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Bile Acid Nuclear Receptor Farnesoid X Receptor: Therapeutic Target for Nonalcoholic Fatty Liver Disease

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Nonalcoholic fatty liver disease (NAFLD) is one of the causes of fatty liver, occurring when fat is accumulated in the liver without alcohol consumption. NAFLD is the most common liver disorder in advanced countries. NAFLD is a spectrum of pathology involving hepatic steatosis with/without inflammation and nonalcoholic steatohepatitis with accumulation of hepatocyte damage and hepatic fibrosis. Recent studies have revealed that NAFLD results in the progression of cryptogenic cirrhosis that leads to hepatocarcinoma and cardiovascular diseases such as heart failure. The main causes of NAFLD have not been revealed yet, metabolic syndromes including obesity and insulin resistance are widely accepted for the critical risk factors for the pathogenesis of NAFLD. Nuclear receptors (NRs) are transcriptional factors that sense environmental or hormonal signals and regulate expression of genes, involved in cellular growth, development, and metabolism. Several NRs have been reported to regulate genes involved in energy and xenobiotic metabolism and inflammation. Among various NRs, farnesoid X receptor (FXR) is abundantly expressed in the liver and a key regulator to control various metabolic processes in the liver. Recent studies have shown that NAFLD is associated with inappropriate function of FXR. The impact of FXR transcriptional activity in NAFLD is likely to be potential therapeutic strategy, but still requires to elucidate underlying potent therapeutic mechanisms of FXR for the treatment of NAFLD. This article will focus the physiological roles of FXR and establish the correlation between FXR transcriptional activity and the pathogenesis of NAFLD.

Keywords: Bile acids and salts; Receptors, cytoplasmic and nuclear; Farnesoid X receptor; Non-alcoholic fatty liver disease; Obeticholic acid

INTRODUCTION

Nonalcoholic fatty liver disease (NAFLD) is characterized by the accumulation of lipid droplets in the hepatocytes without alcohol consumption. NAFLD is a common liver disorder and affects 15% to 40% of the population in United States [1]. According to National Health and Nutrition Examination Survey, the rate of NAFLD in chronic liver disorder has been substantially increasing 47% in 1988 to 1994, 63% in 1999 to 2004, and 75% in 2005 to 2008 [2]. Quite interestingly, metabolic disorders including obesity, type 2 diabetes, chronic kidney disease, hypertension, and colorectal malignant neoplasm also have been largely increasing in the patients with NAFLD, implying of positive correlation between NAFLD and metabolic disorders [3-6]. The spectrum of NAFLD is very wide, covering hepatic steatosis and nonalcoholic steatohepatitis (NASH)

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which resulting in eventually hepatic cirrhosis and hepatocellular carcinoma. Among the NAFLD patients, at least 10% to 20% would develop to NASH, which is a serious condition of liver disorders [7]. To date, no optimal treatment has been established for NAFLD. Thus, it is required to understand the pathogenesis of NAFLD and to determine potential therapeutic target for effective pharmacological treatment for NAFLD.

Nuclear receptors (NRs) are ligand-activated transcriptional factors that broadly regulate genes involved in metabolism, xenobiotics and cellular growth and cycle. NRs are sensitive to many natural and synthetic ligands including steroid hormones, lipids and fatty acids, vitamins, drugs, and various metabolites. Recently, numerous reports have demonstrated that farnesoid X receptor (FXR) is a bile acid NR and regulates hepatic gluconeogenesis and lipogenesis and hepatic inflammation to maintain metabolic homeostasis in the liver [8].

PATHOGENESIS OF NAFLD TO NASH

Though pathogenesis of NAFLD still remains unclear, "two-hit" theory has been proposed to explain the progression [9]. The "first hit" is hepatic steatosis that excessive lipid accumulation in hepatocytes is accompanied by elevated *de novo* lipogenesis and fatty acid uptake. Subsequently, oxidative stress and hepatic inflammation are critical two factors of the "second hit" which cause remarkable hepatic cellular damage. Besides, multiple hits including genetic mutations and intestinal microbiome also account for the progression of NAFLD [10].

FARNESOID X RECEPTOR: BILE ACID NUCLEAR RECEPTOR

FXR belongs to NR superfamily and is a ligand-activated transcriptional regulator, harbouring DNA binding and ligand binding domains [11]. It has been revealed that bile acids are endogenous ligand for FXR and regulate whole body metabolism, including cholesterol/bile acid metabolism, hepatic gluconeogenesis/lipogenesis, and even inflammation [8]. While FXR is abundantly expressed in the liver, intestine, and kidney, low expression of FXR has also been observed in the lung, adipose tissue, and heart [8]. Upon binding with endogenous bile acids, FXR heterodimerizes with its common binding partner, retinoid X receptor (RXR) to bind to the FXR response element (FXRE), in the target gene promoters. Once FXR heterodimerizes with RXR to bind to the FXRE, FXR is able to regulate expressions of its own target genes involved in various biological processes.

MECHANISMS OF FXR IN NAFLD: CHOLESTEROL/BILE ACID METABOLISM

FXR regulates bile acid synthesis by cholesterol catabolism. Bile acid synthesis occurs in the hepatocytes and compose of two pathways, such as classic pathway and alternative pathway [12]. The key enzymes in bile acid synthesis are cholesterol-7α-hydroxylase (CYP7A1) and sterol-27-hydroxylase (CYP27A1), respectively. Converted from cholesterol in the hepatocytes, bile acids are conjugated to taurine and/or glycine and then secreted into the gall bladder. Upon food intake, bile acids are secreted into the small intestine, and 95% of secreted bile acids are recycled and transported back to the liver via portal vein. The circulation of bile acids from the intestine to the liver is called bile acid enterohepatic circulation. Thus, 5% of bile acids excluded in each enterohepatic circulation. To compensate loss of bile acids, the liver synthesizes bile acids from cholesterol to maintain equivalent amount of bile acid pool.

As bile acids have the property of detergent, increasing accumulation of bile acids in the hepatocytes leads to cellular toxicity. To protect bile acids-induced cellular toxicity, bile acids binds to FXR to induces small heterodimer partner, which suppresses CYP7A1 gene expression to reduce the rate of bile acid synthesis in the liver. Besides hepatic regulation of bile acids, enterocytes are also involved in the regulation of bile acid synthesis. Upon bile acid activation in the intestine, FXR induces gene expression of fibroblast growth factor 15/19 (FGF15/19) in the enterocytes. Secreted FGF15/19 from enterocytes travel to the liver via portal vein and bind to FGF receptor 4 in the hepatocytes to activate JNK signaling pathway, which in turn suppresses gene expression of CYP7A1. Therefore, FXR activation in both hepatocytes and enterocytes can negatively regulates bile acid synthesis [12].

MECHANISMS OF FXR IN NAFLD: GLUCOSE/LIPID METABOLISM

Recently, FXR has been revealed to regulate expression of genes involved in glucose and lipid metabolism [13]. Phosphoenolpyruvate carboxykinase (PEPCK) is a well-known vital enzyme that catalyzes a crucial step of gluconeogenesis in the hepatocytes. Animal studies using FXR-null mice have shown that impaired FXR signaling pathway induced insulin resistance and elevated hepatic glucose production. Besides PEPCK, FXR also downregulates expression of glucose-6-phosphatase, a key enzyme to release glucose from the hepatocytes to the circulation

[14-16]. Altogether, FXR plays a key role to regulate glucose homeostasis via suppression of hepatic gluconeogenesis.

Extended studies have reported that FXR-null mice exhibit impaired lipid metabolism. Previous studies have shown that plasma triglyceride (TG) and cholesterol levels are elevated in FXR-null mice. In addition, treatment of FXR agonist, GW4064 in wild type mice largely reduced plasma TGs [17,18]. Furthermore, FXR has been shown to regulate a set of genes involved in lipoprotein metabolism including sterol regulatory element binding transcription factor 1, phospholipid transfer protein, stearoyl-coenzyme A desaturase 1, very low density lipoprotein receptor, apolipoprotein C2, and apolipoprotein E, suggesting that FXR plays a key role to regulate hepatic lipid metabolism [17].

MECHANISMS OF FXR IN NAFLD: INFLAMMATION

In addition to glucose/lipid metabolism, FXR also has anti-inflammatory action in the liver, mainly by supressing nuclear factor-κB signaling pathways. In murine model of NAFLD, FXR agonist GW4064 treatment reduces hepatic proinflammatory cytokine expressions [19]. Furthermore, FXR activation directly reduces lipopolysaccharide-induced proinflammatory cytokine expressions in macrophage [19], suggesting that FXR directly reduces inflammatory responses in immune cells. Consistent with that FXR directly suppresses inflammation, previous reports have shown that severe intestinal inflammation was observed in bile duct obstruction model with FXR-null mice [20]. Altogether, anti-inflammatory property of FXR proposes that FXR activation would be able to inhibit the progression of NASH from NAFLD by suppression of hepatic inflammation.

FXR ACTIVATION: POTENTIAL THERAPEUTIC STRATEGY FOR NAFLD

To date, no effective treatment has been established to manage the progression of NAFLD. As FXR plays critical roles to regulate various hepatic metabolism and inflammation, there is an emerging idea that FXR is an ideal therapeutic target for the treatment of NAFLD. Consistently, numerous natural and synthetic ligands of FXR have shown protective roles in rodent models of NAFLD.

GW4064 is a nonsteroidal FXR synthetic agonist and has been reported to reduce hepatic gluconeogenesis/lipogenesis and weight gain in rodent obese model. FXR activation by GW4064 represses hepatic steatosis by lowering TG and free fatty acid level in the liver, indicating that GW4064 has a therapeutic potential in NAFLD with its property to suppress hepatic lipogenesis [21].

Obeticholic acid (OCA or INT-747, 6α-ethyl-chenodeoxycholic acid) is a semisynthetic derivative of chenodeoxycholic acid found in primary bile acid of human and rodents which is the natural ligand of FXR. Administration of OCA reduces hepatic steatosis and insulin resistance in Zucker obese rats by reducing body weight gain and fat deposit in the liver [22]. OCA treatment largely reduces hepatic gluconeogenesis and lipogenesis, as well as hepatic inflammation. In addition to hepatic inflammation, FXR activation by OCA also reduced intestinal inflammation in experimental colitis rodent model [23]. Recently, administration of OCA has been significantly effective in the patients with type 2 diabetes mellitus and NAFLD (ClinicalTrials.gov, No. NCT00501592). In these patients, OCA treatment significantly improved insulin sensitivity while reduced hepatic damage marker alanine transferase and hepatic fibrosis [24].

Fexaramine is a synthetic FXR agonist and its chemical structure is markedly distinguished from natural bile acids and GW4064. Even target gene expression profile by fexaramine is distinct from those by natural bile acids and GW4064 [25]. Recently, fexaramine has been shown as gut-restricted FXR agonist to activate FXR in the intestine by oral administration [26]. Oral treatment of fexaramine significantly reduced body weight gain and inflammation in rodent obese model. Interestingly, hepatic steatosis and alanine transaminase level were largely decreased by fexaramine treatment, implying that intestinal FXR agonism would be potent therapeutic strategy to treat NAFLD [26].

CONCLUSIONS

FXR plays crucial roles in diverse physiological processes associated with cholesterol/bile acid metabolism, glucose/lipid metabolism, and inflammation, and has beneficial effects to reduce pathogenesis of NAFLD. Numerous studies have proved that FXR agonist or FXR agonism may have therapeutic potentials for the prevention and treatment of NAFLD. However, the role of FXR to regulate cholesterol metabolism is still debatable: the plasma high density lipoprotein (HDL) level was elevated in FXR-null mice, whereas inhibition of FXR transcriptional activity decreased plasma low density lipoprotein level and increased HDL level [27]. Consistently, FXR-null mice were protected from diet-induced obesity, glucose tolerance, and hepatic steatosis [28]. Thus, the physiological impact of FXR agonism

on cholesterol metabolism requires to be elucidated in detail.

In summary, numerous studies on therapeutic potentials of FXR for the treatment of NAFLD have provided new avenues to develop novel therapeutic strategy for NAFLD. Elucidating and understanding the molecular mechanisms during the pathogenesis of NAFLD would promote development of effective treatment for NAFLD. Although effectiveness of FXR activation on NAFLD is still debatable, FXR is an emerging therapeutic molecular target to manage NAFLD. Further attention and clinical evidence about FXR needs to be paid to avoid undesirable contradictory effects.

CONFLICTS OF INTEREST

No potential conflict of interest relevant to this article was reported.

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