant DA, McKeating JA. Hypoxia inducible factors in liver disease and hepatocellular carcinoma: current understanding and future directions. J Hepatol 2014;61(6):1397–1406. doi:10.1016/j.jhep.2014.08.025, PMID:25157983.
[16] Zaorska E, Tomasova L, Koszelewski D, Ostaszewski R, Ufnal M. Hydrogen Sulfide in Pharmacotherapy, Beyond the Hydrogen Sulfide-Donors. Biomolecules 2020;10(2):E323. doi:10.3390/biom10020323, PMID:32085474.
[17] McGettrick AF, O'Neill LAJ. The Role of HIF in Immunity and Inflammation. Cell Metab 2020;32(4):524–536. doi:10.1016/j.cmet.2020.08.002, PMID:32853548.
[18] Infantino V, Santarsiero A, Convertini P, Todisco S, Iacobazzi V. Cancer Cell Metabolism in Hypoxia: Role of HIF-1 as Key Regulator and Therapeutic Target. Int J Mol Sci 2021;22(11):5703. doi:10.3390/ijms22115703, PMID:34071836.
[19] Corrado C, Fontana S. Hypoxia and HIF Signaling: One Axis with Divergent Effects. Int J Mol Sci 2020;21(16):E5611. doi:10.3390/ijms21165611, PMID:32764403.
[20] Papadakis AI, Paraskeva E, Peidis P, Muaddi H, Li S, Raptis L, *et al*. eIF2{alpha} Kinase PKR modulates the hypoxic response by Stat3-dependent transcriptional suppression of HIF-1{alpha}. Cancer Res 2010;70(20):7820–7829. doi:10.1158/0008-5472.CAN-10-0215, PMID:20924113.
[21] Lu X, Ding Y, Liu H, Sun M, Chen C, Yang Y, *et al*. The Role of Hydrogen Sulfide Regulation of Autophagy in Liver Disorders. Int J Mol Sci 2022;23(7):4035. doi:10.3390/ijms23074035, PMID:35409395.
[22] Paul BD, Snyder SH, Kashfi K. Effects of hydrogen sulfide on mitochondrial function and cellular bioenergetics. Redox Biol 2021;38:101772. doi:10.1016/j.redox.2020.101772, PMID:33137711.
[23] Kai S, Tanaka T, Daijo H, Harada H, Kishimoto S, Suzuki K, *et al*. Hydrogen sulfide inhibits hypoxia- but not anoxia-induced hypoxia-inducible factor 1 activation in a von hippel-lindau- and mitochondria-dependent manner. Antioxid Redox Signal 2012;16(3):203–216. doi:10.1089/ars.2011.3882, PMID:22004513.
[24] Wu B, Teng H, Yang G, Wu L, Wang R. Hydrogen sulfide inhibits the translational expression of hypoxia-inducible factor-1α. Br J Pharmacol 2012;167(7):1492–1505. doi:10.1111/j.1476-5381.2012.02113.x, PMID:22831549.
[25] Boye E, Grallert B. eIF2α phosphorylation and the regulation of translation. Curr Genet 2020;66(2):293–297. doi:10.1007/s00294-019-01026-1, PMID:31485739.
[26] Mennerich D, Kubaichuk K, Raza GS, Fuhrmann DC, Herzig KH, Brüne B, *et al*. ER-stress promotes VHL-independent degradation of hypoxia-inducible factors via FBXW1A/βTrCP. Redox Biol 2022;50:102243. doi:10.1016/j.redox.2022.102243, PMID:35074541.
[27] Ma DK, Vozdek R, Bhatla N, Horvitz HR. CYSL-1 interacts with the O2-sensing hydroxylase EGL-9 to promote H2S-modulated hypoxia-induced behavioral plasticity in C. elegans. Neuron 2012;73(5):925–940. doi:10.1016/j.neuron.2011.12.037, PMID:22405203.
[28] Bosch J, Abraldes JG, Fernández M, García-Pagán JC. Hepatic endothelial dysfunction and abnormal angiogenesis: new targets in the treatment of portal hypertension. J Hepatol 2010;53(3):558–567. doi:10.1016/j.jhep.2010.03.021, PMID:20561700.
[29] Li H. Angiogenesis in the progression from liver fibrosis to cirrhosis and hepatocelluar carcinoma. Expert Rev Gastroenterol Hepatol 2021;15(3):217–233. doi:10.1080/17474124.2021.1842732, PMID:33131349.
[30] Kondoh H, Castellvi J, LLeonart ME. Editorial: How Do Metabolism, Angiogenesis, and Hypoxia Modulate Resistance? Front Oncol 2021;11:671222. doi:10.3389/fonc.2021.671222, PMID:33912471.
[31] Chellappan DK, Leng KH, Jia LJ, Aziz NABA, Hoong WC, Qian YC, *et al*. The role of bevacizumab on tumour angiogenesis and in the management of gynaecological cancers: A review. Biomed Pharmacother 2018;102:1127–1144. doi:10.1016/j.biopha.2018.03.061, PMID:29710531.
[32] Yang X, Wang Z, Kai J, Wang F, Jia Y, Wang S, *et al*. Curcumol attenuates liver sinusoidal endothelial cell angiogenesis via regulating Glis-PROX1-HIF-1α in liver fibrosis. Cell Prolif 2020;53(3):e12762. doi:10.1111/cpr.12762, PMID:32119185.
[33] Bhatia M, Gaddam RR. Hydrogen Sulfide in Inflammation: A Novel Mediator and Therapeutic Target. Antioxid Redox Signal 2021;34(17):1368–1377. doi:10.1089/ars.2020.8211, PMID:33138636.
[34] Wang Y, Yu R, Wu L, Yang G. Hydrogen sulfide signaling in regulation of cell behaviors. Nitric Oxide 2020;103:9–19. doi:10.1016/j.niox.2020.07.002, PMID:32682981.
[35] Borkham-Kamphorst E, Weiskirchen R. The PDGF system and its antagonists in liver fibrosis. Cytokine Growth Factor Rev 2016;28:53–61. doi:10.1016/j.cytogfr.2015.10.002, PMID:26547628.
[36] Kimura H. Signalling by hydrogen sulfide and polysulfides via protein S-sulfuration. Br J Pharmacol 2020;177(4):720–733. doi:10.1111/bph.14579,

PMID:30657595.
[37] Mendiola PJ, Naik JS, Gonzalez Bosc LV, Gardiner AS, Birg A, Kanagy NL. Hydrogen Sulfide Actions in the Vasculature. Compr Physiol 2021; 11(4):2467–2488. doi:10.1002/cphy.c200036, PMID:34558672.
[38] Coletta C, Papapetropoulos A, Erdelyi K, Olah G, Módis K, Panopoulos P, *et al*. Hydrogen sulfide and nitric oxide are mutually dependent in the regulation of angiogenesis and endothelium-dependent vasorelaxation. Proc Natl Acad Sci U S A 2012;109(23):9161–9166. doi:10.1073/pnas.1202916109, PMID:22570497.
[39] Altaany Z, Moccia F, Munaron L, Mancardi D, Wang R. Hydrogen sulfide and endothelial dysfunction: relationship with nitric oxide. Curr Med Chem 2014;21(32):3646–3661. doi:10.2174/092986732166614070614 2930, PMID:25005182.
[40] Youness RA, Gad AZ, Sanber K, Ahn YJ, Lee GJ, Khallaf E, *et al*. Targeting hydrogen sulphide signaling in breast cancer. J Adv Res 2021;27:177–190. doi:10.1016/j.jare.2020.07.006, PMID:33318876.
[41] Coletta C, Módis K, Szczesny B, Brunyánszki A, Oláh G, Rios EC, *et al*. Regulation of Vascular Tone, Angiogenesis and Cellular Bioenergetics by the 3-Mercaptopyruvate Sulfurtransferase/H2S Pathway: Functional Impairment by Hyperglycemia and Restoration by DL-α-Lipoic Acid. Mol Med 2015;21:1–14. doi:10.2119/molmed.2015.00035, PMID:25715337.
[42] Sen U, Sathnur PB, Kundu S, Givvimani S, Coley DM, Mishra PK, *et al*. Increased endogenous H2S generation by CBS, CSE, and 3MST gene therapy improves ex vivo renovascular relaxation in hyperhomocysteinemia. Am J Physiol Cell Physiol 2012;303(1):C41–C51. doi:10.1152/ajpcell.00398.2011, PMID:22517358.
[43] Tao BB, Liu SY, Zhang CC, Fu W, Cai WJ, Wang Y, *et al*. VEGFR2 functions as an H2S-targeting receptor protein kinase with its novel Cys1045-Cys1024 disulfide bond serving as a specific molecular switch for hydrogen sulfide actions in vascular endothelial cells. Antioxid Redox Signal 2013;19(5):448–464. doi:10.1089/ars.2012.4565, PMID:23199280.
[44] Saha S, Chakraborty PK, Xiong X, Dwivedi SK, Mustafi SB, Leigh NR, *et al*. Cystathionine β-synthase regulates endothelial function via protein S-sulfhydration. FASEB J 2016;30(1):441–456. doi:10.1096/fj.15-278648, PMID:26405298.
[45] Sluzalska KD, Slawski J, Sochacka M, Lampart A, Otlewski J, Zakrzewska M. Intracellular partners of fibroblast growth factors 1 and 2 - implications for functions. Cytokine Growth Factor Rev 2021;57:93–111. doi:10.1016/j.cytogfr.2020.05.004, PMID:32475760.
[46] Liu L, You Z, Yu H, Zhou L, Zhao H, Yan X, *et al*. Mechanotransduction-modulated fibrotic microniches reveal the contribution of angiogenesis in liver fibrosis. Nat Mater 2017;16(12):1252–1261. doi:10.1038/nmat5024, PMID:29170554.
[47] Wang Y, Liu D, Zhang T, Xia L. FGF/FGFR Signaling in Hepatocellular Carcinoma: From Carcinogenesis to Recent Therapeutic Intervention. Cancers (Basel) 2021;13(6):1360. doi:10.3390/cancers13061360, PMID: 33802841.
[48] Kolluru GK, Bir SC, Yuan S, Shen X, Pardue S, Wang R, *et al*. Cystathionine γ-lyase regulates arteriogenesis through NO-dependent monocyte recruitment. Cardiovasc Res 2015;107(4):590–600. doi:10.1093/cvr/cvv198, PMID:26194202.
[49] Sun HJ, Wu ZY, Nie XW, Wang XY, Bian JS. Implications of hydrogen sulfide in liver pathophysiology: Mechanistic insights and therapeutic potential. J Adv Res 2021;27:127–135. doi:10.1016/j.jare.2020.05.010, PMID:33318872.
[50] Huc T, Jurkowska H, Wróbel M, Jaworska K, Onyszkiewicz M, Ufnal M. Colonic hydrogen sulfide produces portal hypertension and systemic hypotension in rats. Exp Biol Med (Maywood) 2018;243(1):96–106. doi: 10.1177/1535370217741869, PMID:29130338.
[51] Wang C, Han J, Xiao L, Jin CE, Li DJ, Yang Z. Role of hydrogen sulfide in portal hypertension and esophagogastric junction vascular disease. World J Gastroenterol 2014;20(4):1079–1087. doi:10.3748/wjg.v20.i4.1079, PMID:24574782.
[52] Rosmorduc O, Housset C. Hypoxia: a link between fibrogenesis, angiogenesis, and carcinogenesis in liver disease. Semin Liver Dis 2010;30(3):258–270. doi:10.1055/s-0030-1255355, PMID:20665378.
[53] Cai J, Hu M, Chen Z, Ling Z. The roles and mechanisms of hypoxia in liver fibrosis. J Transl Med 2021;19(1):186. doi:10.1186/s12967-021-02854-x, PMID:33933107.
[54] Gao L, Yang X, Li Y, Wang Z, Wang S, Tan S, *et al*. Curcumol inhibits KLF5-dependent angiogenesis by blocking the ROS/ERK signaling in liver sinusoidal endothelial cells. Life Sci 2021;264:118696. doi:10.1016/j.lfs.2020.118696, PMID:33157090.
[55] Gana JC, Serrano CA, Ling SC. Angiogenesis and portal-systemic collaterals in portal hypertension. Ann Hepatol 2016;15(3):303–313. doi:10.5604/16652681.1198799, PMID:27049484.
[56] Dong G, Lin XH, Liu HH, Gao DM, Cui JF, Ren ZG, *et al*. Intermittent hypoxia alleviates increased VEGF and pro-angiogenic potential in liver cancer cells. Oncol Lett 2019;18(2):1831–1839. doi:10.3892/ol.2019.10486, PMID:31423251.
[57] Gioia S, Nardelli S, Riggio O, Faccioli J, Ridola L. Cognitive Impairement in Non-Cirrhotic Portal Hypertension: Highlights on Physiopathology, Diagnosis and Management. J Clin Med 2021;11(1):101. doi:10.3390/jcm11010101, PMID:35011842.
[58] Nicoară-Farcău O, Rusu I, Stefănescu H, Tanțău M, Badea RI, Procopeț B. Diagnostic challenges in non-cirrhotic portal hypertension - porto sinusoidal vascular disease. World J Gastroenterol 2020;26(22):3000–3011. doi:10.3748/wjg.v26.i22.3000, PMID:32587444.
[59] Coriat R, Gouya H, Mir O, Ropert S, Vignaux O, Chaussade S, *et al*. Reversible decrease of portal venous flow in cirrhotic patients: a positive side effect of sorafenib. PLoS One 2011;6(2):e16978. doi:10.1371/journal.pone.0016978, PMID:21340026.
[60] Pinter M, Sieghart W, Reiberger T, Rohr-Udilova N, Ferlitsch A, Peck-Radosavljevic M. The effects of sorafenib on the portal hypertensive syndrome in patients with liver cirrhosis and hepatocellular carcinoma—a pilot study. Aliment Pharmacol Ther 2012;35(1):83–91. doi:10.1111/j.1365-2036.2011.04896.x, PMID:22032637.
[61] Huang L, Jiang S, Shi Y. Tyrosine kinase inhibitors for solid tumors in the past 20 years (2001-2020). J Hematol Oncol 2020;13(1):143. doi:10.1186/s13045-020-00977-0, PMID:33109256.
[62] Peng W, Cheng S, Bao Z, Wang Y, Zhou W, Wang J, *et al*. Advances in the research of nanodrug delivery system for targeted treatment of liver fibrosis. Biomed Pharmacother 2021;137:111342. doi:10.1016/j.biopha.2021.111342, PMID:33581652.
[63] Qu K, Huang Z, Lin T, Liu S, Chang H, Yan Z, *et al*. New Insight into the Anti-liver Fibrosis Effect of Multitargeted Tyrosine Kinase Inhibitors: From Molecular Target to Clinical Trials. Front Pharmacol 2015;6:300. doi:10.3389/fphar.2015.00300, PMID:26834633.
[64] Cheng CC, Chao WT, Shih JH, Lai YS, Hsu YH, Liu YH. Sorafenib combined with dasatinib therapy inhibits cell viability, migration, and angiogenesis synergistically in hepatocellular carcinoma. Cancer Chemother Pharmacol 2021; 88(1):143–153. doi:10.1007/s00280-021-04272-8, PMID:33860837.
[65] Huang W, Xing Y, Zhu L, Zhuo J, Cai M. Sorafenib derivatives-functionalized gold nanoparticles confer protection against tumor angiogenesis and proliferation via suppression of EGFR and VEGFR-2. Exp Cell Res 2021;406(1):112633. doi:10.1016/j.yexcr.2021.112633, PMID:34089726.
[66] Liu L, Cao Y, Chen C, Zhang X, McNabola A, Wilkie D, *et al*. Sorafenib blocks the RAF/MEK/ERK pathway, inhibits tumor angiogenesis, and induces tumor cell apoptosis in hepatocellular carcinoma model PLC/PRF/5. Cancer Res 2006;66(24):11851–11858. doi:10.1158/0008-5472.CAN-06-1377, PMID:17178882.
[67] Motzer RJ, Banchereau R, Hamidi H, Powles T, McDermott D, Atkins MB, *et al*. Molecular Subsets in Renal Cancer Determine Outcome to Checkpoint and Angiogenesis Blockade. Cancer Cell 2020;38(6):803–817.e4. doi:10.1016/j.ccell.2020.10.011, PMID:33157048.
[68] Majumder S, Piguet AC, Dufour JF, Chatterjee S. Study of the cellular mechanism of Sunitinib mediated inactivation of activated hepatic stellate cells and its implications in angiogenesis. Eur J Pharmacol 2013;705(1-3):86–95. doi:10.1016/j.ejphar.2013.02.026, PMID:23454556.
[69] Lin HC, Huang YT, Yang YY, Lee PC, Hwang LH, Lee WP, *et al*. Beneficial effects of dual vascular endothelial growth factor receptor/fibroblast growth factor receptor inhibitor brivanib alaninate in cirrhotic portal hypertensive rats. J Gastroenterol Hepatol 2014;29(5):1073–1082. doi:10.1111/jgh.12480, PMID:24325631.
[70] Yang YY, Liu RS, Lee PC, Yeh YC, Huang YT, Lee WP, *et al*. Anti-VEGFR agents ameliorate hepatic venous dysregulation/microcirculatory dysfunction, splanchnic venous pooling and ascites of NASH-cirrhotic rat. Liver Int 2014;34(4):521–534. doi:10.1111/liv.12299, PMID:23998651.
[71] Miao Q, Zeng X, Ma G, Li N, Liu Y, Luo T, *et al*. Simvastatin suppresses the proangiogenic microenvironment of human hepatic stellate cells via the Kruppel-like factor 2 pathway. Rev Esp Enferm Dig 2015;107(2):63–71. PMID:25659387.
[72] Marrone G, Russo L, Rosado E, Hide D, García-Cardeña G, García-Pagán JC, *et al*. The transcription factor KLF2 mediates hepatic endothelial protection and paracrine endothelial-stellate cell deactivation induced by statins. J Hepatol 2013;58(1):98–103. doi:10.1016/j.jhep.2012.08.026, PMID:22989565.
[73] Liu Y, Wang Z, Wang J, Lam W, Kwong S, Li F, *et al*. A histone deacetylase inhibitor, largazole, decreases liver fibrosis and angiogenesis by inhibiting transforming growth factor-β and vascular endothelial growth factor signalling. Liver Int 2013;33(4):504–515. doi:10.1111/liv.12034, PMID:23279742.
[74] Chen L, Dai L, Yan D, Zhou B, Zheng W, Yin J, *et al*. Gleevec and Rapamycin Synergistically Reduce Cell Viability and Inhibit Proliferation and Angiogenic Function of Mouse Bone Marrow-Derived Endothelial Progenitor Cells. J Vasc Res 2021;58(5):330–342. doi:10.1159/000515816, PMID:34247157.
[75] Hsu SJ, Lin TY, Wang SS, Chuang CL, Lee FY, Huang HC, *et al*. Endothelin receptor blockers reduce shunting and angiogenesis in cirrhotic rats. Eur J Clin Invest 2016;46(6):572–580. doi:10.1111/eci.12636, PMID:27091078.
[76] Schwabl P, Payer BA, Grahovac J, Klein S, Horvatits T, Mitterhauser M, *et al*. Pioglitazone decreases portosystemic shunting by modulating inflammation and angiogenesis in cirrhotic and non-cirrhotic portal hypertensive rats. J Hepatol 2014;60(6):1135–1142. doi:10.1016/j.jhep.2014.01.025, PMID:24530596.
[77] Li TH, Huang CC, Yang YY, Lee KC, Hsieh SL, Hsieh YC, *et al*. Thalidomide Improves the Intestinal Mucosal Injury and Suppresses Mesenteric Angiogenesis and Vasodilatation by Down-Regulating Inflammasomes-Related Cascades in Cirrhotic Rats. PLoS One 2016;11(1):e0147212. doi:10.1371/journal.pone.0147212, PMID:26820153.
[78] Hsu SJ, Lee JY, Lin TY, Hsieh YH, Huang HC, Lee FY, *et al*. The beneficial effects of curcumin in cirrhotic rats with portal hypertension. Biosci Rep 2017;37(6):BSR20171015. doi:10.1042/BSR20171015, PMID:29162665.