Metabolism Clinical and Experimental 101 (2019) 153994

Contents lists available at ScienceDirect

Metabolism Clinical and Experimental

journal homepage: www.metabolismjournal.com

Fibroblast growth factor 21 in non-alcoholic fatty liver disease

Bradley Tucker [a], Huating Li [b,c,*], Xiaoxue Long [b,c], Kerry-Anne Rye [a], Kwok Leung Ong [a,**]

[a] Lipid Research Group, School of Medical Sciences, UNSW Australia, Sydney, NSW, Australia
[b] Department of Endocrinology and Metabolism, Shanghai Jiao Tong University Affiliated Sixth People's Hospital, Shanghai, China
[c] Shanghai Key Laboratory of Diabetes Mellitus, Shanghai Clinical Center of Diabetes, Shanghai, China

ARTICLE INFO

Article history:
Received 15 August 2019
Accepted 4 October 2019

Keywords:
Chronic liver disease
Fibroblast growth factor-21
Non-alcoholic fatty liver disease
Non-alcoholic steatohepatitis

ABSTRACT

Non-alcoholic fatty liver disease (NAFLD) encompasses a spectrum of pathologies ranging from uncomplicated hepatic fat accumulation to a state of lobular inflammation and hepatocyte ballooning, known as non-alcoholic steatohepatitis (NASH). Currently, there are no reliable biomarkers or effective therapeutic options established for NAFLD. Nevertheless, there are several molecular targets in the pipeline, of which fibroblast growth factor 21 (FGF21) is one. FGF21 is secreted primarily from liver and has a plethora of metabolic functions. Pre-clinical and epidemiological studies indicate a relationship between circulating FGF21 levels and hepatic fat content in both mice and humans. Moreover, animal studies have clearly shown that aberrant FGF21 signalling is a key pathological step in the development and progression of NAFLD. A recent Phase II clinical trial demonstrated that administration of an FGF21 analogue significantly reduced hepatic fat in subjects with NASH. As such, FGF21 provides a novel target for future biomarker and therapeutic studies. This review appraises preclinical data to outline the current understanding of FGF21 function in both normal hepatic function and NAFLD. Epidemiological evidence is explored to delineate the relationship between circulating FGF21 levels and NAFLD in humans. Finally, we review the therapeutic effects of FGF21 in the treatment of NAFLD.

Contents

Abbreviations: BAT, brown adipose tissue; cAMP, cyclic adenosine monophosphate; CNS, central nervous system; CVD, cardiovascular disease; EPAC, exchange protein directly activated by cyclic adenosine monophosphate; FFA, free fatty acid; FGF21, fibroblast growth factor 21; FGFR, fibroblast growth factor receptor; IL, interleukin; HCC, hepatocellular carcinoma; HIV, human immunodeficiency virus; HSC, hepatic stellate cell; NAFLD, non-alcoholic fatty liver disease; NASH, non-alcoholic steatohepatitis; PKA, protein kinase A; PPAR, peroxisome proliferator-activated receptor; ROS, reactive oxygen species; TG, triglyceride; TNF, tumour necrosis factor; VLDL, very-low-density lipoprotein; VLDLR, very-low-density lipoprotein receptor; WAT, white adipose tissue.

* Correspondence to: H. Li, Department of Endocrinology and Metabolism, Shanghai Jiao Tong University Affiliated Sixth People's Hospital, 200233, Shanghai, China.
** Correspondence to: K. L. Ong, School of Medical Sciences, University of New South Wales, Sydney, NSW 2052, Australia.
E-mail addresses: huarting99@sjtu.edu.cn (H. Li), kwokleung.ong@unsw.edu.au (K.L. Ong).

https://doi.org/10.1016/j.metabol.2019.153994
0026-0495/

1. Introduction

Non-alcoholic fatty liver disease (NAFLD) is estimated to effect one in four individuals globally and is now the leading cause of chronic liver disease [1]. NAFLD is defined as ≥5% hepatic fat content (steatosis) in the absence of a secondary cause, such as excessive alcohol use, viral infection or autoimmune disease [2]. Although the definition of excessive alcohol use is not uniform throughout the literature, current best practice guidelines define excessive alcohol consumption as >21 and >14 standard drinks per week for males and females, respectively [3]. NAFLD encompasses a spectrum of pathologies ranging from uncomplicated hepatic fat accumulation (simple steatosis) to a state of lobular inflammation and hepatocyte ballooning, known as non-alcoholic steatohepatitis (NASH). Patients with NASH experience accelerated hepatic fibrosis and up to 20% will develop widespread, irreversible fibrosis (cirrhosis) ultimately requiring liver transplantation [3,4]. NAFLD is currently the second most common indication for liver transplantation in the United States and is expected to surpass chronic hepatitis C for the top spot in coming years [5,6]. Moreover, steatosis and/or steatohepatitis is the most common reason for hepatic donor disqualification [7].

Unsurprisingly, NAFLD patients have an increased risk of hepatocellular carcinoma (HCC), particularly those who develop cirrhosis. However, the ramifications of NAFLD spread beyond the liver, to the heart, pancreas and kidneys. There is substantial evidence of a causal relationship between NAFLD and cardiovascular disease (CVD) [8–11]. Importantly, CVD is currently the leading cause of death in patients with NAFLD. Considering the two conditions share the majority of their risk factors (i.e. obesity, dyslipidaemia, hypertension, inflammation and insulin resistance) the association is expected. A recent meta-analysis concluded that NAFLD is an independent risk factor for determining cardiovascular events [12].

As the burden of NAFLD and its sequelae progresses, the need for a population-based screening method increases. Currently, a combination of liver enzymes and ultrasound are used for the diagnosis of NAFLD. However, the use of liver enzymes for the diagnosis of NAFLD is grossly inaccurate [13]. Ultrasound provides a more reliable diagnostic method, yet is limited by its sensitivity, user-dependence and relative cost [13]. Controlled attenuation parameter score assessment with the FibroScan® provides a more sensitive and specific alternative to traditional ultrasound [14]. Nevertheless, the cost and availability of this novel technique limits its application. Other imaging modalities, such as magnetic resonance spectroscopy and computed tomography are highly accurate, however, these options are not readily accessible nor cost-effective for such a prevalent disease [13]. Due to the lack of reliable biomarkers for NAFLD, patients frequently remain undiagnosed. Development of an effective biomarker would, not only streamline diagnosis, but help identify high-risk patients for treatment, monitor progression and provide vital information regarding prognostication. In cases where a diagnosis is made, treatment is currently limited to lifestyle adaptations, symptom management and cardiovascular risk reduction [3]. Evidently, there is an underwhelming supply of biomarkers and treatment options for patients with NAFLD. Nevertheless, several promising options are in the pipeline, of which, fibroblast growth factor 21 (FGF21) is one.

This review will provide an overview of FGF21 function and discuss its role in the pathogenesis and progression of NAFLD. Furthermore, epidemiological data is analysed to evaluate the relationship between circulating FGF21 levels and NAFLD in humans. Finally, by drawing from animal studies and human clinical trials we explore the therapeutic potential of FGF21 for the treatment of NAFLD.

2. NAFLD Pathogenesis

The development and progression of NAFLD is a multifactorial process, which requires contribution from genetic, physiological and environmental factors. Obesity and insulin resistance are the principal pathological mechanisms underpinning the development of NAFLD (Fig. 1). In fact, the prevalence of NAFLD is >75% in patients with type 2 diabetes mellitus and >90% in morbidly obese patients [1]. Broadly, obesity and insulin resistance promote hepatic steatosis by increasing free fatty acid (FFA) delivery to the liver. Obesity is the result of an energy imbalance, commonly due to excessive consumption of dietary fats, which induces adipocyte dysfunction; thereby increasing FFA release, promoting inflammation and altering adipokine expression (Fig. 1) [15]. Insulin resistance and the accompanying hyperinsulinemia activate lipogenic pathways that further increase FFA biosynthesis [16]. Chronic insulin resistance increases lipolysis in adipocytes, increases hepatic de novo lipogenesis, decreases hepatocyte β-oxidation and decreases very-low-density lipoprotein (VLDL) production and secretion from the liver. This ultimately results in triglyceride (TG) accumulation in the liver and disruption of hepatic lipid homeostasis [17]. Increased intrahepatic TG accumulation promotes gluconeogenesis and de novo lipogenesis, thereby worsening insulin resistance and the oversupply of FFAs [15]. This process can continue in an unrelenting cycle for many years.

Epidemiological data suggests that one-third of individuals with simple hepatic steatosis progress to NASH [1]. The exact reason why some progress and other do not, is not completely understood. However, lipotoxicity has been identified as a common underlying mechanism. Lipotoxicity occurs when the liver's ability to esterify FFAs is overwhelmed, leading to hepatic FFA accumulation [18]. FFAs are toxic to cells as they induce inflammation, endoplasmic reticulum stress, mitochondrial dysfunction and reactive oxygen species (ROS) formation [18,19]. Worsening parenchymal injury promotes the infiltration of innate (macrophages) and adaptive (B- and T-lymphocytes) immune cells, which further exacerbate lobular inflammation and degeneration [20,21]. Simultaneously, oxidative modification of host antigens triggers $CD4^+$ T-cell activation, leading to further activation of M1 macrophages and hepatic fibrogenesis [20]. Ultimately, the liver becomes diffusely fibrotic and the risk of decompensation and hepatocellular carcinoma (HCC) increases dramatically (Fig. 1) [18].

3. FGF21

In humans, 22 members of fibroblast growth factor family have been identified. FGF21 is a hormone-like FGF commonly expressed in the liver, adipose tissue and pancreas [22]. Circulating FGF21 is predominantly liver-derived [22]. Hepatic FGF21 expression is primarily regulated by nutritional stress – specifically starvation. Hepatic FGF21 expression is regulated by the peroxisome proliferator-activated receptor (PPAR)-α pathway and the exchange protein directly activated by

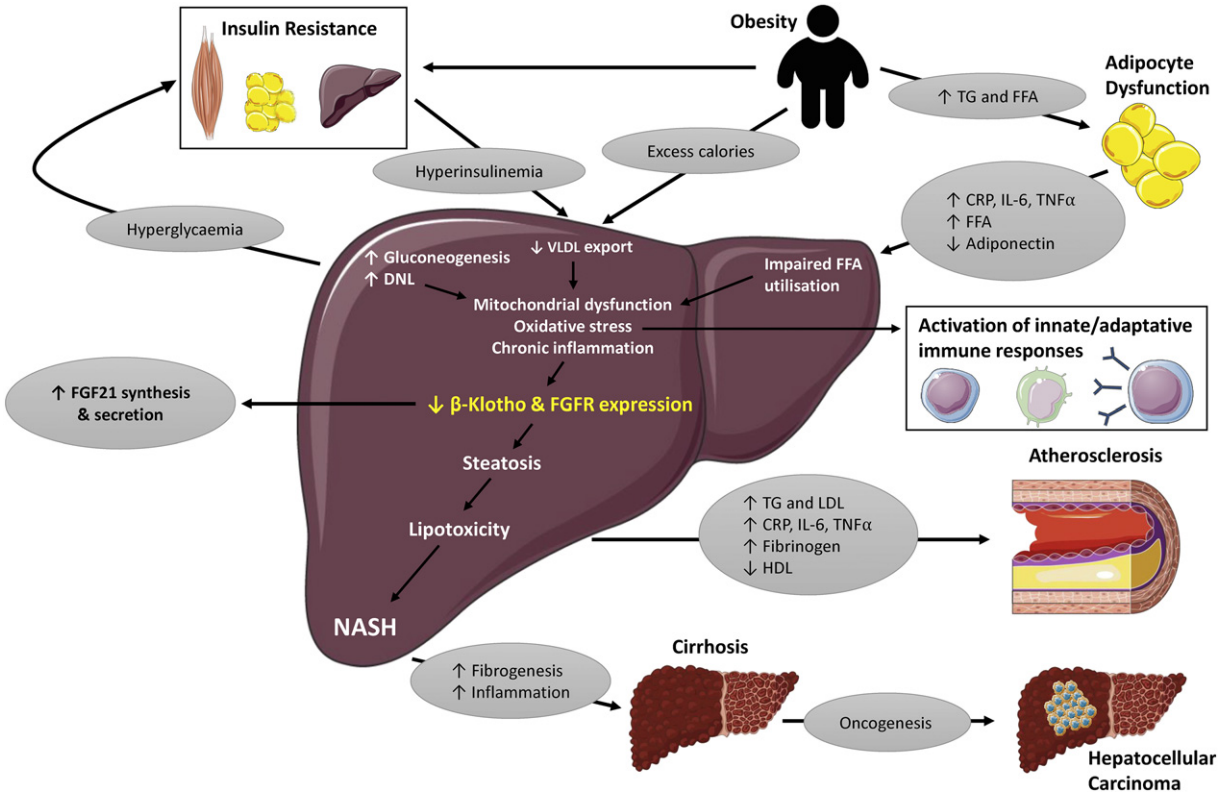

Fig. 1. Pathogenesis of NAFLD. Obesity and insulin resistance drive steatosis by promoting adipocyte dysfunction, hyperglycaemia and hyperinsulinemia. Oxidative damage and chronic inflammation suppress β-klotho and FGFR expression, leading to a compensatory increase in FGF21 synthesis and secretion. Steatosis impairs normal hepatocellular functions and lipid metabolites begin to accumulate. Lipotoxicity and oxidative damage stimulate innate and adaptative immune responses leading to activation of hepatic stellate cells (HSCs). The chronic inflammatory state associated with NASH predisposes individuals to cardiovascular disease; whereas, activation of HSCs induces fibrosis and can lead to cirrhosis and hepatocellular carcinoma. Abbreviations: CRP, C-reactive protein; DNL, de novo lipogenesis; ER, endoplasmic reticulum; FC, free cholesterol); FFA, free fatty acid; HDL, high-density lipoprotein; IL-6, interleukin-6; LDL, low-density lipoprotein; TG, triglycerides; TNFα, tumour necrosis factor α; VLDL, very-low-density lipoprotein.

cyclic adenosine monophosphate (EPAC)/protein kinase A (PKA) pathway. The PPARα pathway is activated by FFAs and/or protein insufficiency, which increases FGF21 gene expression [23]. The EPAC/PKA pathway is activated by stimulation of the hepatic glucagon receptor, triggering a signalling cascade that results in activation of the PKA and EPAC branches of the cyclic adenosine monophosphate (cAMP) pathway [24]. As such, EPAC/PKA signalling increases FGF21 gene expression and secretion, via pre- and post-transcriptional mechanisms [24].

FGF21 acts in an endocrine, paracrine and autocrine-like fashion via fibroblast growth factor receptors (FGFRs). There are four FGFRs: FGFR1–4. FGF21 is known to interact with FGFR1–3 [25]. Binding of FGF21 to FGFRs requires the co-receptor β-Klotho, a transmembrane glycoprotein. β-klotho expression is tissue-specific, and expressed predominantly in the liver and adipose tissue [25]. A β-klotho independent pathway for FGF21 signalling was proposed by Tomiyama et al. (2010), however, there is little evidence supporting this theory [26].

4. Metabolic Functions of FGF21

4.1. Overview

FGF21 is involved in many metabolic processes including insulin sensitivity, glucose and lipid metabolism, and energy homeostasis. The metabolic effects of FGF21 were first described by Kharitonenkov et al. (2005) who demonstrated that FGF21 stimulation enhances glucose uptake in adipocytes by increasing GLUT1 expression [27]. In the same study, daily administration of FGF21 to mice markedly reduced plasma glucose and insulin levels. These investigators also found that transgenic *Fgf21*$^{+/+}$ mice have enhanced insulin sensitivity, reduced hepatic steatosis and increased brown adipose tissue (BAT) mass [27]. This seminal paper brought FGF21 into the spotlight as a key regulator of glucose homeostasis.

4.2. FGF21 and Peripheral Metabolism

Ketogenic diets are commonly employed to challenge metabolic pathways. Consumption of a ketogenic diet restricts carbohydrate intake and switches the primary energy source to ketones – a product of fatty acid metabolism. Typically, mice that are fed a ketogenic diet, lose weight and have elevated circulating FGF21 levels. In contrast, *Fgf21*$^{-/-}$ mice gain weight and develop significant hepatic steatosis following exposure to a ketogenic diet [28]. This abnormal response to the ketogenic diet is associated with a reduction in β-hydroxybutyrate levels, indicating that FGF21 is required for fatty acid oxidation [28]. This effect has been corroborated by Potthoff et al. (2009), who showed that *Fgf21*$^{+/+}$ transgenic mice have markedly increased hepatic β-oxidation [29]. Adding to this, deficiency of FGF21 has been shown to impair ketogenesis [28]. FGF21 also suppresses hepatic lipogenesis in an autocrine and paracrine manner, therefore, redirecting fatty acids to β-oxidation [30]. FGF21 additionally influences glucose production by activating PPAR-γ coactivator 1α, which regulates hepatic glucose and lipid metabolism [29]. Similar to a ketogenic diet, amino acid deprivation causes a robust increase in FGF21 expression. Under these conditions, *Fgf21*$^{-/-}$ mice have a reduced ability to induce lipolysis and repress lipogenesis [31]. Moreover, *Fgf21*$^{-/-}$ mice have impaired upregulation of uncoupling protein 1 – a BAT-specific, inner mitochondrial membrane transport protein essential for non-shivering thermogenesis [31]. When taken together, these studies indicate that FGF21 is essential for proper lipid and glucose metabolism. They also suggest that

interference of this pathway can have deleterious effects on metabolic homeostasis.

Adding to its beneficial metabolic profile, FGF21 promotes insulin sensitisation and enhances β-cell function. The induction of FGF21, whether by transgenic overexpression or therapeutic administration of FGF21, protects mice against insulin resistance and lowers systemic insulin levels [32–34]. Similarly, *Fgf21*$^{-/-}$ mice tend to be hyperinsulinemic and have diminished insulin sensitivity, especially in the liver [35,36]. The insulin sensitising effects of FGF21 are, at least in part, mediated by its ability to promote the healthy expansion of subcutaneous adipose tissue [36]. In diabetic *db*/*db* mice, FGF21 expression is elevated in the liver and adipose tissue, but is downregulated in the pancreatic islets [37]. It is this downregulation of FGF21 in the pancreatic islets that promotes β-cell dysfunction [37]. *Fgf21*$^{-/-}$ mice develop islet hyperplasia, β-cell proliferation and perivascular T lymphocytic inflammation that ultimately leads to increased insulin synthesis [38,39]. When pancreatic FGF21 expression is increased by adeno-associated virus gene therapy technique, islet morphology and function dramatically improves [37]. Moreover, FGF21 can restore β-cell function and reduce cytokine-induced β-cell apoptosis [40].

FGF21 is involved in a complex interplay with the adipokines, leptin and adiponectin. In mice lacking either of these adipokines, the beneficial metabolic effects of FGF21 are diminished [41,42]. In vitro, leptin treatment of hepatocytes increases FGF21 expression [43], which is required to maintain leptin sensitivity [44]. On the other hand, stimulation of adipocytes with FGF21 augments adiponectin production [41]. In fact, chronic exposure to increased FGF21 levels raises plasma adiponectin levels in mice [34,45], and humans [46–48]. The 'FGF21-adipokine axis' is not completely understood, although it is clear that functional adipokine expression is essential for FGF21 to exert its glycaemic and insulin sensitising effects. Future studies examining this relationship would provide valuable information and markedly improve understanding of the role of FGF21 in metabolic diseases, such as NAFLD.

Energy expenditure is partially regulated by FGF21. FGF21 administration reduces body weight and increases energy expenditure in mice [49]. This effect is partially due to the action of FGF21 on adipose tissue. In BAT, FGF21 upregulates thermogenic genes [49]. In white adipose tissue (WAT), FGF21 promotes 'browning'; a process whereby BAT-specific genes are upregulated, and tissue specific energy consumption is increased [50]. Browning of WAT is known to improve metabolic parameters and has been proposed as a therapeutic target for metabolic disorders [51]. This increased energy expenditure, at least partially explains the increased longevity seen in *Fgf21*$^{+/+}$ mice [34].

4.3. Centrally Acting FGF21

Beyond its role in the periphery, FGF21 also crosses the blood-brain barrier by simple diffusion and acts on the central nervous system (CNS) [52]. A continuous low dose intracerebroventricular infusion of recombinant human FGF21 can increase energy expenditure and improves insulin sensitivity in obese rats [53]. Human FGF21 is not detected in the peripheral circulation of these rats, which suggests that the metabolic effects of FGF21 were regulated via the CNS. Centrally acting FGF21 can increase energy expenditure, promote 'browning' of WAT and activate BAT in mice [54]. β-klotho and several FGFRs, are expressed in the CNS [55]. The transgenic overexpression of FGF21 in the livers of mice increases systemic corticosterone levels, reduces basal plasma insulin levels and alters circadian periodicity. These effects are abrogated by CNS-specific deletion of β-klotho [55].

In humans, FGF21 is detectable in the cerebrospinal fluid and the level positively correlates with plasma FGF21 levels. Moreover, cerebrospinal fluid FGF21 levels positively correlate with body mass index (BMI), fat mass and plasma insulin [56]. Taken together, these data highlight a central pathway through which FGF21 acts to exert its beneficial metabolic effects. Consideration of this CNS pathway for FGF21 signalling should be an important aspect of future studies aiming to develop FGF21 mimetics for NAFLD and other metabolic diseases.

5. FGF21 in NAFLD

5.1. Overview

As FGF21 is a liver derived, metabolically active hormone, it is conceivable that FGF21 is implicated in the pathobiology of NAFLD. In fact, there is an abundance of preclinical evidence demonstrating that aberrant FGF21 signalling is, at least partially, responsible for the pathogenesis and progression of NAFLD [9,57]. In humans, observational studies have demonstrated that circulating FGF21 levels are elevated in subjects with NAFLD [58]; thus, indicating a state of potential 'FGF21 resistance'.

5.2. Preclinical Evidence of the Role of FGF21 in Hepatic Fat Accumulation

In contrast to the beneficial metabolic effects of FGF21 in animal studies, the studies in Table 1 show that both systemic FGF21 levels and hepatic expression of FGF21 is increased in patients with NAFLD – suggesting that the function of FGF21 is impaired. The term 'FGF21 resistance' was coined by Fisher et al. (2010) who demonstrated that obese mice have reduced hepatic and WAT expression of the FGF21 receptor complex, and that this was associated with a blunted response following the administration of exogenous FGF21 [59]. These results indicate the presence of FGF21 resistance, similar to that of insulin in type 2 diabetes mellitus. Although the existence of FGF21 resistance in obesity remains a contentious issue [60], it appears to play a distinct role in NAFLD. An elegant study by Rusli et al. in which mice were maintained on a medium-fat diet and hepatic steatosis and fibrosis were monitored for up to 26 months [61] established that NAFLD develops in the later stages of life in this species, as is the case in humans. In these mice, plasma FGF21 levels correlated positively with hepatic fat content, yet negatively with hepatic expression of β-klotho, FGFR2 and FGFR4; implying some degree of FGF21 resistance. The suppression of hepatic β-klotho expression is thought to be driven by inflammatory cytokines including interleukin (IL)-1β, IL-6 and tumour necrosis factor (TNF)-α that are commonly elevated in NAFLD [62].

Mechanistically, it appears that the elevation of FGF21 levels in NAFLD patients is the product of dysfunctional PPARα signalling. Mice with NAFLD have an augmented response to PPARα agonism, resulting in enhanced FGF21 expression when compared to non-NAFLD controls [61]. In addition to FGF21, other genes that regulate lipogenesis and cholesterol metabolism are also up-regulated in NAFLD [61]. In the setting of NAFLD, PPARα is activated by intrahepatic fatty acids [63] and sustained activation of this dysfunctional signalling pathway results in the elevated FGF21 levels that are characteristic of individuals with NAFLD.

Due to the central role of FGF21 in lipid metabolism, resistance to this hormone can have deleterious effects on the liver. Circulating FFAs levels are elevated in patients with NAFLD, and hepatic FFA deposition is responsible for approximately 60% of hepatic TG accumulation [64]. In mice, the administration of FGF21 acutely reduces circulating FFA levels by suppressing hormone-sensitive lipase and perilipin expression in WAT [59]. Additionally, FGF21 increases the deposition and catabolism of TG-rich lipoproteins in WAT and BAT, thus reducing plasma TGs [65]. Expression of the hepatic VLDL receptor (VLDLR), responsible for hepatic lipid uptake, is greatly increased in subjects with NAFLD [66]. Hepatic VLDLR expression is also increased in *Fgf21*$^{-/-}$ mice and the restoration of FGF21 in these mice by human recombinant FGF21 administration inhibits endoplasmic-reticulum stress-induced VLDLR expression and reduces hepatic steatosis [66]. FGF21 also alters the hepatic expression of genes involved in hepatic lipid metabolism: it upregulates anti-lipogenic genes (*Lepr* and *Igfbp2)* and represses genes involved in lipid synthesis (*Scd1* and *Gck)* [67].

Table 1
Studies examining the relationship of FGF21 with NAFLD in humans.

Population	Key findings	Reference
Overweight participants ($n = 187$) • No NAFLD (n = 41) • NAFLD ($n = 146$) ○ NASH ($n = 94$)	• Circulating FGF21 inversely correlated with insulin sensitivity in skeletal muscle, adipose tissue, but not the liver • Circulating FGF21 was the highest in patients with NASH • Circulating FGF21 increased with worsening fibrosis and necroinflammation; but not with worsening steatosis	[85]
Non-obese HIV-infected ($n = 73$) • No/mild steatosis ($n = 43$) • Severe steatosis ($n = 30$)	• Circulating FGF21 was higher in those with severe steatosis compared to those with no/mild steatosis • FGF21 is an independent predictor of severe hepatic steatosis • Circulating FGF21 ≥ 51 pg/mL was diagnostic of severe steatosis, with an accuracy of 64%	[111]
Hepatitis B-positive ($n = 415$) • NAFLD ($n = 192$) • No NAFLD ($n = 223$)	• Circulating FGF21 increased progressively with increasing severity of steatosis. • FGF21 is an independent predictor of severe hepatic steatosis • Circulating FGF21 relatively unaffected by degree of fibrosis	[110]
Chinese population ($n = 565$) • No NAFLD ($n = 442$) • NAFLD ($n = 123$)	• Circulating FGF21 increased in patients with NAFLD • Baseline FGF21 predicted onset of simple steatosis • Baseline FGF21 could not predict the onset of NASH, resolution of steatosis or progression of steatosis to NASH	[84]
Morbidly obese females (n = 56) • No NAFLD ($n = 17$) • NAFLD ($n = 39$)	• Circulating FGF21 was higher in patients with a higher steatosis grade • Circulating FGF21 did not differ significantly between patients with and without fibrosis	[102]
Chinese population ($n = 270$) • NAFLD ($n = 179$) • No NAFLD ($n = 91$)	• Circulating FGF21 was elevated in NAFLD patients • Circulating FGF21 was the highest in patients with NASH	[82]
Paediatric participants ($n = 107$) • No NAFLD ($n = 23$) • NAFLD ($n = 84$)	• Circulating FGF21 was lower in NAFLD patients than controls • Circulating FGF21 was lower in NASH compared to simple steatosis • Higher FGF21 levels were associated with lower probability of NASH • Hepatic β-klotho expression was reduced in patients with NAFLD	[89]
Adolescent participants ($n = 217$) • No NAFLD ($n = 138$) • NAFLD ($n = 79$)	• Circulating FGF21 was higher in obese patients and those with higher HFC • Circulating FGF21 correlated positively with visceral fat, HFC and ALT • In NASH patients, circulating FGF21 correlated positively with NAFLD activity score	[91]
Chinese population ($n = 220$) • No NAFLD ($n = 74$) • NAFLD (n = 146)	• Circulating FGF21 was higher in NAFLD patients than controls • Circulating FGF21 had weak association with lobular inflammation • Circulating FGF21 was the highest in patients with NASH	[81]
Paediatric participants ($n = 100$) • No NAFLD ($n = 48$) • NAFLD ($n = 52$)	• Circulating FGF21 was increased in obese children and correlated positively with BMI • Circulating FGF21 positively correlated with FFA and leptin levels • Circulating FGF21 did not significantly differ between children with or without metabolic syndrome or NAFLD	[90]
Chinese population ($n = 99$) • No NAFLD ($n = 62$) • NAFLD ($n = 37$)	• Circulating FGF21 was elevated in those with T2DM and/or NAFLD • Circulating FGF21 positively correlated with CRP levels in T2DM patients	[125]
Participants with abnormal glucose metabolism and NAFLD (n = 138)	• Circulating FGF21 increased progressively with HFC for HFC quartiles 1–3 yet decreased in the fourth HFC quartile • Circulating FGF21 was the strongest factor associated with HFC in patients with abnormal glucose metabolism.	[126]
Chinese population ($n = 348$) • No NAFLD ($n = 124$) • NAFLD ($n = 224$)	• Circulating FGF21 was elevated in NAFLD patients • Hepatic FGF21 expression increased in participants with steatosis • Circulating and hepatic FGF21 levels correlated positively with HFC • Circulating FGF21 correlated positively with waist circumference, ALT, GGT, total cholesterol, triglycerides and HDL cholesterol	[80]
Overweight participants ($n = 159$) • No NAFLD ($n = 77$) • NAFLD ($n = 82$)	• Circulating FGF21 was elevated in NAFLD patients • Circulating FGF21 correlated positively with steatosis score • No association was found between FGF21 and NASH	[83]
Total participants (n = 21) • No NAFLD (n = 6) • NAFLD (n = 15)	• Circulating FGF21 correlated positively with BMI • Circulating FGF21 was elevated in NAFLD patients • Hepatic FGF21 expression was elevated in simple steatosis, but not NASH. • Circulating FGF21 was not altered by nutritional challenges (i.e. OGTT or ketogenic diet)	[79]

Abbreviations: ALT, alanine aminotransferase; BMI, body mass index; CRP, C-reactive protein; FFA, free fatty acid; GGT, gamma glutamyltransferase; HDL, high-density lipoprotein; HFC, hepatic fat content; HIV, human immunodeficiency virus; OGTT, oral glucose tolerance test; NAFLD, non-alcoholic fatty liver disease; NASH, non-alcoholic steatohepatitis; T2DM, type 2 diabetes mellitus.

The autophagic degradation of intracellular lipid droplets (lipophagy), has recently been identified as another pathway of interest in the pathogenesis of NAFLD [68]. Reduced lipophagy is associated with endoplasmic reticulum stress, insulin resistance and hepatic steatosis [68,69]. Inhibition of autophagy predisposes hepatocytes to lipid accumulation [70]. Moreover, knockdown of the essential autophagy genes, *Atg14* and *Tfeb*, promotes excessive hepatic lipid accumulation in mice [71,72]. A comprehensive study by Zhu et al. has reported that FGF21 induces lipophagy by increasing expression of key autophagic genes, sequestration of lipids within autophagosomes and autophagic flux; therefore, correcting hepatic lipid metabolism [73]. A similar mechanism whereby FGF21 deficiency reduces autophagy has recently been proposed for pancreatic islets [74]. Taken together, these studies suggest that FGF21 resistance may impair autophagic mechanisms within hepatocytes, thereby increasing susceptibility to NAFLD.

Excess sugar intake, particularly fructose, can have an important impact on hepatic metabolism. A recent review suggests that excessive sugar consumption predisposes individuals to NAFLD by enhancing lipogenesis and impairing fatty acid oxidation [75]. FGF21 production is stimulated by the ingestion of either fructose or glucose, and it may regulate simple sugar intake, via a negative feedback loop involving the liver and hypothalamus [76]. Sugar intake is increased in *Fgf21*$^{-/-}$

mice, and is suppressed by overexpression of FGF21, which acts on neurons in the hypothalamus to reduce 'sweet-seeking' behaviour and meal size [77]. The spike in FGF21 secretion following a carbohydrate load also acts as a mechanism for peripheral disposal of excess energy. Increases in circulating FGF21 post-feeding correlate with increased hepatic lipogenesis and peripheral glucose secretion [78]. As such, impaired FGF21 signalling, would lead to unopposed carbohydrate consumption with inadequate peripheral disposal of the excess energy.

5.3. Clinical Evidence of the Relationship between FGF21 and NAFLD in Humans

Table 1 shows a summary of clinical studies reporting on the relationship of circulating FGF21 levels with NAFLD in humans. In a cohort of 21 participants, Dushay et al first reported that both the plasma concentration and hepatic expression of FGF21 is elevated in patients with NAFLD [79] – a finding supported by several subsequent cross-sectional studies [80–83]. In a 3-year longitudinal analysis of 565 Chinese participants, elevated serum FGF21 levels predicted the onset of simple steatosis, but not the onset of NASH or the resolution of steatosis [84]. In contrast to the vast majority of existing evidence, a recent study by Barb et al. found no difference in the plasma FGF21 level between those with and without NAFLD [85]. Reasons for this disparity may include the lack of a lean control group and differences in diagnostic methods. The control cohort in the study by Barb et al. had a mean plasma FGF21 of 325 pg/mL, which is much higher than those levels (<200 pg/mL) previously reported in heathy subjects [80,81,83,84]. Circulating FGF21 levels have also been positively associated with NAFLD risk factors, including BMI, waist circumference, C-reactive protein, insulin resistance and the metabolic syndrome [79,80,85–87].

Within the paediatric population, the relationship between circulating FGF21 levels and NAFLD appears to be even more complex, potentially because of different metabolic regulatory mechanisms. A recent study of a large paediatric population found that the correlation of FGF21 levels with obesity, insulin resistance and metabolic syndrome are the inverse of that found in the adult population, with lower FGF21 levels in children with metabolic dysfunction [88]. Hence, FGF21-deficiency, as opposed to FGF21 resistance may predispose children to insulin resistance and its sequelae. In support of this hypothesis, Alisi and colleagues (2013) reported that a lower serum FGF21 level is associated with an increased risk of NASH in children [89]. In the same study, lower hepatic β-klotho expression was associated with NASH and increased circulating FGF21 levels. In contrast, two earlier studies yielded conflicting results. The first of these studies did not find an association between FGF21 levels and NAFLD in pre-pubertal children or in subjects in the early stages of puberty [90]. The second study reported a robust positive association between FGF21 levels and hepatic fat content in adolescents [91]. As such, it is apparent that age and potentially pubertal factors (e.g. adipokine and sex-hormone levels) modify the relationship of FGF21 levels with NAFLD.

A meta-analysis pooling results from a total of 1724 subjects, confirmed that circulating FGF21 levels are elevated in patients with NAFLD [58]. Additionally, this analysis pooled results from four studies with data on the diagnostic accuracy of FGF21 in NASH – reporting the overall sensitivity and specificity as 0.62 and 0.78, respectively. The diagnostic accuracy of FGF21 for NAFLD, without NASH, was not calculated. This pooled-analysis provides an interesting overview of FGF21 in NAFLD; however, the study is limited by its relatively small sample size, marked cross-study heterogeneity, predominantly single ethnic population, inadequate subgroup analyses, and pooling of data from studies in paediatric and non-paediatric populations in a single analysis.

5.4. Preclinical Evidence of the Role of FGF21 in NASH

Arguably, isolated steatosis is not detrimental to hepatic function. The esterification and deposition of TGs in the liver is a compensatory response to the high levels of hepatic FFAs [18]. However, this compensatory response can be overwhelmed leading to the accumulation of fatty acids in the liver and lipotoxicity [18,19] – a process that is exacerbated by FGF21 deficiency [92]. *Fgf21*$^{-/-}$ mice have significantly reduced acyl CoA synthetase activity, which inhibits the hepatic conversion of FFAs to acyl CoA [92]. This leads to accumulation of FFAs that induce a lipotoxic effect and promote the development of NASH. Treatment of *Fgf21*$^{-/-}$ mice with exogenous FGF21 restores acyl CoA synthetase activity and attenuates the development of NASH [92]. Furthermore, apoptosis is enhanced in *Fgf21*$^{-/-}$ mice due to prolonged lipotoxicity [93].

NASH is principally an inflammatory condition in which activation of pro-inflammatory pathways induces fibrogenesis, the first stage of cirrhosis. There is strong evidence indicating that FGF21 has an anti-inflammatory effect, both within the liver and systemically. Mice with global FGF21 deficiency have an augmented inflammatory response, increased hepatic macrophage infiltration, and increased expression of pro-inflammatory and pro-fibrotic cytokines [93]. Increasing FGF21 production in mice by adeno-associated viral vector-mediated gene therapy inhibits hepatic macrophage infiltration [94]. Moreover, pharmacological doses of FGF21 suppress pro-inflammatory cytokine levels in the liver, plasma and WAT [95,96]. It is not clear how FGF21 induces this anti-inflammatory effect, however, two potential pathways have been identified. First, FGF21 can exert an anti-inflammatory effect in macrophages by inhibiting nuclear factor kB (NF-kB) activation and stimulating the nuclear factor erythroid 2-related factor 2 (Nrf2) signalling pathway [97]. Second, FGF21 protects hepatocytes from inflammatory insults by diminishing the negative effects of the pro-inflammatory cytokine IL-1β [62].

A fine balance between the generation and elimination of reactive oxygen species (ROS) is essential for maintenance of normal cellular function. Mitochondrial dysfunction and augmented ROS production is evident in hepatocytes that are overloaded with FFAs [98]. ROS also have deleterious effects on hepatocellular homeostasis by inducing lipid peroxidation, hepatic stellate cell (HSC) stimulation, hepatocyte apoptosis and activation of both the innate and adaptive immune responses [98]. Under normal physiological conditions, FGF21 reduces oxidative stress by upregulating Nrf2-mediated antioxidant capacity [97]. In mice, administration of exogenous FGF21 improves hepatic and adipocyte mitochondrial function [99,100], while FGF21-deficency increases hepatic ROS accumulation, which can be alleviated by the replenishment of FGF21 [101]. Therefore, it is ostensible that FGF21 resistance promotes NAFLD by enhancing oxidative stress.

5.5. Clinical Evidence of the Relationship of FGF21 With NASH in Humans

Although both subsets of NAFLD, simple steatosis and NASH differ significantly in their pathophysiological mechanisms – simple steatosis being relatively benign hepatic lipid accumulation and NASH, an inflammatory disorder driven by lipotoxicity. As such, these differences can influence the relationship of circulating FGF21 levels with NAFLD. An early report indicated that both serum and hepatic FGF21 levels are lower in patients with NASH compared to those with simple steatosis [79]. However, this finding has since been refuted in subsequent studies. In a cohort of 179 patients with NAFLD and 91 healthy controls, a linear relationship between circulating FGF21 levels and NAFLD progression was reported, with the highest FGF21 levels found in subjects with NASH [82]. Additionally, FGF21 levels correlate positively with the degree of necro-inflammation (hepatocyte ballooning and lobular inflammation) and fibrosis in patients with NASH [85]. FGF21 levels also correlate positively with cytokeratin-18 [85]; a marker of hepatocyte apoptosis and fibrosis. On the contrary, a cross-sectional study of 56 morbidly obese females undergoing bariatric surgery [102] failed to demonstrate any association between plasma FGF21 and pathological features of NASH, suggesting that gender, body fat and other co-morbidities may modify this association.

6. FGF21 in Secondary Steatosis

Secondary steatosis (i.e. preceded by excessive alcohol consumption and/or viral infection) does not fall under the umbrella of NAFLD. Nevertheless, FGF21 also appears to be involved in the pathobiology of these important causes of steatosis.

6.1. Alcoholic Fatty Liver Disease

Ethanol is a potent inducer of FGF21 [103]. In humans, 0.9 g/kg ethanol consumption causes a 40-fold increase in serum FGF21 levels [103], possibly as a negative feedback mechanism to reduce alcohol consumption by reducing the activity of dopaminergic reward pathways [104]. Independent of its aforementioned effect on alcohol preference, FGF21 protects against alcoholic liver disease by limiting hepatotoxicity [103,105]. Furthermore, recombinant FGF21 has been shown to ameliorate ethanol-induced liver damage by improving hepatic metabolism in mice [106]. Large epidemiological studies are required to delineate the relationship of circulating FGF21 levels with alcoholic fatty liver disease in humans.

6.2. Infection-association Steatosis

Chronic infections, such as human immunodeficiency virus (HIV) and hepatitis, predispose individuals to hepatic steatosis [107]. On the other hand, steatosis has been suggested to accelerate fibrosis in patients chronically infected with hepatitis C [108]. In three independent studies, circulating FGF21 levels have demonstrated a positive

Table 2
FGF21 augmentation in NAFLD.

Reference	Population and intervention	Effect
Humans		
[48]	Obese participants with biopsy confirmed NASH ($n = 75$) Intervention: subcutaneous pegbelfermin	• Improved lipid profile • Reduced hepatocyte damage and fibrosis • Increased systemic adiponectin levels • Reduced hepatic fat content
[46]	Obese participants with T2DM Intervention: subcutaneous pegbelfermin	• No effect on HbA1c • Improved lipid profile • Reduced hepatocyte damage and fibrosis • Increased systemic adiponectin levels
[123]	Obese participants with hypertriglyceridaemia ± T2DM (n = 107) Intervention: intravenous PF-05231023	• No effect on body weight • Improved lipid profile • Dose-dependent increase in adiponectin • No effect on fasting plasma glucose
[47]	Obese participants with T2DM ($n = 47$) Intervention: subcutaneous LY	• Improved lipid profile • Reduced fasting insulin • Decreased body weight • Increased systemic adiponectin levels
Mice		
[95]	Diet-induce obesity Intervention: human FGF21	• Corrected cognitive impairment and anxiety • Reduced body weight, blood glucose and HbA1c • Improved lipid profile • Suppressed local and systemic inflammation
[96]	Methionine choline-deficient diet Intervention: PsTag600-FGF21	• Reduced body weight • Improved lipid profile and glucose utilisation • Reduced circulating FFA and liver enzymes • Reduced hepatic cholesterol and triglyceride content • Suppressed hepatic inflammation and fibrosis
[32]	Choline-deficient high-fat diet Intervention: PsTag600-FGF21	• Reduced body weight, • Improved lipid profile and glucose utilisation • Reversed hepatic steatosis • Attenuated local and systemic inflammation • Increased adiponectin levels • Decreased liver enzyme levels
[117]	High-fat diet obesity Intervention: native FGF21	• Reduced fasting insulin and free fatty acids • Improved glucose utilisation • Attenuated hepatic lipid accumulation • Increased energy expenditure • Increased insulin sensitivity (particularly in BAT)
[100]	Leptin-deficient +- methionine and choline-deficient diet Intervention: LY2405319	• Suppressed liver enzymes and inflammation • Reduced oxidative stress and lipid peroxidation • Reduced hepatic steatosis and fibrosis • Enhanced mitochondrial function • Improved insulin sensitivity and energy expenditure
[119]	Dimethylnitrosamine-induced hepatic fibrogenesis Intervention: human FGF21	• Attenuated hepatic fibrosis by inhibiting HSC activation • Promoted apoptosis of activated HSCs • Reduced hepatic inflammation by inhibiting NF-kB induction
[73]	Monosodium L-glutamate-induced NAFLD Intervention: native FGF21	• Reduced body weight • Improved lipid profile • Suppressed intracellular hepatocyte lipid accumulation • Increased energy expenditure and glucose utilisation • Suppressed expression of fat synthesis genes
[92]	Methionine and choline-deficient diet Intervention: human FGF21	• Reduced hepatic steatosis and peroxidative damage • Increased fatty acid β-oxidation • Inhibited pro-inflammatory and pro-fibrotic gene expression

Abbreviations: BAT, brown adipose tissue; FFA, free fatty acid; HbA1c, Haemoglobin A1C; HSC, hepatic stellate cell; NAFLD, non-alcoholic fatty liver disease; NASH, non-alcoholic steatohepatitis; NF-kB, nuclear factor kappa-light-chain-enhancer of activated B cells; T2DM, type 2 diabetes mellitus.

correlation with the degree of hepatic steatosis in individuals with chronic HIV, hepatitis B or hepatitis C [109–111]. In patients chronically infected with hepatitis B, circulating FGF21 levels correlated inversely with the extent of hepatic fibrosis [110]. Therefore, as the liver becomes fibrotic, its ability to synthesize and secrete FGF21 is likely to be diminished.

7. FGF21 in Hepatocarcinogenesis

Hepatocellular carcinoma (HCC) is a potentially fatal complication of NAFLD. There is mounting evidence suggesting that aberrant FGF21 signalling may predispose individuals with NAFLD to hepatocarcinogenesis. Zhang et al. first noted that reduced FGF21 levels were associated with cancerous hyperproliferation and atypical oncogene signalling in the liver [112]. It has since been reported that plasma FGF21 levels correlate positively with transcriptional changes related to hepatocarcinogenesis, and that FGF21 deficiency compromises tumour suppressor signalling [61,93]. Recently, *Fgf21*$^{-/-}$ mice have been shown to have a 13-fold increased risk of HCC when fed an obesogenic diet for 52-weeks [113]. This latest report provides strong evidence for a causal role of aberrant FGF21 signalling in hepatocarcinogenesis. Large-scale epidemiological studies investigating this association would provide valuable insight into the significance of this relationship in humans.

8. Therapeutic Potential of FGF21 Modulation in NAFLD

Currently, there are no effective therapies for the treatment of NAFLD. A recent systematic review concluded that, due to the low level of evidence available, the effectiveness of pharmacological treatment in NAFLD is unclear and recommendations cannot be made for or against [114]. Nevertheless, there are several experimental treatments in the pipeline, including FGF21 (Table 2). Utilising preclinical evidence, Fig. 2 summarises the therapeutic effects of FGF21 for the treatment of NAFLD.

As FGF21 has a short half-life, and poor instability and bioavailability [25], several FGF21 mimetics have been developed in recent years. Typical pharmacological doses of FGF21 range from 100 to 1000 ng/mL, while endogenous FGF21 levels are usually <30 ng/mL. Furthermore, FGF21 analogues often have a longer half-life and duration of action than the native protein [115], which may explain why their pharmacological effects are not always congruent with the physiological actions of endogenous FGF21.

Overexpression or administration of exogenous FGF21 has a profound effect in animal models of NAFLD. Multiple studies have demonstrated that supraphysiological doses of FGF21 can reverse steatosis, suppress hepatic and systemic inflammation, and limit fibrosis [32,92,96] through different pathways. FGF21 reduces hepatic expression of lipogenic genes and increases the expression of genes responsible for energy expenditure [73]. FGF21 also dramatically improves glucose metabolism by enhancing insulin sensitivity and secretion and promoting glucose uptake in skeletal muscle and BAT [116,117]. Administration of a recombinant FGF21 variant can protect against NASH by improving mitochondrial function, preventing oxidative stress and reducing chronic inflammation in obese mice [100]. FGF21 also has anti-fibrotic effects in the liver. LY2405319, a FGF21 analogue, reduces hepatic fibrosis in mice by reducing succinate signalling and α-SMA expression in HSCs [118]. In vitro, FGF21 inhibits HSC activation and promotes apoptosis of activated HSCs [119]. Although not yet assessed in an animal model of NAFLD, β-klotho/FGFR1-specific agonists may provide a novel method for mimicking FGF21 function while minimising off-target effects. Such a therapeutic approach has been shown to vastly improve metabolic parameters in non-human primates [120], and ameliorate diabetes and obesity in mice [121].

Despite the abundance of preclinical evidence supporting the use of FGF21 mimetics for the treatment of NAFLD, there is only one human

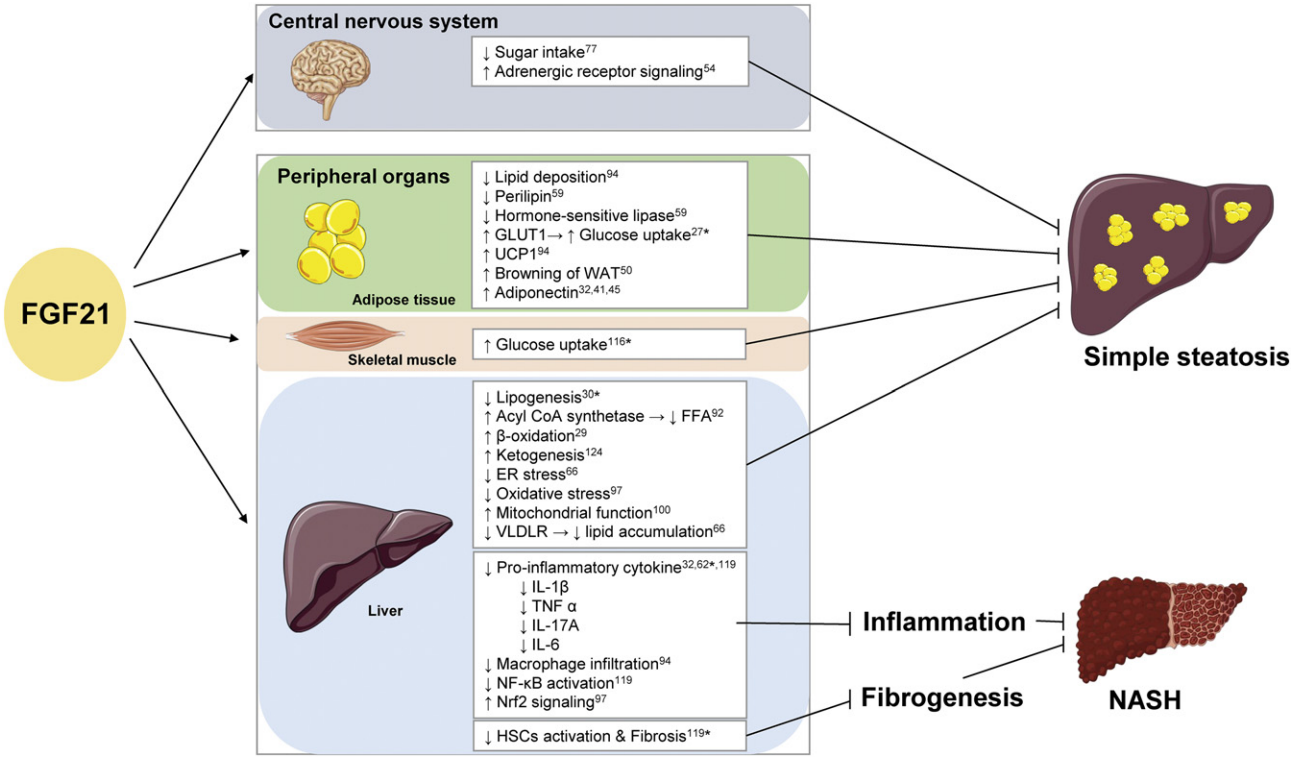

Fig. 2. Preclinical evidence of the therapeutic effects of FGF21 for the treatment of NAFLD. FGF21 can act in both peripheral organs and the central nervous system as a regulator of multiple processes to improve simple steatosis and NASH, such as lipogenesis, glucose uptake, β-oxidation, inflammation, and fibrosis [27,29,30,32,41,45,50,54,59,62,66,77,92,94,97,100,116,119,124]. Abbreviations: AT, adipose tissue; FGF21, fibroblast growth factor 21; GLUT1, glucose transporter 1; UCP1,uncoupling protein 1; WAT, white adipose tissue; FFA, free fatty acid; ER, endoplasmic reticulum; VLDLR, very-low-density lipoprotein receptor; IL-1β, interleukin-1β; TNFα, tumour necrosis factor α; IL-17A, interleukin-17A; IL-6, interleukin-6; HSC, hepatic stellate cell; NF-κB, nuclear factor κB; Nrf2, nuclear factor erythroid 2-related factor 2. *Studies performed in vitro.

trial that has tested this hypothesis. In this study, obese participants with biopsy-confirmed NASH were randomised to either placebo or once-daily subcutaneous pegbelfermin (a polyethylene glycol-conjugated recombinant analogue of human FGF21 with a prolonged half-life) for 16 weeks [48]. Overall, pegbelfermin was well tolerated and provided a clinically significant reduction in hepatic fat content. Additionally, pegbelfermin increased adiponectin levels, improved lipid profiles and reduced markers of hepatocellular damage [48]. Further Phase IIb trials are currently being conducted to corroborate these findings (NCT03486912 and NCT03486899). Pegbelfermin and LY2405319 have also been trialled in obese patients with type 2 diabetes mellitus that are at high risk of NAFLD [47,122,123]. In these studies, the FGF21 analogues improved metabolic parameters, such as the lipid profile and circulating insulin and adiponectin levels.

9. Conclusion

The incidence of NAFLD is rising rapidly. Currently, diagnostic pathways are suboptimal and there is a lack of effective management options. There have been recent improvements in the understanding of NAFLD pathogenesis, although a number of pathways remain ill-defined. Although there appears to be a distinct relationship between FGF21 and NAFLD, it remains to be determined whether FGF21 is a robust biomarker for NAFLD. FGF21 is a promising target for drug development, particularly in advanced NAFLD. We believe that optimisation of FGF21 mimetics will greatly reduce NASH-associated mortality and potentially provide a curative intervention for hepatic steatosis and inflammation.

Author Contributions

BT performed the literature search and drafted the manuscript. HL, XL, KAR and KLO provided critical intellectual contributions. All Authors have read and approved the final version of the manuscript.

Funding

KLO was supported by the Australian National Health and Medical Research Council (NHMRC) Career Development Fellowship (1122854). KLO received a Grant-in-Aid (G 12S 6681) and the NSW CVRN Research Development Project Grant (100715) both from the National Heart Foundation of Australia to investigate the role of FGF21 in atherosclerosis and different CVD risk factors. HL was supported by the General Program of National Natural Science Foundation of China (NSFC) (81870598), NSFC-NHMRC joint research grant (81561128016), and Municipal Human Resources Development Program for Outstanding Young Talents in Medical and Health Sciences in Shanghai (2017YQ009).

Declaration of Competing Interest

The authors declare no conflict of interest.

References

[1] Younossi ZM, Koenig AB, Abdelatif D, Fazel Y, Henry L, Wymer M. Global epidemiology of nonalcoholic fatty liver disease-meta-analytic assessment of prevalence, incidence, and outcomes. Hepatology (Baltimore, Md) 2016;64(1):73–84.
[2] Brunt EM, Wong VWS, Nobili V, Day CP, Sookoian S, Maher JJ, et al. Nonalcoholic fatty liver disease. Nat Rev Dis Primers 2015;1:15080.
[3] Chalasani N, Younossi Z, Lavine JE, Charlton M, Cusi K, Rinella M, et al. The diagnosis and management of nonalcoholic fatty liver disease: practice guidance from the American Association for the Study of Liver Diseases. Hepatology 2018;67(1):328–57.
[4] Fazel Y, Koenig AB, Sayiner M, Goodman ZD, Younossi ZM. Epidemiology and natural history of non-alcoholic fatty liver disease. Metabolism 2016;65(8):1017–25.
[5] Khan RS, Newsome PN. Non-alcoholic fatty liver disease and liver transplantation. Metabolism 2016;65(8):1208–23.
[6] Pais R, Barritt AS, Calmus Y, Scatton O, Runge T, Lebray P, et al. NAFLD and liver transplantation: current burden and expected challenges. J Hepatol 2016;65(6):1245–57.
[7] Minervini MI, Ruppert K, Fontes P, Volpes R, Vizzini G, de Vera ME, et al. Liver biopsy findings from healthy potential living liver donors: reasons for disqualification, silent diseases and correlation with liver injury tests. J Hepatol 2009;50(3):501–10.
[8] Adams LA, Anstee QM, Tilg H, Targher G. Non-alcoholic fatty liver disease and its relationship with cardiovascular disease and other extrahepatic diseases. Gut 2017;66(6):1138–53.
[9] Katsiki N, Mantzoros C. Fibroblast growth factor 21: A role in cardiometabolic disorders and cardiovascular risk prediction? Metabolism 2019;93:iii–v.
[10] Katsiki N, Mikhailidis DP, Mantzoros CS. Non-alcoholic fatty liver disease and dyslipidemia: an update. Metabolism 2016;65(8):1109–23.
[11] Lonardo A, Sookoian S, Pirola CJ, Targher G. Non-alcoholic fatty liver disease and risk of cardiovascular disease. Metabolism 2016;65(8):1136–50.
[12] Wu S, Wu F, Ding Y, Hou J, Bi J, Zhang Z. Association of non-alcoholic fatty liver disease with major adverse cardiovascular events: a systematic review and meta-analysis. Sci Rep 2016;6:33386.
[13] Dyson JK, Anstee QM, McPherson S. Non-alcoholic fatty liver disease: a practical approach to diagnosis and staging. Frontline Gastroenterol 2014;5(3):211–8.
[14] Eddowes PJ, Sasso M, Allison M, Tsochatzis E, Anstee QM, Sheridan D, et al. Accuracy of FibroScan controlled attenuation parameter and liver stiffness measurement in assessing steatosis and fibrosis in patients with nonalcoholic fatty liver disease. Gastroenterology 2019;156(6):1717–30.
[15] Buzzetti E, Pinzani M, Tsochatzis EA. The multiple-hit pathogenesis of non-alcoholic fatty liver disease (NAFLD). Metabolism 2016;65(8):1038–48.
[16] Hazlehurst JM, Woods C, Marjot T, Cobbold JF, Tomlinson JW. Non-alcoholic fatty liver disease and diabetes. Metabolism 2016;65(8):1096–108.
[17] Choi S, Diehl A. Hepatic triglyceride synthesis and nonalcoholic fatty liver disease. Curr Opin Lipidol 2008;19(3):295–300.
[18] Peverill W, Powell L, Skoien R. Evolving concepts in the pathogenesis of NASH: beyond steatosis and inflammation. Int J Mol Sci 2014;15(5):8591–638.
[19] Ibrahim SH, Kohli R, Gores GJ. Mechanisms of lipotoxicity in NAFLD and clinical implications. J Pediatr Gastroenterol Nutr 2011;53(2):131–40.
[20] Sutti S, Jindal A, Locatelli I, Vacchiano M, Gigliotti L, Bozzola C, et al. Adaptive immune responses triggered by oxidative stress contribute to hepatic inflammation in NASH. Hepatology 2014;59(3):886–97.
[21] Wu Z, Xu J, Tan J, Song Y, Liu L, Zhang F, et al. Mesenteric adipose tissue B lymphocytes promote local and hepatic inflammation in non-alcoholic fatty liver disease mice. J Cell Mol Med 2019;23(5):3375–85.
[22] Markan KR, Naber MC, Ameka MK, Anderegg MD, Mangelsdorf DJ, Kliewer SA, et al. Circulating FGF21 is liver derived and enhances glucose uptake during refeeding and overfeeding. Diabetes 2014;63(12):4057–63.
[23] Lundåsen T, Hunt MC, Nilsson L-M, Sanyal S, Angelin B, Alexson SEH, et al. PPARalpha is a key regulator of hepatic FGF21. Biochem Biophys Res Commun 2007;360(2):437–40.
[24] Cyphert HA, Alonge KM, Ippagunta SM, Hillgartner FB. Glucagon stimulates hepatic FGF21 secretion through a PKA-and EPAC-dependent posttranscriptional mechanism. PLoS One 2014;9(4):e94996.
[25] Sonoda J, Chen MZ, Baruch A. FGF21-receptor agonists: an emerging therapeutic class for obesity-related diseases. Horm Mol Biol Clin Invest 2017;30(2).
[26] Tomiyama K-i, Maeda R, Urakawa I, Yamazaki Y, Tanaka T, Ito S, et al. Relevant use of Klotho in FGF19 subfamily signaling system in vivo. Proc Natl Acad Sci U S A 2010;107(4):1666–71.
[27] Kharitonenkov A, Shiyanova TL, Koester A, Ford AM, Micanovic R, Galbreath EJ, et al. FGF-21 as a novel metabolic regulator. J Clin Invest 2005;115(6):1627–35.
[28] Badman MK, Koester A, Flier JS, Kharitonenkov A, Maratos-Flier E. Fibroblast growth factor 21-deficient mice demonstrate impaired adaptation to ketosis. Endocrinology 2009;150(11):4931–40.
[29] Potthoff MJ, Inagaki T, Satapati S, Ding X, He T, Goetz R, et al. FGF21 induces PGC-1alpha and regulates carbohydrate and fatty acid metabolism during the adaptive starvation response. Proc Natl Acad Sci U S A 2009;106(26):10853–8.
[30] Zhang Y, Lei T, Huang JF, Wang SB, Zhou LL, Yang ZQ, et al. The link between fibroblast growth factor 21 and sterol regulatory element binding protein 1c during lipogenesis in hepatocytes. Mol Cell Endocrinol 2011;342(1–2):41–7.
[31] De Sousa-Coelho AL, Relat J, Hondares E, Pérez-Martí A, Ribas F, Villarroya F, et al. FGF21 mediates the lipid metabolism response to amino acid starvation. J Lipid Res 2013;54(7):1786–97.
[32] Bao L, Yin J, Gao W, Wang Q, Yao W, Gao X. A long-acting FGF21 alleviates hepatic steatosis and inflammation in a mouse model of non-alcoholic steatohepatitis partly through an FGF21-adiponectin-IL17A pathway. Br J Pharmacol 2018;175(16):3379–93.
[33] Kim HW, Lee JE, Cha JJ, Hyun YY, Kim JE, Lee MH, et al. Fibroblast growth factor 21 improves insulin resistance and ameliorates renal injury in db/db mice. Endocrinology 2013;154(9):3366–76.
[34] Zhang Y, Xie Y, Berglund ED, Coate KC, He TT, Katafuchi T, et al. The starvation hormone, fibroblast growth factor-21, extends lifespan in mice. Elife 2012;1:e00065.
[35] Gong Q, Hu Z, Zhang F, Cui A, Chen X, Jiang H, et al. Fibroblast growth factor 21 improves hepatic insulin sensitivity by inhibiting mammalian target of rapamycin complex 1 in mice. Hepatology 2016;64(2):425–38.
[36] Li H, Wu G, Fang Q, Zhang M, Hui X, Sheng B, et al. Fibroblast growth factor 21 increases insulin sensitivity through specific expansion of subcutaneous fat. Nat Commun 2018;9(1):272.
[37] Pan Y, Wang B, Zheng J, Xiong R, Fan Z, Ye Y, et al. Pancreatic fibroblast growth factor 21 protects against type 2 diabetes in mice by promoting insulin expression and

secretion in a PI3K/Akt signaling-dependent manner. J Cell Mol Med 2019;23(2):1059–71.
[38] Singhal G, Fisher FM, Chee MJ, Tan TG, El Ouaamari A, Adams AC, et al. Fibroblast growth factor 21 (FGF21) protects against high fat diet induced inflammation and islet hyperplasia in pancreas. PLoS One 2016;11(2):e0148252.
[39] So WY, Cheng Q, Xu A, Lam KSL, Leung PS. Loss of fibroblast growth factor 21 action induces insulin resistance, pancreatic islet hyperplasia and dysfunction in mice. Cell Death Dis 2015;6(3):e1707.
[40] Wente W, Efanov AM, Brenner M, Kharitonenkov A, Köster A, Sandusky GE, et al. Fibroblast growth factor-21 improves pancreatic β-cell function and survival by activation of extracellular signal–regulated kinase 1/2 and Akt signaling pathways. Diabetes 2006;55(9):2470–8.
[41] Lin Z, Tian H, Lam KS, Lin S, Hoo Ruby CL, Konishi M, et al. Adiponectin mediates the metabolic effects of FGF21 on glucose homeostasis and insulin sensitivity in mice. Cell Metab 2013;17(5):779–89.
[42] Véniant MM, Hale C, Helmering J, Chen MM, Stanislaus S, Busby J, et al. FGF21 promotes metabolic homeostasis via white adipose and leptin in mice. PLoS One 2012;7(7):e40164.
[43] Asrih M, Veyrat-Durebex C, Poher A-L, Lyautey J, Rohner-Jeanrenaud F, Jornayvaz FR. Leptin as a potential regulator of FGF21. Cell Physiol Biochem 2016;38(3):1218–25.
[44] Müller TD, Sullivan LM, Habegger K, Yi C-X, Kabra D, Grant E, et al. Restoration of leptin responsiveness in diet-induced obese mice using an optimized leptin analog in combination with exendin-4 or FGF21. J Pept Sci 2012;18(6):383–93.
[45] Holland William L, Adams Andrew C, Brozinick Joseph T, Bui Hai H, Miyauchi Y, Kusminski Christine M, et al. An FGF21-adiponectin-ceramide axis controls energy expenditure and insulin action in mice. Cell Metab 2013;17(5):790–7.
[46] Charles ED, Neuschwander-Tetri BA, Frias JP, Kundu S, Luo Y, Tirucherai GS, et al. Pegbelfermin (BMS-986036), PEGylated FGF21, in patients with obesity and type 2 diabetes: results from a randomized phase 2 study. Obesity 2019;27(1):41–9.
[47] Gaich G, Chien JY, Fu H, Glass LC, Deeg MA, Holland WL, et al. The effects of LY2405319, an FGF21 analog, in obese human subjects with type 2 diabetes. Cell Metab 2013;18(3):333–40.
[48] Sanyal A, Charles ED, Neuschwander-Tetri BA, Loomba R, Harrison SA, Abdelmalek MF, et al. Pegbelfermin (BMS-986036), a PEGylated fibroblast growth factor 21 analogue, in patients with non-alcoholic steatohepatitis: a randomised, double-blind, placebo-controlled, phase 2a trial. Lancet 2018;392(10165):2705–17.
[49] Coskun T, Bina HA, Schneider MA, Dunbar JD, Hu CC, Chen Y, et al. Fibroblast growth factor 21 corrects obesity in mice. Endocrinology 2008;149(12):6018–27.
[50] Fisher FM, Kleiner S, Douris N, Fox EC, Mepani RJ, Verdeguer F, et al. FGF21 regulates PGC-1α and browning of white adipose tissues in adaptive thermogenesis. Genes Dev 2012;26(3):271–81.
[51] Bartelt A, Heeren J. Adipose tissue browning and metabolic health. Nat Rev Endocrinol 2014;10(1):24–36.
[52] Hsuchou H, Pan W, Kastin AJ. The fasting polypeptide FGF21 can enter brain from blood. Peptides 2007;28(12):2382–6.
[53] Sarruf DA, Thaler JP, Morton GJ, German J, Fischer JD, Ogimoto K, et al. Fibroblast growth factor 21 action in the brain increases energy expenditure and insulin sensitivity in obese rats. Diabetes 2010;59(7):1817–24.
[54] Douris N, Stevanovic DM, Fisher FM, Cisu TI, Chee MJ, Nguyen NL, et al. Central fibroblast growth factor 21 browns white fat via sympathetic action in male mice. Endocrinology 2015;156(7):2470–81.
[55] Bookout AL, de Groot MHM, Owen BM, Lee S, Gautron L, Lawrence HL, et al. FGF21 regulates metabolism and circadian behavior by acting on the nervous system. Nat Med 2013;19(9):1147–52.
[56] Tan BK, Hallschmid M, Adya R, Kern W, Lehnert H, Randeva HS. Fibroblast growth factor 21 (FGF21) in human cerebrospinal fluid. Diabetes 2011;60(11):2758–62.
[57] Liu J, Xu Y, Hu Y, Wang G. The role of fibroblast growth factor 21 in the pathogenesis of non-alcoholic fatty liver disease and implications for therapy. Metabolism 2015;64(3):380–90.
[58] He L, Deng L, Zhang Q, Guo J, Zhou J, Song W, et al. Diagnostic value of CK-18, FGF-21, and related biomarker panel in nonalcoholic fatty liver disease: a systematic review and meta-analysis. Biomed Res Int 2017(2017) 9729107.
[59] Fisher FM, Chui PC, Antonellis PJ, Bina HA, Kharitonenkov A, Flier JS, et al. Obesity is a fibroblast growth factor 21 (FGF21)-resistant state. Diabetes 2010;59(11):2781–9.
[60] Hale C, Chen MM, Stanislaus S, Chinookoswong N, Hager T, Wang M, et al. Lack of overt FGF21 resistance in two mouse models of obesity and insulin resistance. Endocrinology 2012;153(1):69–80.
[61] Rusli F, Deelen J, Andriyani E, Boekschoten MV, Lute C, van den Akker EB, et al. Fibroblast growth factor 21 reflects liver fat accumulation and dysregulation of signalling pathways in the liver of C57BL/6J mice. Sci Rep 2016;6:30484.
[62] Lee KJ, Jang YO, Cha S-K, Kim MY, Park K-S, Eom YW, et al. Expression of fibroblast growth factor 21 and β-Klotho regulates hepatic fibrosis through the nuclear factor-κB and c-Jun N-terminal kinase pathways. Gut and Liver 2018;12(4):449–56.
[63] Chakravarthy MV, Pan Z, Zhu Y, Tordjman K, Schneider JG, Coleman T, et al. "New" hepatic fat activates PPARα to maintain glucose, lipid, and cholesterol homeostasis. Cell Metab 2005;1(5):309–22.
[64] Utzschneider KM, Kahn SE. The role of insulin resistance in nonalcoholic fatty liver disease. J Clin Endocrinol Metab 2006;91(12):4753–61.
[65] Schlein C, Talukdar S, Heine M, Fischer Alexander W, Krott Lucia M, Nilsson Stefan K, et al. FGF21 lowers plasma triglycerides by accelerating lipoprotein catabolism in white and brown adipose tissues. Cell Metab 2016;23(3):441–53.
[66] Zarei M, Barroso E, Palomer X, Dai J, Rada P, Quesada-López T, et al. Hepatic regulation of VLDL receptor by PPARβ/δ and FGF21 modulates non-alcoholic fatty liver disease. Mol Metab 2018;8:117–31.
[67] Emanuelli B, Vienberg SG, Smyth G, Cheng C, Stanford KI, Arumugam M, et al. Interplay between FGF21 and insulin action in the liver regulates metabolism. J Clin Invest 2014;124(2):515–27.
[68] Czaja MJ. Function of autophagy in nonalcoholic fatty liver disease. Dig Dis Sci 2016;61(5):1304–13.
[69] Yang L, Li P, Fu S, Calay ES, Hotamisligil GS. Defective hepatic autophagy in obesity promotes ER stress and causes insulin resistance. Cell Metab 2010;11(6):467–78.
[70] Singh R, Kaushik S, Wang Y, Xiang Y, Novak I, Komatsu M, et al. Autophagy regulates lipid metabolism. Nature 2009;458(7242):1131–5.
[71] Settembre C, De Cegli R, Mansueto G, Saha PK, Vetrini F, Visvikis O, et al. TFEB controls cellular lipid metabolism through a starvation-induced autoregulatory loop. Nat Cell Biol 2013;15(6):647–58.
[72] Xiong X, Tao R, DePinho RA, Dong XC. The autophagy-related gene 14 (Atg14) is regulated by forkhead box O transcription factors and circadian rhythms and plays a critical role in hepatic autophagy and lipid metabolism. J Biol Chem 2012;287(46):39107–14.
[73] Zhu S, Wu Y, Ye X, Ma L, Qi J, Yu D, et al. FGF21 ameliorates nonalcoholic fatty liver disease by inducing autophagy. Mol Cell Biochem 2016;420(1–2):107–19.
[74] Cheng STW, Li SYT, Leung PS. Fibroblast growth factor 21 stimulates pancreatic islet autophagy via inhibition of AMPK-mTOR signaling. Int J Mol Sci 2019;20(10):2517.
[75] Jensen T, Abdelmalek MF, Sullivan S, Nadeau KJ, Green M, Roncal C, et al. Fructose and sugar: a major mediator of non-alcoholic fatty liver disease. J Hepatol 2018;68(5):1063–75.
[76] Dushay JR, Toschi E, Mitten EK, Fisher FM, Herman MA, Maratos-Flier E. Fructose ingestion acutely stimulates circulating FGF21 levels in humans. Mol Metab 2014;4(1):51–7.
[77] von Holstein-Rathlou S, BonDurant L, Peltekian L, Naber MC, Yin TC, Claflin KE, et al. FGF21 mediates endocrine control of simple sugar intake and sweet taste preference by the liver. Cell Metab 2016;23(2):335–43.
[78] Lundsgaard A-M, Fritzen AM, Sjøberg KA, Myrmel LS, Madsen L, Wojtaszewski JFP, et al. Circulating FGF21 in humans is potently induced by short term overfeeding of carbohydrates. Mol Metab 2017;6(1):22–9.
[79] Dushay J, Chui PC, Gopalakrishnan GS, Varela-Rey M, Crawley M, fM Fisher, et al. Increased fibroblast growth factor 21 in obesity and nonalcoholic fatty liver disease. Gastroenterology 2010;139(2):456–63.
[80] Li H, Fang Q, Gao F, Fan J, Zhou J, Wang X, et al. Fibroblast growth factor 21 levels are increased in nonalcoholic fatty liver disease patients and are correlated with hepatic triglyceride. J Hepatol 2010;53(5):934–40.
[81] Shen J, Chan HL-Y, Wong GL-H, Choi PC-L, Chan AW-H, Chan H-Y, et al. Non-invasive diagnosis of non-alcoholic steatohepatitis by combined serum biomarkers. J Hepatol 2012;56(6):1363–70.
[82] Yang M, Xu D, Liu Y, Guo X, Li W, Guo C, et al. Combined serum biomarkers in non-invasive diagnosis of non-alcoholic steatohepatitis. PLoS One 2015;10(6):e0131664.
[83] Yilmaz Y, Eren F, Yonal O, Kurt R, Aktas B, Celikel CA, et al. Increased serum FGF21 levels in patients with nonalcoholic fatty liver disease. Eur J Clin Invest 2010;40(10):887–92.
[84] Wu G, Li H, Fang Q, Zhang J, Zhang M, Zhang L, et al. Complementary role of fibroblast growth factor 21 and cytokeratin 18 in monitoring the different stages of non-alcoholic fatty liver disease. Sci Rep 2017;7(1):5095.
[85] Barb D, Bril F, Kalavalapalli S, Cusi K. Plasma fibroblast growth factor 21 is associated with severity of nonalcoholic steatohepatitis in patients with obesity and type 2 diabetes. J Clin Endocrinol Metab 2019;104(8):3327–36.
[86] Chen C, Cheung BMY, Tso AWK, Wang Y, Law LSC, Ong KL, et al. High plasma level of fibroblast growth factor 21 is an independent predictor of type 2 diabetes: a 5.4-year population-based prospective study in Chinese subjects. Diabetes Care 2011;34(9):2113–5.
[87] Lakhani I, Gong M, Wong WT, Bazoukis G, Lampropoulos K, Wong SH, et al. Fibroblast growth factor 21 in cardio-metabolic disorders: a systematic review and meta-analysis. Metabolism 2018;83:11–7.
[88] Li G, Yin J, Fu J, Li L, Grant SFA, Li C, et al. FGF21 deficiency is associated with childhood obesity, insulin resistance and hypoadiponectinaemia: the BCAMS study. Diabetes Metab 2017;43(3):253–60.
[89] Alisi A, Ceccarelli S, Panera N, Prono F, Petrini S, De Stefanis C, et al. Association between serum atypical fibroblast growth factors 21 and 19 and pediatric nonalcoholic fatty liver disease. PLoS One 2013;8(6):e67160.
[90] Reinehr T, Woelfle J, Wunsch R, Roth CL. Fibroblast growth factor 21 (FGF-21) and its relation to obesity, metabolic syndrome, and nonalcoholic fatty liver in children: a longitudinal analysis. J Clin Endocrinol Metab 2012;97(6):2143–50.
[91] Giannini C, Feldstein AE, Santoro N, Kim G, Kursawe R, Pierpont B, et al. Circulating levels of FGF-21 in obese youth: associations with liver fat content and markers of liver damage. J Clin Endocrinol Metab 2013;98(7):2993–3000.
[92] Fisher FM, Chui PC, Nasser IA, Popov Y, Cunniff JC, Lundasen T, et al. Fibroblast growth factor 21 limits lipotoxicity by promoting hepatic fatty acid activation in mice on methionine and choline-deficient diets. Gastroenterology 2014;147(5)(1073-83.e6).
[93] Liu X, Zhang P, Martin RC, Cui G, Wang G, Tan Y, et al. Lack of fibroblast growth factor 21 accelerates metabolic liver injury characterized by steatohepatities in mice. Am J Cancer Res 2016;6(5):1011–25.
[94] Jimenez V, Jambrina C, Casana E, Sacristan V, Muñoz S, Darriba S, et al. FGF21 gene therapy as treatment for obesity and insulin resistance. EMBO Mol Med 2018;10:e8791.
[95] Wang Q, Yuan J, Yu Z, Lin L, Jiang Y, Cao Z, et al. FGF21 attenuates high-fat diet-induced cognitive impairment via metabolic regulation and anti-inflammation of obese mice. Mol Neurobiol 2018;55(6):4702–17.

[96] Yin J, Bao L, Chen R, Gao W, Gao X, Yao W. Enhanced expression and distinctive characterization of a long-acting FGF21 and its potential to alleviate nonalcoholic steatohepatitis. Biochimie 2018;151:166–75.
[97] Yu Y, Bai F, Liu Y, Yang Y, Yuan Q, Zou D, et al. Fibroblast growth factor (FGF21) protects mouse liver against d-galactose-induced oxidative stress and apoptosis via activating Nrf2 and PI3K/Akt pathways. Mol Cell Biochem 2015;403(1):287–99.
[98] Polimeni L, Del Ben M, Baratta F, Perri L, Albanese F, Pastori D, et al. Oxidative stress: new insights on the association of non-alcoholic fatty liver disease and atherosclerosis. World J Hepatol 2015;7(10):1325–36.
[99] Chau MDL, Gao J, Yang Q, Wu Z, Gromada J. Fibroblast growth factor 21 regulates energy metabolism by activating the AMPK–SIRT1–PGC-1α pathway. Proc Natl Acad Sci 2010;107(28):12553–8.
[100] Lee JH, Kang YE, Chang JY, Park KC, Kim H-W, Kim JT, et al. An engineered FGF21 variant, LY2405319, can prevent non-alcoholic steatohepatitis by enhancing hepatic mitochondrial function. Am J Transl Res 2016;8(11):4750–63.
[101] Ye D, Wang Y, Li H, Jia W, Man K, Lo CM, et al. Fibroblast growth factor 21 protects against acetaminophen-induced hepatotoxicity by potentiating peroxisome proliferator-activated receptor coactivator protein-1α-mediated antioxidant capacity in mice. Hepatology 2014;60(3):977–89.
[102] Waluga M, Kukla M, Zorniak M, Kajor M, Liszka L, Dyaczynski M, et al. Fibroblast growth factor-21 and omentin-1 hepatic mRNA expression and serum levels in morbidly obese women with non-alcoholic fatty liver disease. J Physiol Pharmacol 2017;68(3):363–74.
[103] Desai BN, Singhal G, Watanabe M, Stevanovic D, Lundasen T, fM Fisher, et al. Fibroblast growth factor 21 (FGF21) is robustly induced by ethanol and has a protective role in ethanol associated liver injury. Mol Metab 2017;6(11):1395–406.
[104] Talukdar S, Owen BM, Song P, Hernandez G, Zhang Y, Zhou Y, et al. FGF21 regulates sweet and alcohol preference. Cell Metab 2016;23(2):344–9.
[105] Chen X, Ward SC, Cederbaum AI, Xiong H, Lu Y. Alcoholic fatty liver is enhanced in CYP2A5 knockout mice: the role of the PPARα-FGF21 axis. Toxicology 2017;379:12–21.
[106] Zhu S, Ma L, Wu Y, Ye X, Zhang T, Zhang Q, et al. FGF21 treatment ameliorates alcoholic fatty liver through activation of AMPK-SIRT1 pathway. Acta Biochim Biophys Sin 2014;46(12):1041–8.
[107] Kneeman JM, Misdraji J, Corey KE. Secondary causes of nonalcoholic fatty liver disease. Therap Adv Gastroenterol 2012;5(3):199–207.
[108] Leandro G, Mangia A, Hui J, Fabris P, Rubbia-Brandt L, Colloredo G, et al. Relationship between steatosis, inflammation, and fibrosis in chronic hepatitis C: a meta-analysis of individual patient data. Gastroenterology 2006;130(6):1636–42.
[109] Kukla M, Berdowska A, Stygar D, Gabriel A, Mazur W, Łogiewa-Bazger B, et al. Serum FGF21 and RBP4 levels in patients with chronic hepatitis C. Scand J Gastroenterol 2012;47(8–9):1037–47.
[110] Mak L-Y, Lee C-H, Cheung K-S, Wong DK-H, Liu F, Hui RW-H, et al. Association of adipokines with hepatic steatosis and fibrosis in chronic hepatitis B patients on long-term nucleoside analogue. Liver Int 2019;39(1):1217–25.
[111] Praktiknjo M, Djayadi N, Mohr R, Schierwagen R, Bischoff J, Dold L, et al. Fibroblast growth factor 21 is independently associated with severe hepatic steatosis in non-obese HIV-infected patients. Liver Int 2019;39(8):1514–20.
[112] Zhang Q, Li Y, Liang T, Lu X, Liu X, Zhang C, et al. Loss of FGF21 in diabetic mouse during hepatocellular carcinogenetic transformation. Am J Cancer Res 2015;5(5):1762–74.
[113] Singhal G, Kumar G, Chan S, Fisher FM, Ma Y, Vardeh HG, et al. Deficiency of fibroblast growth factor 21 (FGF21) promotes hepatocellular carcinoma (HCC) in mice on a long term obesogenic diet. Mol Metab 2018;13:56–66.
[114] Lombardi R, Onali S, Thorburn D, Davidson BR, Gurusamy KS, Tsochatzis E. Pharmacological interventions for non-alcohol related fatty liver disease (NAFLD). Cochrane Database Syst Rev 2017;3:CD011640.
[115] Kliewer SA, Mangelsdorf DJ. A dozen years of discovery: insights into the physiology and pharmacology of FGF21. Cell Metab 2019;29(2):246–53.
[116] Mashili FL, Austin RL, Deshmukh AS, Fritz T, Caidahl K, Bergdahl K, et al. Direct effects of FGF21 on glucose uptake in human skeletal muscle: implications for type 2 diabetes and obesity. Diabetes Metab Res Rev 2011;27(3):286–97.
[117] Mottillo EP, Desjardins EM, Fritzen AM, Zou VZ, Crane JD, Yabut JM, et al. FGF21 does not require adipocyte AMP-activated protein kinase (AMPK) or the phosphorylation of acetyl-CoA carboxylase (ACC) to mediate improvements in whole-body glucose homeostasis. Mol Metab 2017;6(6):471–81.
[118] Le CT, Nguyen G, Park SY, Choi DH, Cho E-H. LY2405319, an analog of fibroblast growth factor 21 ameliorates α-smooth muscle actin production through inhibition of the succinate—G-protein couple receptor 91 (GPR91) pathway in mice. PLoS One 2018;13(2):e0192146.
[119] Xu P, Zhang Y, Liu Y, Yuan Q, Song L, Liu M, et al. Fibroblast growth factor 21 attenuates hepatic fibrogenesis through TGF-β/smad2/3 and NF-κB signaling pathways. Toxicol Appl Pharmacol 2016;290:43–53.
[120] Smith R, Duguay A, Bakker A, Li P, Weiszmann J, Thomas MR, et al. FGF21 can be mimicked in vitro and in vivo by a novel anti-FGFR1c/β-Klotho bispecific protein. PLoS One 2013;8(4):e61432.
[121] Foltz IN, Hu S, King C, Wu X, Yang C, Wang W, et al. Treating diabetes and obesity with an FGF21-mimetic antibody activating the βKlotho/FGFR1c receptor complex. Sci Transl Med. 2012;4(162):(162ra53).
[122] Charles ED, Neuschwander-Tetri BA, Pablo Frias J, Kundu S, Luo Y, Tirucherai GS, et al. Pegbelfermin (BMS-986036), PEGylated FGF21, in patients with obesity and type 2 diabetes: results from a randomized phase 2 study. Obesity (Silver Spring) 2019;27(1):41–9.
[123] Kim AM, Somayaji VR, Dong JQ, Rolph TP, Weng Y, Chabot JR, et al. Once-weekly administration of a long-acting fibroblast growth factor 21 analogue modulates lipids, bone turnover markers, blood pressure and body weight differently in obese people with hypertriglyceridaemia and in non-human primates. Diabetes Obes Metab 2017;19(12):1762–72.
[124] Inagaki T, Dutchak P, Zhao G, Ding X, Gautron L, Parameswara V, et al. Endocrine regulation of the fasting response by PPARalpha-mediated induction of fibroblast growth factor 21. Cell Metab 2007;5(6):415–25.
[125] Li X, Fan X, Ren F, Zhang Y, Shen C, Ren G, et al. Serum FGF21 levels are increased in newly diagnosed type 2 diabetes with nonalcoholic fatty liver disease and associated with hsCRP levels independently. Diabetes Res Clin Pract 2011;93(1):10–6.
[126] Yan H, Xia M, Chang X, Xu Q, Bian H, Zeng M, et al. Circulating fibroblast growth factor 21 levels are closely associated with hepatic fat content: a cross-sectional study. PLoS One 2011;6(9):e24895.