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•Review•

Emerging mechanisms of non-alcoholic steatohepatitis and novel drug therapies

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[ABSTRACT] Non-alcoholic fatty liver disease (NAFLD) has become a leading cause of chronic liver disease globally. It initiates with simple steatosis (NAFL) and can progress to the more severe condition of non-alcoholic steatohepatitis (NASH). NASH often advances to end-stage liver diseases such as liver fibrosis, cirrhosis, and hepatocellular carcinoma (HCC). Notably, the transition from NASH to end-stage liver diseases is irreversible, and the precise mechanisms driving this progression are not yet fully understood. Consequently, there is a critical need for the development of effective therapies to arrest or reverse this progression. This review provides a comprehensive overview of the pathogenesis of NASH, examines the current therapeutic targets and pharmacological treatments, and offers insights for future drug discovery and development strategies for NASH therapy.

[KEY WORDS] Metabolic dysfunction; Drug development; Bile acids; Liver-gut axis; Inflammation; Fibrosis
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Introduction

Non-alcoholic fatty liver disease (NAFLD) is a prevalent hepatic condition characterized by excessive lipid accumulation in the liver, independent of significant alcohol consumption or other identifiable causes of liver injury [1-3]. NAFLD encompasses a spectrum of liver diseases, ranging from simple steatosis (NAFL) to the more severe and progressive form, non-alcoholic steatohepatitis (NASH) [4]. NASH is distinguished by liver inflammation and fibrosis, which can ultimately lead to irreversible liver damage, including cirrhosis and hepatocellular carcinoma (HCC) [5]. Although new nomenclatures such as metabolic dysfunction-associated fatty liver disease (MAFLD) or metabolic dysfunction-associated steatotic liver disease (MASLD) have been proposed, they remain controversial due to neglecting alcohol consumption and failing to encompass all fatty liver patients [6-8]. Consequently, this review will adhere to the original term, NAFLD.

The global burden of NASH has been rapidly increasing, significantly impacting public health [9, 10]. The prevalence of NASH varies by region, with global estimates ranging from 4% to 7% [11]. In high-risk populations, such as individuals with obesity or diabetes, prevalence rates can reach 30% to 40% [12, 13]. NASH is also associated with a higher risk of mortality, primarily due to liver-related complications and cardiovascular diseases [14-16]. The progression from NASH to liver fibrosis, cirrhosis, and HCC is considered irreversible, underscoring the importance of early detection and intervention [17]. Currently, there is only one internationally approved pharmacological therapy for NASH, which has unignorable adverse effects [18]. Lifestyle modifications, including weight loss, exercise, and dietary changes, remain the cornerstone of management [19, 20]. However, these interventions may not be sufficient for all patients, highlighting the urgent need for effective therapeutic strategies.

This review aims to provide a comprehensive overview of the pathogenesis of NASH and the latest therapeutic targets and drugs. By examining recent discoveries in NASH development, we emphasize the urgency of identifying novel drug targets. Specifically, we explore therapeutic approaches that regulate metabolic disorders, oxidative stress, the gut-liver axis, and pathways involving inflammation, fibrosis, and

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cell injury. Additionally, traditional Chinese medicine (TCM) has shown promise in treating NASH but faces limitations due to unclear therapeutic mechanisms and indications. We categorize the mechanisms underlying the potential therapeutic effects of various TCM treatments for NASH to clarify their therapeutic properties. By consolidating this information, we aim to contribute to the development of effective therapeutic interventions for NASH, ultimately improving patient outcomes and reducing the socio-economic burden of this progressive liver disease.

NASH Pathogenesis and Drug Development

Several well-established contributors to the pathogenesis of NASH are illustrated in Fig. 1. Firstly, unhealthy lifestyle factors have been identified as major contributors to the increasing prevalence of NASH [21]. Complex interactions between a Western diet, sedentary behavior, fatigue, and disrupted circadian rhythm provide novel insights for therapeutic interventions [22]. Secondly, disruptions in glucose and lipid metabolism, oxidative stress response, inflammation and fibrosis pathways, and the liver-gut axis play critical roles in the development of NASH [23]. Additionally, genome-wide association studies (GWAS) have identified genetic variants associated with NASH susceptibility, such as *PNPLA3* and *TM6SF2* mutations, which result in abnormal triglyceride accumulation in hepatocytes, ultimately triggering NASH [24, 25].

Given the urgent need for effective and safe treatments, the number of clinical trials dedicated to NASH has been

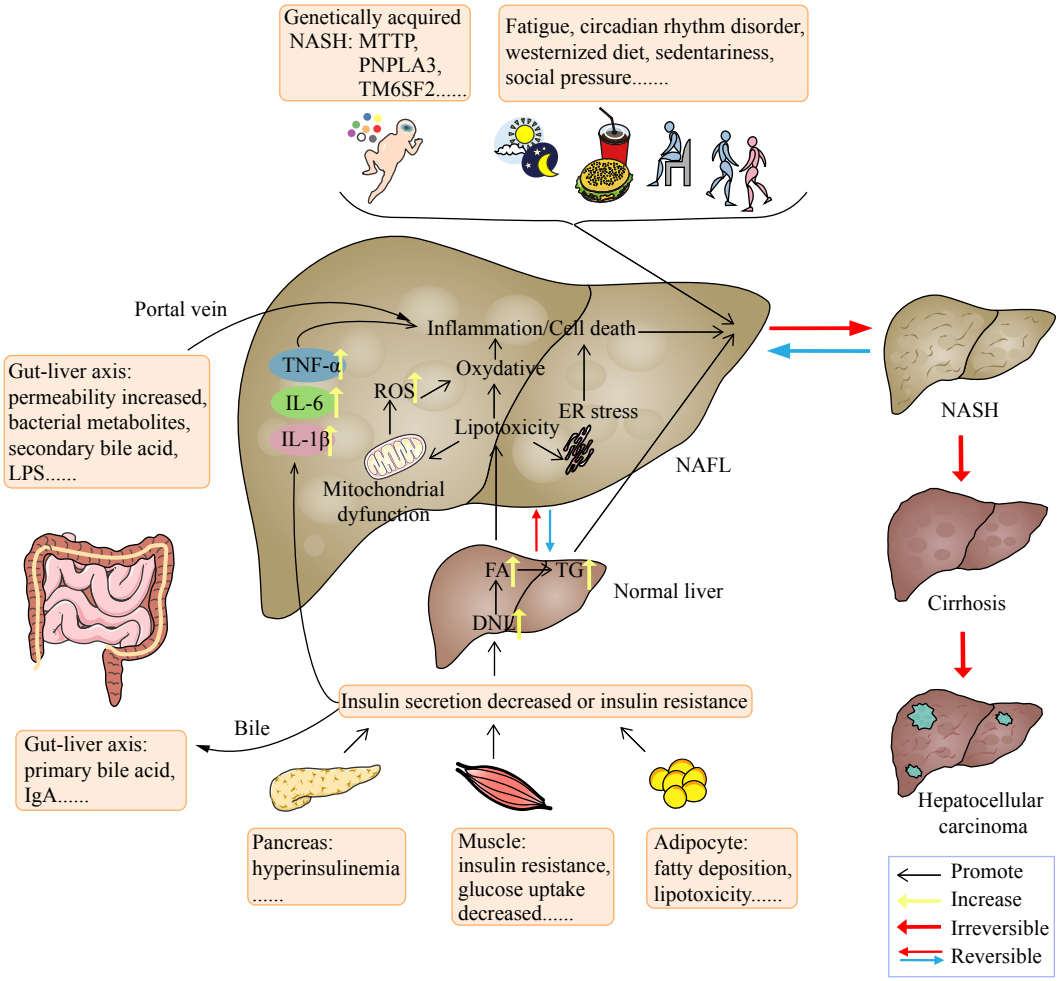

Fig. 1 Well-known contributors to NASH pathogenesis. Excess lipid synthesis or uptake in a normal liver leads to fat accumulation, causing NAFL. Accumulated fats cause lipotoxicity, damaging hepatocytes and triggering inflammatory responses. Lipotoxicity induces inflammation, further driving the progression from NAFL to NASH. As NASH worsens, it leads to cirrhosis and eventually develops into HCC. The progression from NASH to cirrhosis and HCC is irreversible, so treatment strategies should target the progression from NAFL to NASH to prevent progression. Certain genetic mutations increase susceptibility to NASH. Additionally, unhealthy lifestyles, such as high-fat, high-sugar diets and lack of exercise, significantly increase the risk of NASH. Disorders in bile acid metabolism and imbalances in gut microbiota promote the development of NASH. Insulin-sensitive organs produce adipokines and myokines, which enhance liver inflammation and oxidative stress, ultimately leading to NASH. Yellow arrows indicate upregulation in NAFL/NASH, unidirectional red arrows indicate irreversible processes, and red and blue arrows indicate reversible processes. DNL, *de novo* lipogenesis; FA, fatty acid; TG, triglyceride; ER, endoplasmic reticulum; TNF-α, tumor necrosis factor-alpha; IL, interleukin; ROS, reactive oxygen species.

steadily increasing. An analysis of clinical trial data reveals a substantial 1050 registered trials worldwide, focusing on NASH and its associated conditions. Among these, phase II clinical trials are the most numerous (318), followed by phase I trials (160). Notably, the number of phase III clinical trials is relatively low, with only 74 trials in progress. Additionally, 71 trials are currently in the phase IV stage, primarily involving small-sample randomized controlled trials (RCTs) evaluating the therapeutic potential of approved drugs for NASH indications (ClinicalTrials.gov). This review explores potential therapeutic interventions targeting NASH pathogenesis, with the goal of providing insights for new drug development and clinical treatment strategies.

Metabolic dysregulation and potential therapeutic strategies

The "two-hit theory" is widely recognized as the classical pathogenic mechanism of NASH [26]. The first hit primarily involves excessive fat accumulation in hepatocytes, which is associated with insulin resistance. In normal circumstances, insulin promotes the uptake and utilization of glucose. However, in the presence of insulin resistance, elevated blood glucose levels further stimulate insulin secretion, leading to increased synthesis and release of fatty acids. These fatty acids are then converted into triglycerides and stored in the liver, triggering and accelerating the occurrence of NASH (Fig. 2) [27].

Glucagon-like peptide 1 (GLP-1) plays a critical role in enhancing insulin secretion and suppressing appetite, while dipeptidyl peptidase-4 (DPP-4) degrades GLP-1, reducing its biological activity [28-30]. In a phase II study, 39% of patients treated with the GLP-1 analog liraglutide experienced a clear resolution of NASH without severe adverse reactions, compared to only 9% in the placebo group (NCT01237119). Another GLP-1 analog, semaglutide, also showed a significantly higher percentage of patients achieving NASH resolution during a 72-week trial (NCT02970942). However, a recent study assessing the efficacy of semaglutide in NASH patients with compensated cirrhosis found no statistically significant improvements in fibrosis or NASH resolution, with more frequent gastrointestinal adverse reactions reported (NCT03987451). In a phase II trial involving patients with type 2 diabetes mellitus (T2DM) and NASH, tirzepatide, a dual agonist of glucose-dependent insulinotropic polypeptide (GIP) and GLP-1 receptors, significantly reduced liver enzyme levels and increased adiponectin secretion, offering a promising new approach for treating NASH (NCT03131687). Another phase II trial is currently evaluating tirzepatide's safety and efficacy as a once-weekly treatment for NASH (NCT04166773). Additional GLP-1 agonists, including exenatide and efinopegdutide, have also demonstrated efficacy in reducing liver fat content [31-33]. Furthermore, vildagliptin, a DPP-4 inhibitor, has been shown to alleviate liver fibrosis in diabetic rats with NASH by modulating insulin resistance, inflammation, and oxidative stress [34].

Glucose transport and metabolism are crucial for energy production in the body, attracting significant research attention [35]. Glucose transporters (GLUTs), specifically sodium-independent GLUTs and sodium-dependent glucose transporters (SGLTs), play vital roles in glucose metabolism [36]. Overexpression of these transporters leads to increased glucose reabsorption in renal tubules, consequently reducing urinary glucose excretion [37]. This excessive glucose uptake by hepatocytes enhances fatty acid synthesis and fat accumulation [38, 39]. Furthermore, excessive SGLT activity disrupts intracellular sodium levels, causing apoptosis and facilitating the progression from NAFL to NASH [40]. SGLT inhibitors, such as dapagliflozin, empagliflozin, and canagliflozin, have been extensively used as antidiabetic drugs [41-43]. These inhibitors also show beneficial effects on NASH in diabetic rats by reducing oxidative stress, hepatic steatosis, inflammation, and fibrosis through the mitigation of glucotoxicity [44, 45]. In a RCT, dapagliflozin improved hepatic steatosis in patients with T2DM and NAFLD. Notably, in NASH patients with significant hepatic fibrosis, dapagliflozin significantly reduced liver stiffness, serum alanine transaminase (ALT), and gamma-glutamyl transpeptidase levels [46]. The efficacy and safety of dapagliflozin in NASH are currently being evaluated in a phase III clinical trial (NCT03723252). Licogliflozin, a novel SGLT inhibitor, has demonstrated significant efficacy in patients with obesity and T2DM [47]. A 12-week proof-of-concept study involving participants with NASH showed that high-dose licogliflozin significantly decreased serum ALT levels and liver fat content, indicating its potential in halting NASH progression (NCT03205150) [48, 49].

Key enzymes involved in *de novo* lipogenesis (DNL) offer promising therapeutic strategies for the prevention and treatment of NASH [50]. Inhibiting ATP-citrate lyase (ACLY), an enzyme that catalyzes the conversion of citrate to acetyl-CoA and oxaloacetate, has been shown to reduce DNL and increase fatty acid oxidation (FAO) in a mouse model of NASH induced by a high-fat, high-fructose diet [51]. The ACLY inhibitor bempedoic acid effectively suppresses hepatic stellate cell (HSC) activation and fibrosis markers [52]. Clinical development of ACLY inhibitors, such as BGT-002, is ongoing, with promising preclinical efficacy demonstrated in rodent and primate models of NASH. A phase Ib/IIa study evaluating the safety and tolerability of oral BGT-002 in NASH patients has commenced (ChiCTR2300068706). Acetyl-coenzyme A carboxylase (ACC), which converts acetyl-CoA to malonyl-CoA, represents the first committed step in DNL. Clesacostat, an oral ACC inhibitor, demonstrated a 50%−60% reduction in liver fat content in adult NASH patients in a phase IIa study by Pfizer (NCT03248882). Combining clesacostat with the diacylglycerol acyltransferase 2 (DGAT2) inhibitor ervogastat further reduces liver stiffness and prevents the increase in serum triglycerides caused by clesacostat alone (NCT03776175). Firsocostat, a liver-targeted ACC allosteric inhibitor, significantly improves hepatic steatosis in NASH patients when combined with semaglutide and cilofexor (NCT03987074). Another novel ACC1/2 inhibitor, WZ66, has shown excellent results in preclinical

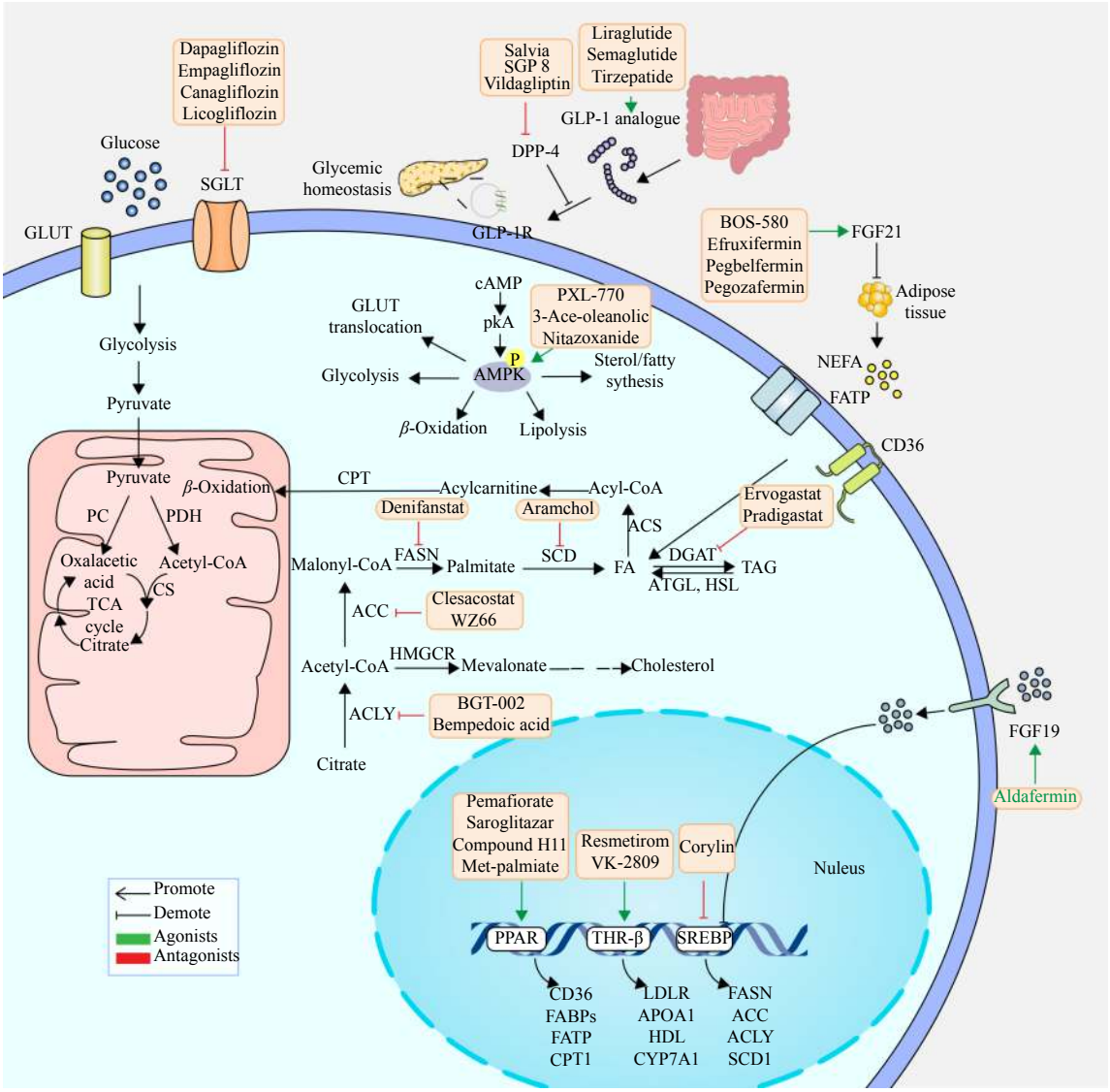

Fig. 2 Metabolic dysfunction and relevant drug strategies in NASH pathogenesis. GLUT and SGLT facilitate glucose uptake into cells. DPP4 degrades GLP-1, which reduces blood glucose levels by enhancing insulin secretion and inhibiting gastric emptying. AMPK activation promotes energy production and inhibits energy consumption. The TCA cycle generates ATP by breaking down carbohydrates, fats, and proteins, providing energy to cells. In adipose tissue and the liver, enzymes such as, ACC, FASN, ACLY, SCD, and ACSL, are involved in fatty acid synthesis and metabolism. DGAT converts these fatty acids into triglycerides stored in the liver. SREBPs, PPARs, and THR-β also play crucial roles in lipid metabolism and liver health, and their dysregulation collectively contributes to the progression of NASH. ACLY, ATP-citrate lyase; ACC, acetyl-coenzyme A carboxylase; FASN, fatty acid synthase; SCD, stearoyl-CoA desaturase; DGAT, diacylglycerol acyltransferase; ACS, Acyl-CoA synthetase; CPT, carnitine palmitoyltransferase; GLP, glucagon-like peptide; SGLT, sodium-dependent glucose transporter; FGF, fibroblast growth factor; NEFA, non-esterified fatty acid; FATP, fatty acid transport protein; THR, thyroid hormone receptor; PPAR, peroxisome proliferator-activated receptor; SREBP, sterol regulatory element-binding protein; TCA, tricarboxylic acid; PC, pyruvate carboxylase; PDH, pyruvate dehydrogenase.

diet-induced NASH mouse models [53]. Denifanstat, a reversible fatty acid synthase (FASN) inhibitor, improves liver fat content and fibrosis markers in NASH patients (NCT03938246, NCT04906421). Other FASN inhibitors, such as TVB-3664 and TVB-3166, have shown efficacy in improving liver steatosis and fibrosis scores in diet-induced NASH mouse models, indicating their potential for treating NASH [54]. Aramchol, an inhibitor of stearoyl-CoA desaturase (SCD1), has demonstrated efficacy in improving NASH and fibrosis in mouse models [55]. It also exhibited dose-dependent efficacy in clinical trials for treating NASH and liver fibrosis, with excellent safety and tolerability (NCT02279524, NCT04104321). Selective inhibition of acyl-CoA synthetase long-chain family member 4 (ACSL4) by abemaciclib reduces liver fat content, serum AST, and liver collagen deposition in diet-induced NASH mice [56]. BMS-963272, a selective monoacylglycerol acyltransferase 2 (MGAT2) inhibitor, reduces inflammation and fibrosis in NASH mouse models without

causing diarrhea which is often associated with selective DGAT1 inhibitors, making MGAT2 inhibitors a promising therapeutic strategy for NASH [57]. Ervogastat, a selective DGAT2 inhibitor, is being evaluated as a monotherapy or in combination with clesacostat in NASH patients (NCT04399538). Additionally, other proteins indirectly regulate lipid synthesis. For example, HSP90β, rather than HSP90α, is overexpressed in the liver of NAFLD patients and obese mice. Selective inhibition of HSP90β by corylin specifically promotes the degradation of mSREBPs, thereby improving obesity-induced fatty liver and NASH progression [58].

Thyroid hormone (TH) and its nuclear receptors (THR) have emerged as promising targets in the development and progression of NASH [59-61]. Resmetirom (brand name Rezdiffra), a once-daily oral THR-β selective agonist, is well-known as the first innovative drug approved by FDA and only one on the market till now for the treatment of NASH, paving a significant milestone in the history of NASH treatment [18]. The approval of resmetirom is primarily based on its excellent phase III clinical results, which show excellent achievement of both primary histological endpoints of NASH resolution and fibrosis alleviation (NCT03900429) [62]. However, side effects such as diarrhea, nausea, and itching cannot be ignored [63]. VK2809, another THR-β agonist, has shown effectiveness, safety, and tolerability in NASH patients, with marked improvements in liver fibrosis (NCT04173065). Unlike resmetirom, VK2809 is a prodrug that is converted into the active compound VK2809A by CYP3A4, providing liver-restricted activity and presenting a promising future treatment option for NASH [64].

AMP-activated protein kinase (AMPK) activation enhances glucose uptake and stimulates glycogen synthesis, thereby inhibiting DNL and promoting FAO, which reduces liver fat accumulation, improves insulin resistance, and lowers blood glucose levels [65, 66]. Additionally, AMPK exerts anti-inflammatory and anti-fibrotic effects, offering potential relief for liver damage in NASH [67]. PXL770, the first direct AMPK activator undergoing human efficacy evaluation, has shown benefits for hepatic steatosis, ballooning degeneration, inflammation, and fibrosis [68]. Although a phase IIa clinical trial assessing PXL770 in NAFLD patients did not meet its primary endpoint, a re-analysis indicated a higher response rate among NASH patients with comorbid T2DM, showing significant improvements in liver fat content, ALT/AST, fasting blood glucose, and reduced inflammation and fibrosis (NCT03763877). Therefore, a 52-week phase IIb clinical investigation in NASH patients with comorbid T2DM or prediabetes is planned (ClinicalTrials.gov). Other AMPK activators, including PF-06409577, have demonstrated their capacity to inhibit DNL, reduce hepatic lipid levels, and decrease fibrosis marker expression in preclinical NASH models [69]. Nitazoxanide, a broad-spectrum thiazolide antiprotozoal and antiparasitic agent, also activates hepatocyte AMPK, reduces blood and liver lipid levels, and ameliorates liver pathology in the early stages of NASH [70].

Peroxisome proliferator-activated receptors (PPARs) play a crucial role in regulating lipid metabolism and offer promising therapeutic targets for NASH. PPARα, predominantly expressed in the liver, promotes FAO and enhances the expression of fibroblast growth factor 21 (FGF21), thereby improving insulin sensitivity and glucose homeostasis [71-74]. The zinc finger transcription factor early growth response 1 (EGR1) interacts with PPARα and accelerates NASH progression by inhibiting PPARα-dependent FAO gene transcription through the recruitment of the corepressor NGFI-A binding protein 1 (NAB1) [75]. Furthermore, extrahepatic PPARα, particularly in the intestine, induces the expression of fatty acid-binding protein 1 (FABP1), enhancing dietary fatty acid uptake and exacerbating NASH progression [76]. PPARγ is involved in adipocyte differentiation, insulin sensitivity, and glucose uptake, while PPARβ/δ plays a role in lipid metabolism and inflammation regulation [77, 78]. Dysregulation of PPAR expression can lead to insulin resistance and fatty acid accumulation, underscoring the importance of modulating PPAR activity to prevent and treat NASH [79]. Pemafibrate, a selective PPARα agonist [80], reduces plasma triglycerides, residual cholesterol, very low-density lipoprotein (VLDL) cholesterol, and apolipoprotein C3 (APOC3) in patients with cardiovascular disease, indicating its potential for NASH treatment (NCT03071692). However, a phase II clinical trial in NAFLD patients did not show a significant reduction in liver fat content (NCT03350165). The combination of pemafibrate with the SGLT2 inhibitor tofogliflozin has shown promise in reducing the progression from NAFL to NASH in a mouse model [81]. Currently, a phase II clinical trial is underway to evaluate the safety and efficacy of the pemafibrate-tofogliflozin combination in NASH patients, with results expected in 2025 (NCT05327127). Interestingly, the beneficial effects of the PPARα antagonist GW6471 in NASH mice are thought to be associated with intestinal mechanisms [76]. Pioglitazone, a PPARγ agonist, has significantly improved liver histology in patients with T2DM and NASH (NCT00994682, NCT01068444). Due to adverse effects linked to its *S*-isomer, the deuterium-stabilized *R*-isomer of pioglitazone, PXL065, has been developed to mitigate these adverse reactions [82]. Its effectiveness and safety in NASH patients are being assessed in clinical trials (NCT04321343). The dual agonist ZLY032, targeting PPARδ and free fatty acid receptor 1 (FFAR1), has significantly improved CCl_4-induced liver fibrosis in mice [83]. Saroglitazar, a PPARα/γ dual agonist, has shown beneficial effects in preclinical studies and a phase II clinical trial in NAFL patients (NCT03061721) and is currently under evaluation in another phase II trial for NASH treatment (NCT03863574) [84]. Lanifibranor, a non-selective PPAR agonist, has completed a phase II clinical trial for NASH treatment, demonstrating reductions in serum liver enzymes and improvements in lipid, inflammation, and fibrosis biomarkers (NCT03008070). A phase III clinical trial for lanifibranor in NASH management is currently in progress (NCT04849728). Triazolone derivat-

ives, which are potent PPARα/δ dual agonists, such as compound H11, have shown strong inhibitory effects on lipid accumulation and fibrosis in diet-induced NASH mouse models [85].

The FGF family consists of 22 polypeptide hormones that play diverse roles in embryonic development, cell proliferation, immune regulation, and metabolic homeostasis [86]. Notably, FGF19 and FGF21 are crucial in regulating glucolipid metabolism and insulin sensitivity, positioning them as potential therapeutic targets for NASH [87-90]. Pegbelfermin, a human FGF21 protein variant, has shown potential in improving metabolic parameters and alleviating hepatic fibrosis in individuals with obesity, T2DM, and NASH [91] (NCT02413372). However, it failed to meet the primary endpoints of improving the NASH clinical research network fibrosis score by at least one stage without worsening NASH at week 48, leading to the discontinuation of its development (NCT03486899, NCT03486912). Other human FGF21 analogs, including BOS-580 [92], efruxifermin [93], and pegozafermin, are currently being evaluated in clinical trials for safety, tolerability, and dose-response relationships in NASH patients, with promising initial results (NCT04880031, NCT04767529, NCT04929483). Aldafermin, an FGF19 analog, improves insulin sensitivity and metabolic balance by inhibiting cholesterol 7*α*-hydroxylase (CYP7A1) [94]. It achieved its primary endpoint by improving liver fibrosis by at least one stage without worsening NASH, although it did not reach the prespecified significance for dose responses (NCT03912532). Despite these advances, the overall efficacy of FGF-based therapies for NASH remains uncertain.

Oxidative stress: ER stress and mitochondrial dysfunction

Oxidative stress plays a critical role as the "second hit" in NASH pathogenesis, leading to the production of reactive oxygen species (ROS), mitochondrial dysfunction, and endoplasmic reticulum (ER) stress, which collectively accelerate disease progression [95, 96]. Mitochondria are essential for fatty acid oxidation, ATP synthesis, and ROS generation [97]. High-fat diet (HFD)-induced obesity impairs mitochondrial fatty acid oxidation in the liver, resulting in the accumulation of lipotoxic substances [98]. This lipid overload burdens mitochondria, causing electron flux overload, electron leakage, and increased ROS production [99]. Additionally, alterations in circular RNA expression within the mitochondria of NASH patients, particularly the downregulation of circRNA ATP5B regulator (SCAR), further contribute to increased ROS production and fibroblast activation [100]. In NASH livers, antioxidant enzyme activities, such as glutathione peroxidase and catalase, are significantly reduced [101-103]. Excessive ROS production leads to the formation of oxidized mitochondrial DNA (mtDNA) fragments, which are released into the cytoplasm. These mtDNA fragments act as danger-associated molecular patterns (DAMPs) and bind to NLRP3 and cGAS, triggering inflammatory responses [104-106]. Targeting mtDNA and its interactions with NLRP3 and cGAS could offer innovative approaches to anti-inflammatory therapy in NASH [107].

The ER is involved in lipid synthesis and protein folding [108]. Excessive lipid accumulation disrupts ER function, triggering the unfolded protein response (UPR), which leads to cellular apoptosis and NASH progression [109]. ER stress sensors, including ATF6, IRE1α, and PERK, play crucial roles in UPR signaling [110]. Under normal conditions, the immunoglobulin-binding protein (BiP) binds to these sensors, keeping them inactive. During ER stress, BiP exhibits a higher affinity for misfolded proteins, exacerbating hepatic steatosis, inflammation, and fibrosis in NASH patients [111]. Disruption of ER-mitochondria interactions and calcium exchange are also early events in diet-induced insulin resistance and NASH development [112].

Dietary intake of the endogenous polyamine spermidine reduces ROS production and rescues mitochondrial dysfunction caused by NASH, indicating the potential for NASH prevention and treatment [113]. Reduced *α*-tocopherol levels in NASH patients suggest the potential benefits of vitamin E (VE), an antioxidant containing *α*-tocopherol [114, 115]. In the methionine-choline-deficient (MCD) diet-induced NASH mouse model, VE supplementation effectively alleviates liver injury by reducing glutathione (GSH) consumption and suppressing the activation of HSCs and COL1α1 expression [116]. Combining VE with other drugs, such as pioglitazone, has been shown to reduce NASH severity in preclinical and clinical studies (NCT00063622, NCT01002547). Additionally, a combination therapy of natural compounds hydroxytyrosol (HXT) and VE demonstrated promising results in a mouse model of CCl_4 plus Western diet-induced NASH fibrosis by interfering with the nuclear translocation/activation of SMAD2/3 transcription factors [117]. A study on biopsy-confirmed NAFLD children showed that HXT and VE improved oxidative stress, insulin resistance, and steatosis (NCT02842567). Other antioxidants are also being investigated for their potential in NASH treatment. Thioredoxin-interacting protein (TXNIP) inhibitors, such as SRI-37330, reduce hepatic and serum triglyceride levels and improve insulin resistance in animal models [118].

Inhibiting Kelch-like ECH-associated protein 1 (KEAP1)-induced degradation of nuclear factor erythroid 2-related factor 2 (NRF2) activates a major endogenous antioxidant pathway. However, existing KEAP1 inhibitors, also known as NRF2 activators, such as sulforaphane, have weak selectivity for KEAP1. A multiple proteolysis targeting chimera (PROTAC)-based approach using 2-cyano-3,12-dioxooleana-1, 9(11)-dien-28-oic acid (CDDO) has shown increased selectivity and strong antioxidant activity, reducing liver fat content and inflammatory infiltration in NASH mice (Fig. 3) [119].

Inflammation and fibrosis

Inflammation and fibrosis are critical drivers in the progression from NAFL to NASH, ultimately leading to liver cirrhosis and HCC [120]. Therefore, anti-inflammatory and anti-fibrotic therapies are crucial for treating NASH. Activated Toll-like receptor 4 (TLR4) initiates the nuclear translocation of NF-*κ*B, which triggers the production of inflammatory

factors such as IL-1β and TNF-α [121-123]. This inflammatory cascade further activates HSCs and the Notch pathway, exacerbating liver fibrosis [124]. Despite these insights, a phase II clinical trial with the TLR4 antagonist JKB-121 did not show significant improvement in liver fat content and inflammation in NASH patients (NCT02442687). While other TLR4 inhibitors like TAK-242 and eritoran have shown promise in animal studies, developing effective TLR4 antagonists for hu-

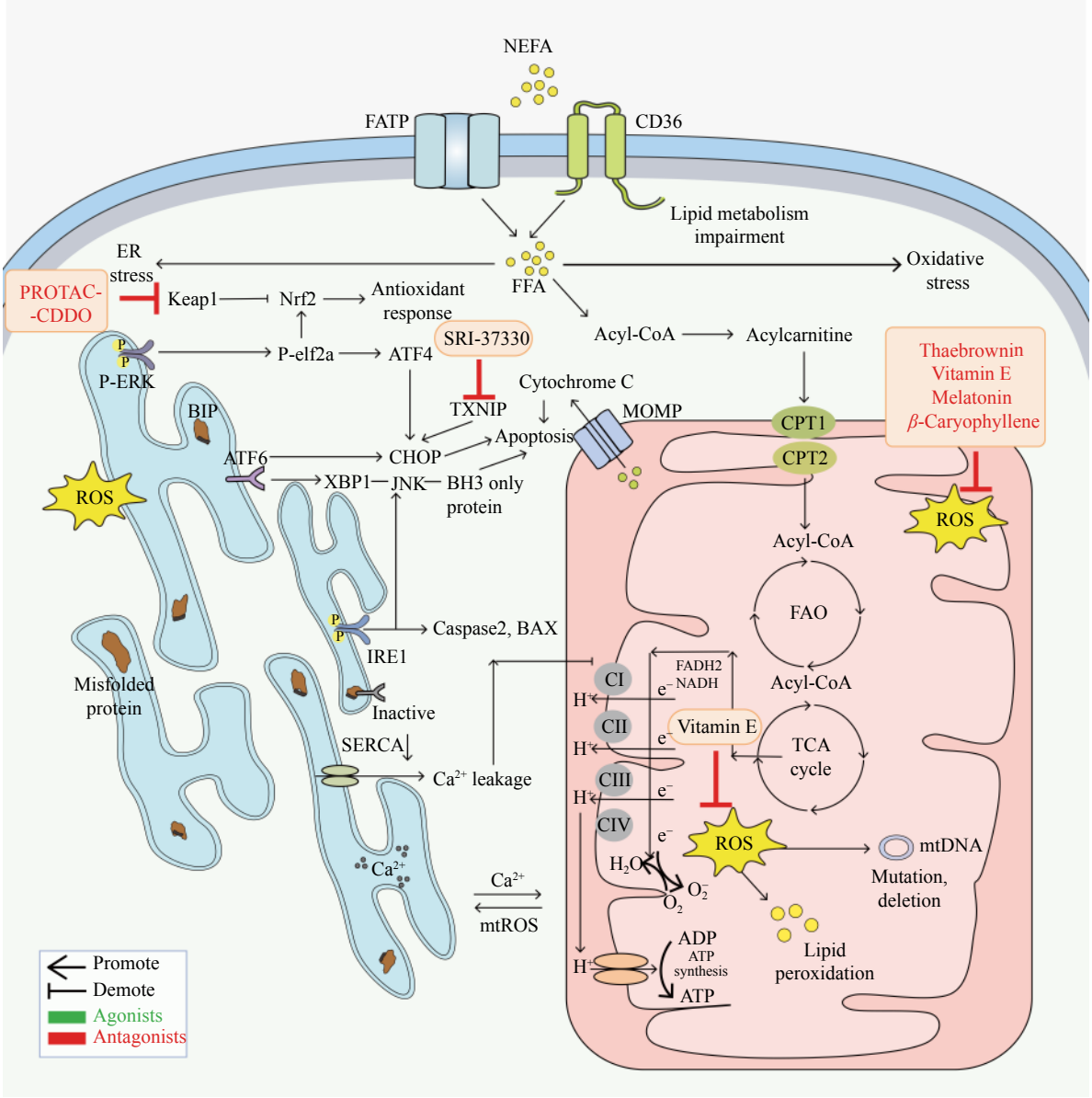

Fig. 3 Developing NASH therapies targeting ER stress and mitochondrial dysfunction. NASH begins with lipid overload, increasing lipid absorption and storage in hepatocytes *via* FATP and CD36. Excess lipids undergo peroxidation, generating ROS that damage cell membranes and organelles. KEAP1 inhibits NRF2 from entering the nucleus, thereby blocking the activation of antioxidant genes. ER stress activates PERK, which phosphorylates eIF2*α*, activating ATF4 to regulate stress response proteins like TXNIP, exacerbating oxidative stress. RE1 splices XBP1 to produce active XBP1 that regulates protein folding and degradation. ATF6 promotes CHOP expression, leading to cell apoptosis. Mitochondrial dysfunction results from lipid overload, elevating acylcarnitine levels, impairing *β*-oxidation, reducing TCA cycle efficiency, and decreasing ATP production and energy supply. Increased ROS levels damage mitochondrial DNA and function. Bcl-2 regulates mitochondrial membrane permeability; BH3 proteins promote MOMP, releasing CytC into the cytoplasm and activating the caspase cascade, resulting in apoptosis. ER stress increases Ca^{2+} release, damaging mitochondria and promoting CytC release and apoptosis. CD36, cluster of differentiation 36; KEAP1, Kelch-like protein 1; NRF2, nuclear factor erythroid 2-related factor 2; P-elf2a, phospho-eukaryotic initiation factor 2; ATF4, activating transcription factor 4; BiP, binding immunoglobulin protein; XBP1, X-box-binding protein 1; CHOP, CCAAT/enhancer-binding protein homologous protein; JNK, c-Jun *N*-terminal kinase; BH3, BCL2 homology region 3; IRE1, inositol-requiring protein-1; SERCA, sarco-endoplasmic reticulum Ca^{2+}-ATPase; BAX, Bcl-2-associated X protein; MOMP, mitochondrial outer membrane permeabilization; TXNIP, thioredoxin interacting protein.

man NASH treatment remains challenging [125-127]. Selective phosphodiesterase 4 (PDE4) inhibitors exhibit potential anti-inflammatory and anti-fibrotic effects by elevating cAMP levels [128]. Roflumilast, an FDA-approved PDE4 inhibitor for chronic obstructive pulmonary disease, has been shown to improve hepatic steatosis and dyslipidemia in mouse models and reduce serum liver enzymes in NASH patients, with good tolerability (NCT01703260) [129]. Activation of the A3 adenosine receptor (A3AR) has also been found to reduce inflammation and improve liver function in NASH animal models [130]. Namodenoson, an A3AR agonist, has demonstrated dose-dependent reductions in serum liver enzymes in NASH patients in phase II studies (NCT02927314, NCT04697810). The inflammatory chemokine receptors CCR2 and CCR5 are highly expressed in NASH and contribute to hepatic inflammation [131]. Cenicriviroc, a dual CCR2/5 antagonist, is currently being evaluated for efficacy and safety in NASH patients with liver fibrosis (NCT03028740). Leronlimab, a monoclonal antibody targeting CCR5, has consistently shown reductions in liver fat and hepatic inflammatory markers (NCT04521114). LYS006, a leukotriene A4 hydrolase (LTA4H) inhibitor, effectively inhibits the biosynthesis of pro-inflammatory leukotriene B4 (LTB4). In a phase II clinical trial, LYS006, alone or in combination with the farnesoid X receptor (FXR) agonist tropifexor, significantly reduced liver fat content in NASH patients, although some reported gastrointestinal discomfort and dizziness (NCT04147195). Annexin A5 (AnxA5) has shown promise in reversing metabolic reprogramming in activated macrophages, facilitating the shift of liver macrophages from a pro-inflammatory M1 phenotype to an anti-inflammatory M2 phenotype. This shift effectively mitigates NASH symptoms, including steatosis, inflammation, and hepatocyte necrosis, in HFD-induced mice [132].

Angiotensin II receptor inhibition by losartan has been shown to inhibit TGF-β signaling, reduce collagen deposition, and improve liver fibrosis [133, 134]. Lysyl oxidase-like 2 (LOXL2) promotes collagen cross-linking and inhibits collagen degradation, playing a crucial role in liver fibrosis progression [135] and plays a pivotal role in liver fibrosis progression [136-139]. In a thioacetamide (TAA)-induced liver fibrosis model, blocking LOXL2 with a monoclonal antibody significantly reduced collagen cross-linking and alleviated fibrosis [140]. Autotaxin (ATX) is an enzyme that hydrolyzes lysophosphatidylcholine (LPC) to generate lysophosphatidic acid (LPA) [141], which plays a role in NASH [142, 143]. ATX inhibitors, such as PAT-505 and CPD17, have shown significant reductions in liver fibrosis in NASH models, indicating a promising approach for treating liver fibrosis in NASH patients [144]. Gal-3, a member of the *β*-galactoside-binding protein family, activates inflammasomes in hepatic macrophages and promotes liver fibrosis by activating HSCs in the injured liver [145]. Belapectin, a Gal-3 inhibitor, has been shown to reverse liver fibrosis and cirrhosis in preclinical studies [146] and is currently under investigation in a phase IIb/III clinical trial for NASH-induced cirrhosis, with good safety and tolerability reported (NCT02462967). CCL24 activates fibroblasts and recruits immune cells, acting as a critical regulator in fibrotic diseases [147]. The monoclonal antibody CM-101, which targets CCL24, has demonstrated improvements in liver fibrosis-related biomarkers in NASH patients (NCT05824156). Cyclophilin B is a vital enzyme involved in collagen protein folding, synthesis, secretion, and the formation of fibrotic matrices [148]. The cyclophilin B inhibitor rencofilstat has achieved its primary endpoint of improving liver function, as well as several secondary endpoints, including reductions in liver injury and fibrosis-associated biomarkers, in NASH patients with stage 3 fibrosis in a phase II clinical trial (NCT05461105) (Fig. 4).

Liver-gut axis

The liver-gut axis plays a crucial role in the development of NASH, functioning as a bidirectional communication network between the liver and the intestine [149]. Primary bile acids secreted by the liver conjugate with taurine or glycine to form conjugated bile acids, which are actively transported into bile by the bile salt export pump (BSEP) and released into the duodenum [150]. Most unconjugated bile acids are absorbed by intestinal epithelial cells *via* the apical sodium-dependent bile acid transporter (ASBT), then secreted into the portal vein and enter hepatocytes *via* the sodium/taurocholate cotransporting polypeptide (NTCP) and sodium-independent organic anion transporter polypeptide (OATP) [151]. Studies have shown that higher concentrations of bile acids correlate with more severe hepatic fibrosis [152]. Additionally, bile acids act as signaling molecules through the FXR and Takeda G protein-coupled receptor 5 (TGR5) pathways, participating in the metabolism of glucose, lipids, and cholesterol, thus regulating the development of NASH [153-155].

Obeticholic acid (OCA), an FXR agonist, has made significant strides in NASH therapeutics as the first drug to enter phase III clinical trials worldwide (clinicaltrials.gov). An ongoing study involving 2480 patients with non-cirrhotic NASH and liver fibrosis has shown promising interim results. After 18 months of OCA treatment, a higher percentage of patients experienced fibrosis improvement without worsening of NASH (NCT02548351). However, final results are pending, and the interim data are insufficient to support a new drug application. The FDA requires completion of the long-term phase of the study before any resubmission of OCA for NASH treatment. Recent studies suggest that FXR agonists have limited efficacy in NASH patients due to FXR SUMOylation, which renders HSCs insensitive to OCA and other FXR agonists. SUMOylation inhibitors enhance OCA's effectiveness by rescuing FXR signaling and reducing fibrosis in NASH mouse models [156]. Additionally, FXR acetylation under liver injury conditions hinders its nuclear translocation, leading to increased cytoplasmic retention and degradation mediated by the carboxyl terminus of HSP70 interacting protein. SIRT1 activators mitigate FXR acetylation and degradation, suggesting that combining SIRT1 activators

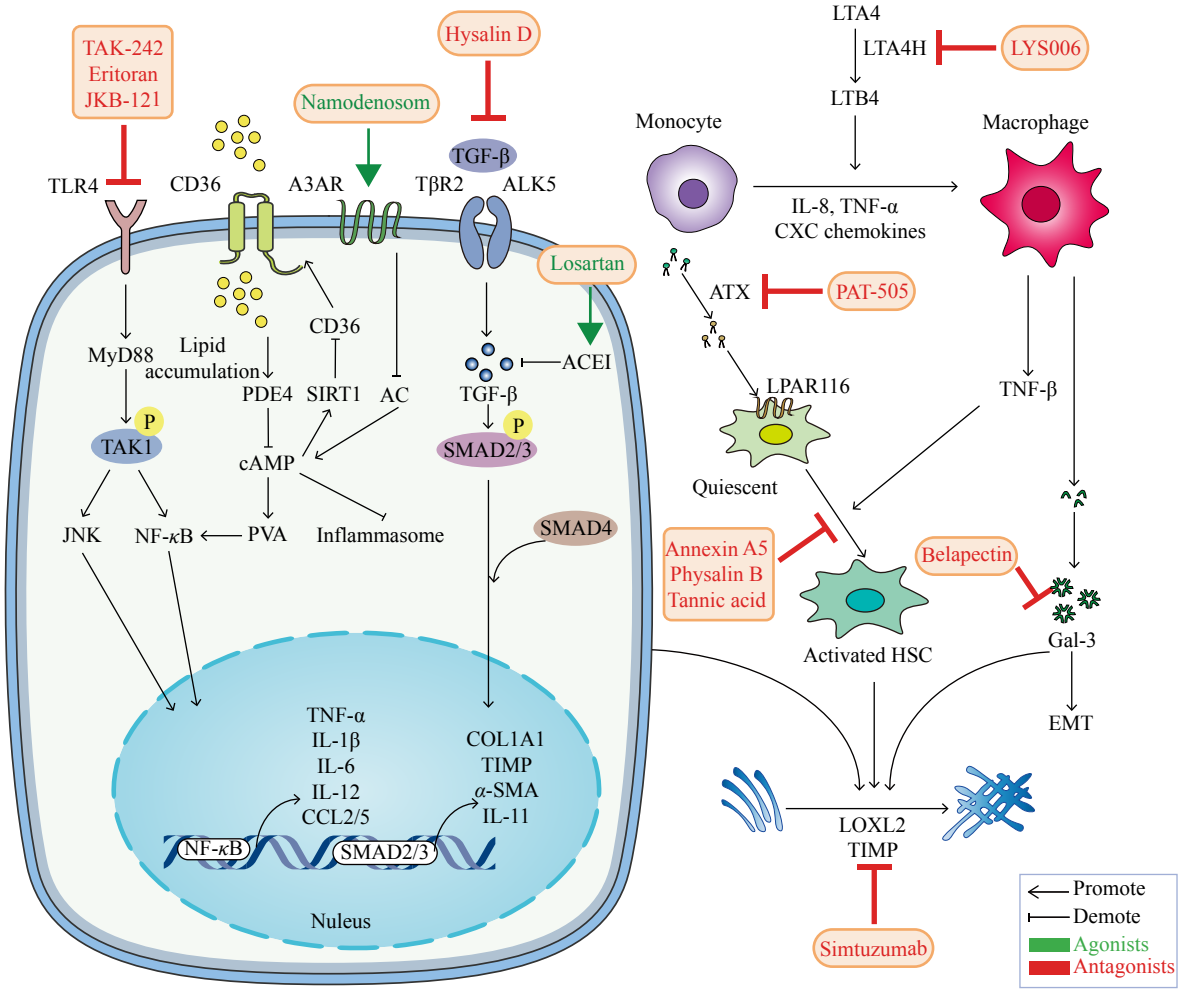

Fig. 4 Drugs regulating the inflammatory response and fibrosis in NASH. TLR4 recognizes pathogen-associated molecular patterns and activates MyD88, which, in turn, activates JNK and NF-κB. NF-κB translocates to the nucleus, initiating the transcription of inflammatory cytokines. CD36 is responsible for the uptake of fatty acids. When overexpressed, it leads to fat accumulation and inflammation. PDE4 degrades cAMP, inhibiting its anti-inflammatory effects. Regulation of PVA and SIRT1 promotes CD36 reutilization. A3AR activation of AC increases cAMP levels, which inhibits NF-κB activity and reduces inflammation. TGFβ binds to TβR2, activating ALK5, which phosphorylates SMAD2/3. These combine with SMAD4 and translocate to the nucleus to transcribe fibrosis-related genes. LTA4-LTB4 converts monocytes to macrophages, producing TNFα and Gal3, driving EMT. ATX binds to LPAR116, activating hepatic stellate cells to produce collagen, leading to liver fibrosis. TLR4, Toll-like receptor 4; A3AR, A3 adenosine receptor; TGFβ, transforming growth factor beta; ALK5, TGFβ activin receptor like kinase 5; TAK1, transforming growth factor-β activated kinase 1; NF-κB, nuclear factor kappa-light-chain-enhancer of activated B cells; PDE4, phosphodiesterase 4; SIRT1, NAD-dependent deacetylase sirtuin-1; AC, adenylate cyclase; ACE, angiotensin-converting enzyme; LTA4, leukotriene A4 methyl ester; LPAR, lysophosphatidic acid receptor; EMT, epithelial-mesenchymal transition; LOXL2, lysyl oxidase-like protein 2; TIMP, tissue inhibitor of metalloproteinase.

with FXR agonists could be promising [157]. OCA-induced changes in gut microbiota lead to lipid peroxidation, which impairs its anti-fibrotic effect, indicating the need for co-administration of OCA with lipid peroxidation inhibitors [158]. Despite clinical challenges, OCA's efficacy remains significant, necessitating further investigation to mitigate side effects and prolong its action [159]. Other FXR agonists, such as MET-642 and cilofexor, are also showing promise in phase II clinical trials by reducing liver fat content in NASH patients (NCT04773964, NCT02854605). INT-767, a dual FXR/TGR5 agonist, dose-dependently improves NASH severity by reducing inflammatory cell infiltration and fibrotic biomarkers [160]. Comparative studies between OCA and INT-767 in NASH mice revealed that INT-767 exhibits superior efficacy [161].

Dysregulation of transporters, including BSEP, ASBT, NTCP, and organic solute transporter alpha/beta (OSTα/β), disrupts bile acid homeostasis, leading to liver fat accumulation, inflammation, and fibrosis [162]. Volixibat, an ASBT inhibitor, increases fecal bile acid levels and reduces hepatocyte enlargement, hepatic triglycerides, and cholesterol esters in NASH mice induced by HFD [163]. However, phase II clinical trials showed that volixibat failed to reduce liver fat content or protect against liver injury (NCT02787304). Similarly,

another ASBT inhibitor, elobixibat, did not significantly improve ALT levels or reduce liver fat content in NASH patients in a recent phase II clinical trial (NCT04006145). Nevertheless, combining the ASBT inhibitor GSK672 with FGF15 activation showed synergistic effects in suppressing the reabsorption of intestinal bile acids and reducing liver bile acid synthesis, resulting in a more pronounced reduction in the bile acid pool and improved therapeutic outcomes in NASH mice [164]. Hepalatide, an NTCP agonist, significantly decreased serum triglycerides, cholesterol, and ALT levels in diet-induced NASH mice by mitigating inflammation, hepatic steatosis, cell apoptosis, and fibrosis [165].

The progression of NASH is also linked to changes in gut microbiota composition, which compromise the intestinal barrier, allowing harmful substances and bacteria to enter the bloodstream. This interference with liver glucose/lipid metabolism and bile synthesis ultimately promotes NASH development [166-168]. *Akkermansia muciniphila* (*A. muciniphila*), a mucin-degrading bacterium found in the mucus layer, is significantly reduced in obese patients [169]. Oral administration of *A. muciniphila* improves hepatic steatosis, decreases inflammation in NAFL, and prevents NASH by regulating γδT17 cell activation and macrophage polarization [170]. A phase II clinical trial is currently evaluating the efficacy and safety of lyophilized fecal microbiota transplantation capsules containing *A. muciniphila* along with two other probiotics (*A. soehngenii* and *B. animalis* subsp.) for treating NASH patients (NCT05821010). Nobiletin has been shown to suppress the progression of diet-induced NASH in mice by reversing gut microbiota dysbiosis, primarily by increasing Bacteroidetes [171]. Other probiotics, such as *Lactobacillus lactis* and *Pediococcus pentosaceus*, have demonstrated potential in improving diet-induced NAFLD in mice [172]. Clinical studies are exploring the use of *P. pentosaceus*, *L. lactis*, or *L. helveticus* for treating NASH patients (NCT04555434). Additionally, combinations of probiotics like *Lactobacillus rhamnosus* GG, *Bifidobacterium breve* BR03, and *Lactobacillus plantarum*, along with compounds such as glutamine, quercetin, vitamin E, curcumin, silybin, and pectin, are under investigation for NASH treatment (NCT04781933) [173-175]. Further research is necessary to fully understand these interventions and determine their long-term efficacy and safety in clinical settings (Fig. 5).

Cell death and pro-survival of hepatocytes

Cell death is an intrinsic aspect of both physiological and pathological processes. Various cell death modalities, such as apoptosis, pyroptosis, autophagy, and ferroptosis, are clinically associated with the progression of NASH and cirrhosis [176]. Apoptosis, a form of programmed cell death, is commonly observed in the hepatocytes of NASH patients [177]. Inhibition of apoptotic proteases caspase 2/3 reduces NASH development in mice by mitigating apoptosis [178, 179]. Pharmacological agents that inhibit apoptosis also attenuate the pathological progression of NASH. Selonsertib, a small-molecule inhibitor of apoptosis signal-regulated kinase 1 (ASK1), alleviates liver fibrosis in patients with F2/3 liver fibrosis in phase II clinical trials (NCT02781584). Combination therapy involving the apoptosis inhibitor IDN-655 and OCA has shown promise in attenuating liver fibrosis in NASH mice [180]. In contrast to apoptosis, ferroptosis is an iron-dependent, non-apoptotic form of cell death linked to the emergence of steatohepatitis inflammation in the early stages of NASH [181, 182]. Treatment with ferroptosis inhibitors, such as liproxstatin-1 or the iron chelator deferiprone, has shown positive results in experimental models [183]. Impaired autophagy, characterized by reduced autophagy in liver sinusoidal endothelial cells and mitochondrial autophagy, has been observed in NASH patients [184-186]. Activation of autophagy, either by overexpressing the autophagy-related protein 7 (ATG7) or using the mTOR inhibitor rapamycin, successfully reduces hepatic steatosis in obese mouse models [187]. However, it is worth noting that overexpression of ATG3 increases hepatocyte lipid accumulation, suggesting the need to consider its independent roles when targeting autophagy for NASH treatment [188]. Pyroptosis, a form of cell death characterized by the activation of pro-inflammatory cells, is also implicated in NASH [189]. In mice fed an MCD diet, reduced expression of gasdermin D (GSDMD) slows NASH pathology [190]. The GLP-1 analog exenatide, commonly used to treat T2DM, attenuates NASH caused by the MCD diet by suppressing pyroptosis signaling pathways [191]. These findings highlight the potential of targeting cell death pathways as a promising therapeutic strategy to inhibit NASH progression (Fig. 6).

Pro-survival signals and receptors, such as the TRK receptor family (TrkA, TrkB, and TrkC), are implicated in the progression of NASH. These receptors are activated by neurotrophin proteins: nerve growth factor (NGF) binds to TrkA, brain-derived neurotrophic factor (BDNF) and neurotrophic factor 4 (NT-4) bind to TrkB, and neurotrophic factor 3 (NT-3) binds to TrkC [192, 193]. The initiation of the TRK pathway occurs when a neurotrophin binds to a TRK receptor on the cell surface, leading to receptor dimerization [194]. In healthy hepatocytes, TrkB expression is mostly absent. However, our research has shown that a truncated isoform of TrkB (TrkB-T1) is significantly elevated in a diet-induced mouse model of NASH. This elevation is conserved between NASH mice and clinical patients. Our experiments demonstrate that increased expression of TrkB-T1 promotes inflammatory signaling and stress-induced cell death in a cell-autonomous manner. Furthermore, BDNF reduces TrkB-T1 expression in hepatocyte membranes, protecting mice from diet-induced NASH [195]. Similarly, TrkC expression is absent in normal liver tissue but is induced in HSCs of NASH patients. The use of LOXO-195, a highly specific TrkC kinase domain inhibitor, reduces the interaction between TrkC and NT-3. This inhibition prevents the activation of human HSCs and reverses advanced murine NASH fibrosis [196]. These findings highlight the critical role of TRK receptors and their pathways in the progression of NASH and suggest potential therapeutic strategies by targeting these pathways.

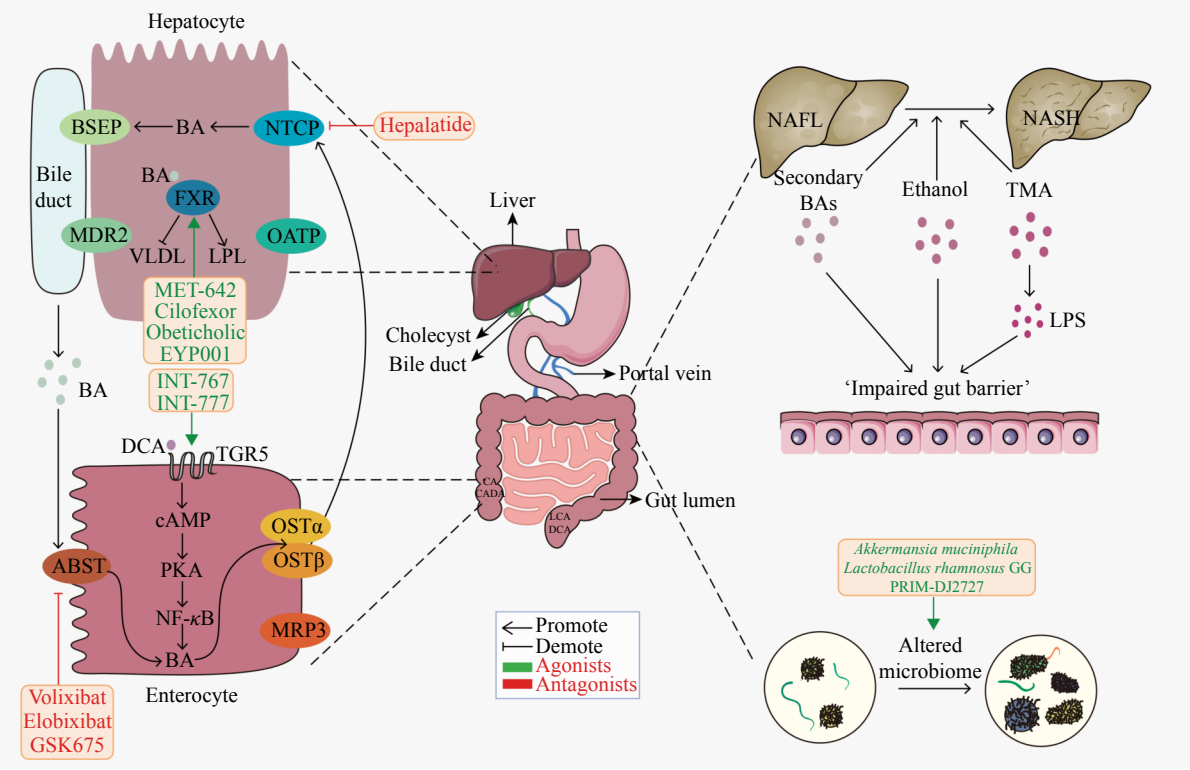

Fig. 5 Drugs targeting gut–liver axis in NASH. Bile acids are produced in the liver and help in the digestion and absorption of fats in the intestine. After their action, they are reabsorbed in the intestine and transported back to the liver through the portal vein. Key proteins involved in this process include BESP, MDR2, NTCP, OATP, ABST, OSTα/β, and MRP3. FXR is activated by bile acids, leading to decreased liver lipid production and inflammation. TGR5 helps with bile acid reabsorption and energy expenditure. Excessive secondary bile acids, ethanol, and trimethylamine (TMA) harm the intestinal barrier, making it more permeable and allowing toxins to enter the liver, which worsens NASH. In individuals with NASH, an imbalanced gut microbiota further compromises the intestinal barrier, leading to increased liver inflammation and fibrosis. BESP, bile salt export pump; MDR2, multidrug resistance protein 2; NTCP, sodium/taurocholate cotransporting polypeptide; OATP, organic anion transporter polypeptide; ABST, sodium-dependent bile acid transporter; OSTα/β, organic solute transporter alpha/beta; DCA, deoxycholic acid; BA, bile acid; TGR5, Takeda G protein-coupled receptor 5.

Others emerging strategies

Extracellular vesicles (EVs) are lipid membrane-bound vesicles released into the extracellular space, playing a crucial role in mediating communication between the liver and other organs by carrying bioactive molecules such as proteins, lipids, DNA, and RNA [197]. In NASH patients, circulating EVs exhibit altered levels and cargo composition compared to healthy individuals, contributing to the activation of HSCs and macrophages, fibrosis, and the occurrence of HCC [198-200]. Primary hepatocytes of mice fed a short-term HFD express higher intracellular levels of let-7e-5p and miR-210-3a, whose elevation in EVs drives triglyceride accumulation in adipose tissue, adapting to metabolic changes induced by lipid overload [201]. Additionally, exosomes rich in miR-28-5p secreted by hepatocytes promote NF-κB transcriptional activity in macrophages and stimulate the expression of inflammatory factors, establishing serum miR-28-5p as a biomarker for NASH diagnosis [202]. Enhancing the uptake of beneficial EVs or counteracting the effects of EVs that promote NASH progression is an emerging therapeutic strategy [203-205].

Mesenchymal stem cells (MSCs), originating from the mesoderm, are multipotent stem cells widely distributed throughout the body [206]. MSCs can promote endogenous tissue regeneration and protect pancreatic β cell function, thereby improving NASH progression caused by insulin resistance [207]. Furthermore, MSCs inhibit inflammation, reduce hepatocyte apoptosis, and suppress hepatic fibrosis and cirrhosis by secreting cytokines such as IL-10 and hepatocyte growth factor (HGF) [208, 209]. Human umbilical cord MSC-derived exosomes (hUC-MSC exosomes) have been found to alleviate liver inflammation, lipid deposition, and macrophage infiltration induced by high-fat, high-cholesterol (HFHC) or MCD diets [210-212]. Similarly, MSCs from human bone marrow improve NASH in immune-deficient mice induced by HFD. Additionally, donor MSCs deliver mitochondria to recipients, enhancing lipid oxidation and restoring metabolic homeostasis [213]. Given their self-replication, low immunogenicity, and high activity, MSCs have gained significant attention for NASH therapy.

Sex hormone replacement therapy has been explored as a potential treatment for NASH, considering the significant gender differences in disease severity and prevalence [214].

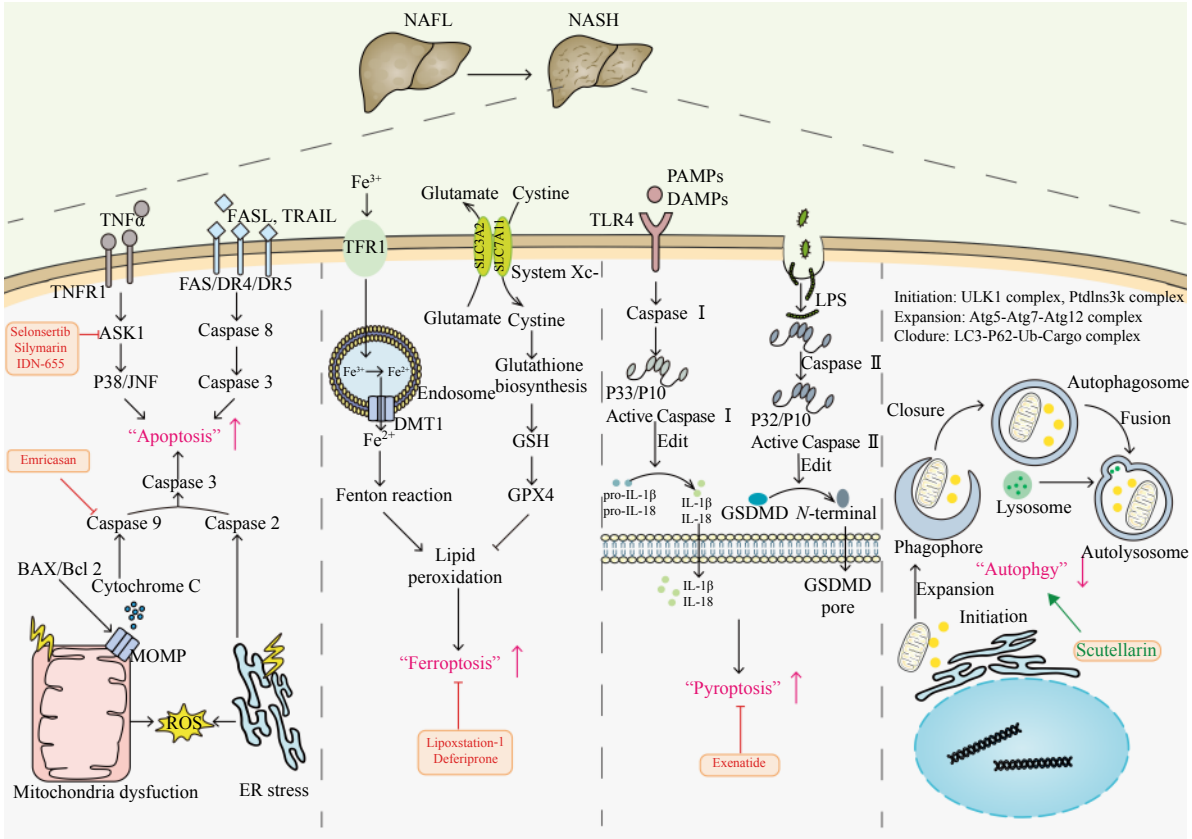

Fig. 6 Programmed cell death and relevant drug strategies in the pathogenesis of NASH. Activation of the TNFR1 receptor by TNFα engages ASK1, P38, and JNK, leading to apoptosis. Stimulation of FAS or DR4/DR5 receptors initiates caspase 8 and caspase 3, also resulting in apoptosis. An elevated BAX/Bcl2 ratio promotes cytochrome C release from mitochondria, which binds with caspase 9 to form the apoptosome, further advancing NASH progression. The TFR1-DMT1 pathway regulates ferroptosis, with TFR1 responsible for iron uptake and DMT1 for iron transport. System Xc−, composed of SLC7A11 and SLC3A2, mediates cystine uptake, converting it into glutathione. GPX4 uses GSH to inhibit lipid peroxidation, preventing NASH. Pyroptosis is highly inflammatory. Activation of the TLR4 receptor induces caspase 1 activation, which cleaves GSDMD to form P33/P10 fragments, causing IL-18 release and cell membrane rupture. LPS activates caspase 2, which subsequently activates GSDMD, leading to cell membrane perforation and an inflammatory response, exacerbating NASH. Autophagy begins from the ER, forming autophagosomes that merge with lysosomes to form autolysosomes. It is believed that autophagy ameliorates NASH progression. FASL, factor-related apoptosis ligand; TRAIL, tumor necrosis factor-related apoptosis-inducing ligand; TFR1, transferrin receptor 1; GSH, glutathione; GPX4, glutathione peroxidase 4; DMT1, divalent metal transporter 1; PAMPs, pathogen-associated molecular patterns; DAMPs, damage-associated molecular patterns; LPS, lipopolysaccharide; GSDMD, gasdermin D; ULK1, Unc-51 like kinase 1.

Men are more prone to develop severe liver fibrosis in NASH compared to premenopausal women [215]. Research has demonstrated a correlation between low testosterone levels and the severity of NASH in men [216]. Therefore, restoring hormone levels within the normal range, such as testosterone replacement therapy (TRT) in males and estradiol supplementation in females [217-219], may benefit liver function by reducing hepatic fat accumulation and improving NASH-related metabolic markers. These interventions require evaluation for efficacy and safety in clinical trials (NCT01919-294) [220-223]. The anti-NASH therapeutic drugs currently in clinical and preclinical stages have been summarized in Tables S1 and S2.

Application of TCM in the treatment of NASH

TCM, renowned for its efficacy and safety, plays a vital role in the development of hepatoprotective drugs. A meta-analysis comparing randomized clinical trials using TCM alone or in combination with placebos, ursodeoxycholic acid, insulin sensitizers, lipid-lowering agents, or antioxidants, found that TCM is more effective than Western medicine in reducing serum ALT levels and alleviating radiographic steatosis in the treatment of NAFLD [224].

Ginsenosides, the primary bioactive components of ginseng, significantly reduce hyperlipidemia. A four-week oral administration of ginsenosides in rats significantly improved HFD-induced liver weight gain and insulin resistance by increasing GLP-1 levels in the portal vein and intestine [225, 226]. Recent advancements have combined ginsenosides with albumin to address poor bioavailability, effectively delaying HFD-induced hepatic steatosis and fibrosis in mice [227].

IAVPGEVA (SGP8), an octapeptide derived from soybean 11S globulin hydrolysis, alleviates diet-induced liver in-

jury and metabolic disorders in NASH mice by inhibiting DPP4 activity [228]. Natural compounds like those found in Salvia improve insulin resistance and hepatic steatosis induced by high-density lipoprotein cholesterol in mice [229, 230]. *Cyclocarya paliurus*, rich in triterpenoids, reduces diet-induced insulin resistance and hepatic steatosis in mice [231-233]. Methyl palmitate, an endogenous PPAR transcription activator, mitigates MCD-induced NASH progression in mice by reducing hepatic steatosis, inflammation, and fibrosis [234].

Baicalin and scutellarin reduce diet-induced NASH by enhancing glucose reabsorption in adipocytes *via* phosphorylation of AMPK and AKT [235-237]. Catalpol enhances skeletal muscle mitochondrial function by activating AMPK-mediated mitochondrial biogenesis [238]. Black quinoa polyphenols (BQPs) alleviate insulin resistance in HepG2 cells by modulating the IRS1/PI3K/Akt/GLUTs signaling pathway, potentially providing new treatments for NASH [239].

Andrographolide reduces HFD-induced weight gain and hepatic fat accumulation in mice by downregulating SREBP target genes, thereby improving serum lipid levels and insulin/glucose sensitivity [240]. Berberine, a natural alkaloid from *Coptis chinensis* and other Chinese herbs, effectively reduces hepatic lipid accumulation and treats NASH in rodent models and patients [241, 242]. Curcumin, a natural polyphenol, inhibits lipolysis and prevents hepatic insulin resistance by suppressing ER stress in adipose tissue [174, 243].

Osthole, an AMPK activator, combats NASH by regulating mitochondrial homeostasis and inhibiting NLRP3 inflammasome activation [244, 245]. *β*-Glycyrrhetinic acid, a bioactive metabolite of glycyrrhizin (GL), alleviates liver inflammation and injury by stabilizing lysosomal membranes, inhibiting cathepsin B, suppressing mitochondrial cytochrome C release, and reducing FFA-induced oxidative stress [246]. Physalin B improves MCD diet-induced NASH by stimulating autophagy and activating P62-KEAP1-NRF2 antioxidant signaling [247]. Schisandra lignans extract significantly reduces liver phosphatidylethanolamines (PEs) and intracellular ROS, suggesting its potential for treating NASH [248]. Antioxidants such as melatonin and theabrownin have also been shown to reduce inflammation, oxidative stress, and fibrosis in NASH models [249, 250].

Silymarin exerts anti-NASH effects by reducing histone deacetylase 2 (HDAC2) activity, enhancing FXR transcription, and promoting FGF-15/19 expression in the ileum [251]. Large-scale trials are currently evaluating silymarin's potential to reduce liver fibrosis (NCT02006498). Magnesium glycyrrhizinate (MGIG), a derivative of *Glycyrrhiza glabra*, has anti-inflammatory effects by inhibiting TLR4 and is widely used in treating inflammatory liver diseases [252].

Sappanone A, a natural PDE4 inhibitor from *Caesalpinia sappan* heartwood, alleviates CCl_4-induced liver fibrosis in mice [253]. Physalin D (PD), a kaferol lactone from Solanaceae plants, alleviates HSC activation and liver fibrosis by blocking TGF-β/Smad signaling [254]. Hypericin K (HK) interacts with DEAD-box protein 5 (DDX5) and prevents its degradation, significantly reducing lipid accumulation, inflammation, and fibrosis in NASH mouse models [255]. Compounds such as 3-tigloyl-khasenegasin F (a mexicanolide-type limonoid derivative) and tannic acid inhibit HSC activation and fibrosis marker expression, potentially improving liver fibrosis [256, 257].

Terminalia bellirica, a traditional Tibetan medicine, restores CDAHFD-induced gut microbiota structural disorders by reducing *Intestinimonas*, *Lachnoclostridium*, and *Lachnospirace* while increasing *Akkermansia* and *Bifidobacterium*, thereby alleviating liver lipid accumulation and cell necrosis in mice [258].

TCM is a valuable source for developing new anti-NASH drugs, but the bioavailability of these compounds often poses a challenge, potentially limiting their efficacy. For instance, resveratrol's concentration in the liver can be insufficient due to low bioavailability. Enhancing bioavailability through structural modification is a key area of research. Additionally, the multi-target nature of TCM adds complexity, necessitating clinical trials to further determine their efficacy for NASH. TCMs currently under clinical evaluation for the treatment of NASH are summarized in Table S3.

Perspectives

The lack of effective diagnostic tools for NASH in clinical trials presents a significant challenge [259, 260]. Although tissue biopsy is considered the gold standard for NASH diagnosis, it is invasive, time-consuming, and high-risk [261, 262]. Transient elastography (TE) has shown high sensitivity in evaluating liver cirrhosis, but its accuracy is compromised in obese patients [263-266]. Currently, NASH diagnosis relies primarily on imaging, clinical history, and biochemical indicators, often leading to late-stage diagnosis and missed opportunities for early intervention [267, 268]. The complex pathogenesis of NASH contributes to scientific disputes and a lack of clarity regarding patient heterogeneity and disease subtypes. This knowledge gap among clinicians results in inconsistent diagnoses, even when analyzing the same pathological specimen [269, 270]. Additionally, translating beneficial effects from preclinical animal studies to human trials has proven difficult, contributing to clinical research failures [271, 272]. Recruiting subjects for clinical trials also poses challenges, as it is difficult to match participants with similar backgrounds regarding drug treatment, disease stages, and physical fitness levels [273, 274].

Recently, a CRISPR-based screening platform using APOB and MTTP mutant organoids has been established to identify regulators and targets of lipid metabolism and assess NAFLD risk genes. This platform pinpointed FADS2 (a rate-limiting enzyme for polyunsaturated fatty acid biosynthesis) as a crucial regulator of lipid metabolism. This novel approach demonstrates the feasibility of liver-like organs in elucidating the early pathogenesis of NASH and suggests new drug targets for liver steatosis, providing promising avenues for drug discovery and development [275]. Utilizing organoids

and metabolomics to explore the pathogenesis of NASH is expected to yield more efficient and insightful results.

NASH arises from a complex interplay of lifestyle factors and genetic predisposition, leading to dysregulated glucose and lipid metabolism, hepatic fat accumulation, and insulin resistance. This metabolic imbalance increases fatty acid oxidation and ROS production, causing oxidative stress and hepatocyte damage. Oxidative stress activates inflammatory pathways, resulting in hepatic inflammation and fibrosis. Additionally, gut microbiota dysbiosis heightens intestinal permeability, allowing bacterial products to enter the bloodstream and activate the hepatic TLR4 signaling pathway, further intensifying inflammation. Cell death *via* apoptosis and necrosis releases cellular contents, perpetuating a vicious cycle of inflammation. Single therapeutic agents targeting specific mechanisms often fail to reverse NASH pathogenesis, making combination therapy a crucial strategy. Innovations such as drug repurposing (e.g., metformin) and emerging therapies (e.g., cell therapy) offer potential breakthroughs. Managing NASH concurrently with diabetes, cardiovascular diseases, and other metabolic syndromes through comprehensive treatment approaches represents a promising direction.

Conclusion

Developing effective treatments for NASH has become a significant challenge for scientists worldwide. Despite only one drug being approved so far, recent years have seen considerable progress in understanding the disease's mechanisms, laying a solid foundation for future drug development. We propose the following therapeutic strategies for NASH:

1. Multi-target intervention: combining drugs with different mechanisms to simultaneously regulate bile acid metabolism, glucose and lipid metabolism, oxidative stress, and inflammation. For instance, the combination of the GLP-1 analog semaglutide, the FXR agonist cilofexor, and the ACC inhibitor firsocostat has proven significantly more effective than monotherapy [276].

2. Personalized treatment: developing individualized treatment plans based on the patient's genetic background, lifestyle, and disease status. For example, patients with *PNPLA3* gene mutations may require specialized approaches to regulate fat metabolism, while those with gut microbiota imbalances might benefit from probiotics or prebiotics.

3. Comprehensive management: inaddition to pharmacotherapy, lifestyle modifications such as dietary control and increased physical activity are essential components of NASH management.

As NASH is a chronic liver disease requiring long-term medication, ensuring drug safety is paramount. TCM, with its long history of clinical use and multi-target effects, has demonstrated considerable safety and efficacy. We anticipate that TCM will make significant contributions to the treatment of NASH.

Supporting Information

Supporting information of this paper can be requested by sending E-mails to the corresponding authors.

References

[1] Negi CK, Babica P, Bajard L, *et al*. Insights into the molecular targets and emerging pharmacotherapeutic interventions for nonalcoholic fatty liver disease [J]. *Metabolism*, 2022, **126**: 154925.

[2] Abdelmalek MF. Nonalcoholic fatty liver disease: another leap forward [J]. *Nat Rev Gastroenterol Hepatol*, 2021, **18**(2): 85-86.

[3] Lazarus JV, Mark HE, Anstee QM, *et al*. Advancing the global public health agenda for NAFLD: a consensus statement [J]. *Nat Rev Gastroenterol Hepatol*, 2022, **19**(1): 60-78.

[4] Bugianesi E, Petta S. Nafld/Nash [J]. *J Hepatol*, 2022, **77**(2): 549-550.

[5] Loomba R, Friedman SL, Shulman GI. Mechanisms and disease consequences of nonalcoholic fatty liver disease [J]. *Cell*, 2021, **184**(10): 2537-2564.

[6] Younossi ZM, Alqahtani SA, Alswat K, *et al*. Global survey of stigma among physicians and patients with nonalcoholic fatty liver disease [J]. *J Hepatol*, 2024, **80**(3): 419-430.

[7] Kokkorakis M, Boutari C, Katsiki N, *et al*. From non-alcoholic fatty liver disease (NAFLD) to steatotic liver disease (SLD): an ongoing journey towards refining the terminology for this prevalent metabolic condition and unmet clinical need [J]. *Metabolism*, 2023, **147**: 155664.

[8] Kim GA, Moon JH, Kim W. Critical appraisal of metabolic dysfunction-associated steatotic liver disease: implication of Janus-faced modernity [J]. *Clin Mol Hepatol*, 2023, **29**(4): 831-843.

[9] Devarbhavi H, Asrani SK, Arab JP, *et al*. Global burden of liver disease: 2023 update [J]. *J Hepatol*, 2023, **79**(2): 516-537.

[10] Allen AM, Lazarus JV, Younossi ZM. Healthcare and socioeconomic costs of NAFLD: a global framework to navigate the uncertainties [J]. *J Hepatol*, 2023, **79**(1): 209-217.

[11] Younossi ZM, Golabi P, Paik JM, *et al*. The global epidemiology of nonalcoholic fatty liver disease (NAFLD) and nonalcoholic steatohepatitis (NASH): a systematic review [J]. *Hepatology*, 2023, **77**(4): 1335-1347.

[12] Liu J, Ayada I, Zhang X, *et al*. Estimating global prevalence of metabolic dysfunction-associated fatty liver disease in overweight or obese adults [J]. *Clin Gastroenterol Hepatol*, 2022, **20**(3): e573-e582.

[13] Quek J, Chan KE, Wong ZY, *et al*. Global prevalence of non-alcoholic fatty liver disease and non-alcoholic steatohepatitis in the overweight and obese population: a systematic review and meta-analysis [J]. *Lancet Gastroenterol Hepatol*, 2023, **8**(1): 20-30.

[14] Simon TG, Roelstraete B, Hagstrom H, *et al*. Non-alcoholic fatty liver disease and incident major adverse cardiovascular events: results from a nationwide histology cohort [J]. *Gut*, 2022, **71**(9): 1867-1875.

[15] Shang Y, Nasr P, Widman L, *et al*. Risk of cardiovascular disease and loss in life expectancy in NAFLD [J]. *Hepatology*, 2022, **76**(5): 1495-1505.

[16] Chung GE, Jeong SM, Cho EJ, *et al*. Association of fatty liver index with all-cause and disease-specific mortality: a nationwide cohort study [J]. *Metabolism*, 2022, **133**: 155222.

[17] Mishra A, Castaneda TR, Bader E, *et al*. Triantennary GalNAc molecular imaging probes for monitoring hepatocyte function in a rat model of nonalcoholic steatohepatitis [J]. *Adv Sci (Weinh)*, 2020, **7**(24): 2002997.
[18] Keam SJ. Resmetirom: first approval [J]. *Drugs*, 2024, **84**(6): 729-735.
[19] Geerts A, Lefere S. Bariatric surgery for non-alcoholic fatty liver disease: indications and post-operative management [J]. *Clin Mol Hepatol*, 2023, **29**(Suppl): S276-S285.
[20] Hwang J, Hwang H, Shin H, *et al*. Bariatric intervention improves metabolic dysfunction-associated steatohepatitis in patients with obesity: a systematic review and meta-analysis [J]. *Clin Mol Hepatol*, 2024, **30**(3): 561-576.
[21] Cigrovski BM, Bilic-Curcic I, Mrzljak A, *et al*. NAFLD and physical exercise: ready, steady, go [J]. *Front Nutr*, 2021, **8**: 734859.
[22] Younossi ZM, Zelber-Sagi S, Henry L, *et al*. Lifestyle interventions in nonalcoholic fatty liver disease [J]. *Nat Rev Gastroenterol Hepatol*, 2023, **20**(11): 708-722.
[23] Lee KC, Wu PS, Lin HC. Pathogenesis and treatment of nonalcoholic steatohepatitis and its fibrosis [J]. *Clin Mol Hepatol*, 2023, **29**(1): 77-98.
[24] Linden D, Romeo S. Therapeutic opportunities for the treatment of NASH with genetically validated targets [J]. *J Hepatol*, 2023, **79**(4): 1056-1064.
[25] Huang G, Wallace DF, Powell EE, *et al*. Gene variants implicated in steatotic liver disease: opportunities for diagnostics and therapeutics [J]. *Biomedicines*, 2023, **11**(10): 2809.
[26] Utzschneider KM, Kahn SE. Review: the role of insulin resistance in nonalcoholic fatty liver disease [J]. *J Clin Endocrinol Metab*, 2006, **91**(12): 4753-4761.
[27] Sakurai Y, Kubota N, Yamauchi T, *et al*. Role of insulin resistance in MAFLD [J]. *Int J Mol Sci*, 2021, **22**(8): 4156.
[28] Zhou J, Cai X, Huang X, *et al*. A novel glucagon-like peptide-1/glucagon receptor dual agonist exhibits weight-lowering and diabetes-protective effects [J]. *Eur J Med Chem*, 2017, **138**: 1158-1169.
[29] Lai ZW, Li C, Liu J, *et al*. Discovery of highly potent DPP-4 inhibitors by hybrid compound design based on linagliptin and alogliptin [J]. *Eur J Med Chem*, 2014, **83**: 547-560.
[30] Zhou J, Ni S, Zhang H, *et al*. Synthesis and bioactivity evaluation of dipeptidyl peptidase IV resistant glucagon-like peptide-1 analogues [J]. *Protein Pept Lett*, 2010, **17**(10): 1290-1295.
[31] Romero-Gomez M, Lawitz E, Shankar RR, *et al*. A Phase 2a active-comparator-controlled study to evaluate the efficacy and safety of efinopegdutide in patients with nonalcoholic fatty liver disease [J]. *J Hepatol*, 2023, **79**(4): 888-897.
[32] Menekse B, Batman A. Effect of exenatide on nonalcoholic steatohepatitis and inflammation-related indices in diabetic patients with non-alcoholic fatty liver disease [J]. *Metab Syndr Relat Disord*, 2023, **21**(4): 205-213.
[33] Mahalingam S, Bellamkonda R, Arumugam MK, *et al*. Glucagon-like peptide 1 receptor agonist, exendin-4, reduces alcohol-associated fatty liver disease [J]. *Biochem Pharmacol*, 2023, **213**: 115613.
[34] Hendawy AS, El-Lakkany NM, Mantawy EM, *et al*. Vildagliptin alleviates liver fibrosis in NASH diabetic rats *via* modulation of insulin resistance, oxidative stress, and inflammatory cascades [J]. *Life Sci*, 2022, **304**: 120695.
[35] Zou J, Zhu X, Xiang D, *et al*. LIX1-like protein promotes liver cancer progression *via* miR-21-3p-mediated inhibition of fructose-1,6-bisphosphatase [J]. *Acta Pharm Sin B*, 2021, **11**(6): 1578-1791.
[36] Du D, Liu C, Qin M, *et al*. Metabolic dysregulation and emerging therapeutical targets for hepatocellular carcinoma [J]. *Acta Pharm Sin B*, 2022, **12**(2): 558-580.
[37] Gnudi L, Thomas SM, Viberti G. Mechanical forces in diabetic kidney disease: a trigger for impaired glucose metabolism [J]. *J Am Soc Nephrol*, 2007, **18**(8): 2226-2232.
[38] Sedzikowska A, Szablewski L. Human glucose transporters in renal glucose homeostasis [J]. *Int J Mol Sci*, 2021, **22**(24): 13522.
[39] Vallon V, Thomson SC. Targeting renal glucose reabsorption to treat hyperglycaemia: the pleiotropic effects of SGLT2 inhibition [J]. *Diabetologia*, 2017, **60**(2): 215-225.
[40] Fiorentino TV, De Vito F, Suraci E, *et al*. Augmented duodenal levels of sodium/glucose co-transporter 1 are associated with higher risk of nonalcoholic fatty liver disease and noninvasive index of liver fibrosis [J]. *Diabetes Res Clin Pract*, 2022, **185**: 109789.
[41] Dwibedi C, Ekstrom O, Brandt J, *et al*. Randomized open-label trial of semaglutide and dapagliflozin in patients with type 2 diabetes of different pathophysiology [J]. *Nat Metab*, 2024, **6**(1): 50-60.
[42] Laffel LM, Danne T, Klingensmith GJ, *et al*. Efficacy and safety of the SGLT2 inhibitor empagliflozin versus placebo and the DPP-4 inhibitor linagliptin versus placebo in young people with type 2 diabetes (DINAMO): a multicentre, randomised, double-blind, parallel group, phase 3 trial [J]. *Lancet Diabetes Endocrinol*, 2023, **11**(3): 169-181.
[43] Fu EL, Wexler DJ, Cromer SJ, *et al*. SGLT-2 inhibitors, GLP-1 receptor agonists, and DPP-4 inhibitors and risk of hyperkalemia among people with type 2 diabetes in clinical practice: population based cohort study [J]. *BMJ*, 2024, **385**: e078483.
[44] Hazem RM, Ibrahim AZ, Ali DA, *et al*. Dapagliflozin improves steatohepatitis in diabetic rats *via* inhibition of oxidative stress and inflammation [J]. *Int Immunopharmacol*, 2022, **104**: 108503.
[45] Bellanti F, Lo Buglio A, Dobrakowski M, *et al*. Impact of sodium glucose cotransporter-2 inhibitors on liver steatosis/fibrosis/inflammation and redox balance in non-alcoholic fatty liver disease [J]. *World J Gastroenterol*, 2022, **28**(26): 3243-3257.
[46] Shimizu M, Suzuki K, Kato K, *et al*. Evaluation of the effects of dapagliflozin, a sodium-glucose co-transporter-2 inhibitor, on hepatic steatosis and fibrosis using transient elastography in patients with type 2 diabetes and non-alcoholic fatty liver disease [J]. *Diabetes Obes Metab*, 2019, **21**(2): 285-292.
[47] He YL, Haynes W, Meyers CD, *et al*. The effects of licogliflozin, a dual SGLT1/2 inhibitor, on body weight in obese patients with or without diabetes [J]. *Diabetes Obes Metab*, 2019, **21**(6): 1311-1321.
[48] Tan S, Ignatenko S, Wagner F, *et al*. Licogliflozin versus placebo in women with polycystic ovary syndrome: a randomized, double-blind, phase 2 trial [J]. *Diabetes Obes Metab*, 2021, **23**(11): 2595-2599.
[49] Harrison SA, Manghi FP, Smith WB, *et al*. Licogliflozin for nonalcoholic steatohepatitis: a randomized, double-blind, placebo-controlled, phase 2a study [J]. *Nat Med*, 2022, **28**(7): 1432-1438.
[50] Esler WP, Cohen DE. Pharmacologic inhibition of lipogenesis for the treatment of NAFLD [J]. *J Hepatol*, 2024, **80**(2):

362-377.

[51] Zhang L, Hu W, Qiu Z, *et al*. Opportunities and challenges for inhibitors targeting citrate transport and metabolism in drug discovery [J]. *J Med Chem*, 2023, **66**(14): 9229-9250.

[52] Morrow MR, Batchuluun B, Wu J, *et al*. Inhibition of ATP-citrate lyase improves NASH, liver fibrosis, and dyslipidemia [J]. *Cell Metab*, 2022, **34**(6): 919-936.e8.

[53] Gao YS, Qian MY, Wei QQ, *et al*. WZ66, a novel acetyl-CoA carboxylase inhibitor, alleviates nonalcoholic steatohepatitis (NASH) in mice [J]. *Acta Pharmacol Sin*, 2020, **41**(3): 336-347.

[54] O'farrell M, Duke G, Crowley R, *et al*. FASN inhibition targets multiple drivers of NASH by reducing steatosis, inflammation and fibrosis in preclinical models [J]. *Sci Rep*, 2022, **12**(1): 15661.

[55] Bhattacharya D, Basta B, Mato JM, *et al*. Aramchol downregulates stearoyl CoA-desaturase 1 in hepatic stellate cells to attenuate cellular fibrogenesis [J]. *JHEP Rep*, 2021, **3**(3): 100237.

[56] Duan J, Wang Z, Duan R, *et al*. Therapeutic targeting of hepatic ACSL4 ameliorates NASH in mice [J]. *Hepatology*, 2022, **75**(1): 140-153.

[57] Cheng D, Zinker BA, Luo Y, *et al*. MGAT2 inhibitor decreases liver fibrosis and inflammation in murine NASH models and reduces body weight in human adults with obesity [J]. *Cell Metab*, 2022, **34**(11): 1732-1748.e5.

[58] Zheng ZG, Zhang X, Liu XX, *et al*. Inhibition of HSP90beta improves lipid disorders by promoting mature SREBPs degradation *via* the ubiquitin-proteasome system [J]. *Theranostics*, 2019, **9**(20): 5769-5783.

[59] Cusi K. Selective agonists of thyroid hormone receptor beta for the treatment of NASH [J]. *N Engl J Med*, 2024, **390**(6): 559-561.

[60] Sinha RA, Yen PM. Metabolic messengers: thyroid hormones [J]. *Nat Metab*, 2024, **6**(4): 639-650.

[61] Marino L, Kim A, Ni B, *et al*. Thyroid hormone action and liver disease, a complex interplay [J]. *Hepatology*, 2023, DOI: 10.1097/HEP.0000000000000551. Online ahead of print.

[62] Harrison SA, Bedossa P, Guy CD, *et al*. A Phase 3, randomized, controlled trial of resmetirom in NASH with liver fibrosis [J]. *N Engl J Med*, 2024, **390**(6): 497-509.

[63] Dutta D, Kamrul-Hasan ABM, Mondal E, *et al*. Role of resmetirom, a liver-directed, thyroid hormone receptor beta-selective agonist, in managing nonalcoholic steatohepatitis: a systematic review and meta-analysis [J]. *Endocr Pract*, 2024, **30**(7): 631-638.

[64] Luong XG, Stevens SK, Jekle A, *et al*. Regulation of gene transcription by thyroid hormone receptor beta agonists in clinical development for the treatment of non-alcoholic steatohepatitis (NASH) [J]. *PLoS One*, 2020, **15**(12): e0240338.

[65] Qiu B, Lawan A, Xirouchaki CE, *et al*. MKP1 promotes non-alcoholic steatohepatitis by suppressing AMPK activity through LKB1 nuclear retention [J]. *Nat Commun*, 2023, **14**(1): 5405.

[66] Jurado-Aguilar J, Barroso E, Bernard M, *et al*. GDF15 activates AMPK and inhibits gluconeogenesis and fibrosis in the liver by attenuating the TGF-beta1/SMAD3 pathway [J]. *Metabolism*, 2024, **152**: 155772.

[67] Steinberg GR, Hardie DG. New insights into activation and function of the AMPK [J]. *Nat Rev Mol Cell Biol*, 2023, **24**(4): 255-272.

[68] Cusi K, Alkhouri N, Harrison SA, *et al*. Efficacy and safety of PXL770, a direct AMP kinase activator, for the treatment of non-alcoholic fatty liver disease (STAMP-NAFLD): a randomised, double-blind, placebo-controlled, phase 2a study [J]. *Lancet Gastroenterol Hepatol*, 2021, **6**(11): 889-902.

[69] Esquejo RM, Salatto CT, Delmore J, *et al*. Activation of liver AMPK with PF-06409577 corrects NAFLD and lowers cholesterol in rodent and primate preclinical models [J]. *EBioMedicine*, 2018, **31**: 122-132.

[70] Li F, Jiang M, Ma M, *et al*. Anthelmintics nitazoxanide protects against experimental hyperlipidemia and hepatic steatosis in hamsters and mice [J]. *Acta Pharm Sin B*, 2022, **12**(3): 1322-1338.

[71] Iroz A, Montagner A, Benhamed F, *et al*. A specific ChREBP and PPARalpha cross-talk is required for the glucose-mediated FGF21 response [J]. *Cell Rep*, 2017, **21**(2): 403-416.

[72] Hinds TD, Kipp ZA, Xu M, *et al*. Adipose-specific PPARalpha knockout mice have increased lipogenesis by PASK-SREBP1 signaling and a polarity shift to inflammatory macrophages in white adipose tissue [J]. *Cells*, 2021, **11**(1): 4.

[73] Fougerat A, Schoiswohl G, Polizzi A, *et al*. ATGL-dependent white adipose tissue lipolysis controls hepatocyte PPARalpha activity [J]. *Cell Rep*, 2022, **39**(10): 110910.

[74] Sun Y, Zhang L, Jiang Z. The role of peroxisome proliferator-activated receptors in the regulation of bile acid metabolism [J]. *Basic Clin Pharmacol Toxicol*, 2024, **134**(3): 315-324.

[75] Guo Y, Miao X, Sun X, *et al*. Zinc finger transcription factor Egf1 promotes non-alcoholic fatty liver disease [J]. *JHEP Rep*, 2023, **5**(6): 100724.

[76] Yan T, Luo Y, Yan N, *et al*. Intestinal peroxisome proliferator-activated receptor alpha-fatty acid-binding protein 1 axis modulates nonalcoholic steatohepatitis [J]. *Hepatology*, 2023, **77**(1): 239-255.

[77] Tian Q, Wang M, Wang X, *et al*. Identification of an alternative ligand-binding pocket in peroxisome proliferator-activated receptor gamma and its correlated selective agonist for promoting beige adipocyte differentiation [J]. *MedComm (2020)*, 2024, **5**(7): e650.

[78] Qiu YY, Zhang J, Zeng FY, *et al*. Roles of the peroxisome proliferator-activated receptors (PPARs) in the pathogenesis of nonalcoholic fatty liver disease (NAFLD) [J]. *Pharmacol Res*, 2023, **192**: 106786.

[79] Staels B, Butruille L, Francque S. Treating NASH by targeting peroxisome proliferator-activated receptors [J]. *J Hepatol*, 2023, **79**(5): 1302-1316.

[80] Bentanachs R, Miro L, Sanchez RM, *et al*. Pemafibrate abrogates SLD in a rat experimental dietary model, inducing a shift in fecal bile acids and microbiota composition [J]. *Biomed Pharmacother*, 2024, **177**: 117067.

[81] Murakami K, Sasaki Y, Asahiyama M, *et al*. Selective PPAR-alpha modulator pemafibrate and sodium-glucose cotransporter 2 inhibitor tofogliflozin combination treatment improved histopathology in experimental mice model of non-alcoholic steatohepatitis [J]. *Cells*, 2022, **11**(4): 720.

[82] Jacques V, Bolze S, Hallakou-Bozec S, *et al*. Deuterium-stabilized (R)-pioglitazone (PXL065) is responsible for pioglitazone efficacy in NASH yet exhibits little to no PPARgamma activity [J]. *Hepatol Commun*, 2021, **5**(8): 1412-1425.

[83] Li Z, Zhou Z, Hu L, *et al*. ZLY032, the first-in-class dual FFA1/PPARδ agonist, improves glucolipid metabolism and alleviates hepatic fibrosis [J]. *Pharmacol Res*, 2020, **159**: 105035.

[84] Bandyopadhyay S, Samajdar SS, Das S. Effects of saroglitaz-

ar in the treatment of non-alcoholic fatty liver disease or non-alcoholic steatohepatitis: a systematic review and meta-analysis [J]. *Clin Res Hepatol Gastroenterol*, 2023, **47**(7): 102174.

[85] Feng Z, Xiang J, Liu H, *et al*. Design, synthesis, and biological evaluation of triazolone derivatives as potent PPARalpha/delta dual agonists for the treatment of nonalcoholic steatohepatitis [J]. *J Med Chem*, 2022, **65**(3): 2571-2592.

[86] Jin L, Yang R, Geng L, *et al*. Fibroblast growth factor-based pharmacotherapies for the treatment of obesity-related metabolic complications [J]. *Annu Rev Pharmacol Toxicol*, 2023, **63**: 359-382.

[87] Das SJ, Pathak B, Dutta S, *et al*. Role of serum endotoxin, FGF19, TLR2, TNF-alpha, IL-12 and IL-10 in NAFLD-associated T2DM pathogenesis: insights into Th1 bias and protective mechanisms [J]. *Indian J Gastroenterol*, 2024, DOI: 10.1007/s12664-024-01597-z. Online ahead of print.

[88] Bao L, Yin J, Gao W, *et al*. A long-acting FGF21 alleviates hepatic steatosis and inflammation in a mouse model of non-alcoholic steatohepatitis partly through an FGF21-adiponectin-IL17A pathway [J]. *Br J Pharmacol*, 2018, **175**(16): 3379-3393.

[89] Wang L, Mazagova M, Pan C, *et al*. YIPF6 controls sorting of FGF21 into COPII vesicles and promotes obesity [J]. *Proc Natl Acad Sci U S A*, 2019, **116**(30): 15184-15193.

[90] Yin J, Bao L, Chen R, *et al*. Enhanced expression and distinctive characterization of a long-acting FGF21 and its potential to alleviate nonalcoholic steatohepatitis [J]. *Biochimie*, 2018, **151**: 166-175.

[91] Charles ED, Neuschwander-Tetri BA, Pablo FJ, *et al*. Pegbelfermin (BMS-986036), PEGylated FGF21, in patients with obesity and type 2 diabetes: results from a randomized phase 2 study [J]. *Obesity (Silver Spring)*, 2019, **27**(1): 41-49.

[92] Raptis DD, Mantzoros CS, Polyzos SA. Fibroblast growth factor-21 as a potential therapeutic target of nonalcoholic fatty liver disease [J]. *Ther Clin Risk Manag*, 2023, **19**: 77-96.

[93] Kinne AS, Tillman EJ, Abdeen SJ, *et al*. Noncompetitive immunoassay optimized for pharmacokinetic assessments of biologically active efruxifermin [J]. *J Pharm Biomed Anal*, 2023, **232**: 115402.

[94] Sanyal AJ, Ling L, Beuers U, *et al*. Potent suppression of hydrophobic bile acids by aldafermin, an FGF19 analogue, across metabolic and cholestatic liver diseases [J]. *JHEP Rep*, 2021, **3**(3): 100255.

[95] Fromenty B, Roden M. Mitochondrial alterations in fatty liver diseases [J]. *J Hepatol*, 2023, **78**(2): 415-429.

[96] Chen FH, Zhang LB, Qiang L, *et al*. Reactive oxygen species-mitochondria pathway involved in LYG-202-induced apoptosis in human hepatocellular carcinoma HepG(2) cells [J]. *Cancer Lett*, 2010, **296**(1): 96-105.

[97] Greatorex S, Kaur S, Xirouchaki CE, *et al*. Mitochondria- and NOX4-dependent antioxidant defense mitigates progression to nonalcoholic steatohepatitis in obesity [J]. *J Clin Invest*, 2023, **134**(3): e162533.

[98] Pan A, Sun XM, Huang FQ, *et al*. The mitochondrial beta-oxidation enzyme HADHA restrains hepatic glucagon response by promoting beta-hydroxybutyrate production [J]. *Nat Commun*, 2022, **13**(1): 386.

[99] Peng S, Wang Y, Zhou Y, *et al*. Rare ginsenosides ameliorate lipid overload-induced myocardial insulin resistance *via* modulating metabolic flexibility [J]. *Phytomedicine*, 2019, **58**: 152745.

[100] Zhao Q, Liu J, Deng H, *et al*. Targeting mitochondria-located circRNA SCAR alleviates NASH *via* reducing mROS output [J]. *Cell*, 2020, **183**(1): 76-93.e22.

[101] Baskol G, Baskol M, Kocer D. Oxidative stress and antioxidant defenses in serum of patients with non-alcoholic steatohepatitis [J]. *Clin Biochem*, 2007, **40**(11): 776-780.

[102] Hwang I, Uddin MJ, Pak ES, *et al*. The impaired redox balance in peroxisomes of catalase knockout mice accelerates nonalcoholic fatty liver disease through endoplasmic reticulum stress [J]. *Free Radic Biol Med*, 2020, **148**: 22-32.

[103] Zhou TJ, Zhang MM, Liu DM, *et al*. Glutathione depletion and dihydroorotate dehydrogenase inhibition actuated ferroptosis-augment to surmount triple-negative breast cancer [J]. *Biomaterials*, 2024, **305**: 122447.

[104] Wang Y, Hu LF, Cui PF, *et al*. Pathologically responsive mitochondrial gene therapy in an allotopic expression-independent manner cures leber's hereditary optic neuropathy [J]. *Adv Mater*, 2021, **33**(41): e2103307.

[105] Liu X, Zhou W, Zhang X, *et al*. Dimethyl fumarate ameliorates dextran sulfate sodium-induced murine experimental colitis by activating Nrf2 and suppressing NLRP3 inflammasome activation [J]. *Biochem Pharmacol*, 2016, **112**: 37-49.

[106] Ding Q, Tang W, Li X, *et al*. Mitochondrial-targeted brequinar liposome boosted mitochondrial-related ferroptosis for promoting checkpoint blockade immunotherapy in bladder cancer [J]. *J Control Release*, 2023, **363**: 221-234.

[107] Miao Z, Wang W, Miao Z, *et al*. Role of selenoprotein W in participating in the progression of non-alcoholic fatty liver disease [J]. *Redox Biol*, 2024, **71**: 103114.

[108] Li Y, Yang J, Chen MH, *et al*. Ilexgenin A inhibits endoplasmic reticulum stress and ameliorates endothelial dysfunction *via* suppression of TXNIP/NLRP3 inflammasome activation in an AMPK dependent manner [J]. *Pharmacol Res*, 2015, **99**: 101-115.

[109] Ji C, Kaplowitz N, Lau MY, *et al*. Liver-specific loss of glucose-regulated protein 78 perturbs the unfolded protein response and exacerbates a spectrum of liver diseases in mice [J]. *Hepatology*, 2011, **54**(1): 229-239.

[110] Sundaram A, Appathurai S, Plumb R, *et al*. Dynamic changes in complexes of IRE1alpha, PERK, and ATF6alpha during endoplasmic reticulum stress [J]. *Mol Biol Cell*, 2018, **29**(11): 1376-1388.

[111] Yang Q, Chen X, Zhang Y, *et al*. The E3 ubiquitin ligase ring finger protein 5 ameliorates NASH through ubiquitin-mediated degradation of 3-hydroxy-3-methylglutaryl CoA reductase degradation protein 1 [J]. *Hepatology*, 2021, **74**(6): 3018-3036.

[112] Beaulant A, Dia M, Pillot B, *et al*. Endoplasmic reticulum-mitochondria miscommunication is an early and causal trigger of hepatic insulin resistance and steatosis [J]. *J Hepatol*, 2022, **77**(3): 710-722.

[113] Zhou J, Pang J, Tripathi M, *et al*. Spermidine-mediated hypusination of translation factor EIF5A improves mitochondrial fatty acid oxidation and prevents non-alcoholic steatohepatitis progression [J]. *Nat Commun*, 2022, **13**(1): 5202.

[114] Jensen SK, Lauridsen C. Alpha-tocopherol stereoisomers [J]. *Vitam Horm*, 2007, **76**: 281-308.

[115] Jeon D, Son M, Shim J. Dynamics of serum retinol and alpha-tocopherol levels according to non-alcoholic fatty liver disease status [J]. *Nutrients*, 2021, **13**(5): 1720.

[116] Phung N, Pera N, Farrell G, *et al*. Pro-oxidant-mediated hepatic fibrosis and effects of antioxidant intervention in murine

dietary steatohepatitis [J]. *Int J Mol Med*, 2009, **24**(2): 171-180.
[117] Panera N, Braghini MR, Crudele A, *et al*. Combination treatment with hydroxytyrosol and vitamin E improves NAFLD-related fibrosis [J]. *Nutrients*, 2022, **14**(18): 3791.
[118] Thielen LA, Chen J, Jing G, *et al*. Identification of an anti-diabetic, orally available small molecule that regulates TXNIP expression and glucagon action [J]. *Cell Metab*, 2020, **32**(3): 353-65.e8.
[119] Qi M, Zhong H, Cheng Z, *et al*. Discovery of NAFLD-improving agents by promoting the degradation of Keap1 [J]. *J Med Chem*, 2023, **66**(13): 9184-9200.
[120] Duan Y, Yang Y, Zhao S, *et al*. Crosstalk in extrahepatic and hepatic system in NAFLD/NASH [J]. *Liver Int*, 2024, **44**(8): 1856-1871.
[121] Khanmohammadi S, Kuchay MS. Toll-like receptors and metabolic (dysfunction)-associated fatty liver disease [J]. *Pharmacol Res*, 2022, **185**: 106507.
[122] Sun BL, Sun X, Kempf CL, *et al*. Involvement of eNAMPT/TLR4 inflammatory signaling in progression of non-alcoholic fatty liver disease, steatohepatitis, and fibrosis [J]. *FASEB J*, 2023, **37**(3): e22825.
[123] Wang Y, Li M, Chen L, *et al*. Natural killer cell-derived exosomal miR-1249-3p attenuates insulin resistance and inflammation in mouse models of type 2 diabetes [J]. *Signal Transduct Target Ther*, 2021, **6**(1): 409.
[124] Yu J, Zhu C, Wang X, *et al*. Hepatocyte TLR4 triggers inter-hepatocyte Jagged1/Notch signaling to determine NASH-induced fibrosis [J]. *Sci Transl Med*, 2021, **13**(599): 1692.
[125] Liu S, Wu J, Chen P, *et al*. TAK-242 ameliorates hepatic fibrosis by regulating the liver-gut axis [J]. *Biomed Res Int*, 2022, **2022**: 4949148.
[126] Hsieh YC, Lee KC, Wu PS, *et al*. Eritoran attenuates hepatic inflammation and fibrosis in mice with chronic liver injury [J]. *Cells*, 2021, **10**(6): 1562.
[127] Sharifnia T, Antoun J, Verriere TG, *et al*. Hepatic TLR4 signaling in obese NAFLD [J]. *Am J Physiol Gastrointest Liver Physiol*, 2015, **309**(4): G270-G278.
[128] Lugnier C. The complexity and multiplicity of the specific cAMP phosphodiesterase family: PDE4, open new adapted therapeutic approaches [J]. *Int J Mol Sci*, 2022, **23**(18): 10616.
[129] Hermann R, Nassr N, Lahu G, *et al*. Steady-state pharmacokinetics of roflumilast and roflumilast *N*-oxide in patients with mild and moderate liver cirrhosis [J]. *Clin Pharmacokinet*, 2007, **46**(5): 403-416.
[130] Etzion O, Bareket-Samish A, Yardeni D, *et al*. Namodenoson at the crossroad of metabolic dysfunction-associated steatohepatitis and hepatocellular carcinoma [J]. *Biomedicines*, 2024, **12**(4): 848.
[131] Lefere S, Devisscher L, Tacke F. Targeting CCR2/5 in the treatment of nonalcoholic steatohepatitis (NASH) and fibrosis: opportunities and challenges [J]. *Expert Opin Investig Drugs*, 2020, **29**(2): 89-92.
[132] Xu F, Guo M, Huang W, *et al*. Annexin A5 regulates hepatic macrophage polarization *via* directly targeting PKM2 and ameliorates NASH [J]. *Redox Biol*, 2020, **36**: 101634.
[133] Li X, Ding Z, Wu Z, *et al*. Targeting the TGF-β signaling pathway for fibrosis therapy: a patent review (2015-2020) [J]. *Expert Opin Ther Pat*, 2021, **31**(8): 723-743.
[134] Gu L, Zhu Y, Lee M, *et al*. Angiotensin II receptor inhibition ameliorates liver fibrosis and enhances hepatocellular carcinoma infiltration by effector T cells [J]. *Proc Natl Acad Sci U S A*, 2023, **120**(19): e2300706120.
[135] Altinbas A, Holmes JA, Salloum S, *et al*. LOXL-2 and TNC-C are markers of liver fibrogenesis in HCV/HIV-, HIV- and HCV-infected patients [J]. *Biomark Med*, 2022, **16**(11): 839-846.
[136] Klepfish M, Gross T, Vugman M, *et al*. LOXL2 inhibition paves the way for macrophage-mediated collagen degradation in liver fibrosis [J]. *Front Immunol*, 2020, **11**: 480.
[137] Barry-Hamilton V, Spangler R, Marshall D, *et al*. Allosteric inhibition of lysyl oxidase-like-2 impedes the development of a pathologic microenvironment [J]. *Nat Med*, 2010, **16**(9): 1009-1017.
[138] Dongiovanni P, Meroni M, Baselli GA, *et al*. Insulin resistance promotes lysyl oxidase like 2 induction and fibrosis accumulation in non-alcoholic fatty liver disease [J]. *Clin Sci (Lond)*, 2017, **131**(12): 1301-1315.
[139] Cosgrove D, Dufek B, Meehan DT, *et al*. Lysyl oxidase like-2 contributes to renal fibrosis in Col4alpha3/Alport mice [J]. *Kidney Int*, 2018, **94**(2): 303-314.
[140] Ikenaga N, Peng ZW, Vaid KA, *et al*. Selective targeting of lysyl oxidase-like 2 (LOXL2) suppresses hepatic fibrosis progression and accelerates its reversal [J]. *Gut*, 2017, **66**(9): 1697-1708.
[141] Yan T, Yan N, Xia Y, *et al*. Hepatocyte-specific CCAAT/enhancer binding protein alpha restricts liver fibrosis progression [J]. *J Clin Invest*, 2024, **134**(7): 166731.
[142] Magkrioti C, Galaris A, Kanellopoulou P, *et al*. Autotaxin and chronic inflammatory diseases [J]. *J Autoimmun*, 2019, **104**: 102327.
[143] Iwadare T, Kimura T, Okumura T, *et al*. Serum autotaxin is a prognostic indicator of liver-related events in patients with non-alcoholic fatty liver disease [J]. *Commun Med (Lond)*, 2024, **4**(1): 73.
[144] Booijink R, Salgado-Polo F, Jamieson C, *et al*. A type IV autotaxin inhibitor ameliorates acute liver injury and nonalcoholic steatohepatitis [J]. *EMBO Mol Med*, 2022, **14**(9): e16333.
[145] Ezhilarasan D. Unraveling the pathophysiologic role of galectin-3 in chronically injured liver [J]. *J Cell Physiol*, 2023, **238**(4): 673-686.
[146] Li H, Cao Z, Wang L, *et al*. Chronic high-fat diet induces galectin-3 and TLR4 to activate NLRP3 inflammasome in NASH [J]. *J Nutr Biochem*, 2023, **112**: 109217.
[147] Greenman R, Segal-Salto M, Barashi N, *et al*. CCL24 regulates biliary inflammation and fibrosis in primary sclerosing cholangitis [J]. *JCI Insight*, 2023, **8**(12): e162270.
[148] Ishikawa Y, Vranka JA, Boudko SP, *et al*. Mutation in cyclophilin B that causes hyperelastosis cutis in American quarter horse does not affect peptidylprolyl *cis-trans* isomerase activity but shows altered cyclophilin B-protein interactions and affects collagen folding [J]. *J Biol Chem*, 2012, **287**(26): 22253-22265.
[149] Yu Q, Jiang Z, Zhang L. Bile acid regulation: a novel therapeutic strategy in non-alcoholic fatty liver disease [J]. *Pharmacol Ther*, 2018, **190**: 81-90.
[150] Soroka CJ, Boyer JL. Biosynthesis and trafficking of the bile salt export pump, BSEP: therapeutic implications of BSEP mutations [J]. *Mol Aspects Med*, 2014, **37**: 3-14.
[151] Thomas C, Pellicciari R, Pruzanski M, *et al*. Targeting bile-acid signalling for metabolic diseases [J]. *Nat Rev Drug Discov*, 2008, **7**(8): 678-693.
[152] Puri P, Daita K, Joyce A, *et al*. The presence and severity of

nonalcoholic steatohepatitis is associated with specific changes in circulating bile acids [J]. *Hepatology*, 2018, **67**(2): 534-548.
[153] Wang H, Ge C, Zhou J, *et al*. Noncanonical farnesoid X receptor signaling inhibits apoptosis and impedes liver fibrosis [J]. *EBioMedicine*, 2018, **37**: 322-333.
[154] Hao H, Cao L, Jiang C, *et al*. Farnesoid X receptor regulation of the NLRP3 inflammasome underlies cholestasis-associated sepsis [J]. *Cell Metab*, 2017, **25**(4): 856-867.e5.
[155] Guan B, Tong J, Hao H, *et al*. Bile acid coordinates microbiota homeostasis and systemic immunometabolism in cardiometabolic diseases [J]. *Acta Pharm Sin B*, 2022, **12**(5): 2129-2149.
[156] Zhou J, Cui S, He Q, *et al*. SUMOylation inhibitors synergize with FXR agonists in combating liver fibrosis [J]. *Nat Commun*, 2020, **11**(1): 240.
[157] Cui S, Hu H, Chen A, *et al*. SIRT1 activation synergizes with FXR agonism in hepatoprotection *via* governing nucleocytoplasmic shuttling and degradation of FXR [J]. *Acta Pharm Sin B*, 2023, **13**(2): 559-576.
[158] Zhuge A, Li S, Yuan Y, *et al*. Microbiota-induced lipid peroxidation impairs obeticholic acid-mediated antifibrotic effect towards nonalcoholic steatohepatitis in mice [J]. *Redox Biol*, 2023, **59**: 102582.
[159] Wang K, Zhang Y, Wang G, *et al*. FXR agonists for MASH therapy: lessons and perspectives from obeticholic acid [J]. *Med Res Rev*, 2024, **44**(2): 568-586.
[160] Hu YB, Liu XY, Zhan W. Farnesoid X receptor agonist INT-767 attenuates liver steatosis and inflammation in rat model of nonalcoholic steatohepatitis [J]. *Drug Des Devel Ther*, 2018, **12**: 2213-2221.
[161] Anfuso B, Tiribelli C, Adorini L, *et al*. Obeticholic acid and INT-767 modulate collagen deposition in a NASH *in vitro* model [J]. *Sci Rep*, 2020, **10**(1): 1699.
[162] Lai J, Luo L, Zhou T, *et al*. Alterations in circulating bile acids in metabolic dysfunction-associated steatotic liver disease: a systematic review and meta-analysis [J]. *Biomolecules*, 2023, **13**(9): 1356.
[163] Salic K, Kleemann R, Wilkins-Port C, *et al*. Apical sodium-dependent bile acid transporter inhibition with volixibat improves metabolic aspects and components of non-alcoholic steatohepatitis in Ldlr$^{-/-}$ leiden mice [J]. *PLoS One*, 2019, **14**(6): e0218459.
[164] Matye DJ, Wang H, Luo W, *et al*. Combined ASBT inhibitor and FGF15 treatment improves therapeutic efficacy in experimental nonalcoholic steatohepatitis [J]. *Cell Mol Gastroenterol Hepatol*, 2021, **12**(3): 1001-1019.
[165] Liu XJ, Liu C, Zhu LY, *et al*. Hepalatide ameliorated progression of nonalcoholic steatohepatitis in mice [J]. *Biomed Pharmacother*, 2020, **126**: 110053.
[166] Demir M, Lang S, Hartmann P, *et al*. The fecal mycobiome in non-alcoholic fatty liver disease [J]. *J Hepatol*, 2022, **76**(4): 788-799.
[167] Forlano R, Mullish BH, Roberts LA, *et al*. The intestinal barrier and its dysfunction in patients with metabolic diseases and non-alcoholic fatty liver disease [J]. *Int J Mol Sci*, 2022, **23**(2): 662.
[168] Qian M, Hu H, Yao Y, *et al*. Coordinated changes of gut microbiome and lipidome differentiates nonalcoholic steatohepatitis (NASH) from isolated steatosis [J]. *Liver Int*, 2020, **40**(3): 622-637.
[169] Depommier C, Everard A, Druart C, *et al*. Supplementation with *Akkermansia muciniphila* in overweight and obese human volunteers: a proof-of-concept exploratory study [J]. *Nat Med*, 2019, **25**(7): 1096-1103.
[170] Han Y, Ling Q, Wu L, *et al*. *Akkermansia muciniphila* inhibits nonalcoholic steatohepatitis by orchestrating TLR2-activated gammadeltaT17 cell and macrophage polarization [J]. *Gut Microbes*, 2023, **15**(1): 2221485.
[171] Li SZ, Zhang NN, Yang X, *et al*. Nobiletin ameliorates nonalcoholic fatty liver disease by regulating gut microbiota and myristoleic acid metabolism [J]. *J Agric Food Chem*, 2023, **71**(19): 7312-7323.
[172] Yu JS, Youn GS, Choi J, *et al*. *Lactobacillus lactis* and *Pediococcus pentosaceus*-driven reprogramming of gut microbiome and metabolome ameliorates the progression of non-alcoholic fatty liver disease [J]. *Clin Transl Med*, 2021, **11**(12): e634.
[173] Yang T, Wang Y, Cao X, *et al*. Targeting mTOR/YY1 signaling pathway by quercetin through CYP7A1-mediated cholesterol-to-bile acids conversion alleviated type 2 diabetes mellitus induced hepatic lipid accumulation [J]. *Phytomedicine*, 2023, **113**: 154703.
[174] Sun Q, Niu Q, Guo Y, *et al*. Regulation on citrate influx and metabolism through inhibiting SLC13A5 and ACLY: a novel mechanism mediating the therapeutic effects of curcumin on NAFLD [J]. *J Agric Food Chem*, 2021, **69**(31): 8714-8725.
[175] Wu J, Lou YG, Yang XL, *et al*. Silybin regulates P450s activity by attenuating endoplasmic reticulum stress in mouse nonalcoholic fatty liver disease [J]. *Acta Pharmacol Sin*, 2023, **44**(1): 133-144.
[176] Shojaie L, Iorga A, Dara L. Cell death in liver diseases: a review [J]. *Int J Mol Sci*, 2020, **21**(24): 9682.
[177] Lei Z, Yu J, Wu Y, *et al*. CD1d protects against hepatocyte apoptosis in non-alcoholic steatohepatitis [J]. *J Hepatol*, 2024, **80**(2): 194-208.
[178] Kim JY, Garcia-Carbonell R, Yamachika S, *et al*. ER stress drives lipogenesis and steatohepatitis *via* caspase-2 activation of S1P [J]. *Cell*, 2018, **175**(1): 133-145.e15.
[179] Thapaliya S, Wree A, Povero D, *et al*. Caspase 3 inactivation protects against hepatic cell death and ameliorates fibrogenesis in a diet-induced NASH model [J]. *Dig Dis Sci*, 2014, **59**(6): 1197-1206.
[180] Zhou J, Huang N, Guo Y, *et al*. Combined obeticholic acid and apoptosis inhibitor treatment alleviates liver fibrosis [J]. *Acta Pharm Sin B*, 2019, **9**(3): 526-536.
[181] Feng G, Byrne CD, Targher G, *et al*. Ferroptosis and metabolic dysfunction-associated fatty liver disease: is there a link [J]. *Liver Int*, 2022, **42**(7): 1496-1502.
[182] Tsurusaki S, Tsuchiya Y, Koumura T, *et al*. Hepatic ferroptosis plays an important role as the trigger for initiating inflammation in nonalcoholic steatohepatitis [J]. *Cell Death Dis*, 2019, **10**(6): 449.
[183] Tong J, Lan XT, Zhang Z, *et al*. Ferroptosis inhibitor liproxstatin-1 alleviates metabolic dysfunction-associated fatty liver disease in mice: potential involvement of PANoptosis [J]. *Acta Pharmacol Sin*, 2023, **44**(5): 1014-1028.
[184] Ruart M, Chavarria L, Camprecios G, *et al*. Impaired endothelial autophagy promotes liver fibrosis by aggravating the oxidative stress response during acute liver injury [J]. *J Hepatol*, 2019, **70**(3): 458-469.
[185] Yamada T, Murata D, Adachi Y, *et al*. Mitochondrial stasis reveals p62-mediated ubiquitination in parkin-independent mitophagy and mitigates nonalcoholic fatty liver disease [J].

Cell Metab, 2018, **28**(4): 588-604.e5.
[186] Hammoutene A, Biquard L, Lasselin J, *et al*. A defect in endothelial autophagy occurs in patients with non-alcoholic steatohepatitis and promotes inflammation and fibrosis [J]. *J Hepatol*, 2020, **72**(3): 528-538.
[187] Qian H, Chao X, Williams J, *et al*. Autophagy in liver diseases: a review [J]. *Mol Aspects Med*, 2021, **82**: 100973.
[188] Da Silva LN, Fondevila MF, Novoa E, *et al*. Inhibition of ATG3 ameliorates liver steatosis by increasing mitochondrial function [J]. *J Hepatol*, 2022, **76**(1): 11-24.
[189] Gaul S, Leszczynska A, Alegre F, *et al*. Hepatocyte pyroptosis and release of inflammasome particles induce stellate cell activation and liver fibrosis [J]. *J Hepatol*, 2021, **74**(1): 156-167.
[190] Xu B, Jiang M, Chu Y, *et al*. Gasdermin D plays a key role as a pyroptosis executor of non-alcoholic steatohepatitis in humans and mice [J]. *J Hepatol*, 2018, **68**(4): 773-782.
[191] Liu Y, Wang DW, Wang D, *et al*. Exenatide attenuates nonalcoholic steatohepatitis by inhibiting the pyroptosis signaling pathway [J]. *Front Endocrinol (Lausanne)*, 2021, **12**: 663039.
[192] Khotskaya YB, Holla VR, Farago AF, *et al*. Targeting TRK family proteins in cancer [J]. *Pharmacol Ther*, 2017, **173**: 58-66.
[193] Amatu A, Sartore-Bianchi A, Bencardino K, *et al*. Tropomyosin receptor kinase (TRK) biology and the role of NTRK gene fusions in cancer [J]. *Ann Oncol*, 2019, **30**(Suppl_8): viii5-viii15.
[194] Ahmed F, Hristova K. Dimerization of the Trk receptors in the plasma membrane: effects of their cognate ligands [J]. *Biochem J*, 2018, **475**(22): 3669-3685.
[195] Xiong J, Liu T, Mi L, *et al*. hnRNPU/TrkB defines a chromatin accessibility checkpoint for liver injury and nonalcoholic steatohepatitis pathogenesis [J]. *Hepatology*, 2020, **71**(4): 1228-1246.
[196] Wang S, Li K, Pickholz E, *et al*. An autocrine signaling circuit in hepatic stellate cells underlies advanced fibrosis in nonalcoholic steatohepatitis [J]. *Sci Transl Med*, 2023, **15**(677): eadd3949.
[197] Yu P, Han Y, Meng L, *et al*. The incorporation of acetylated LAP-TGF-beta1 proteins into exosomes promotes TNBC cell dissemination in lung micro-metastasis [J]. *Mol Cancer*, 2024, **23**(1): 82.
[198] Povero D, Panera N, Eguchi A, *et al*. Lipid-induced hepatocyte-derived extracellular vesicles regulate hepatic stellate cell *via* microRNAs targeting PPAR-gamma [J]. *Cell Mol Gastroenterol Hepatol*, 2015, **1**(6): 646-663.e4.
[199] Liu X, Tan S, Liu H, *et al*. Hepatocyte-derived MASP1-enriched small extracellular vesicles activate HSCs to promote liver fibrosis [J]. *Hepatology*, 2023, **77**(4): 1181-1197.
[200] Gao H, Jin Z, Bandyopadhyay G, *et al*. Aberrant iron distribution *via* hepatocyte-stellate cell axis drives liver lipogenesis and fibrosis [J]. *Cell Metab*, 2022, **34**(8): 1201-1213.e5.
[201] Zhao Y, Zhao MF, Jiang S, *et al*. Liver governs adipose remodelling *via* extracellular vesicles in response to lipid overload [J]. *Nat Commun*, 2020, **11**(1): 719.
[202] Zhang W, Lu J, Feng L, *et al*. Sonic hedgehog-heat shock protein 90beta axis promotes the development of nonalcoholic steatohepatitis in mice [J]. *Nat Commun*, 2024, **15**(1): 1280.
[203] Song J, Song B, Yuan L, *et al*. Multiplexed strategies toward clinical translation of extracellular vesicles [J]. *Theranostics*, 2022, **12**(15): 6740-6761.
[204] Gao J, Wei B, De Assuncao TM, *et al*. Hepatic stellate cell autophagy inhibits extracellular vesicle release to attenuate liver fibrosis [J]. *J Hepatol*, 2020, **73**(5): 1144-1154.
[205] Tong L, Zhang S, Liu Q, *et al*. Milk-derived extracellular vesicles protect intestinal barrier integrity in the gut-liver axis [J]. *Sci Adv*, 2023, **9**(15): eade5041.
[206] Wei X, Yang X, Han ZP, *et al*. Mesenchymal stem cells: a new trend for cell therapy [J]. *Acta Pharmacol Sin*, 2013, **34**(6): 747-754.
[207] Sun Y, Shi H, Yin S, *et al*. Human mesenchymal stem cell derived exosomes alleviate type 2 diabetes mellitus by reversing peripheral insulin resistance and relieving beta-cell destruction [J]. *ACS Nano*, 2018, **12**(8): 7613-7628.
[208] Wang H, Zheng R, Chen Q, *et al*. Mesenchymal stem cells microvesicles stabilize endothelial barrier function partly mediated by hepatocyte growth factor (HGF) [J]. *Stem Cell Res Ther*, 2017, **8**(1): 211.
[209] Du J, Jiang Y, Liu X, *et al*. HGF secreted by menstrual blood-derived endometrial stem cells ameliorates non-alcoholic fatty liver disease through downregulation of hepatic Rnf186 [J]. *Stem Cells*, 2023, **41**(2): 153-168.
[210] Kang Y, Song Y, Luo Y, *et al*. Exosomes derived from human umbilical cord mesenchymal stem cells ameliorate experimental non-alcoholic steatohepatitis *via* Nrf2/NQO-1 pathway [J]. *Free Radic Biol Med*, 2022, **192**: 25-36.
[211] Shi Y, Yang X, Wang S, *et al*. Human umbilical cord mesenchymal stromal cell-derived exosomes protect against MCD-induced NASH in a mouse model [J]. *Stem Cell Res Ther*, 2022, **13**(1): 517.
[212] Yang F, Wu Y, Chen Y, *et al*. Human umbilical cord mesenchymal stem cell-derived exosomes ameliorate liver steatosis by promoting fatty acid oxidation and reducing fatty acid synthesis [J]. *JHEP Rep*, 2023, **5**(7): 100746.
[213] Nickel S, Christ M, Schmidt S, *et al*. Human mesenchymal stromal cells resolve lipid load in high fat diet-induced non-alcoholic steatohepatitis in mice by mitochondria donation [J]. *Cells*, 2022, **11**(11): 1829.
[214] Doycheva I, Erickson D, Watt KD. Growth hormone deficiency and NAFLD: an overlooked and underrecognized link [J]. *Hepatol Commun*, 2022, **6**(9): 2227-2237.
[215] Burra P, Bizzaro D, Gonta A, *et al*. Clinical impact of sexual dimorphism in non-alcoholic fatty liver disease (NAFLD) and non-alcoholic steatohepatitis (NASH) [J]. *Liver Int*, 2021, **41**(8): 1713-1733.
[216] Weng C, Shao Z, Xiao M, *et al*. Association of sex hormones with non-alcoholic fatty liver disease: an observational and mendelian randomization study [J]. *Liver Int*, 2024, **44**(5): 1154-1166.
[217] Galmes-Pascual BM, Martinez-Cignoni MR, Moran-Costoya A, *et al*. 17beta-Estradiol ameliorates lipotoxicity-induced hepatic mitochondrial oxidative stress and insulin resistance [J]. *Free Radic Biol Med*, 2020, **150**: 148-160.
[218] Araujo LC, Cruz AG, Camargo FN, *et al*. Estradiol protects female ApoE KO mice against western-diet-induced non-alcoholic steatohepatitis [J]. *Int J Mol Sci*, 2023, **24**(12): 9845.
[219] Li M, Cai Y, Chen X, *et al*. Tamoxifen induced hepatic steatosis in high-fat feeding rats through SIRT1-Foxo1 suppression and LXR-SREBP1c activation [J]. *Toxicol Res*, 2022, **11**(4): 673-682.
[220] Al-Qudimat A, Al-Zoubi RM, Yassin AA, *et al*. Testosterone treatment improves liver function and reduces cardiovascular risk: a long-term prospective study [J]. *Arab J Urol*, 2021, **19**(3): 376-386.

[221] Gagliano-Juca T, Basaria S. Testosterone replacement therapy and cardiovascular risk [J]. *Nat Rev Cardiol*, 2019, **16**(9): 555-574.
[222] Li L, Wang H, Yao Y, *et al*. The sex steroid precursor dehydroepiandrosterone prevents nonalcoholic steatohepatitis by activating the AMPK pathway mediated by GPR30 [J]. *Redox Biol*, 2021, **48**: 102187.
[223] Che Q, Xiao X, Xu J, *et al*. 17beta-Estradiol promotes endometrial cancer proliferation and invasion through IL-6 pathway [J]. *Endocr Connect*, 2019, **8**(7): 961-968.
[224] Shi KQ, Fan YC, Liu WY, *et al*. Traditional Chinese medicines benefit to nonalcoholic fatty liver disease: a systematic review and meta-analysis [J]. *Mol Biol Rep*, 2012, **39**(10): 9715-9722.
[225] Liu C, Hu MY, Zhang M, *et al*. Association of GLP-1 secretion with anti-hyperlipidemic effect of ginsenosides in high-fat diet fed rats [J]. *Metabolism*, 2014, **63**(10): 1342-1351.
[226] Xiao N, Yang LL, Yang YL, *et al*. Ginsenoside Rg5 inhibits succinate-associated lipolysis in adipose tissue and prevents muscle insulin resistance [J]. *Front Pharmacol*, 2017, **8**: 43.
[227] Yue C, Li D, Fan S, *et al*. Long-term and liver-selected ginsenoside C−K nanoparticles retard NAFLD progression by restoring lipid homeostasis [J]. *Biomaterials*, 2023, **301**: 122291.
[228] Ma P, Ou H, Sun Z, *et al*. IAVPGEVA: orally available DPP4-targeting soy glycinin derived octapeptide with therapeutic potential in nonalcoholic steatohepatitis [J]. *J Agric Food Chem*, 2024, **72**(13): 7167-7178.
[229] Leng J, Chen MH, Zhou ZH, *et al*. Triterpenoids-enriched extract from the aerial parts of *Salvia miltiorrhiza* regulates macrophage polarization and ameliorates insulin resistance in high-fat fed mice [J]. *Phytother Res*, 2017, **31**(1): 100-107.
[230] Wang W, Xu AL, Li ZC, *et al*. Combination of probiotics and *Salvia miltiorrhiza* polysaccharide alleviates hepatic steatosis *via* gut microbiota modulation and insulin resistance improvement in high fat-induced NAFLD mice [J]. *Diabetes Metab J*, 2020, **44**(2): 336-348.
[231] Lin Z, Wu ZF, Jiang CH, *et al*. The chloroform extract of *Cyclocarya paliurus* attenuates high-fat diet induced non-alcoholic hepatic steatosis in sprague dawley rats [J]. *Phytomedicine*, 2016, **23**(12): 1475-1483.
[232] Zhao MG, Sheng XP, Huang YP, *et al*. Triterpenic acids-enriched fraction from *Cyclocarya paliurus* attenuates non-alcoholic fatty liver disease *via* improving oxidative stress and mitochondrial dysfunction [J]. *Biomed Pharmacother*, 2018, **104**: 229-239.
[233] Zheng X, Zhao MG, Jiang CH, *et al*. Triterpenic acids-enriched fraction from *Cyclocarya paliurus* attenuates insulin resistance and hepatic steatosis *via* PI3K/Akt/GSK3beta pathway [J]. *Phytomedicine*, 2020, **66**: 153130.
[234] Zhang L, Li HX, Pan WS, *et al*. Administration of methyl palmitate prevents non-alcoholic steatohepatitis (NASH) by induction of PPAR-alpha [J]. *Biomed Pharmacother*, 2019, **111**: 99-108.
[235] Xu J, Li Y, Lou M, *et al*. Baicalin regulates SirT1/STAT3 pathway and restrains excessive hepatic glucose production [J]. *Pharmacol Res*, 2018, **136**: 62-73.
[236] Yang LL, Xiao N, Liu J, *et al*. Differential regulation of baicalin and scutellarin on AMPK and Akt in promoting adipose cell glucose disposal [J]. *Biochim Biophys Acta Mol Basis Dis*, 2017, **1863**(2): 598-606.
[237] Gao Y, Liu J, Hao Z, *et al*. Baicalin ameliorates high fat diet-induced nonalcoholic fatty liver disease in mice *via* adenosine monophosphate-activated protein kinase-mediated regulation of SREBP1/Nrf2/NF-kappaB signaling pathways [J]. *Phytother Res*, 2023, **37**(6): 2405-2418.
[238] Xu DQ, Li CJ, Jiang ZZ, *et al*. The hypoglycemic mechanism of catalpol involves increased AMPK-mediated mitochondrial biogenesis [J]. *Acta Pharmacol Sin*, 2020, **41**(6): 791-799.
[239] Cao M, Wang J, Jiang X, *et al*. Phenolic constituents from black quinoa alleviate insulin resistance in HepG2 cells *via* regulating IRS1/PI3K/Akt/GLUTs signaling pathways [J]. *J Agric Food Chem*, 2023, **71**(48): 18780-18791.
[240] Ding L, Li J, Song B, *et al*. Andrographolide prevents high-fat diet-induced obesity in C57BL/6 mice by suppressing the sterol regulatory element-binding protein pathway [J]. *J Pharmacol Exp Ther*, 2014, **351**(2): 474-483.
[241] Li A, Liu Q, Li Q, *et al*. Berberine reduces pyruvate-driven hepatic glucose production by limiting mitochondrial import of pyruvate through mitochondrial pyruvate carrier 1 [J]. *EBioMedicine*, 2018, **34**: 243-255.
[242] Zhang Y, Wen J, Liu D, *et al*. Demethylenetetrahydroberberine alleviates nonalcoholic fatty liver disease by inhibiting the NLRP3 inflammasome and oxidative stress in mice [J]. *Life Sci*, 2021, **281**: 119778.
[243] Yan C, Zhang Y, Zhang X, *et al*. Curcumin regulates endogenous and exogenous metabolism *via* Nrf2-FXR-LXR pathway in NAFLD mice [J]. *Biomed Pharmacother*, 2018, **105**: 274-281.
[244] Zhao X, Xue J, Xie M. Osthole inhibits oleic acid/lipopolysaccharide-induced lipid accumulation and inflammatory response through activating PPARalpha signaling pathway in cultured hepatocytes [J]. *Exp Gerontol*, 2019, **119**: 7-13.
[245] Jiang X, Lu Z, Zhang Q, *et al*. Osthole: a potential AMPK agonist that inhibits NLRP3 inflammasome activation by regulating mitochondrial homeostasis for combating rheumatoid arthritis [J]. *Phytomedicine*, 2023, **110**: 154640.
[246] Wu X, Zhang L, Gurley E, *et al*. Prevention of free fatty acid-induced hepatic lipotoxicity by 18beta-glycyrrhetinic acid through lysosomal and mitochondrial pathways [J]. *Hepatology*, 2008, **47**(6): 1905-1915.
[247] Zhu X, Ye S, Yu D, *et al*. Physalin B attenuates liver fibrosis *via* suppressing LAP2alpha-HDAC1-mediated deacetylation of the transcription factor GLI1 and hepatic stellate cell activation [J]. *Br J Pharmacol*, 2021, **178**(17): 3428-3447.
[248] Xue L, Liu K, Yan C, *et al*. Schisandra lignans ameliorate nonalcoholic steatohepatitis by regulating aberrant metabolism of phosphatidylethanolamines [J]. *Acta Pharm Sin B*, 2023, **13**(8): 3545-3560.
[249] Miguel FM, Picada JN, Da Silva JB, *et al*. Melatonin attenuates inflammation, oxidative stress, and DNA damage in mice with nonalcoholic steatohepatitis induced by a methionine- and choline-deficient diet [J]. *Inflammation*, 2022, **45**(5): 1968-1984.
[250] Zhen Q, Liang Q, Wang H, *et al*. Theabrownin ameliorates liver inflammation, oxidative stress, and fibrosis in MCD diet-fed C57BL/6J mice [J]. *Front Endocrinol (Lausanne)*, 2023, **14**: 1118925.
[251] Bai Y, Zhang J, Li J, *et al*. Silibinin, a commonly used therapeutic agent for non-alcohol fatty liver disease, functions through upregulating intestinal expression of fibroblast growth factor 15/19 [J]. *Br J Pharmacol*, 2024, DOI: 10.1111/bph.16431. Online ahead of print.
[252] Jiang W, Xu S, Guo H, *et al*. Magnesium isoglycyrrhizinate

prevents the nonalcoholic hepatic steatosis *via* regulating energy homeostasis [J]. *J Cell Mol Med*, 2020, **24**(13): 7201-7213.
[253] Wang YZ, Wang YL, Che HJ, *et al*. Sappanone A: a natural PDE4 inhibitor with dual anti-inflammatory and antioxidant activities from the heartwood of *Caesalpinia sappan* L. [J]. *J Ethnopharmacol*, 2023, **304**: 116020.
[254] Xiang D, Zou J, Zhu X, *et al*. Physalin D attenuates hepatic stellate cell activation and liver fibrosis by blocking TGF-beta/Smad and YAP signaling [J]. *Phytomedicine*, 2020, **78**: 153294.
[255] Zhang Y, Ye S, Lu W, *et al*. RNA helicase DEAD-box protein 5 alleviates nonalcoholic steatohepatitis progression *via* tethering TSC complex and suppressing mTORC1 signaling [J]. *Hepatology*, 2023, **77**(5): 1670-1687.
[256] Yang T, Wu E, Zhu X, *et al*. TKF, a mexicanolide-type limonoid derivative, suppressed hepatic stellate cells activation and liver fibrosis through inhibition of the YAP/Notch3 pathway [J]. *Phytomedicine*, 2022, **107**: 154466.
[257] Chu X, Wang H, Jiang YM, *et al*. Ameliorative effects of tannic acid on carbon tetrachloride-induced liver fibrosis *in vivo* and *in vitro* [J]. *J Pharmacol Sci*, 2016, **130**(1): 15-23.
[258] Zhang B, Luo X, Han C, *et al*. *Terminalia bellirica* ethanol extract ameliorates nonalcoholic fatty liver disease in mice by amending the intestinal microbiota and faecal metabolites [J]. *J Ethnopharmacol*, 2023, **305**: 116082.
[259] Crossan C, Majumdar A, Srivastava A, *et al*. Referral pathways for patients with NAFLD based on non-invasive fibrosis tests: diagnostic accuracy and cost analysis [J]. *Liver Int*, 2019, **39**(11): 2052-2060.
[260] Gerges SH, Wahdan SA, Elsherbiny DA, *et al*. Non-alcoholic fatty liver disease: an overview of risk factors, pathophysiological mechanisms, diagnostic procedures, and therapeutic interventions [J]. *Life Sci*, 2021, **271**: 119220.
[261] Castera L, Friedrich-Rust M, Loomba R. Noninvasive assessment of liver disease in patients with nonalcoholic fatty liver disease [J]. *Gastroenterology*, 2019, **156**(5): 1264-1281.e4.
[262] Zhou JH, Cai JJ, She ZG, *et al*. Noninvasive evaluation of nonalcoholic fatty liver disease: current evidence and practice [J]. *World J Gastroenterol*, 2019, **25**(11): 1307-1326.
[263] Long MT, Noureddin M, Lim JK. AGA clinical practice update: diagnosis and management of nonalcoholic fatty liver disease in lean individuals: expert review [J]. *Gastroenterology*, 2022, **163**(3): 764-774.e1.
[264] Nogami A, Yoneda M, Iwaki M, *et al*. Diagnostic comparison of vibration-controlled transient elastography and MRI techniques in overweight and obese patients with NAFLD [J]. *Sci Rep*, 2022, **12**(1): 21925.
[265] Ali AH, Al Juboori A, Petroski GF, *et al*. The utility and diagnostic accuracy of transient elastography in adults with morbid obesity: a prospective study [J]. *J Clin Med*, 2022, **11**(5): 1201.
[266] Starekova J, Hernando D, Pickhardt PJ, *et al*. Quantification of liver fat content with CT and MRI: state of the art [J]. *Radiology*, 2021, **301**(2): 250-262.
[267] Powell EE, Wong VW, Rinella M. Non-alcoholic fatty liver disease [J]. *Lancet*, 2021, **397**(10290): 2212-2224.
[268] Perakakis N, Stefanakis K, Mantzoros CS. The role of omics in the pathophysiology, diagnosis and treatment of non-alcoholic fatty liver disease [J]. *Metabolism*, 2020, **111S**: 154320.
[269] Younossi ZM. Non-alcoholic fatty liver disease: a global public health perspective [J]. *J Hepatol*, 2019, **70**(3): 531-544.
[270] Hardy T, Oakley F, Anstee QM, *et al*. Nonalcoholic fatty liver disease: pathogenesis and disease spectrum [J]. *Annu Rev Pathol*, 2016, **11**: 451-496.
[271] Kumar V, Xin X, Ma J, *et al*. Therapeutic targets, novel drugs, and delivery systems for diabetes associated NAFLD and liver fibrosis [J]. *Adv Drug Deliv Rev*, 2021, **176**: 113888.
[272] Harrison SA, Allen AM, Dubourg J, *et al*. Challenges and opportunities in NASH drug development [J]. *Nat Med*, 2023, **29**(3): 562-573.
[273] Takyar V, Nath A, Beri A, *et al*. How healthy are the "Healthy volunteers"? Penetrance of NAFLD in the biomedical research volunteer pool [J]. *Hepatology*, 2017, **66**(3): 825-833.
[274] Brunt EM, Kleiner DE, Carpenter DH, *et al*. NAFLD: reporting histologic findings in clinical practice [J]. *Hepatology*, 2021, **73**(5): 2028-2038.
[275] Hendriks D, Brouwers JF, Hamer K, *et al*. Engineered human hepatocyte organoids enable CRISPR-based target discovery and drug screening for steatosis [J]. *Nat Biotechnol*, 2023, **41**(11): 1567-1581.
[276] Alkhouri N, Herring R, Kabler H, *et al*. Safety and efficacy of combination therapy with semaglutide, cilofexor and firsocostat in patients with non-alcoholic steatohepatitis: a randomised, open-label phase II trial [J]. *J Hepatol*, 2022, **77**(3): 607-618.

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