Antiangiogenic therapy for portal hypertension in liver cirrhosis: Current progress and perspectives

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Abstract

Developing medicines for hemodynamic disorders that are characteristic of cirrhosis of the liver is a relevant problem in modern hepatology. The increase in hepatic vascular resistance to portal blood flow and subsequent hyperdynamic circulation underlie portal hypertension (PH) and promote its progression, despite the formation of portosystemic collaterals. Angiogenesis and vascular bed restructurization play an important role in PH pathogenesis as well. In this regard, strategic directions in the therapy for PH in cirrhosis include selectively decreasing hepatic vascular resistance while preserving or increasing portal blood flow, and correcting hyperdynamic circulation and pathological angiogenesis. The aim of this review is to describe the mechanisms of angiogenesis in PH and the methods of antiangiogenic therapy. The PubMed database, the Google Scholar retrieval system, and the reference lists from related articles were used to search for relevant publications. Articles corresponding to the aim of the review were selected for 2000-2017 using the keywords: "liver cirrhosis", "portal hypertension", "pathogenesis", "angiogenesis", and "antiangiogenic therapy". Antiangiogenic therapy for PH was the inclusion criterion. In this review, we have described angiogenesis inhibitors and their mechanism of action in relation to PH. Although most of them were studied

only in animal experiments, this selective therapy for abnormally growing newly formed vessels is pathogenetically reasonable to treat PH and associated complications.

Key words: Liver cirrhosis; Portal hypertension; Pathogenesis; Angiogenesis; Antiangiogenic therapy

Core tip: This review describes the role of angiogenesis in the pathogenesis of portal hypertension in liver cirrhosis and the prospects of antiangiogenic therapy. The analysis of the data showed that angiogenesis plays an important role in the pathogenesis of cirrhosis and accompanies portal hypertension, underlying its development and causing related complications. Although most of angiogenesis inhibitors were studied only in animal experiments, this selective therapy for abnormally growing newly formed vessels is pathogenetically reasonable to treat portal hypertension and associated complications.

INTRODUCTION

Developing medicines to treat hemodynamic disorders that are characteristic of liver cirrhosis and promote portal hypertension (PH) and related complications is a relevant problem in modern hepatology. In accordance with the current clinical recommendations, nonselective β-adrenoblockers are the drugs of choice[1]. However, their influence on portal pressure is variable. A number of studies showed that they did not lead to a clinically significant decrease in portal pressure, and the weakening of their therapeutic effect was noted in 50%-70% of cases in the long-term period. Also, the question of the appropriateness of using nonselective β-adrenergic blockers in patients with decompensated cirrhosis has not been finally resolved[2].

Ideally, the pharmacotherapy of PH should lessen the severity of morphofunctional disorders in the liver, contributing to the reduction of the vascular resistance to portal blood flow. Also, it should successfully correct a hyperdynamic circulatory state. As a result, the hepatic venous pressure gradient (HVPG), the most accurate equivalent of portal pressure, should be reduced to less than 12 mmHg or be 20% lower than an original value. In addition, it is necessary to avoid arterial hypotension and at the same time reduce the influx of splanchnic

blood into the portal vein, keeping unchanged the portal blood flow, which participates in liver perfusion[3].

Angiogenesis plays an important role in the pathogenesis of many chronic liver diseases, including fibrosis, cirrhosis, and hepatocellular carcinoma[4]. It can also accompany PH, underlying its development and causing related complications. Indeed, the newly formed blood vessels, which bypass sinusoids in response to the gross morphofunctional rearrangement of the liver in cirrhosis, fail to provide oxygen and nutrients to the tissues, which worsens the course of the disease and increases hepatic vascular resistance to portal blood flow[5]. Further progression of PH is a consequence of complex processes including angiogenesis, vascular remodeling, and endothelial dysfunction, which contribute to splanchnic congestion, portosystemic shunt formation, and a hyperdynamic circulatory state[6] (Figure 1). From this, it can be inferred that antiangiogenic therapy, which is selectively aimed at suppressing newly formed vessels' formation and growth, is a pathogenetically grounded method of treating PH and associated complications[7].

The efforts to develop angiogenesis inhibitors began in the 1970s at Harvard University under the guidance of Judah Folkman. The drugs were actively introduced into clinical practice a decade after the first were developed[8].

INHIBITORS OF INTRAHEPATIC ANGIOGENESIS

One of the two main mechanisms in the formation of new blood vessels in the liver in cirrhosis is associated with an increased expression of pro-angiogenic growth factors, cytokines, and matrix metalloproteinases in the presence of chronic inflammation. Proinflammatory mediators produced by Kupffer cells, mast cells, and leukocytes may manifest an angiogenic response at the expense of hypoxia-inducible factor-1α (HIF-1α) induction and increased transcription activity[9]. HIF-1α activates hepatic stellate cells (HSC), which leads to the development of various angiogenic and fibrogenic factors, promoting both angiogenesis and liver fibrosis[10]. At the same time, diffuse fibrosis, the formation of regenerative nodules, and also the capillarization of sinusoids cause an increase in hepatic vascular resistance and impair oxygen delivery to liver cells[11]. Accumulation of HIFs, in particular HIF-1α, increases the expression of vascular endothelial growth factor (VEGF), angiopoietin-1 (Ang1), and their related receptors on activated HSC. This leads to recruitment and stimulation of sinusoidal endothelial cells (SEC), which stabilizes the newly formed vessels and ensures their strength. In turn, SEC produce platelet-derived growth factor (PDGF) and transforming growth factor-β1 (TGF-β1), contributing to the recruitment and migration of HSC, a process that involves reactive oxygen species-dependent activation of the extracellular
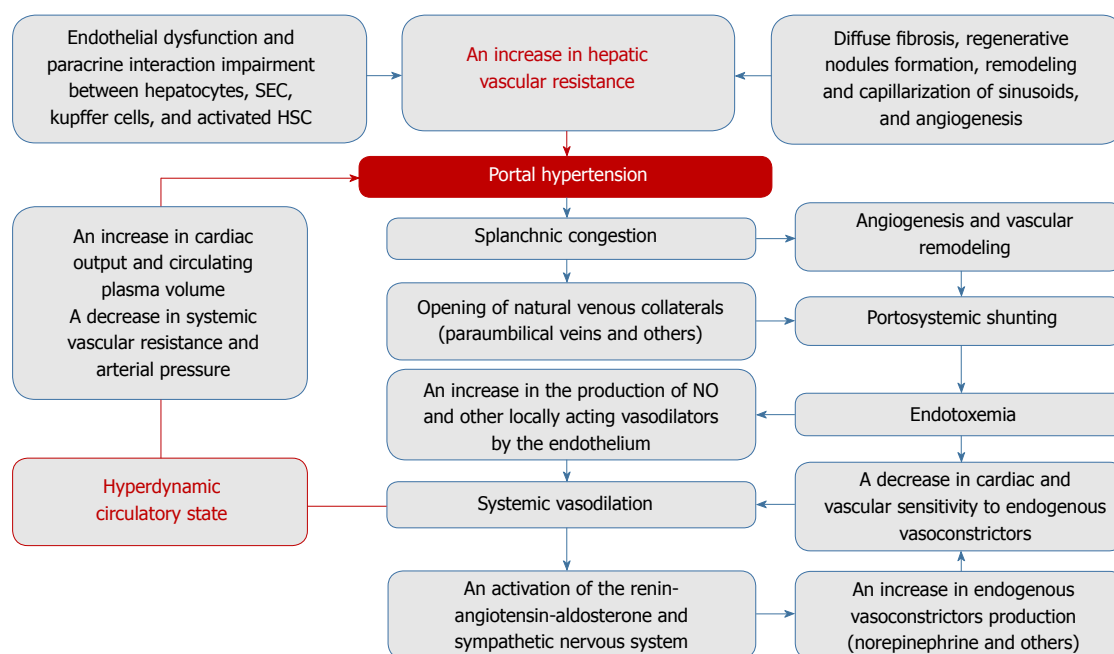
Figure 1 Potential mechanisms of portal hypertension pathogenesis in cirrhosis. The newly formed blood vessels, which bypass sinusoids in response to the gross morphofunctional rearrangement of the liver in cirrhosis, fail to provide oxygen and nutrients to the tissues. With endothelial dysfunction and impaired paracrine interaction between hepatocytes, sinusoidal endothelial cells (SEC), Kupffer cells, and activated hepatic stellate cells (HSC), this increases hepatic vascular resistance to portal blood flow. Further progression of portal hypertension is a consequence of complex processes including angiogenesis, vascular remodeling, and endothelial dysfunction, which contribute to splanchnic congestion, systemic vasodilation, and portosystemic shunt formation. The subsequent hyperdynamic circulatory state worsens the course of the disease.

signal-regulated kinase (ERK) pathway and c-Jun NH2-terminal kinases (JNK) followed by HIF-1α-dependent synthesis of VEGF[12].

Tyrosine kinase inhibitors

The introduction of antiangiogenic therapy into hepatological practice began with the treatment of hepatocellular carcinoma, a well-vascularized tumor that needs intense angiogenic activity for its development[13]. The most studied drug used for this purpose is sorafenib, a multi-targeted inhibitor of receptor and non-receptor tyrosine kinases, which are responsible for transmitting various signals to cells, including proliferative stimuli. The antitumor and antiangiogenic effect of sorafenib is achieved mainly through the suppression of the Raf/MEK/ERK signaling pathway and blockade of signaling from the receptors of VEGF (VEGFR), PDGF (PDGFR), and c-kit (SCFR)[14].

Experimental studies have shown the antiangiogenic effect of sorafenib during the early stage of hepatic fibrosis[15]. In animals with various models of cirrhosis, it had positive effects on some pathogenetic pathways of fibrogenesis and angiogenesis in the liver by blocking the receptor tyrosine kinases located on the surface of HSC, the expression of which, especially VEGFR and PDGFR, was increased[16] (Figure 2): (1) The suppression of activated HSC proliferation and the activation of apoptosis; (2) the inhibition of cyclin D1 and cyclin-dependent kinase 4 (Cdk-4) with a simultaneous increase in the expression of Fas, Fas-L, and Caspase-3, and a decrease in the ratio of

Bcl-2 to Bax; (3) an increase in the ratio of matrix metalloproteinases to the tissue inhibitor of matrix metalloproteinases, and also a decrease in the synthesis of collagen by HSC; (4) the inhibition of phosphorylation of ERK, Akt, and ribosomal protein kinase S6 with a molecular mass of 70 kDa (p70S6K)[17]; and (5) the disturbance of the Kruppel-like factor 6–Ang1–fibronectin molecular triad functioning[18]. Sorafenib decreased the severity of inflammation, fibrogenesis, and angiogenesis in rats with biliary cirrhosis, which led to a reduction in hepatic vascular resistance to portal blood flow[19].

Another multi-targeted tyrosine kinase inhibitor sunitinib is less studied but known to block VEGFR1/2/3, PDGFR-α/β, fibroblast growth factor receptor (FGFR), and c-kit signaling[20]. In addition, an *in vitro* study by Majumder *et al*[21] showed that sunitinib can slow HSC collagen synthesis by 47%, reduce HSC contractility by 65%, and decrease cellular migration by 28%, as well as inhibit the angiogenic capacity of SEC.

Branivib is a double inhibitor of VEGFR and FGFR signaling. It significantly suppressed intrahepatic angiogenesis and reduced PH in rats with biliary cirrhosis[22]. Additionally, it improved blood circulation in the liver and hindered the formation of ascites in rats with liver cirrhosis caused by nonalcoholic steatohepatitis[23].

Statins

The positive effect of statins on hepatic fibro- and angiogenesis in cirrhosis is associated with the induction
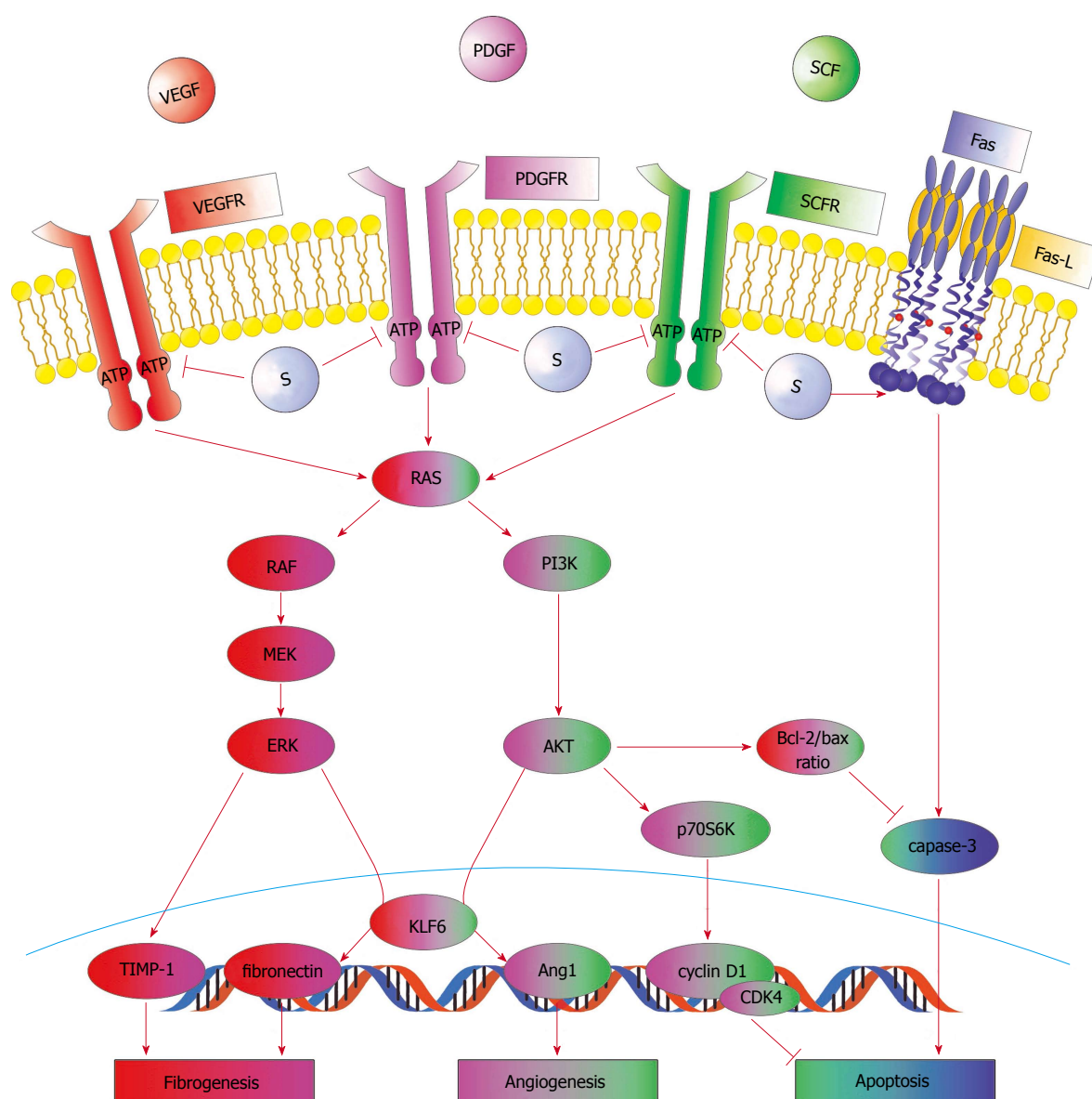
Figure 2 Positive effects of sorafenib on some pathogenetic pathways of fibrogenesis and angiogenesis in the liver. Sorafenib (S) blocks the ATP-binding site of the vascular endothelial growth factor receptor (VEGFR), platelet-derived growth factor receptor (PDGFR), and stem cell growth factor receptor (SCFR) tyrosine kinases located on the surface of hepatic stellate cells (HSC), inhibiting the two main cellular pathways of the RAS protein. At the same time, sorafenib increases the expression of Fas and its ligand. This decreases the severity of fibrogenesis and angiogenesis and increases apoptosis, leading to a reduction in hepatic vascular resistance to portal blood flow.

of KLF2 in SEC[24]. KLF2 is a member of a family of widely expressed transcription factors that regulate cell and tissue growth. KLF2 is well represented in the vascular endothelium and is necessary for the normal development of vessels; in addition, it is a well-known antiangiogenic factor that modulates the severity of many endothelial vasoprotective genes[25]. KLF2 can effectively inhibit HIF-1α, reducing the expression of such proangiogenic factors as VEGF and Ang2[26].

The mechanical stimuli generated by shear stress are the main physiological impulse for triggering and maintaining endothelial KLF2 expression[27]. In the cirrhotic liver, KLF2 expression was elevated in both SEC[28] and activated HSC[29]. This serves as a compensatory mechanism aimed at eliminating

vascular dysfunction and preventing angiogenesis by suppressing the proliferation and migration of SEC, as well as downregulating the ERK1/2 signaling pathway to inhibit the formation of tubular structures[30].

In an *in-vitro* study conducted by Miao *et al*[31], simvastatin eliminated the pro-angiogenic environment for TGF-β-activated HSC as a result of the following processes: (1) The reduction of cell migration and proliferation; (2) the inhibition of the α-smooth muscle actin expression, and the elevation of mRNA and KLF2 levels in HSC; (3) an increase in the production of endothelial nitric oxide synthase (eNOS) and suppression of the various proangiogenic proteins expression in HSC, such as VEGF, HIF-1α, and pro-inflammatory nuclear factor kappa B (NF-κB); and (4) the reduction

of the hyperactivity of interferon γ, which participates in angiogenesis. In rats with CCL4-induced liver cirrhosis, it was noted that statins (atorvastatin, mevastatin, simvastatin, and lovastatin) enhanced the effect of KLF2. By doing this, they deactivated SEC and reduced the severity of fibrosis and associated angiogenesis, thereby exerting a positive effect on PH[32].

Rifaximin
Endotoxemia, which is due to the translocation of gram-negative bacteria from the intestine, plays an important role in the pathogenesis of both cirrhosis and associated complications[33]. During the development of cirrhosis, bacterial lipopolysaccharide influences Kupffer cells and HSC. Nevertheless, SEC is affected first. Toll-like receptors 4 (TLR4), which are located on their surface and capable of binding bacterial lipopolysaccharide, is involved in fibrosis-associated angiogenesis. These receptors manifest such properties through the related cytosolic adapter protein MyD88, which is involved in the production of extracellular protease regulating the invasive ability of SEC[34].

In mice with biliary cirrhosis, it was shown that rifaximin, a nonabsorbable antibiotic with broad antimicrobial activity against aerobic and anaerobic gram-negative bacteria, reduced the severity of fibrosis and angiogenesis in the liver by inhibiting bacterial lipopolysaccharide binding to TLR4. As a consequence, it reduced PH[35]. This drug is already used to treat hepatic encephalopathy. It has an acceptable safety profile when applied in patients with chronic liver diseases and is approved by the US Food and Drug Administration[36]. The experimental study[35] may be a basis for evaluation of rifaximin in other complications of cirrhosis.

Largazole
The histone deacetylase inhibitor largazole is a natural compound derived from marine cyanobacteria Symploca sp. With a strong antiproliferative and cytotoxic effect, it has a wide spectrum but differential activity against several different lines of cancer cells[37]. In addition, in experimental studies *in vitro* and *in vivo*, largazole attenuated the severity of liver fibrosis and associated angiogenesis through numerous independent mechanisms: (1) The reduction of VEGF production by HSC; (2) the inhibition of VEGF-stimulated HSC proliferation; (3) the downregulation of TGF-β1- and VEGF-induced Akt phosphorylation in activated HSC, as well as the downregulation of VEGFR2-dependent p38MAPK phosphorylation in SEC; and (4) the suppression of CD34, VEGF, and VEGFR2 expression[38]. The ability of largazole to affect the main fibrogenic and angiogenic pathways in the cirrhotic liver can be used to test its effectiveness in PH.

Ribavirin
In addition to antiviral activity against certain DNA- and RNA-containing viruses, ribavirin may have a positive effect on the morphological changes underlying the development of cirrhosis[39]. In addition, at therapeutic concentrations, it is able to inhibit angiogenesis both *in vitro* and *in vivo*, which is due to the inhibition of inosine-5'-monophosphate dehydrogenase 1 activity and a decrease in tetrahydrobiopterin, NO, and cGMP levels in SEC[40].

INHIBITORS OF EXTRAHEPATIC ANGIOGENESIS
Disturbances of organ and systemic hemodynamics and the development of portosystemic collateral circulation in PH begin with splanchnic vasodilation and neovascularization caused by hypoxia of intestinal mucosa and pro-inflammatory cytokines, chemokines, and angiogenic factors, such as VEGF, PDGF, the placental growth factor (PlGF), and others[41]. It was traditionally thought that portosystemic shunts are formed when increased portal pressure "opens" pre-existing vessels in the areas of embryonic connection between the portal and systemic circulation. This paradigm was challenged by Fernandez, who first reported that portosystemic collaterals in PH are formed due to active angiogenesis. It was shown in an animal model of prehepatic PH induced by partial portal vein ligation that the blockade of VEGFR-2 with anti-VEGFR-2 monoclonal antibody for 5-7 d and inhibition of VEGF/VEGFR-2 signalization using autophosphorylation inhibitor VEGFR-2 for 5 d after the operation resulted in 50% reduction of portosystemic collateral vessel formation[42,43]. Blockade of NAD(P)H also contributed to this owing to the reduced splanchnic expression of VEGF, VEGFR-2, and CD31[44].

It should be noted that VEGF is of the greatest importance only at the initial stages of angiogenesis, when it activates endothelial cell proliferation and the subsequent formation of endothelial tubules. Vascular maturation is modulated mainly by PDGF, which regulates the introduction of endothelial tubules into the population of intramural cells and pericytes, thus stabilizing the newly formed vasculature[45]. The simultaneous suppression of the signaling caused by both VEGF and PDGF appears more promising than suppressing them individually.

Tyrosine kinase inhibitors
Fernandez et al[46] studied the combined effect of rapamycin (mTOR inhibitor) and glivec (tyrosine protein kinase inhibitor) on VEGF and PDGF signaling, respectively, in rats with extrahepatic PH caused by partial portal vein ligation and with well-developed portosystemic collateral circulation. It was noted that rapamycin and glivec in combination markedly reduced the splanchnic neovascularization and pericyte coverage of new vessels through the decreased expression of VEGF, VEGFR2, CD31, PDGF, PDGFR-β, and α-smooth muscle actin. In addition, there was a reduction of

portal pressure and blood flow along the superior mesenteric artery by 40% and 30% from the baseline level, respectively.

Similar results were obtained by Mejias *et al*[19], who found that multi-kinase inhibitor sorafenib triggered blockade of VEGF and PDGF signaling transduction and the Raf/MEK/ERK signaling pathways. Sorafenib significantly reduced intraorgan and systemic blood flow, and increased splanchnic neovascularization by 80% and portosystemic shunting by 18%. This led to a reduction in hepatic vascular resistance and decrease in portal pressure by 25% from the baseline. It was also noted that the positive effect of sorafenib on PH was more significant when it was combined with propranolol[47].

Somatostatin and its synthetic analogs
Somatostatin is a cyclic 14-amino acid peptide, which is secreted by nerve, endocrine, and enteroendocrine cells in the hypothalamus and digestive system (in the stomach, intestine, and pancreatic δ-cells). Somatostatin and its synthetic analogs (octreotide, vapreotide, and others) are used in patients with cirrhosis to treat bleeding from esophageal varices by affecting both intra- and extrahepatic mechanisms of PH[48].

The ability of octreotide to inhibit cell proliferation and neovascularization through the high-affinity somatostatin subtype receptor 2 (SSTR2) was an impetus for studying its antiangiogenic properties in various diseases[49]. In studies involving rats with extrahepatic PH caused by partial portal vein ligation, octreotide significantly weakened the expression of VEGF and CD31 in the internal organs, reduced the development of splanchnic neovascularization by 64%, and lessened the severity of a portosystemic collateral circulation by 16%. At the same time, its angioinhibitory effect manifested only in the first four days of the experiment and completely disappeared after a week, as PH progressed. This is possibly due to a decrease in SSTR2 expression in mucosa, intestinal vessels, and portosystemic collaterals[50].

Spironolactone
Pathophysiological disturbances inherent to PH underlie the occurrence of ascites in cirrhosis. Systemic arterial vasodilation and the activation of various neurohormonal pathways, including the renin-angiotensin-aldosterone system, caused renal dysfunction. This decreases Na^+ and water excretion and reduces the glomerular filtration rate. The drug of choice for treatment is spironolactone, an antagonist of aldosterone, a mineralocorticoid, that mediates the reabsorption of Na^+ and water in the distal part of the nephron[51]. In addition to the important role in maintaining water-salt metabolism, aldosterone has angiogenic properties. In particular, it enhances ischemia-induced neovascularization[52], stimulates pathological angiogenesis in the retina[53], and promotes

the proliferation of endothelial cells of the heart[54] by activating angiotensin II signaling. At the same time, its antagonist spironolactone inhibits these processes both *in vitro* and *in vivo*[55]. In rats with biliary cirrhosis, spironolactone significantly reduced the degree of mesenteric angiogenesis and portosystemic shunting by suppressing the VEGF signal transduction pathway[56].

N-acetylcysteine
Because hypoxia serves as the main inducer of angiogenesis both under physiological and pathological conditions, angiogenesis inhibitors may be drugs with antioxidant properties. One of them is N-acetylcysteine, which is a derivative of amino acid cysteine, the thiol groups of which directly interact with electrophilic groups of free radicals. N-acetylcysteine can also enhance the activity of glutathione-S-transferase, glutathione peroxidase, glutathione reductase, and a number of other enzymes involved in maintaining the oxidant/antioxidant balance[57].

Long-term application of N-acetylcysteine in rats with biliary cirrhosis lessened oxidative stress in the mesentery of the small intestine, reduced the level of circulating inflammatory cytokines, and inhibited mesenteric angiogenesis by decreasing angiogenic marker expression (VEGF, VEGFR2, Ang1, and CD31). This eventually improved splanchnic and systemic hemodynamics.

In addition, N-acetylcysteine inhibited VEGF-induced endothelial tubule formation and endothelial cell migration by suppressing tumor necrosis factor-α (TNF-α) and Akt/eNOS/NO angiogenic signaling cascade *in vitro*. It also reduced the number of reactive oxygen species (including reactive compounds of thiobarbituric acid and malondialdehyde) and inflammatory cytokines in the human umbilical vein endothelial cell supernatant[58].

Endothelin receptor blockers
Endothelin-1 (ET-1) is one of the mediators whose synthesis is enhanced in conditions of tissue hypoxia. It belongs to the endothelin family, which includes two more homologous oligopeptides (ET-2 and ET-3). Endothelins are the products of the proteolysis of their precursor "large endothelin", which is driven by the endothelin-converting enzyme. They act through two types of G-protein-coupled receptors: type A (ET_A) and type B (ET_B). Type B (ET_B) has two isoforms: ET_{B1} and ET_{B2}. ET_A are located primarily on membranes of vascular smooth muscle cells, whereas ET_B are present on both endothelial and smooth muscle cells.

ET-1, the most studied potent vasoconstrictor, is produced by vascular endothelial and smooth muscle cells. It is directly involved in intra- and extrahepatic mechanisms of PH pathogenesis, and its circulating level is increased in cirrhosis because of "large endothelin" hyperproduction and increased expression of endothelin-converting enzyme[59]. Experimental studies have shown that ET-1 induces angiogenic responses in

cultured endothelial cells through endothelial ET_B-type receptors and, in combination with VEGF, stimulates neovascularization *in vivo*[60]. The nonselective endothelin receptor blocker bosentan and the selective ET_A receptor blocker ambrisentan reduced the degree of mesenteric angiogenesis and portosystemic shunting in rats with biliary cirrhosis by suppressing inducible nitric oxide synthase (iNOS), cyclooxygenase 2, VEGF and VEGFR2, and Akt signaling[61].

Pioglitazone

Pioglitazone, a potent selective agonist of peroxisome proliferator-activated receptors-γ (PPAR-γ), is able to reduce the level of systemic inflammation in patients with a high cardiovascular disease risk. It blocks the activity of pro-inflammatory genes by post-transcriptional modification of their products (by attaching small SUMO proteins to them) and suppresses NF-κB expression by transrepression. All PPAR isomers (PPAR-α, PPAR-β/-δ, and PPAR-γ) are anti-inflammatory nuclear transcription factors and NF-κB antagonists. Dominant negative mutation of PPAR-γ leads to systemic inflammation and rapid development of related diseases: arterial hypertension, atherosclerosis, type 2 diabetes, nonalcoholic steatohepatitis, psoriasis, and premature aging[62].

In addition to systemic inflammation reduction, PPAR-γ agonists are also capable of inhibiting oxidative stress and angiogenesis[63]. In rats with models of biliary cirrhosis and extrahepatic PH caused by partial portal vein ligation, pioglitazone reduced the degree of portosystemic shunting by 22%-30% by suppressing angiogenic and pro-inflammatory cytokines, chemokines, and growth factors (VEGF, PDGF, and PlGF)[64].

Thalidomide

Thalidomide, a glutamic acid derivative with anti-angiogenic, anti-inflammatory, and immunomodulatory properties, is able to hinder TNF-α/interleukin-1β production for which activated immune cells are responsible[65]. It was also shown in rats with biliary cirrhosis that thalidomide blocked the TNFα-VEGF-NOS-NO pathway by downregulating elevated inflammasome expression in the intestinal and mesenteric tissues, which weakened mesenteric angiogenesis and portosystemic shunting[66].

Polyphenols

The possibility of influencing the pathogenetic mechanisms of extrahepatic angiogenesis was found in polyphenols, the chemicals of plant origin with a strong antioxidant effect.

The tea catechins extracted from the dried leaves of *Camellia sinensis* reduced the severity of mesenteric angiogenesis and portosystemic shunting in rats with biliary cirrhosis by reducing HIF-1α expression, Akt signaling, and VEGF synthesis[67].

2'-hydroxyflavonoid, which is contained in citruses, prevented the formation of new splanchnic vessels and portosystemic collaterals in rats with thioacetamide-induced liver cirrhosis by downregulating apoptosis[68].

The long-term use of curcumin, a polyphenol extracted from turmeric roots, improved the course of PH in liver cirrhosis by positively affecting liver fibrosis and reducing portal influx. These effects were achieved through inhibiting mesenteric angiogenesis and restoring mesenteric vessel contractility, as well as decreasing the degree of portosystemic collateral circulation and hyperdynamic circulatory state. Moreover, its favorable effects on the splanchnic and systemic blood flow included the suppression of VEGF, cyclooxygenase 2, and eNOS[69].

CLINICAL EXPERIENCE OF ANTIANGIOGENIC THERAPY FOR PORTAL HYPERTENSION

The effect of the drugs described above was studied only in experiments involving animals (Tables 1 and 2), and only tyrosine kinase inhibitors were tested as an antiangiogenic therapy in patients with cirrhosis and PH.

Coriat *et al*[70] were the first to assess the effect of sorafenib on the portal and systemic hemodynamics, in seven patients with cirrhosis and hepatocellular carcinoma. Five of them had Child–Turcotte–Pugh (CTP) class A, and two had CTP class B. Sorafenib was administered for one month at 400 mg twice a day. In one patient, this was first reduced to 400 mg once a day and then to 400 mg every two days because side effects appeared. A decrease in portal blood flow by at least 36% was noted, while no changes in blood flow were found in the azygos vein and abdominal aorta.

In a pilot study, Pinter *et al*[71] investigated the effects of sorafenib on HVPG and systemic hemodynamics, as well as the expression of mRNA genes involved in fibrogenesis, angiogenesis, and inflammation in the liver in 13 patients suffering from cirrhosis and hepatocellular carcinoma (10 patients had CTP class A and three patients had CTP class B). The drug was administered at 400 mg twice a day for two weeks. Four of the 11 patients with PH (eight of whom had clinically significant PH) had a reduction in HVPG by more than 20% from the baseline values and, in addition, a decrease in the levels of mRNA, VEGF, PDGF, PlGF, RhoA kinase, and TNF-α.

However, the results were not as optimistic in a randomized double-blind placebo-controlled study that assessed the effects of sorafenib administered at 400 mg twice a day on HVPG in nine patients with cirrhosis and hepatocellular carcinoma[72].

The main drawback of tyrosine kinase inhibitors is hepatotoxicity. A study of possibilities of their selective delivery to target cells, in particular, HSC, seems to be a promising direction in solving this problem.

CONCLUSION

Advances in understanding the pathogenesis of PH in cirrhosis stimulated the development of new methods

Table 1 Drugs that can inhibit intrahepatic angiogenesis in portal hypertension

Ref.	Drugs	Experimental models	Effects
Liu *et al*[15], Qu *et al*[16], Wang *et al*[17], Thabut *et al*[18], Mejias *et al*[19]	Sorafenib	Biliary cirrhosis, non-alcoholic steatohepatitis, thioacetamide-, diethylnitrosamine-, dimethylnitrosamine-, and CCl4-induced cirrhosis	Suppresses the Raf/MEK/ERK signaling pathway and blocks the signaling from the VEGFR, PDGFR, and SCFR; therefore, increases apoptosis and decreases inflammation, fibrogenesis, angiogenesis, and hepatic vascular resistance
Tugues *et al*[20], Majumder *et al*[21]	Sunitinib	CCl4-induced cirrhosis and cell cultures (immortalized human activated HSC cell line, human HSC, and isolated primary human liver sinusoidal endothelial cells)	Blocks VEGFR1/2/3, PDGFR-α/β, FGFR, and SCFR; reduces HSC collagen synthesis, contractility, cellular migration, and SEC angiogenic capacity
Lin *et al*[22], Yang *et al*[23]	Brivanib	Biliary cirrhosis, non-alcoholic steatohepatitis	Inhibits VEGFR and FGFR; therefore, suppresses intrahepatic angiogenesis and portal hypertension, improves blood circulation, and hinders ascites formation
Miao *et al*[31], Marrone *et al*[32]	Simvastatin	CCl4-induced cirrhosis and LX-2 cell line	Enhances KLF2, through which deactivates SEC and reduces the severity of fibrosis and associated angiogenesis
Zhu *et al*[35]	Rifaximin	Biliary cirrhosis	Downregulates bacterial lipopolysaccharide binding to TLR4; therefore, reduces the severity of fibrosis and associated angiogenesis
Liu *et al*[37], Liu *et al*[38]	Largazole	Human colorectal carcinoma cells (HCT116, HT29, and HCT15), human HSC, and CCl4-induced cirrhosis	Suppresses the effects of CD34, VEGF, TGF-β1, and VEGFR2, blocking the main fibrogenic and angiogenic pathways
Michaelis *et al*[40]	Ribavirin	Human umbilical vein endothelial cells	Hinders angiogenesis by inhibiting inosine-5'-monophosphate dehydrogenase 1, tetrahydrobiopterin, NO, and cGMP

CCl4: Carbon tetrachloride; VEGFR: Vascular endothelial growth factor receptor; PDGFR: Platelet-derived growth factor receptor; SCFR: Stem cell growth factor receptor; FGFR: Fibroblast growth factor receptor; HSC: Hepatic stellate cells; SEC: Sinusoidal endothelial cells; KLF2: Krüppel-like factor 2; TLR4: Toll-like receptor 4; TGF-β1: Transforming growth factor beta 1; NO: Nitric oxide; cGMP: Cyclic guanosine monophosphate.

Table 2 Drugs that can inhibit extrahepatic angiogenesis in portal hypertension

Ref.	Drugs	Experimental models	Effects
Fernandez *et al*[46]	Rapamycin and glivec	Partial portal vein ligation	Downregulates VEGF, VEGFR2, CD31, PDGF, PDGFR-β, and α-SMA
Mejias *et al*[19]	Sorafenib	Partial portal vein ligation and CCl4-induced cirrhosis	Blocks VEGF, PDGF, and Raf/MEK/ERK signaling pathway; therefore, reduces intraorgan and systemic blood flow, splanchnic neovascularization, portosystemic shunting, hepatic vascular resistance, and portal pressure
Woltering *et al*[49], Mejias *et al*[50]	Somatostatin and its synthetic analogs	Partial portal vein ligation	Reduces VEGF and CD31 expression, splanchnic neovascularization, and portosystemic collateral circulation by blocking SSTR2
Miternique-Grosse *et al*[55]	Spironolactone	Biliary cirrhosis	Suppresses the effects of aldosterone and the VEGF signal transduction pathway
Lee *et al*[58]	N-acetylcysteine	Biliary cirrhosis	Reduces oxidative stress, inflammatory cytokine levels, TNF-α, VEGF, VEGFR2, Ang1, CD31 expression, and suppresses Akt/eNOS/NO pathway
Hsu *et al*[61]	Bosentan and ambrisentan	Biliary cirrhosis	Block endothelin receptors and suppress iNOS, cyclooxygenase 2, VEGF, VEGFR2, and Akt signaling
Schwabl *et al*[64]	Pioglitazone	Biliary cirrhosis	Downregulates inflammatory genes and NF-κB expression, suppresses angiogenic and pro-inflammatory cytokines, chemokines, and growth factors (VEGF, PDGF, and PIGF)
Li *et al*[66]	Thalidomide	Biliary cirrhosis	Hinders TNF-α/interleukin-1β production and blocks the TNFα-VEGF-NOS-NO pathway
Hsu *et al*[67]	Catechins of Camellia sinensis	Biliary cirrhosis	Reduce HIF-1α expression, Akt signaling, and VEGF synthesis
Hsin *et al*[68]	2'-hydroxyflavonoid	Thioacetamide-induced liver cirrhosis	Downregulates apoptosis
Hsu *et al*[69]	Curcumin	Biliary cirrhosis	Suppresses VEGF, cyclooxygenase 2, and eNOS

CCl4: Carbon tetrachloride; VEGFR: Vascular endothelial growth factor receptor; PDGFR: Platelet-derived growth factor receptor; α-SMA: Alpha smooth muscle actin; SSTR2: Somatostatin receptor type 2; TNF-α: Tumor necrosis factor alpha; Ang1: Angiopoietin 1; eNOS: Endothelial nitric oxide synthase; NO: Nitric oxide; iNOS: Inducible nitric oxide synthase; NF-κB: Factor kappa-light-chain-enhancer of activated B cells; PIGF: Placental growth factor; HIF-1α: Hypoxia-inducible factor-1 alpha.

for its pharmacotherapy. Currently, the drugs of choice are nonselective β-blockers. Nevertheless, their use is not recommended during the subclinical stage of the disease, when the most justified treatment is etiotropic and pathogenetic and aimed at, for example, affecting fibro- and angiogenesis in the liver, as well as angiogenesis underlying the formation of portosystemic shunts. Etiogenic approach, as part of a complex correction of pathophysiological disorders that contribute to the development of PH, may be the key to

success in preventing related complications.

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REFERENCES

1   **de Franchis R**; Baveno VI Faculty. Expanding consensus in portal hypertension: Report of the Baveno VI Consensus Workshop: Stratifying risk and individualizing care for portal hypertension. *J Hepatol* 2015; **63**: 743-752 [PMID: 26047908 DOI: 10.1016/j.jhep.2015.05.022]

2   **Garbuzenko DV**. Contemporary concepts of the medical therapy of portal hypertension under liver cirrhosis. *World J Gastroenterol* 2015; **21**: 6117-6126 [PMID: 26034348 DOI: 10.3748/wjg.v21.i20.6117]

3   **Berzigotti A**, Bosch J. Pharmacologic management of portal hypertension. *Clin Liver Dis* 2014; **18**: 303-317 [PMID: 24679496 DOI: 10.1016/j.cld.2013.12.003]

4   **Coulon S**, Heindryckx F, Geerts A, Van Steenkiste C, Colle I, Van Vlierberghe H. Angiogenesis in chronic liver disease and its complications. *Liver Int* 2011; **31**: 146-162 [PMID: 21073649 DOI: 10.1111/j.1478-3231.2010.02369.x]

5   **Garbuzenko DV**, Arefyev NO, Belov DV. Mechanisms of adaptation of the hepatic vasculature to the deteriorating conditions of blood circulation in liver cirrhosis. *World J Hepatol* 2016; **8**: 665-672 [PMID: 27326313 DOI: 10.4254/wjh.v8.i16.665]

6   **Garbuzenko DV**, Arefyev NO, Belov DV. Restructuring of the vascular bed in response to hemodynamic disturbances in portal hypertension. *World J Hepatol* 2016; **8**: 1602-1609 [PMID: 28083082 DOI: 10.4254/wjh.v8.i36.1602]

7   **Rosmorduc O**. Antiangiogenic therapies in portal hypertension: a breakthrough in hepatology. *Gastroenterol Clin Biol* 2010; **34**: 446-449 [PMID: 20630674 DOI: 10.1016/j.gcb.2010.05.007]

8   **Folkman J**. Angiogenesis: an organizing principle for drug discovery? *Nat Rev Drug Discov* 2007; **6**: 273-286 [PMID: 17396134 DOI: 10.1038/nrd2115]

9   **Chaparro M**, Sanz-Cameno P, Trapero-Marugan M, Garcia-Buey L, Moreno-Otero R. Mechanisms of angiogenesis in chronic inflammatory liver disease. *Ann Hepatol* 2007; **6**: 208-213 [PMID: 18007549]

10 **Lemoinne S**, Cadoret A, El Mourabit H, Thabut D, Housset C. Origins and functions of liver myofibroblasts. *Biochim Biophys Acta* 2013; **1832**: 948-954 [PMID: 23470555 DOI: 10.1016/j.bbadis.2013.02.019]

11 **Yokomori H**, Oda M, Yoshimura K, Hibi T. Enhanced expressions of apelin on proliferative hepatic arterial capillaries in human cirrhotic liver. *Hepatol Res* 2012; **42**: 508-514 [PMID: 22502744 DOI: 10.1111/j.1872-034X.2011.00945.x]

12 **Novo E**, Povero D, Busletta C, Paternostro C, di Bonzo LV, Cannito S, Compagnone A, Bandino A, Marra F, Colombatto S, David E, Pinzani M, Parola M. The biphasic nature of hypoxia-induced directional migration of activated human hepatic stellate cells. *J Pathol* 2012; **226**: 588-597 [PMID: 21959987 DOI: 10.1002/path.3005]

13 **Dufour JF**. Anti-angiogenic therapy for HCC. *Minerva Gastroenterol Dietol* 2012; **58**: 81-86 [PMID: 22419006]

14 **Llovet JM**, Ricci S, Mazzaferro V, Hilgard P, Gane E, Blanc JF, de Oliveira AC, Santoro A, Raoul JL, Forner A, Schwartz M, Porta C, Zeuzem S, Bolondi L, Greten TF, Galle PR, Seitz JF, Borbath I, Häussinger D, Giannaris T, Shan M, Moscovici M, Voliotis D, Bruix J; SHARP Investigators Study Group. Sorafenib in advanced hepatocellular carcinoma. *N Engl J Med* 2008; **359**: 378-390 [PMID: 18650514 DOI: 10.1056/NEJMoa0708857]

15 **Liu L**, You Z, Yu H, Zhou L, Zhao H, Yan X, Li D, Wang B, Zhu L, Xu Y, Xia T, Shi Y, Huang C, Hou W, Du Y. Mechanotransduction-
modulated fibrotic microniches reveal the contribution of angiogenesis in liver fibrosis. *Nat Mater* 2017; **16**: 1252-1261 [PMID: 29170554 DOI: 10.1038/nmat5024]

16 **Qu K**, Huang Z, Lin T, Liu S, Chang H, Yan Z, Zhang H, Liu C. New Insight into the Anti-liver Fibrosis Effect of Multitargeted Tyrosine Kinase Inhibitors: From Molecular Target to Clinical Trials. *Front Pharmacol* 2016; **6**: 300 [PMID: 26834633 DOI: 10.3389/fphar.2015.00300]

17 **Wang Y**, Gao J, Zhang D, Zhang J, Ma J, Jiang H. New insights into the antifibrotic effects of sorafenib on hepatic stellate cells and liver fibrosis. *J Hepatol* 2010; **53**: 132-144 [PMID: 20447716 DOI: 10.1016/j.jhep.2010.02.027]

18 **Thabut D**, Routray C, Lomberk G, Shergill U, Glaser K, Huebert R, Patel L, Masyuk T, Blechacz B, Vercnocke A, Ritman E, Ehman R, Urrutia R, Shah V. Complementary vascular and matrix regulatory pathways underlie the beneficial mechanism of action of sorafenib in liver fibrosis. *Hepatology* 2011; **54**: 573-585 [PMID: 21567441 DOI: 10.1002/hep.24427]

19 **Mejias M**, Garcia-Pras E, Tiani C, Miquel R, Bosch J, Fernandez M. Beneficial effects of sorafenib on splanchnic, intrahepatic, and portocollateral circulations in portal hypertensive and cirrhotic rats. *Hepatology* 2009; **49**: 1245-1256 [PMID: 19137587 DOI: 10.1002/hep.22758]

20 **Tugues S**, Fernandez-Varo G, Muñoz-Luque J, Ros J, Arroyo V, Rodés J, Friedman SL, Carmeliet P, Jiménez W, Morales-Ruiz M. Antiangiogenic treatment with sunitinib ameliorates inflammatory infiltrate, fibrosis, and portal pressure in cirrhotic rats. *Hepatology* 2007; **46**: 1919-1926 [PMID: 17935226 DOI: 10.1002/hep.21921]

21 **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**: 86-95 [PMID: 23454556 DOI: 10.1016/j.ejphar.2013.02.026]

22 **Lin HC**, Huang YT, Yang YY, Lee PC, Hwang LH, Lee WP, Kuo YJ, Lee KC, Hsieh YC, Liu RS. 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**: 1073-1082 [PMID: 24325631 DOI: 10.1111/jgh.12480]

23 **Yang YY**, Liu RS, Lee PC, Yeh YC, Huang YT, Lee WP, Lee KC, Hsieh YC, Lee FY, Tan TW, Lin HC. Anti-VEGFR agents ameliorate hepatic venous dysregulation/microcirculatory dysfunction, splanchnic venous pooling and ascites of NASH-cirrhotic rat. *Liver Int* 2014; **34**: 521-534 [PMID: 23998651 DOI: 10.1111/liv.12299]

24 **Bieker JJ**. Krüppel-like factors: three fingers in many pies. *J Biol Chem* 2001; **276**: 34355-34358 [PMID: 11443140 DOI: 10.1074/jbc.R100043200]

25 **Taniguchi H**, Jacinto FV, Villanueva A, Fernandez AF, Yamamoto H, Carmona FJ, Puertas S, Marquez VE, Shinomura Y, Imai K, Esteller M. Silencing of Kruppel-like factor 2 by the histone methyltransferase EZH2 in human cancer. *Oncogene* 2012; **31**: 1988-1994 [PMID: 21892211 DOI: 10.1038/onc.2011.387]

26 **Kawanami D**, Mahabeleshwar GH, Lin Z, Atkins GB, Hamik A, Haldar SM, Maemura K, Lamanna JC, Jain MK. Kruppel-like factor 2 inhibits hypoxia-inducible factor 1alpha expression and function in the endothelium. *J Biol Chem* 2009; **284**: 20522-20530 [PMID: 19491109 DOI: 10.1074/jbc.M109.025346]

27 **Doddaballapur A**, Michalik KM, Manavski Y, Lucas T, Houtkooper RH, You X, Chen W, Zeiher AM, Potente M, Dimmeler S, Boon RA. Laminar shear stress inhibits endothelial cell metabolism via KLF2-mediated repression of PFKFB3. *Arterioscler Thromb Vasc Biol* 2015; **35**: 137-145 [PMID: 25359860 DOI: 10.1161/ATVBAHA.114.304277]

28 **Gracia-Sancho J**, Russo L, García-Calderó H, García-Pagán JC, García-Cardeña G, Bosch J. Endothelial expression of transcription factor Kruppel-like factor 2 and its vasoprotective target genes in the normal and cirrhotic rat liver. *Gut* 2011; **60**: 517-524 [PMID: 21112949 DOI: 10.1136/gut.2010.220913]

29 **Das A**, Shergill U, Thakur L, Sinha S, Urrutia R, Mukhopadhyay

D, Shah VH. Ephrin B2/EphB4 pathway in hepatic stellate cells stimulates Erk-dependent VEGF production and sinusoidal endothelial cell recruitment. *Am J Physiol Gastrointest Liver Physiol* 2010; **298**: G908-G915 [PMID: 20338920 DOI: 10.1152/ajpgi.00510.2009]

30   **Zeng XQ**, Li N, Pan DY, Miao Q, Ma GF, Liu YM, Tseng YJ, Li F, Xu LL, Chen SY. Kruppel-like factor 2 inhibit the angiogenesis of cultured human liver sinusoidal endothelial cells through the ERK1/2 signaling pathway. *Biochem Biophys Res Commun* 2015; **464**: 1241-1247 [PMID: 26212440 DOI: 10.1016/j.bbrc.2015.07.113]

31   **Miao Q**, Zeng X, Ma G, Li N, Liu Y, Luo T, Lian J, Chen S. Simvastatin suppresses the proangiogenic microenvironment of human hepatic stellate cells via the Kruppel-like factor 2 pathway. *Rev Esp Enferm Dig* 2015; **107**: 63-71 [PMID: 25659387]

32   **Marrone G**, Russo L, Rosado E, Hide D, García-Cardeña G, García-Pagán JC, Bosch J, Gracia-Sancho J. The transcription factor KLF2 mediates hepatic endothelial protection and paracrine endothelial-stellate cell deactivation induced by statins. *J Hepatol* 2013; **58**: 98-103 [PMID: 22989565 DOI: 10.1016/j.jhep.2012.08.026]

33   **Garbuzenko DV**. [The role of intestinal microflora in the development of complications of hepatic cirrhosis-associated portal hypertension.] *Klin Med* (Mosk) 2007; **85**: 15-19 [PMID: 17926483]

34   **Jagavelu K**, Routray C, Shergill U, O'Hara SP, Faubion W, Shah VH. Endothelial cell toll-like receptor 4 regulates fibrosis-associated angiogenesis in the liver. *Hepatology* 2010; **52**: 590-601 [PMID: 20564354 DOI: 10.1002/hep.23739]

35   **Zhu Q**, Zou L, Jagavelu K, Simonetto DA, Huebert RC, Jiang ZD, DuPont HL, Shah VH. Intestinal decontamination inhibits TLR4 dependent fibronectin-mediated cross-talk between stellate cells and endothelial cells in liver fibrosis in mice. *J Hepatol* 2012; **56**: 893-899 [PMID: 22173161 DOI: 10.1016/j.jhep.2011.11.013]

36   **DuPont HL**. Biologic properties and clinical uses of rifaximin. *Expert Opin Pharmacother* 2011; **12**: 293-302 [PMID: 21226639 DOI: 10.1517/14656566.2011.546347]

37   **Liu Y**, Salvador LA, Byeon S, Ying Y, Kwan JC, Law BK, Hong J, Luesch H. Anticolon cancer activity of largazole, a marine-derived tunable histone deacetylase inhibitor. *J Pharmacol Exp Ther* 2010; **335**: 351-361 [PMID: 20739454 DOI: 10.1124/jpet.110.172387]

38   **Liu Y**, Wang Z, Wang J, Lam W, Kwong S, Li F, Friedman SL, Zhou S, Ren Q, Xu Z, Wang X, Ji L, Tang S, Zhang H, Lui EL, Ye T. 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**: 504-515 [PMID: 23279742 DOI: 10.1111/liv.12034]

39   **Fontaine H**, Vallet-Pichard A, Equi-Andrade C, Nalpas B, Verkarre V, Chaix ML, Lebray P, Sobesky R, Serpaggi J, Kreis H, Pol S. Histopathologic efficacy of ribavirin monotherapy in kidney allograft recipients with chronic hepatitis C. *Transplantation* 2004; **78**: 853-857 [PMID: 15385804 DOI: 10.1097/01.TP.0000128911.87538.AA]

40   **Michaelis M**, Michaelis R, Suhan T, Schmidt H, Mohamed A, Doerr HW, Cinatl J Jr. Ribavirin inhibits angiogenesis by tetrahydrobiopterin depletion. *FASEB J* 2007; **21**: 81-87 [PMID: 17135367 DOI: 10.1096/fj.06-6779com]

41   **Bosch J**, Groszmann RJ, Shah VH. Evolution in the understanding of the pathophysiological basis of portal hypertension: How changes in paradigm are leading to successful new treatments. *J Hepatol* 2015; **62**: S121-S130 [PMID: 25920081 DOI: 10.1016/j.jhep.2015.01.003]

42   **Fernandez M**, Vizzutti F, Garcia-Pagan JC, Rodes J, Bosch J. Anti-VEGF receptor-2 monoclonal antibody prevents portal-systemic collateral vessel formation in portal hypertensive mice. *Gastroenterology* 2004; **126**: 886-894 [PMID: 14988842 DOI: 10.1053/j.gastro.2003.12.012]

43   **Fernandez M**, Mejias M, Angermayr B, Garcia-Pagan JC, Rodés J, Bosch J. Inhibition of VEGF receptor-2 decreases the development of hyperdynamic splanchnic circulation and portal-systemic

collateral vessels in portal hypertensive rats. *J Hepatol* 2005; **43**: 98-103 [PMID: 15893841 DOI: 10.1016/j.jhep.2005.02.022]

44   **Angermayr B**, Fernandez M, Mejias M, Gracia-Sancho J, Garcia-Pagan JC, Bosch J. NAD(P)H oxidase modulates angiogenesis and the development of portosystemic collaterals and splanchnic hyperaemia in portal hypertensive rats. *Gut* 2007; **56**: 560-564 [PMID: 16854998 DOI: 10.1136/gut.2005.088013]

45   **Fernández M**, Semela D, Bruix J, Colle I, Pinzani M, Bosch J. Angiogenesis in liver disease. *J Hepatol* 2009; **50**: 604-620 [PMID: 19157625 DOI: 10.1016/j.jhep.2008.12.011]

46   **Fernandez M**, Mejias M, Garcia-Pras E, Mendez R, Garcia-Pagan JC, Bosch J. Reversal of portal hypertension and hyperdynamic splanchnic circulation by combined vascular endothelial growth factor and platelet-derived growth factor blockade in rats. *Hepatology* 2007; **46**: 1208-1217 [PMID: 17654489 DOI: 10.1002/hep.21785]

47   **D'Amico M**, Mejías M, García-Pras E, Abraldes JG, García-Pagán JC, Fernández M, Bosch J. Effects of the combined administration of propranolol plus sorafenib on portal hypertension in cirrhotic rats. *Am J Physiol Gastrointest Liver Physiol* 2012; **302**: G1191-G1198 [PMID: 22403792 DOI: 10.1152/ajpgi.00252.2011]

48   **Garbuzenko DV**. Current approaches to the management of patients with liver cirrhosis who have acute esophageal variceal bleeding. *Curr Med Res Opin* 2016; **32**: 467-475 [PMID: 26804426 DOI: 10.1185/03007995.2015.1124846]

49   **Woltering EA**. Development of targeted somatostatin-based antiangiogenic therapy: a review and future perspectives. *Cancer Biother Radiopharm* 2003; **18**: 601-609 [PMID: 14503956 DOI: 10.1089/108497803322287691]

50   **Mejias M**, Garcia-Pras E, Tiani C, Bosch J, Fernandez M. The somatostatin analogue octreotide inhibits angiogenesis in the earliest, but not in advanced, stages of portal hypertension in rats. *J Cell Mol Med* 2008; **12**: 1690-1699 [PMID: 18194463 DOI: 10.1111/j.1582-4934.2008.00218.x]

51   **Garbuzenko DV**. The principles of management of patients with liver cirrhosis comlicated by ascites. *Klin Med* (Mosk) 2017; **95**: 789-796

52   **Michel F**, Ambroisine ML, Duriez M, Delcayre C, Levy BI, Silvestre JS. Aldosterone enhances ischemia-induced neovascularization through angiotensin Ⅱ-dependent pathway. *Circulation* 2004; **109**: 1933-1937 [PMID: 15078792 DOI: 10.1161/01.CIR.0000127112.36796.9B]

53   **Wilkinson-Berka JL**, Tan G, Jaworski K, Miller AG. Identification of a retinal aldosterone system and the protective effects of mineralocorticoid receptor antagonism on retinal vascular pathology. *Circ Res* 2009; **104**: 124-133 [PMID: 19038868 DOI: 10.1161/CIRCRESAHA.108.176008]

54   **Gravez B**, Tarjus A, Pelloux V, Ouvrard-Pascaud A, Delcayre C, Samuel J, Clément K, Farman N, Jaisser F, Messaoudi S. Aldosterone promotes cardiac endothelial cell proliferation in vivo. *J Am Heart Assoc* 2015; **4**: e001266 [PMID: 25564371 DOI: 10.1161/JAHA.114.001266]

55   **Miternique-Grosse A**, Griffon C, Siegel L, Neuville A, Weltin D, Stephan D. Antiangiogenic effects of spironolactone and other potassium-sparing diuretics in human umbilical vein endothelial cells and in fibrin gel chambers implanted in rats. *J Hypertens* 2006; **24**: 2207-2213 [PMID: 17053542 DOI: 10.1097/01.hjh.0000249698.26983.4e]

56   **Hsu SJ**, Wang SS, Huo TI, Lee FY, Huang HC, Chang CC, Hsin IF, Ho HL, Lin HC, Lee SD. The Impact of Spironolactone on the Severity of Portal-Systemic Collaterals and Hepatic Encephalopathy in Cirrhotic Rats. *J Pharmacol Exp Ther* 2015; **355**: 117-124 [PMID: 26260462 DOI: 10.1124/jpet.115.225516]

57   **Chikina SYu**. Antioxidant effects of N-acetylcysteine in modern clinical practice. *Jeffektivnaja farmakoterapija* 2011; **32**: 19-24

58   **Lee PC**, Yang YY, Huang CS, Hsieh SL, Lee KC, Hsieh YC, Lee TY, Lin HC. Concomitant inhibition of oxidative stress and angiogenesis by chronic hydrogen-rich saline and N-acetylcysteine treatments improves systemic, splanchnic and hepatic hemodynamics of cirrhotic rats. *Hepatol Res* 2015; **45**: 578-588 [PMID: 24961937 DOI: 10.1111/hepr.12379]

59   **Garbuzenko DV**. Pathophysiological mechanisms and new directions of therapy of portal hypertension at liver cirrhosis. *Klin persp gastrojenterol gepatol* 2010; **6**: 11-20

60   **Salani D**, Taraboletti G, Rosanò L, Di Castro V, Borsotti P, Giavazzi R, Bagnato A. Endothelin-1 induces an angiogenic phenotype in cultured endothelial cells and stimulates neovascularization in vivo. *Am J Pathol* 2000; **157**: 1703-1711 [PMID: 11073829 DOI: 10.1016/S0002-9440(10)64807-9]

61   **Hsu SJ**, Lin TY, Wang SS, Chuang CL, Lee FY, Huang HC, Hsin IF, Lee JY, Lin HC, Lee SD. Endothelin receptor blockers reduce shunting and angiogenesis in cirrhotic rats. *Eur J Clin Invest* 2016; **46**: 572-580 [PMID: 27091078 DOI: 10.1111/eci.12636]

62   **Gerasimenko ND**, Degtyar NI, Racin MS. Systemic inflammation and aging: the role of nuclear transcription factors and therapeutic possibilities (Review of the literature). *Probl starenija i dolgoletija* 2016; **25**: 554-561

63   **Dana N**, Javanmard SH, Rafiee L. Role of peroxisome proliferator-activated receptor alpha and gamma in antiangiogenic effect of pomegranate peel extract. *Iran J Basic Med Sci* 2016; **19**: 106-110 [PMID: 27096071]

64   **Schwabl P**, Payer BA, Grahovac J, Klein S, Horvatits T, Mitterhauser M, Stift J, Boucher Y, Trebicka J, Trauner M, Angermayr B, Fuhrmann V, Reiberger T, Peck-Radosavljevic M. Pioglitazone decreases portosystemic shunting by modulating inflammation and angiogenesis in cirrhotic and non-cirrhotic portal hypertensive rats. *J Hepatol* 2014; **60**: 1135-1142 [PMID: 24530596 DOI: 10.1016/j.jhep.2014.01.025]

65   **Enomoto N**, Takei Y, Hirose M, Ikejima K, Miwa H, Kitamura T, Sato N. Thalidomide prevents alcoholic liver injury in rats through suppression of Kupffer cell sensitization and TNF-alpha production. *Gastroenterology* 2002; **123**: 291-300 [PMID: 12105857 DOI: 10.1053/gast.2002.34161]

66   **Li TH**, Huang CC, Yang YY, Lee KC, Hsieh SL, Hsieh YC, Alan L, Lin HC, Lee SD, Tsai CY. 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**: e0147212 [PMID: 26820153 DOI: 10.1371/journal.pone.0147212]

67   **Hsu SJ**, Wang SS, Hsin IF, Lee FY, Huang HC, Huo TI, Lee WS, Lin HC, Lee SD. Green tea polyphenol decreases the severity of portosystemic collaterals and mesenteric angiogenesis in rats with liver cirrhosis. *Clin Sci* (Lond) 2014; **126**: 633-644 [PMID: 24063570 DOI: 10.1042/CS20130215]

68   **Hsin IF**, Lee JY, Huo TI, Lee FY, Huang HC, Hsu SJ, Wang SS, Ho HL, Lin HC, Lee SD. 2'-Hydroxyflavanone ameliorates mesenteric angiogenesis and portal-systemic collaterals in rats with liver fibrosis. *J Gastroenterol Hepatol* 2016; **31**: 1045-1051 [PMID: 26474184 DOI: 10.1111/jgh.13197]

69   **Hsu SJ**, Lee JY, Lin TY, Hsieh YH, Huang HC, Lee FY, Lin HC, Hou MC, Lee SD. The beneficial effects of curcumin in cirrhotic rats with portal hypertension. *Biosci Rep* 2017; **37**: [PMID: 29162665 DOI: 10.1042/BSR20171015]

70   **Coriat R**, Gouya H, Mir O, Ropert S, Vignaux O, Chaussade S, Sogni P, Pol S, Blanchet B, Legmann P, Goldwasser F. Reversible decrease of portal venous flow in cirrhotic patients: a positive side effect of sorafenib. *PLoS One* 2011; **6**: e16978 [PMID: 21340026 DOI: 10.1371/journal.pone.0016978]

71   **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**: 83-91 [PMID: 22032637 DOI: 10.1111/j.1365-2036.2011.04896.x]

72   **Garcia-Tsao G**, Fallon M, Reddy K, Loo N, Bari K, Augustin S, Ciarleglio M, Deng Y, Taddei TH, Strazzabosco M. Placebo-controlled, randomized, pilot study of the effect of sorafenib on portal pressure in patients with cirrhosis, portal hypertension and ablated hepatocellular carcinoma (HCC). *Hepatology* 2015; **62**: 580A.