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PORTAL HYPERTENSION (J GONZALEZ-ABRALDES AND E TSOCHATZIS, SECTION EDITORS)

Portal Hypertension in NASH: Is It Different from Other Aetiologies?

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Abstract

Purpose of Review In non-alcoholic fatty liver disease (NAFLD), an increased portal pressure is observed before signs of cirrhosis or even inflammation or fibrosis are histologically present. This review describes the differences between the mechanisms of cirrhotic portal hypertension (PHT) and PHT in non-cirrhotic NAFLD.

Recent Findings The increased portal pressure in NAFLD is primarily a result of an increased intrahepatic vascular resistance. Vasodilation is decreased by endothelial dysfunction and the sensitivity to vasoconstrictors is increased. Furthermore, the activation of hepatic stellate cells and the presence of microvascular thrombosis could also be involved in the pathogenesis of PHT in NAFLD.

Summary Although the increased portal pressure in early NAFLD is not considered clinically significant PHT, it might play a role in the pathophysiology of NAFLD. Due to the increased intrahepatic vascular resistance, the hepatic blood flow is impaired and hence the oxygen delivery is decreased, potentially triggering transition to steatohepatitis. The underlying mechanisms of these alterations therefore represent promising targets for pharmacological treatment.

Keywords Non-alcoholic fatty liver disease · Portal hypertension · Intrahepatic vascular resistance · Hypoxia · Cirrhosis

Abbreviations

ACh	Acetylcholine
CBDL	Common bile duct ligation
COX	Cyclooxygenase
eNOS	Endothelial nitric oxide synthase
ET	Endothelin
HABR	Hepatic arterial buffer response
HSC	Hepatic stellate cell
HVPG	Hepatic venous pressure gradient
HFD	High-fat diet
H2S	Hydrogen sulphide
HIF	Hypoxia-inducible factor
IHVR	Intrahepatic vascular resistance
LT	Leukotriene
MCD	Methionine-choline-deficient diet
NO	Nitric oxide
NAFLD	Non-alcoholic fatty liver disease
NASH	Non-alcoholic steatohepatitis
OSA	Obstructive sleep apnoea
PGF	Placental growth factor
PHT	Portal hypertension
PGI2	Prostacyclin
ROS	Reactive oxygen species
TX	Thromboxane
TXAS	Thromboxane synthase
TNF-α	Tumour necrosis factor α
VEGF	Vascular endothelial growth factor

This article is part of the Topical Collection on *Portal Hypertension*

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Introduction

Non-alcoholic fatty liver disease (NAFLD) covers a spectrum of disease that is characterised by the accumulation of fat in the hepatocytes. Without signs of inflammation or fibrosis, it is referred to as isolated steatosis or non-alcoholic fatty liver (NAFL). In some cases, NAFLD progresses to concurrent inflammation and hepatocellular damage, known as non-alcoholic steatohepatitis (NASH). The distinction between NAFL and NASH is based on histology, but some pathophysiological mechanisms (e.g. oxidative stress) can already be

activated in NAFL without histological signs of inflammation. Therefore, NAFLD is considered to be a gradual spectrum instead of strictly separated stages of disease. NASH encompasses an increased risk of fibrosis, cirrhosis and hepatocellular carcinoma [1]. Moreover, NASH is an important risk factor for cardiovascular morbidity and mortality [2].

The presence of portal hypertension (PHT), as known in liver cirrhosis, has also been demonstrated in patients with NAFLD. The increase of portal pressure is already present in patients with NAFL, even in the absence of inflammation or fibrosis [3, 4]. Several studies demonstrated that inflammation does not seem to play a significant role in the development of the increased portal pressure [5, 6]. Moreover, a prospective study showed a correlation between the grade of steatosis and the presence of PHT, and steatosis was identified to be an independent predictive factor of the presence of PHT [3].

The increased portal pressure in this and other studies was, however, on average below the threshold of clinical significance (i.e. < 10 mmHg), thus unlikely to cause complications like ascites or variceal bleeding. Its relevance in NAFLD probably lies in its potential to promote progression of isolated steatosis to NASH and further on [7••], as first suggested by Francque et al. [5, 6]. Therefore, this review describes the differences between cirrhotic PHT and PHT in non-cirrhotic NAFLD.

Pathophysiology of Portal Hypertension in NAFLD

Structural Intrahepatic Vascular Alterations in Steatosis and NASH

The increase of portal pressure is primarily a result of an increased intrahepatic vascular resistance (IHVR). Splanchnic vasodilation and a hyperdynamic circulation may subsequently develop to maintain intrahepatic blood flow. However, these compensatory mechanisms may become insufficient as disease progresses, and by increasing the blood inflow, the portal pressure is increased even further [5]. Although we have shown that some of the latter mechanisms are also present in early NAFLD in an animal model [5], they are probably less relevant in the pathogenesis towards NASH or fibrosis. Therefore, the next paragraphs focus on the intrahepatic vascular alterations underlying the increased IHVR (Table 1).

Structural vascular alterations have been observed in early stages of NAFLD. In both alcoholic and non-alcoholic non-cirrhotic hepatitis, enlarged hepatocytes were suggested to play a role in the increased IHVR [8]. Besides sinusoidal narrowing by swollen hepatocytes due to fat vacuoles and ballooning [9, 10], the regular sinusoidal pattern is replaced by disorganised patterns of irregular and flattened blood vessels with numerous interconnections and blind-ending extensions [6]. These findings were reproduced in mice with NASH by electron microscopic scanning [10]. A recent study reported that the disorganised anatomy of the hepatic vasculature in MCD-fed mice could be improved by the inhibition of angiopoietin-2, a regulator of angiogenesis [11•].

Sinusoids are unique capillary structures without basement membranes and with the presence of fenestrae [12]. In capillarisation, a basement membrane develops, matrix proteins dispose around the hepatic vascular structures and fenestrae disappear [13], eventually contributing to an increased IHVR. Capillarisation might also be present in NASH [9] and liver fibrosis [12], but was not observed in rats with cafeteria diet-induced isolated steatosis with increased portal pressure [4].

Angiogenesis is induced by hypoxia-inducible factors (HIFs) and oxidative stress, which are both already seen as early as in isolated steatosis [14] and before the development of fibrosis [15]. Angiogenesis in the liver is mediated by vascular endothelial growth factor (VEGF). In isolated steatosis, increased levels of serum VEGF and soluble VEGF receptor 1 have been demonstrated [16]. Moreover, angiopoietin-2 was increased both in mice and humans with NASH [11•]. Blocking the VEGF receptor 2 was able to diminish steatosis in a mice with NASH [10]. Otherwise, VEGF can also stimulate the formation of fenestrae and prevent capillarisation [12, 17].

Taken together, structural alterations contribute to the development of an increased IHVR in early NAFLD. Mechanisms of capillarisation and the concomitant development of progressive fibrosis can also occur in NAFLD and seem to be similar to the mechanisms observed in cirrhotic PHT. On the other hand, when it comes to the stage of cirrhosis and cirrhosis-related PHT, fat accumulation and other features of NAFLD can disappear, suggesting other mechanisms are then at play. Whether at the cirrhotic stage structural alterations are much different between NAFLD-associated cirrhosis and cirrhosis of other aetiologies, is currently unknown, as no specific data exist on this issue.

Dynamic Intrahepatic Vascular Alterations in Steatosis and NASH

The endothelium plays an important role in vasoregulation, and when this function is disturbed, the physiological tendency of the hepatic vasculature to dilate is lost. A new balance between increased vasoconstriction and decreased vasodilatory mechanisms will result in net increased vasoconstriction, already to be observed in NAFL [4, 6, 14, 18]. In cirrhosis, endothelial dysfunction is also an important inducer

Table 1 Differences between portal hypertension in early NAFLD, cirrhotic NAFLD and cirrhosis of other aetiologies

		Early NAFLD (F0–F2)	Cirrhotic NAFLD (F3–F4)	Cirrhosis
Static	Hepatocyte size	↑	↑ or =	=
	Capillarisation	=	↑	↑
	Angiogenesis	↑	↑	↑
	Sinusoidal disorganisation	↑	↑	↑
Dynamic	NO level	↓	?	↓
	NO sensitivity	↑ or ↓	?	↓
	NOS activity	↑ or↓	?	?
	Vasodilatory reactivity with H_2S	?	?	↑
	Homocysteine (H_2S precursor)	↑	?	↑
	TXAS expression	↑	?	?
	COX-1 expression	?	?	↑
	COX-derived vasoconstrictor level	?	?	↑
	COX-mediated vasoconstriction	↑	?	?
	PGI_2 level	?	?	↓
	Lipooxygenase activity	↑	?	?
	LT level	?	?	↑
	ET-1 level	↑	?	?
	ET-1 reactivity	↑	?	↑
	α1-adrenergic reactivity	↑	?	↑
	HABR	↑	?	↑
Other	Microthrombi	?	?	↑
	TNF-α	=	↑	↑
	HSC activation	↑ or =	?	↑

↑, increased; ↓, decreased; =, no change; ?, not known

NAFL, non-alcoholic fatty liver; *NASH*, non-alcoholic steatohepatitis; *NO*, nitric oxide; *H_2S*, hydrogen sulphide; *TXAS*, thromboxane synthase; *COX*, cyclooxygenase; *PGI_2*, prostacyclin; *LT*, leukotrienes; *ET-1*, endothelin-1; *HABR*, hepatic artery buffer response; *HSC*, hepatic stellate cell

of an increased IHVR [19]. However, there are several differences in the mechanisms of altered vasoregulation between cirrhosis and early NAFLD.

Vasodilation

Nitric oxide (NO) is the most important vasodilator in the hepatic vasculature. In cirrhosis, the bioavailability of NO is decreased [20], but also the reactivity of hepatic stellate cells (HSCs) to NO is impaired [21].

Studies in early NAFLD, however, show conflicting results regarding NO. Acetylcholine (ACh), a stimulator of endothelium-dependent NO-release, decreased the increased portal pressure less in cafeteria-diet or methionine-choline-deficient diet (MCD)-induced steatotic rat livers compared to controls [4, 6]. The direct administration of NO donor sodium nitroprusside did return the elevated transhepatic pressure gradient to normal [4], but these results were not always confirmed [18]. Decreased levels of NO have been demonstrated in rats with high-fat diet (HFD)-induced steatosis [14]. Protein expression of phosphorylated (i.e. activated) Akt, a protein kinase that induces phosphorylation and activation of endothelial NO synthase (eNOS), and expression of eNOS protein and gene were diminished in steatotic livers [4, 6], but eNOS activity appears to be increased [18].

Besides NO, hydrogen sulphide (H_2S) also stimulates vasodilation, but is less studied in the context of PHT in NAFLD. In bile duct-ligated rats, which developed biliary cirrhosis, H_2S decreased norepinephrine-induced vasoconstriction [22]. H_2S production was disturbed in cirrhotic rats, while homocysteine (a H_2S-precursor) accumulated, induced HSC contraction and impaired endothelial NO-production, hence contributing to the hepatic microvascular dysfunction [23]. Interestingly, increased levels of homocysteine have been demonstrated in NAFLD as well [2], so an imbalance between NO and H_2S bioavailability might also play a role in the

increased IHVR in NAFLD, like in cirrhosis of other aetiologies.

Vasoconstriction

In general, steatotic livers demonstrate hyperreactivity to vasoconstrictors, and the concentration of vasoconstrictors appears to be increased in steatosis [6, 14, 18]. This vasoconstrictor predominance is similar to what is observed in cirrhosis of any aetiology [24].

Cyclooxygenase (COX) converts arachidonic acid to prostanoids and, with help of thromboxane synthase (TXAS), to thromboxane (TX). In cirrhosis, COX-1 has been demonstrated to be overexpressed, leading to increased concentrations of vasoconstrictive mediators [13, 25]. The production of prostacyclin (PGI_2), a vasodilatory COX-derived prostanoid, was reduced in the cirrhotic liver [26]. Research on COX-mediated vasoregulation in NAFLD is limited. TXAS expression was significantly higher in MCD-fed rats compared to controls [6]. Moreover, in a rat model of HFD-induced steatosis, COX inhibition reduced TXB2 (a marker of TXA2 production) and improved the impaired vasodilatory response to ACh [14]. These findings point towards a role of altered COX-mechanisms in NAFLD.

Leukotrienes (LTs) are mediators of inflammation of which some have vasoactive properties [27]. In cirrhotic livers with PHT, the level of LTs was increased [26]. In patients with isolated steatosis or NASH, lipoxygenase activity, which stimulates LT production, was increased as well [28]. Moreover, both inhibition of 5-lipoxygenase and blockage of the cysteinyl-leukotriene receptor in cirrhotic rats decreased the portal pressure [29, 30]. Furthermore, LTs have been suggested to play a role in the progression of NAFLD because of their inflammatory effects, hence are a potential therapeutic target [31].

Endothelin-1 (ET-1) is produced by the endothelium and acts on the ET_A and ET_{B2} receptors to induce vasoconstriction and the ET_{B1} receptor to induce vasodilation [32]. The increased sensitivity of cirrhotic liver vasculature to ET-1 appears to be mediated by the ET_A receptor, as blocking this receptor reduced the portal pressure [33]. In rats with steatosis, the serum levels and hepatic expression of ET-1 were increased [6], and the hepatic vasculature demonstrated hyperreactivity to ET-1 [18]. Besides ET-1, both cirrhotic and steatotic livers appear to exhibit hyperreactivity to α1-adrenergic stimulation by methoxamine [18, 25].

Serum levels of vasoconstrictor angiotensin II are increased in chronic liver diseases [34]. When angiotensin II was blocked in patients with cirrhosis, the wedged hepatic venous pressure was decreased [35]. Blocking the angiotensin receptor in mice with HFD-induced steatosis decreased the degree of steatosis [36], but the effects of angiotensin II on the IHVR in NAFLD have not been studied so far. However, these findings imply that interference with the intrahepatic vascular tone could alter disease progression, as discussed in more detail later on.

As a compensatory mechanism for the decreased portal blood flow to maintain liver blood supply, the arterial inflow increases [37]. This mechanism, which has been well documented in cirrhosis, is known as the hepatic arterial buffer response (HABR) and appears to be already activated in early NAFLD as well [38].

Most data on NAFLD result from models of steatosis, but as the vascular alterations lead to worsening of steatosis and progression of NAFLD, it can be assumed that these vascular alterations will be aggravated in NASH, thus maintaining a vicious cycle of hypoxia and NAFLD progression. Altogether, dynamic alterations causing portal hypertension are more important than static alterations. Between cirrhosis and NAFLD the different mechanisms are more or less the same, though the relative importance of certain pathways shifts.

Hepatic Stellate Cells

In addition to the formation of collagen that eventually can result in fibrosis [39], HSCs have other properties that can contribute to the development of an increased IHVR. Activation of HSCs to myofibroblasts appears to be associated to the degree of steatosis [10]. Myofibroblasts respond to vasoactive mediators [40], which might explain in part the hyperreactivity of the hepatic vasculature to vasoconstrictive agents in pathological circumstances. However, smooth muscle α-actin was not yet increased in animal models of isolated steatosis and PHT [4, 6], so the dynamic cause of the increased IHVR appears not to be exclusively dependent on HSCs.

Thrombosis

Microvascular thrombosis might also play a role in the spectrum of liver disease. Blockage of the microvasculature hinders blood flow, and hence IHVR is increased. Microthrombi have been demonstrated in an animal model of viral hepatitis, and the beneficial effect of anticoagulation therapy on fibrosis was shown in portal hypertensive animal models of biliary and toxic cirrhosis [41•]. In humans, anticoagulation by enoxaparin decreased the risk of portal vein thrombosis, decreased the incidence of liver decompensation and improved survival, presumably by its effects on microthrombi in patients with cirrhosis of mixed aetiology [42]. In NAFLD, this phenomenon as such has been hypothesised as well [43] but not been studied so far, and its relevance in the progression of NAFLD is currently unknown. An increase of prothrombotic factors has, however, been documented, not only in obesity and the metabolic syndrome, but also specifically in relation to the histological severity of NASH [44].

TNF-α

Besides cirrhosis and NAFLD, alcoholic hepatitis is another well-known cause of non-cirrhotic PHT [45]. In alcoholic hepatitis, tumour necrosis factor α (TNF-α) appears to play a role in PHT. The levels of TNF-α were higher, and the administration of anti-TNF-α antibodies was able to significantly reduce the hepatic venous pressure gradient (HVPG) in patients with cirrhosis and alcoholic hepatitis [46]. TNF-α stimulated HSC activation in rat livers [47], which might explain its effect on IHVR and subsequently HVPG. Although TNF-α is involved in NAFLD pathogenesis [48], there are no data on its potential involvement in the increased IHVR in early NAFLD.

Portal Hypertension and the Metabolic Syndrome

NAFLD is associated with the components of the metabolic syndrome [49]. For instance, waist circumference and insulin resistance have been shown to be independent predictors of PHT in overweight patients [50]. Likewise, oesophageal varices and PHT have been associated with the presence of type 2 diabetes mellitus [51].

Diabetes is also associated with advanced fibrosis in NAFLD, although most studies are cross-sectional [52]. Besides, there have been reports of another form of hepatic damage due to diabetes, known as diabetic hepatosclerosis. The existence hereof, based on histological findings of hepatic micro-angiopathy in diabetic patients, has been debated [53]. Moreover, no data have been reported on PHT in diabetic hepatosclerosis. The mechanism through which PHT affects diabetes, or vice versa, appears to be the impairment of hepatic insulin clearance by portosystemic shunting, which leads to hyperinsulinaemia and so suggesting an independent effect of PHT on diabetes [54].

In cirrhosis patients without or with overweight, mean HVPG decreased over a 1-year observation period. The decrease was, however, independent of the type of treatment (placebo or the non-selective beta-blocker timolol). Furthermore, the HVPG did not decrease in patients with obesity. Moreover, both BMI and HVPG were independently associated with the risk of decompensation of cirrhosis [55]. An independent link between obesity and PHT has also been reported in a cohort of 354 NAFLD patients [51]. Moreover, weight loss by diet and exercise resulted in decreased PHT [56]. The reason for this observation might be found in the pro-inflammatory effects of obesity.

Clinical Relevance of Portal Hypertension in (Early) NAFLD

The mechanisms of PHT in the cirrhotic stage of NAFLD and cirrhosis of other aetiologies appear to be similar. The effects of severe fibrosis and the nodular reorganisation of the microscopic hepatic architecture potentially dominate the degree of PHT in such a way, that the subtle differences in pathophysiology of PHT between early NAFLD and other aetiologies become less relevant [57].

As described in the previous paragraphs, the differences between NAFLD or other causes of PHT are situated in the earlier stages of disease. A recent retrospective study demonstrated that clinically significant PHT in NAFLD occurs before they can be classified as cirrhosis by liver biopsy, which was not seen in other etiologies of cirrhosis [58•]. In isolated steatosis, however, the portal pressure remains clinically insignificant (i.e. < 10 mmHg), and is thus unlikely to cause complications. Its relevance probably lies in the fact that it is a marker of an increased IHVR that has the potential to promote disease progression. The increased IHVR that is already present in NAFL impairs the intrahepatic blood flow. The subsequently decreased oxygen delivery through the hepatic vasculature can result in local tissue hypoxia, which then triggers several pathways that ultimately lead to the progression to NASH and fibrosis, which in their turn reduce intrahepatic perfusion and hence perpetuate the whole process [7••].

The property of hypoxia to worsen steatosis and drive the progression to NASH has already been demonstrated in the fields of hepatic (transplantation) surgery and obstructive sleep apnoea (OSA). The hepatic perfusion rate and microcirculation were reduced in steatotic donor livers, with diminished vasoconstrictory responses [59]. Further, steatotic livers appear to be more vulnerable to ischaemia-reperfusion injury compared to normal donor livers [60, 61]. OSA, which induces hypoxia by intermittent nocturnal apnoea, has been demonstrated to provoke the progression of NAFLD [62], whereas animal models of (intermittent) hypoxia have been shown to induce hepatocyte injury, hepatic lipid accumulation and hepatic endothelial dysfunction [63, 64]. Some studies report that the treatment of OSA by continuous positive airway pressure therapy is able to decrease elevated transaminases and liver fat as assessed by imaging [65].

Several hints (other than increased angiogenesis as discussed above) of hypoxia in early NAFLD have been reported in both clinical and experimental research. First, the histological damage in NAFLD such as steatosis, Mallory-Denk bodies and fibrosis first appear in the pericentral zone [66–68], which is most sensitive to hypoxia. In the unique microvasculature in the liver, oxygenated blood accounts for 20–30% of the blood supply, whereas the rest of the blood supply comes from the portal vein [12]. Therefore, the centrolobular liver tissue will be the first zone to suffer from the effects of lowered oxygen tension compared with the periportal zone [69]. The hypoxia marker piminodazole was able to demonstrate the focus of hypoxia in the pericentral region of mouse livers with NASH [70].

HIFs, which are degraded in normoxic circumstances and thus remain present when hypoxia is present, have been detected in isolated steatosis as well [71]. HIF-1α was increased in steatotic mouse livers, above all concentrated in the pericentral liver tissue [72]. Overexpression of HIF-1α and HIF-2α in mice resulted in the development of macrovesicular hepatic steatosis [73], whereas deletion of HIF-2α in steatosis after MCD diet decreased lipid accumulation, NASH and fibrosis [15]. In NAFL/NASH patients, HIF-1α and HIF-2α overexpression has also been demonstrated [15, 74].

Several studies suggest that hypoxia can induce hepatic oxidative stress and thereby stimulate progression of NAFLD [75]. Reactive oxygen species (ROS) can be formed by intermittent hypoxia, whereas dysfunctional mitochondria have been demonstrated in NAFLD, which contribute to ROS and consume more oxygen, further decreasing the remaining oxygen tension [70]. Another pathway through which hypoxia can induce NAFLD progression is related to the glucose metabolism. Hypoxia can induce hepatic and systemic insulin resistance, which is related to the severity of NAFLD [1, 76].

More importantly, hypoxia appears to play a role in the development of fibrosis. The degree of fibrosis in NAFLD is currently the best predictor of mortality [77]. A direct link between fibrogenesis and HIFs has been demonstrated in HIF-1α knockout mice and HIF-2α activated mice [78, 79]. Piminodazole identified hypoxic hepatocytes in fibrotic mouse livers after common bile duct ligation (CBDL), besides elevated levels of HIF-1α. In CBDL mice, elevated mRNA levels of fibrogenic genes were attenuated in HIF-1α deficient mice [80]. Levels of HIF-1α, VEGF, placental growth factor (PGF) mRNA and prolyl-4-hydroxylase-α2 mRNA, an enzyme involved in collagen synthesis, were increased in HSCs after they were submitted to hypoxia [81]. The elevated levels were attenuated in HSCs of HIF-1α deficient mice after hypoxia, pointing towards dependence on HIF-1α [81]. Despite these data, it needs to be kept in mind that fibrosis is a result of multiple and complex pathological processes in steatosis and NASH, which all drive fibrosis progression.

NAFLD can ultimately lead to liver cirrhosis, in which two types can be distinguished (although there is a continuum of disease). On the one hand, NASH patients can demonstrate histological evidence of cirrhosis. On the other hand, cirrhosis can be a result of 'burned-out' NASH, in which cirrhosis is present but the histological sings of NASH (mainly steatosis) have disappeared [82]. In the first case, the vascular alterations that have been demonstrated in early NAFLD might still play a role in the development of PHT and the further progression of the disease, although the effects of fibrosis and cirrhosis itself probably disguise the smaller influences of the NAFLD-specific dynamic vascular alterations. In burned-out NASH, the mechanisms of PHT can be assumed to be similar to other causes of cirrhosis, as the specific characteristics of NAFLD seem to have disappeared. Patients with NAFLD-related cirrhosis tend to be older at the time of decompensation, but when decompensation develops, clinical deterioration appears to be more rapid and with a worse prognosis [83]. Whether this is related to intrinsic differences in the mechanisms of disease progression and PHT in particular, or to factors like age and comorbid conditions, remains currently unknown.

Therapy in NAFLD-Related Portal Hypertension

As mentioned before, the clinical relevance of PHT as such appears to be limited in early NAFLD. However, considering the potential pathophysiological role of the increased IHVR in NAFLD, targeting this mechanism might be an interesting therapeutic option.

In cirrhotic PHT, non-selective beta-blockers and splanchnic vasoconstrictors like terlipressin are frequently used [13]. However, these therapies have not been considered or tested in the context of the PHT early in the development of NAFLD as they mainly focus on the extrahepatic contributors to the observed PHT but target very little the intrahepatic components of the IHVR.

As NO plays an important role in the modulation of the IHVR in cirrhosis and potentially also in NAFLD, NO modulation might be an interesting therapeutic option. NO donors might decrease the IHVR, but can also cause a decrease in the systemic vascular resistance and systemic hypotension [84, 85]. In response to hypotension, water and sodium retention will lead to an overload of effective blood volume which actually might increase portal pressure and worsen complications of PHT [13]. Notwithstanding these difficulties, some NO donors have been studied experimentally. The portal pressure in cirrhotic rats was decreased by AVE9488 and tetrahydrobiopterin, which increase NOS transcription or activity [13, 86], and by liver specific NO-donors NCX-1000 and V-PYRRO/NO [87, 88]. V-PYRRO/NO even decreased steatosis in HFD-fed mice [88].

Blocking angiotensin II decreased the portal pressure in patients with liver cirrhosis [35] and decreased the degree of steatosis in a mouse model of steatosis [36]. These vasoregulatory pathways have hence therapeutic potential.

Statins are currently available to treat dyslipidaemia and are considered safe even in the presence of compensated cirrhosis. They have been demonstrated to have beneficial effects on steatosis, inflammation and fibrosis [89, 90] and were even able to cure NAFLD histologically when they were combined with lifestyle therapy [91]. However, these studies were small, prospective, open-label [89, 91] or cross-sectional retrospective [90], and to our knowledge, no randomised controlled trials have been performed in NAFLD yet. In experimental NASH, statins have demonstrated to be able to decrease the portal pressure [92]. Their effects appear to be partially due to

modulating the IHVR, because the reactivity to vasoregulation was enhanced after treatment with simvastatin [84]. Statins were able to lower the IHVR in patients with cirrhosis and attenuate the increased postprandial portal pressure [93]. Moreover, statins have been demonstrated to diminish angiogenesis [94].

Obeticholic acid is a farnesoid X nuclear receptor (FXR) ligand that is currently studied in phase 3 trials for the treatment of NASH [95]. In rat models of toxic and biliary cirrhosis, obeticholic acid decreased PHT [96]. PX20606, another FXR agonist, was also able to attenuate portal pressure in cirrhotic and pre-sinusoidal PHT. In part, the effect of PX20606 was due to increasing eNOS and downregulating ET-1, thus decreasing the IHVR [97]. PPAR modulation is another interesting therapeutic option in NAFLD. Promising agents are PPAR-α/δ dual agonist elafibranor and panPPAR agonist lanifibranor, which are in phase 3 and phase 2 trials respectively, but their effects on the microcirculation of the liver have not yet been studied. The modulation of PPAR-γ might not be useful in improving the hepatic vasculature, as PPAR-γ knock-out mice still expressed vasoregulatory disturbances [98]. PPAR-α, however, has been demonstrated in hepatic endothelial cells and has the ability to decrease the production of ET-1, and thus can potentially influence the hepatic microcirculation [99].

Still, the drugs described above should be studied in more detail to identify any effects on the early hepatic vascular alterations in NAFLD. Further study is needed to elucidate if these drugs can improve the relative hepatic hypoxia and as a consequence can affect NASH progression.

Conclusions

The pathophysiology of PHT in early NAFLD is different from PHT in cirrhosis. As an increased IHVR might disturb oxygen delivery to the liver and thus cause worsening of steatosis and progression of steatosis to NASH and further on, this might be an interesting therapeutic target. Although some NO modulators, statins and obeticholic acid have shown (mostly preclinical) promising results in terms of lowering the portal pressure and improving NAFLD histologically, more research is necessary to prove the link between the IHVR and progression of NAFLD and the effects of decreasing the IHVR to alter the course of the disease. Whether these NAFLD-specific changes are of significance when the liver evolves to cirrhosis and clinically significant PHT, is insufficiently studied, but besides their impact on further disease progression, their role is probably minor, and the dominant mechanism is presumably comparable to other aetiologies of PHT at this stage.

Compliance with Ethical Standards

Conflict of Interest Denise van der Graaff and Wilhelmus Kwanten each declare no potential conflicts of interest.

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