Review Article

Molecular Crosstalk Between Vitamin D and Non-alcoholic Fatty Liver Disease

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Abstract

Non-alcoholic fatty liver disease (NAFLD) is one of the leading causes of chronic liver disease. The worldwide increasing prevalence of NAFLD has become a cause of concern for clinicians. Furthermore, the molecular mechanism of NAFLD pathogenesis remains poorly understood. Moreover, therapeutic interventions are presently limited. Balanced diet, physical exercise and lifestyle modifications have been recommended. Several studies have revealed that vitamin D deficiency is correlated with NAFLD, and its supplementation may play a vital role in this regard. Sufficient information was obtained from full articles written in the English language, and accessible in PubMed, Google Scholar, Web of Science, and Scopus. The increasing prevalence of vitamin D deficiency remains as a global health risk factor, and this is linked to NAFLD pathogenesis. *In vitro* and *in vivo* studies, and clinical trials have revealed the beneficial role of vitamin D supplementation to control NAFLD. Vitamin D potentially regulates the molecular pathways associated with NAFLD risk factors, such as obesity, insulin resistance, and diabetes. It acts on adipocytes to control free fatty acid (FFA) trafficking, lipogenesis, and inflammation. Similarly, vitamin D acts on hepatocytes to reduce *de novo* lipogenesis and cellular FFA trafficking. Furthermore, it acts on pancreatic β-cells to improve insulin secretion, cell survival, and cellular functions. Vitamin D supplementation improves glucose uptake and insulin sensitivity. In addition, it decreases inflammation and liver injury, and acts on mitochondria to control reactive oxygen species (ROS)-mediated cellular toxicity. Vitamin D deficiency is a major risk factor for NAFLD pathogenesis. Thus, there is an urgent need to conduct molecular level analysis for further discernment.

Introduction

Non-alcoholic fatty liver disease (NAFLD) is one of the emerging

causes of chronic liver disease. Furthermore, this has become a cause of concern for clinicians due to the increasing burden of this disease. The overall global prevalence rate of NAFLD is 32.4%, with a prevalence rate of 39.7% for males and 25.6% for females.[1] The worldwide prevalence of this disease is presently increasing. The common clinicopathological conditions of NAFLD are abnormal accumulation of lipids in the cytoplasm of hepatocytes, and the persistence of abnormal levels of liver enzymes.[2] The histopathological determinants are crucial for characterizing the disease progression.[3] The advanced subtype of NAFLD is characterized as non-alcoholic steatohepatitis (NASH), which can further lead to cirrhosis of the liver. The increasing risk of NAFLD and NASH is associated with the development of hepatocellular carcinoma (HCC). However, NAFLD-associated HCC may be developed with or without cirrhosis.[4] Recently, NAFLD has become more precisely termed as, metabolic dysfunction-associated fatty liver disease (MAFLD). The proposed criteria for MAFLD comprise of the evidence of hepatic steatosis, combined with the presence of overweight/obesity, or metabolic abnormalities or type-2 diabetes mellitus. In this context, MAFLD shows high risk of disease progression, when compared to NAFLD.[5,6] The treatment options for NAFLD remain limited.[7] Dietary restrictions, physical exercise,

lifestyle modifications, and treatment of the metabolic syndrome are recommended to improve the hepatic fibrosis.[8–10] Multiple trials on combination therapies and drugs are presently in the pre-clinical and clinical stages.[11]

The underlying mechanisms of NAFLD is poorly understood. Extensive research is being undertaken to elucidate the close association of NAFLD with metabolic syndrome, insulin resistance, and obesity. Previous studies have revealed that the genetic variation in the rs738409 G allele of the *PNPLA3* (Patatin-like phospholipase domain-containing protein 3) gene that encodes for adiponutrin is associated with NAFLD susceptibility and disease pathogenesis, through increasing liver fat content.[12] *PNPLA3*, which is present in hepatocytes and stellate cells, play major roles in hydrolyzing triglyceride (TG) and polyunsaturated fatty acid transportation to phosphocholine. NAFLD predisposes to T2DM through the elevation of insulin secretion and β-cell failure.[10] In this context, the hepatic diacylglycerol content in the cytoplasm is a good predictor of insulin resistance through the activation of protein kinase C epsilon. Furthermore, metabolic syndrome-associated parameters glucose and triglyceride are overproduced by fatty liver in NAFLD.[13] In addition, NAFLD with metabolic syndrome has been reported in a study population, irrespective of insulin resistance and central obesity.[10,14]

In connection with these metabolic abnormalities, it is important to note that the long-term effect of malnutrition plays a significant role in NAFLD pathogenesis, which induces liver steatosis and insulin resistance.[15] In addition, micronutrients, such as vitamin deficiency, can also be correlated with NAFLD pathogenesis.[16] Recently, vitamin D has received tremendous attention, not only as a part of the proposed supplementation therapy, but also as a key player to regulate molecular interactions and cellular functions in NAFLD.[17–19] The present review focused on vitamin D deficiency and the molecular consequences in the pathogenesis of NAFLD. Additionally, the present study confers the plausible effect of vitamin D on molecular interactions and cellular functions, in order to ameliorate NAFLD pathogenesis and liver dysfunctions.

Vitamin D deficiency and prevalence of NAFLD

Vitamin D is a unique micronutrient, which can be synthesized by the epidermis upon sunlight (290–315 nm) exposure.[20] Vitamin D can be identified into two dominant forms: vitamin D_2 (ergocalciferol) and vitamin D_3 (cholecalciferol). The intestines can more efficiently absorb vitamin D_3, when compared to vitamin D_2.[21] In wild mushrooms, ergosterol is converted to vitamin D_2 in the presence of sunlight. On the other hand, the skin can synthesize vitamin D_3 upon natural or artificial ultraviolet B light irradiation.[22] Food supplements, such as milk, egg yolk, fish and meat (offal, such as liver), are good sources of vitamin D_3, while the plant source of vitamin D_2 is mushroom.[23] The vitamin D_3 obtained from different sources is incorporated into chylomicrons, and subsequently delivered in the lymphatic and venous circulation.[24] Vitamin D, in the form of D_2 or D_3, requires the successive hydroxylation process to produce its active form in the body. Vitamin D produces its biologically active intermediate, 25-hydroxyvitamin D or 25(OH)D, in the liver through 25-hydroxylase, and 1,25-dihydroxyvitamin D (calcitriol), in the kidney through 1α-hydroxylase. 25(OH)D is an active hormone, and this has endocrine functions, such as calcium absorption, maintenance of bone integrity, the regulation of cell growth, etc.[25,26] Serum 25(OH)D is a good indicator for measuring the vitamin D level. A serum or plasma 25(OH)D level of <75 nmol/L (<30 ng/ml) is considered as vitamin D deficiency.[27,28] In

this context, various studies have reported the serum/plasma vitamin D concentration of <20 ng/ml as vitamin D deficiency.[29,30] However, the supplementation dose of vitamin D is presently a subject of debate.[31]

Vitamin D deficiency is intensely and inversely associated with the worldwide prevalence of NAFLD. A population-based observational study conducted from 1990 to 2017 revealed a steep increase in worldwide NAFLD cases (882.1 million).[32] The prevalence rate of NAFLD is greater in East Asia (12.6%), Southeast Asia (14.5%), the Middle East, and North Africa (19.3%). Other systematic review and meta-analysis studies have also published these results.[33,34] On the other hand, the global prevalence of vitamin D deficiency has been reported to be high in Asia, Africa and Europe.[35,36] It has been reported that vitamin D is remarkably deficient in South Asian adults, with a prevalence rate of 68%.[37] In addition, the younger generation was identified to be more prevalent to have vitamin D deficiency, when compared to the elderly population.[38,39] Latitude, seasons, skin pigmentation, the use of sun protection, lifestyle and nutritional variation, and lack of sufficient exposure of skin to sunlight can be the potential reasons for vitamin D deficiency. It is important to note that adolescents and young adults have high risks of developing NAFLD due to obesity, T2DM, smoking, lifestyle, and ethnic variations.[40,41] The studies related to the low serum or plasma vitamin D status in NAFLD patients are presented in Table 1.[32,42–66]

Conceptualization of the mechanisms of NAFLD pathogenesis

The mechanisms underlying NAFLD pathogenesis are quite complicated and multifactorial. Previously, NAFLD pathogenesis was described by the "two hits" hypothesis.[67,68] The first hit explains the accumulation of triglyceride in hepatocytes (i.e. hepatic steatosis) due to insulin resistance, metabolic abnormalities, obesity, and sedentary lifestyle. The second hit describes the development of hepatic fibrosis and liver injury from steatosis. Several triggering factors and cellular conditions have been reported to be involved in the development of fibrosis and liver injury, which include lipid peroxidation, inflammation, drugs, mitochondrial dysfunction, and adipokines (Fig. 1). Recently, NAFLD pathogenesis was explained by the multiple-hit hypothesis.[69] According to this hypothesis, the progression of NAFLD-associated liver toxicity and inflammation is accomplished through multiple mechanisms, which include insulin resistance, nutritional factors, gut microbiota, genetic and epigenetic alterations, adipocyte-specific secretion of steroid hormones, cytokine prostaglandins, fatty acids, and cholesterol.[70] In this context, insulin resistance promotes hepatic lipogenesis, and impairs adipose tissue lipolysis. This generates the pull of free fatty acid (FFA) flux towards the liver, and causes adipose tissue dysfunction with altered adepokine and cytokine secretion.[71] It is noteworthy that 90–95% of patients with NAFLD are associated with simple steatosis, while 5–10% of NAFLD patients arise from NASH, which further lead to cirrhosis and HCC.[69]

Role of vitamin D to control NAFLD risk factors

Metabolic disorders

Role of vitamin D in obesity

According to the Centers for Disease Control and Prevention, a body mass index (BMI) of 25 to <30 is defined as overweight, while a BMI of ≥30 indicates obesity.[72] Excess triglyceride storage

Table 1. Low vitamin D level in NAFLD patients

| Author(s)	Year	Country	Sample size (N) for NAFLD*		Age (mean ± SD or, average or, range) (years)		NAFLD evaluation criteria	Methods of vitamin D estimation in plasma/serum	Level of 25(OH)D (mean ± SD or, average) (ng/mL)		
			Male	Female	Male	Female			Male	Female	Total
Bennouar et al.[42]	2021	Algeria	112	205	51.2 ± 12.6		Fatty Liver Index >60 points	Sequential competitive immunofluoro-assay method by VIDAS®	13.6 ± 7 µg/l	9.4 ± 6.8	10.9 ± 7.2 µg/l
Wang et al.[43]	2022	China	330		40.35 ± 8.14		Guidelines for the prevention and treatment of non-alcoholic fatty liver disease: a 2018 update (https://pubmed.ncbi.nlm.nih.gov/29804393/)	ELISA	Both		≥20 ng/mL
Xing et al.[44]	2022	China	-	234	56.98 ± 13.45		Ultrasonography (hepatorenal echo contrast, liver brightness, deep attenuation, and vascular blurring)	Electrochemiluminescence Immunoassays	Female		15.89 mmol/l
Hamzehzadeh Alamdari et al.[45]	2022	Iran	66		49.98 ± 10.55		Ultrasonography findings	-	Both		8.74 ± 2.47 ng/dl
Gad et al.[32]	2020	Egypt	40		47 ± 9		Ultrasonography findings (bright hepatic texture)	ELISA	Both		16.13 ± 10.23 (ng/mL)
Wang et al.[46]	2021	China	Lean: 163; Obese: 690		Lean: 54.84; Obese: 54.23		Ultrasonography findings as per the standards of the Chinese Liver Disease Association	-	Male		Lean: 60.95 nmol/L; Obese: 59.03 nmol/L
Cordeiro et al.[47]	2017	Brazil	7	43	45.7 ± 6.6	41.3 ± 8.6	Liver Biopsy	HPLC	17.9 ± 9.8 ng/mL	22.9 ± 7.2 ng/mL	-
Sayed et al.[48]	2021	Egypt	50		<60		Ultrasonography findings based on liver echogenicity	ELISA	Both		26.34 ± 11.28 ng/ml

(continued)

Table 1. (continued)

Author(s)	Year	Country	Sample size (N) for NAFLD*		Age (mean ± SD or, average or, range) (years)		NAFLD evaluation criteria	Methods of vitamin D estimation in plasma/serum	Level of 25(OH)D (mean ± SD or, average) (ng /mL)		
			Male	Female	Male	Female			Male	Female	Total
Kasapoglu et al.[49]	2013	Turkey	Stage 1: 133; Stage 2: 106; Stage 3: 99		Stage 1: 51.2 ± 10.5; Stage 2: 53.1 ± 9.7; Stage 3: 56.5 ± 8.9		Ultrasonography findings based on liver steatosis scores	-	Both		Stage 1: 20.0 ± 9.2; Stage 2: 13.3 ± 6.7; Stage 3: 8.8 ± 7.4
Ehrampoush et al.[50]	2019	Iran	745		49 ± 8		Fatty liver index >60	ELISA	Both		15.84 ± 5.50 nmol/L
Ahmed et al.[51]	2016	Egypt	47		11.13 ± 2.7		Ultrasonography findings	ELISA	Both		52.1 ± 41.3 nmol/L
Dasarathy et al.[52]	2017	USA	NAFLD with NASH: 26; NAFLD with HS: 16		NAFLD with NASH: 51.6 ± 13.0; NAFLD with HS: 51.6 6 11.4		Liver biopsy findings as per the NASH CRN scoring system	Direct Competitive Chemiluminescent Assay	Both		NAFLD with NASH: 19.3 ± 5.4 ng/mL; NAFLD with HS: 21.5 ± 4.5 ng/mL
Rageh et al.[53]	2021	Egypt	85		36.1 ± 9.2		Ultrasonography findings	Quantitative Chemiluminescent Microparticle Immunoassay	Both		16.3 ± 7.7
Cai et al.[54]	2020	China	Mild: 191; Moderate: 128; Severe: 66		43.57 ± 5.75		As per NASH Clinical Research Network Pathology Society	-	Both		Mild: 17.23 ± 2.61 nmol/l; Moderate: 15.47 ± 2.38 nmol/l; Severe: 13.83 ± 2.26 nmol/l
Kuçukazman[55]	2014	Turkey	154		46.3 ± 10.7		Ultrasonography findings	Competitive radioimmunoassay	Both		12.3 ± 8.9 ng/dl
Kim et al.[56]	2017	USA	Mild: 1,491; Moderate: 1,707; Severe: 817		Mild: 24.7 ± 0.42; Moderate: 23.7 ± 0.37; Severe: 23.6 ± 0.60		Ultrasonography findings	chemiluminescent immunoassay	Mild: 25.7 ± 0.45 mg/dl; Moderate: 25.0 ± 0.50 mg/dl; Severe: 25.6 ± 0.74 mg/dl	Mild: 23.9 ± 0.50 mg/dl; Moderate: 22.3 ± 0.49 mg/dl; Severe: 20.9 ± 0.68 mg/dl	Mild: 24.7 ± 0.42 mg/dl; Moderate: 23.7 ± 0.37 mg/dl; Severe: 23.6 ± 0.60 mg/dl
Shawky et al.[57]	2018	Egypt	50		49.88 ± 9.126		Ultrasonography findings	ELISA	Both		18.76 ± 14.37 ng/dl

(continued)

Table 1. (continued)

Author(s)	Year	Country	Sample size (N) for NAFLD*		Age (mean ± SD or, average or, range) (years)		NAFLD evaluation criteria	Methods of vitamin D estimation in plasma/serum	Level of 25(OH)D (mean ± SD or, average) (ng/mL)		
			Male	Female	Male	Female			Male	Female	Total
Chakraborty et al.[58]	2019	India	94		18 to 40		Ultrasonography findings	Chemiluminescent assay	Both		17.21 ± 6.34 ng/ml
Hao et al.[59]	2018	China	242		79.98 ± 8.33		Ultrasonography findings	high-performance liquid chromatography/tandem mass spectrometry	Both		11.55 ± 7.66 ng/ml
Mohamed et al.[60]	2017	Saudi Arabia	56		34.6 ± 10.6		Ultrasonography findings	Immunoassay (Abbott Architect i1000 Chemiflex device)	Male		17.45 ± 6.14
Chung et al.[61]	2016	Korea	1,660		53.6 ± 9.5		Ultrasonography findings	Chemiluminescnece immunoassay	Both		21.75 ± 7.54 ng/ml
Alamdari et al.[62]	2022	Iran	33		51.30 ± 10.78		Ultrasonography findings	-	Both		8.19 ± 2.60 ng/dl
Gungor et al.[63]	2020	Turkey	33		1 to 17		Ultrasonography findings		Both		Non-obese NAFLD: 20.83 ± 10.44; Obese NAFLD: 16.69 ± 8.22
Sah et al.[64]	2021	Nepal	70		44.3 ± 12.1		Ultrasonography findings	-	Both		Mild: 22.61 ± 28.07; Moderate: 24.89 ± 26.45; Severe: 17.4 ± 6.4
Wang et al.[65]	2018	China	9,182		Quartile 1: 54.3 ± 13.2; Quartile 2: 54.2 ± 13.2; Quartile 3: 54.6 ± 12.9; Quartile 4: 54.8 ± 12.8		Ultrasonography findings	Chemiluminescence assay	Both		Genetic risk score- Quartile 1 (low) to Quartile 4 (high):; Quartile 1: 41.8 ± 12.9 nmol/L; Quartile 2: 40.4 ± 12.3 nmol/L; Quartile 3: 39.6 ± 12.5 nmol/L; Quartile 4: 38.7 ± 11.9 nmol/L
Lorvand Amiri et al.[66]	2016	Iran	40		39.8 ± 11		Ultrasonography findings	ELISA	Both		9.9 ± 3.9 ng/mL

*The non-NAFLD and healthy controls of the respective studies were excluded. HPLC, High-Performance Liquid Chromatography; NAFLD, non-alcoholic fatty liver disease; SD, standard deviation.
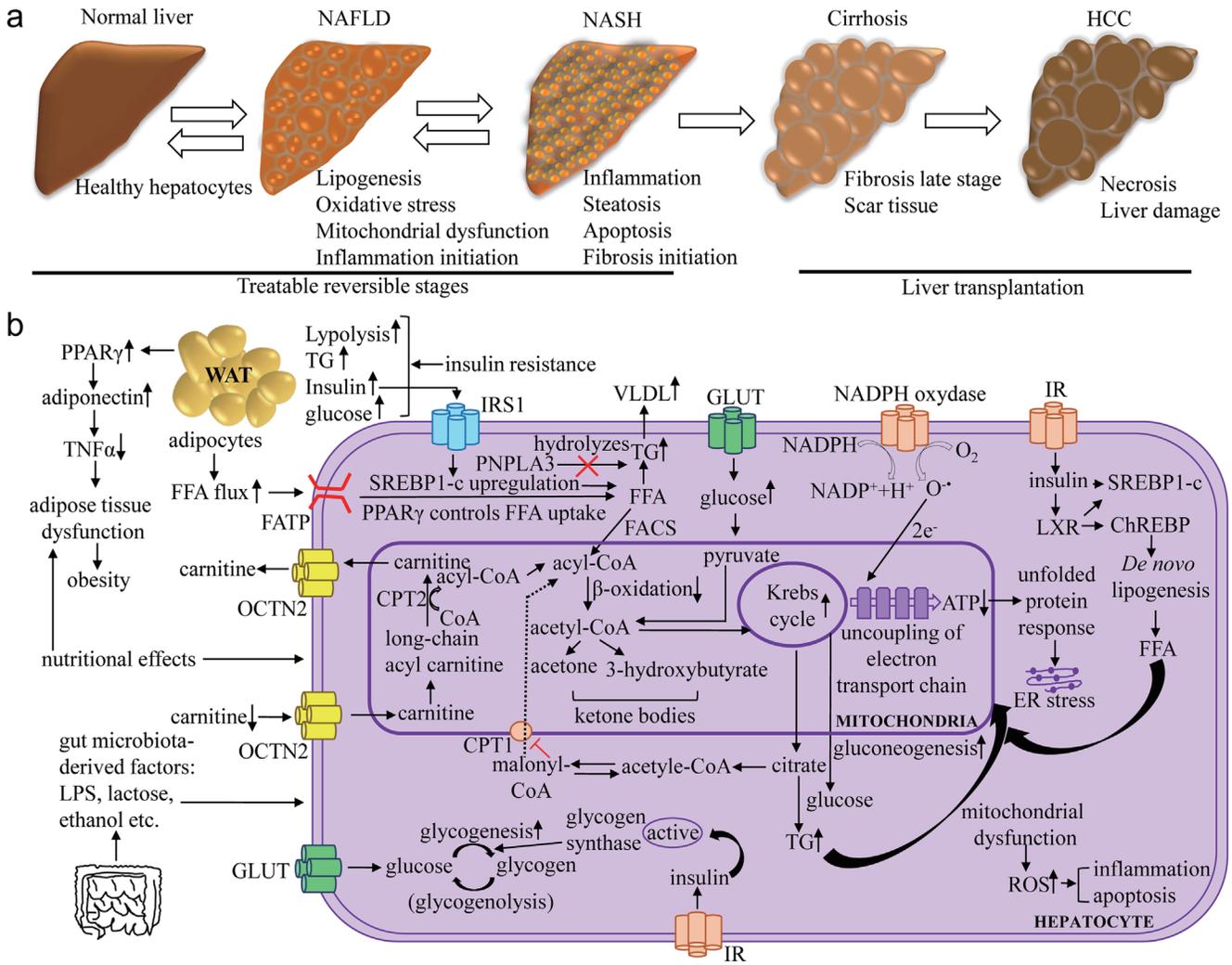
Fig. 1. Different stages of pathophysiological conditions, starting from NAFLD to HCC, can be characterized based on the biological and molecular aspect (Panel A). The molecular understanding of multifactorial NAFLD pathogenesis (panel B). The progression of NAFLD pathogenesis is multifactorial, in which several molecular pathways are involved. The high FFA flux generated by WATs resulted in the functional modulation of hepatocytes by reducing the β-oxidation, and increasing the Krebs cycle and gluconeogenesis. It causes low ATP production, mitochondrial dysfunction, and ER stress. In this context, the high TG and VLDL signature the pathophysiological state of the disease. Insulin stress mediates the increase in glycogenesis and *de novo* lipogenesis inside the hepatocyte. The genetic variation of PNPLA3 effects the hepatic TG hydrolyzation. The less biosynthesis of carnitine resulted in the low transportation of fatty acids to the mitochondria for β-oxidation. The high insulin level promoted the SREBP1-c upregulation, and increased the fatty acids in hepatocytes. As the uncoupling of the electron transport chain took place, the superoxide produced by NADPH oxidase induced liver injury. In addition, gut commensal-released products, such as LPS, lactose and ethanol, contributed to the NAFLD pathogenesis. The symbols "↑", "↓", and "⊥" refer to "high", "low", and "inhibition", respectively. ATP, Adenosine triphosphate; ChREBP, carbohydrate response element binding protein; CPT2, carnitine palmitoyltransferase 2; e⁻, electron; ER, endoplasmic reticulum; FATP, fatty acid-transport protein 1; FFA, free fatty acids; GLUT, glucose transporter; HCC, hepatocellular carcinoma; IR, insulin receptor; IRS, insulin receptor substrate; IRS1, Insulin receptor substrate 1; LPS, lipopolysaccharide; LXR, liver X receptor; NADP, nicotinamide adenine dinucleotide phosphate; NAFLD, non-alcoholic fatty liver disease; NASH, non-alcoholic steatohepatitis; PNPLA3, patatin-like phospholipase domain-containing protein 3; OCTN2, organic cation transporter; PPARγ, peroxisome proliferator- activated receptor gamma; ROS, reactive oxygen species; SREBP1c, sterol regulatory element binding protein 1; TG, triglyceride; TNFα, tumor necrosis factor alpha; WAT, white adipose tissue; VLDL, very low-density lipoprotein.

in white adipose tissues develops into obesity, which subsequently causes fat accumulation in the liver, and FFA release through adipokine secretions.[73,74] Moreover, excessive fat accumulation results in adipocyte hypertrophy, inflammation, insulin resistance, and fibrosis (Fig. 2). These are the key mechanisms for the development of NAFLD.[75] On the other hand, brown adipose tissue or the "browning" of white adipose tissue through adequate fat oxidation is crucial for energy dispersion.[76] Genes that encode peroxisome proliferator-activated receptor gamma (PPARγ), enhanc-

er-binding proteins, sterol regulatory element binding protein 1 (SREBP1), acetyl CoA carboxylase, fatty acid synthase (FAS), and fatty acid binding protein are involved in the regulation of adipogenesis.[74] The activation of the renin-angiotensin system (RAS) is potentially involved in obesity.[77] Thus, it is pivotal to mention that the activation of AMP-activated protein kinase (AMPK) attenuates the pathways and transcription factors related to adipogenesis.[74]

Vitamin D is capable of reducing obesity by regulating the expression of lipogenic genes. It was reported that diabetic mice
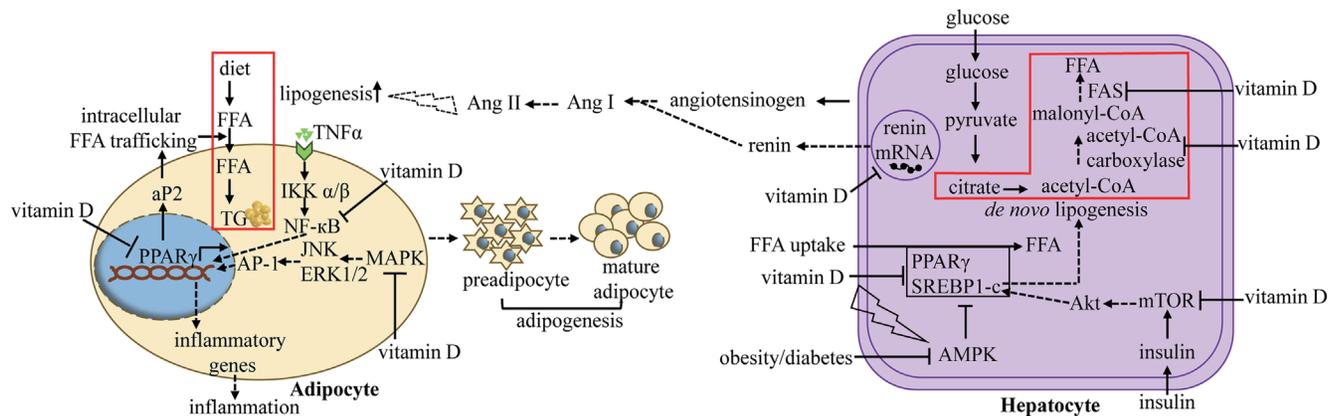
Fig. 2. Vitamin D-induced modulation of molecular pathways linked to obesity. One of the key functions of vitamin D is to restrict aP2-mediated FFA trafficking by inhibiting PPARγ in adipocytes. Vitamin D consequently blocks NFκB and MAPK to reduce PPARγ. The effect of vitamin D on RAS contributes to the reduction in lipogenesis through the inhibition of the renin expression. Furthermore, vitamin D plays a pivotal role in restricting adipocyte differentiation. On the other hand, vitamin D inhibits PPARγ and SREBP1c, which may lead to the control of the *de novo* lipogenesis in hepatocytes. The expression of SREBP1c is decreased by the vitamin D-mediated inhibition of the Akt/mTOR signaling pathway. The effect of vitamin D may directly control the *de novo* lipogenesis by inhibiting two major enzymes (acetyl-CoA carboxylase and FAS), ultimately leading to FFA synthesis. The "↑" and "⊥" refer to "high" and "inhibition", respectively. Akt, serine/threonine-protein kinase; AMPK, AMP-activated protein kinase; Ang I, Angiotensin I; Ang II, Angiotensin II; aP1, activator protein 1; aP2, activator protein 2; ERK1/2, extracellular signal-regulated protein kinase 1/2; FAS, fatty acid synthase; FFA, free fatty acid; IKK α/β, inhibitory-κB Kinase alpha/beta; JNK, c-Jun N-terminal kinases; MAPK, Mitogen-activated protein kinase; mTOR, mammalian target of rapamycin; NFκB, nuclear factor kappa B; PPARγ, Peroxisome proliferator-activated receptor gamma; RAS, renin-angiotensin system; TG, triglyceride.

supplemented with 300 ng/kg of vitamin D presented with a significantly decreased expression of FAS, SREBP1c and PPARγ. In addition, vitamin D supplementation activates AMPK, and blocks the threonine protein kinase B/mammalian target of rapamycin (Akt/mTOR) signaling pathway, resulting hepatic lipid reduction.[78] These study observations are crucial for establishing the beneficial effect of vitamin D supplementation in NAFLD under diabetic conditions. In addition, vitamin D can successfully inhibit nuclear factor-kappa B (NFκB) and mitogen-activated protein kinases, which in turn reduce adipose tissue inflammation.[79–81] It was reported that overweight patients (BMI: 28.3 ± 3.4 kg/m^2) with vitamin D deficiency (18.5 ± 6.4 ng/ml) and hypertension had enhanced RAS activity.[82] RAS contributes to the development of obesity and NAFLD progression, while a RAS inhibitor may be used to stop the development of NAFLD.[83] Vitamin D supplementation (15,000 IU/day for 30 days) can decrease RAS activity.[84] In addition, vitamin D has been shown to be associated with the decrease in plasma angiotensin II and renin mRNA expression levels, which may be correlated with the decrease in lipogenesis and TG accumulation in adipocytes.[85] However, the molecular understanding of the vitamin D-mediated depletion of RAS activity remains poorly understood.

Role of vitamin D in insulin resistance

Insulin resistance is one of the key mechanisms of NAFLD pathogenesis. In NAFLD, both hepatic and adipose tissues present with insulin resistance and decreased insulin sensitivity.[86–88] The liver exhibits inadequate fatty acid oxidation and glucose uptake during NAFLD, and these processes are controlled by adiponectin. Insulin resistance results in the flux of FFA to the liver. In addition, hyperinsulinemia subsequently effects the downstream anabolic process. Vitamin D deficiency is associated with insulin resistance.[89] A recent *in vivo* study reveals that the liver-specific deletion of vitamin D receptor (VDR) and hepatocyte nuclear factor-4 alpha (HNF4α) genes promote insulin resistance, and present histological observations that are similar to human NASH. This study has concluded that vitamin D can downregulate insulin resistance in NAFLD by

interacting with HNF4α, and activating VDR.[19] Vitamin D plays a pivotal role in insulin sensibilization through an anti-inflammatory mechanism, and effects insulin secretion.[90] It was evidenced from various randomized clinical trials that vitamin D supplementation from a minimum 10 weeks to a maximum of 12 months can improve insulin resistance by reducing the homeostatic model assessment for insulin resistance.[19] Besides, vitamin D can improve the fasting blood glucose and serum insulin concentration.[91]

The treatment of human promonocytic cells with vitamin D leads to the transcriptional regulation of the insulin receptor (IR) gene, which results in the stimulation of phosphatidylinositol 3-kinase (PI3K) activity (Fig. 3). This activation leads to the improvement in insulin-stimulated glucose oxidation and cellular transport.[92,93] The binding of PI3K with the pleckstrin homology domain leads to the phosphorylation of phosphoinositide-dependent protein kinase and protein kinase B. This activation induces the regulation of serine/threonine-specific kinase cascades, followed by insulin receptor substrate (IRS) phosphorylation, enhancing insulin sensitivity.[93,94] It was evidenced from an *in vivo* study that the adipocyte specific deletion of p110β (catalytic site of PI3K) can result in adipose tissue insulin resistance, obesity, and liver steatosis.[95] In this context, oncogenic PI3K (PIK3CA^{H1047R}) would promote *de novo* lipogenesis.[96] However, these mechanisms have not yet been established in the context of NAFLD. In addition, vitamin D supplementation (5,000 IU/day for six months) can improve the pancreatic β-cell function in T2DM patients, inducing insulin secretion, and maintaining insulin sensitivity.[97] A high level of VDRs is present in pancreatic β-cells. Thus, an adequate supply of vitamin D can maintain the calcium influx to β-cells, and promote Ca^{2+}-induced insulin secretion.[98] Vitamin D induces PPARγ gene expression.[99] Ligand-activated transcription factor PPARγ plays an indispensable role in increasing insulin sensitivity through regulating insulin signaling. PPARγ competes with VDR for heterodimer formation with the retinoic X receptor. The binding of PPARγ and VDR induces the transcriptional activation of genes responsible for insulin sensitivity in mature adipocytes.[100]
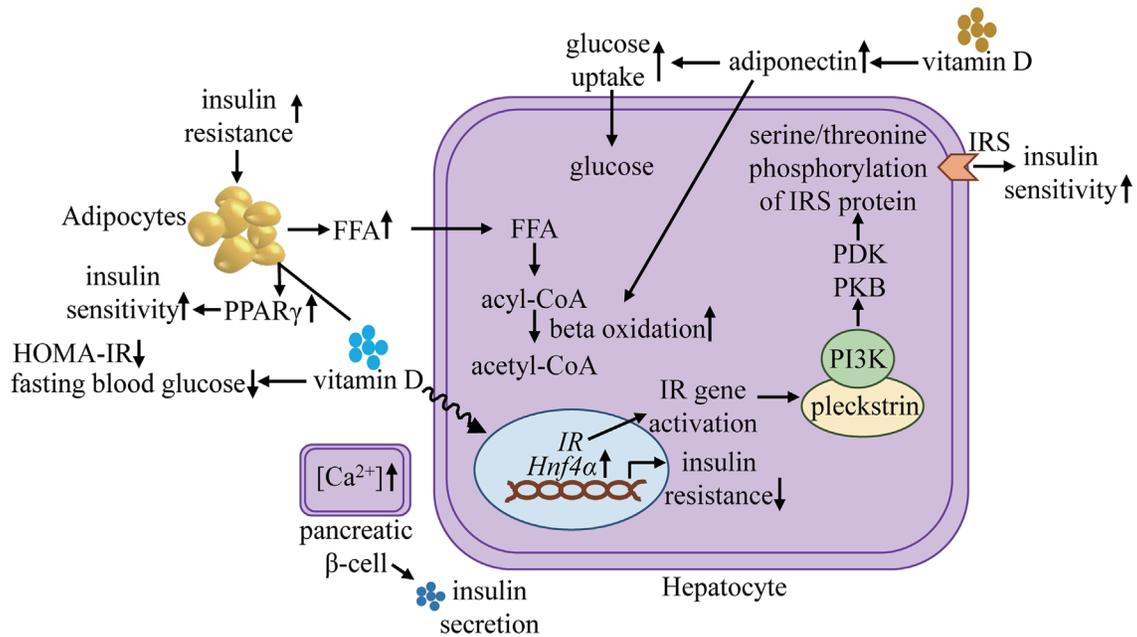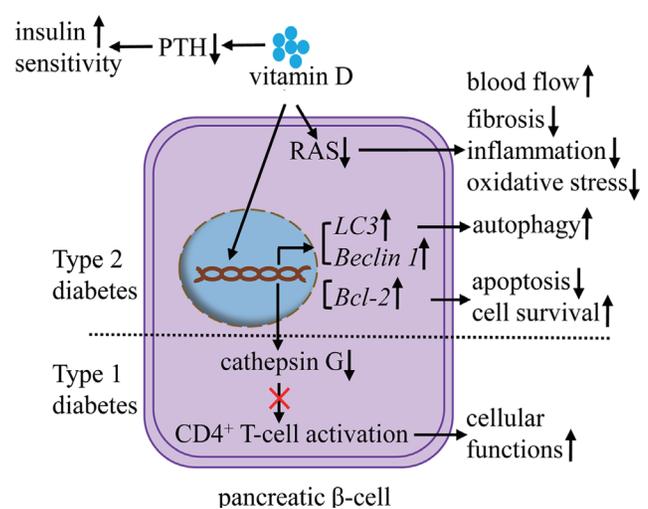
Fig. 3. Molecular targets of vitamin D to regulate insulin resistance mechanism. The major role of vitamin D is to enhance insulin sensitivity by regulating the molecular pathways controlled by adipocytes, pancreatic β-cells, and hepatocytes in the liver. In adipocytes, vitamin D increases the PPARγ expression, which in turn, facilitates insulin sensitivity. In addition, vitamin D acts on pancreatic β-cells to improve insulin secretion. In hepatocytes, vitamin D enhances IR gene activation, followed by the phosphorylation of the IRS protein, which ultimately leads to the induction of insulin sensitivity. In addition, vitamin D induces Hnf4α, and controls insulin resistance. Furthermore, vitamin D increases the adiponectin level to promote the insulin-dependent glucose uptake, and improve glucose homeostasis. Symbols "↑" and "↓" refer to "high" and "low", respectively. Ca^{2+}, calcium ion; FFA, free fatty acid; Hnf4α, hepatocyte nuclear factor 4α; HOMA-IR, homeostatic model assessment for insulin resistance; IR, insulin receptor; IRS, insulin receptor substrate; PDK, phosphoinositide-dependent protein kinase; PI3K, phosphoinositide 3-kinases; PKB, phosphorylate and activate protein kinase B; PPARγ, Peroxisome proliferator-activated receptor gamma.

The activation of PPARγ leads to the maintenance of glucose/lipid uptake and storage, M2 polarization, fatty acid oxidation, and insulin secretion, which in turn, ultimately results in the improvement of insulin sensitivity.[101]

Role of vitamin D in diabetes

Obesity and insulin resistance often lead to the progression of T2DM. The underlying mechanism of T2DM involves the reduction in β-cells mass, and deterioration of β-cell functions.[102] The improper function of β-cells would result in defective or inadequate insulin secretion, and poor glucose homeostasis. As mentioned earlier, vitamin D stimulates pancreatic β-cells to maintain insulin secretion. The effect of vitamin D has already been published, in which INS1E cells were used as an alternative for pancreatic β-cells.[103] It was suggested that 1,25(OH)$_2$D is more effective, when compared to the 25(OH)D type, in inducing insulin secretion.[103] Vitamin D increases autophagy, and reduces the chance of apoptosis of pancreatic β-cells (Fig. 4). It has been demonstrated that treating mouse insulinoma β-cells with 1,25(OH)$_2$D increases the expression of autophagy-related genes *LC3* and *Beclin 1*, while the anti-apoptotic gene *Bcl-2* expression increases after the vitamin D treatment of streptozotocin-treated MIN6 cells.[104] Unlike T2DM, type-1 diabetes mellitus develops due to the autoimmunity-mediated damage of pancreatic β-cells. Vitamin D supplementation restored the functions of pancreatic β-cells in a type-1 diabetic mellitus mice model. It was demonstrated that vitamin D supplementation downregulates the cathepsin G expression, and inhibits CD4+ T cell activation, which in turn, improves β-cell function.[105] Another important mediator of T2DM is RAS, which minimizes insulin secretion by reducing

Fig. 4. The role of vitamin D to regulate molecular pathways linked to diabetes. The major targets of vitamin D are elucidated to improve the pancreatic β-cell function. In T2DM, vitamin D reduces PTH, and recovers insulin sensitivity. Vitamin D also reduces RAS activity, which minimizes fibrosis, inflammation and oxidative stress, but increases blood flow. In addition, vitamin D induces autophagy, and reduces apoptosis, ultimately resulting in the survival of pancreatic β-cells. In type-1 diabetes mellitus, vitamin D reduces the cathepsin G expression, and inhibits CD4+ T-cell activation, which in turn, improves β-cell functions. Symbols "↑" and "↓" refer to "high" and "low", respectively. Bcl2, B-cell lymphoma-2; LC3, microtubule-associated protein 1 light chain 3; PTH, parathyroid hormone; RAS, renin-angiotensin system.
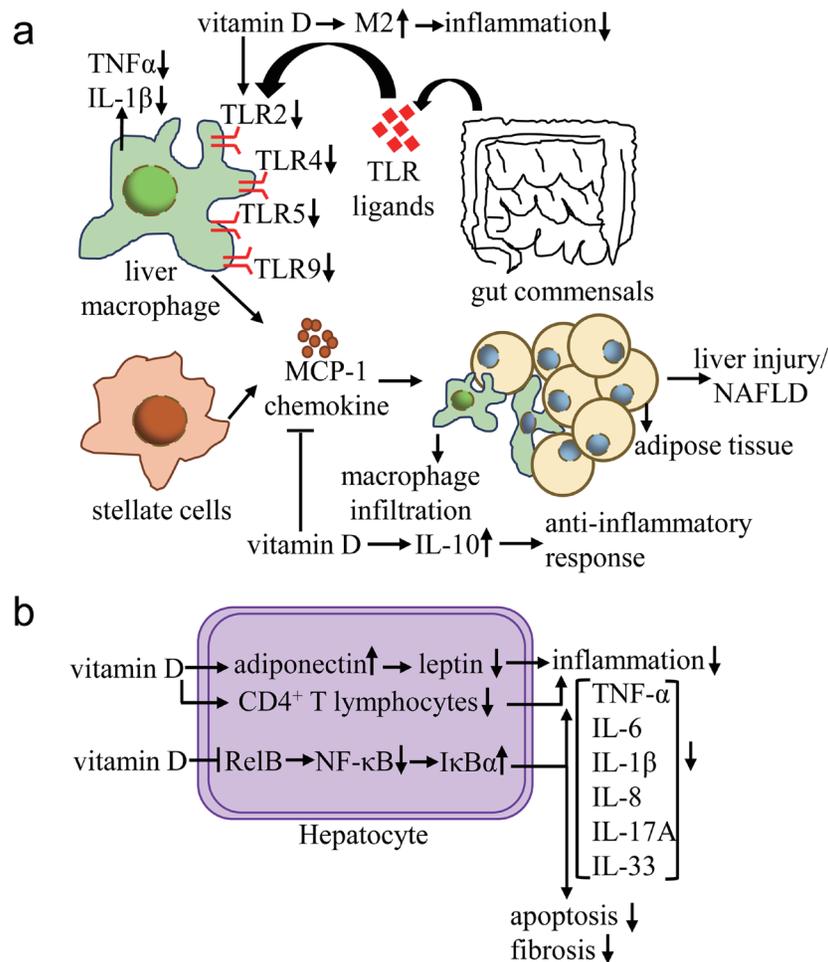
Fig. 5. A complex molecular understanding of the vitamin D-mediated control of liver inflammation. Several cell types, such as adipocytes, hepatocytes, stellate cells, and liver macrophages/Kupffer cells, take part in the process of liver inflammation. Vitamin D downregulates the expression of TLR genes, and decreases the gut microbiota-derived activation of TLR, which further reduces the secretion of proinflammatory cytokines, such as TNFα and IL1β. One of the key mechanisms of vitamin D is to inhibit the secretion of MCP1 chemokines released by both liver macrophages and stellate cells, which in turn, restricts the macrophage infiltration inside adipose tissues, and minimizes liver injury. In addition, vitamin D improves anti-inflammatory response by elevating the IL10 level. In hepatocytes, vitamin D acts to minimize the lectin expression and CD4+ T-cells, which ultimately reduces the chance of liver inflammation. Furthermore, vitamin D inhibits RelB (NFκB subunit), and enhances the IκBα expression to minimize proinflammatory response, fibrosis and apoptosis. Symbols "↑", "↓", and "⊥" refer to "high", "low", and "inhibition", respectively. IκBα, inhibitory κB (IκB) proteins; IL1β, interleukin 1 beta; MCP-1, Monocyte chemoattractant protein 1; NAFLD, non-alcoholic fatty liver disease; RelB, transcription factor RelB (NFκB subunit); TLR, toll-like receptor; TNFα, tumor necrosis factor alpha.

pancreatic blood flow, and promoting islets fibrosis, inflammation, and oxidative stress.[106] It was reported that VDR-knockout (KO) mice presented with an increased expression of RAS components (renin and angiotensinogen), while the vitamin D treatment significantly suppressed the expression of RAS components in isolated KO mice pancreatic islets.[107] Parathyroid hormone (PTH) plays a crucial role in NAFLD. An *in vivo* study revealed that PTH increases the expression of *Ppargc1α* and *Cpt1α* genes linked to lipid β-oxidation, while PTH downregulates the genes (*Pparg*, *Acaca*, and *Fasn*) for *de novo* lipogenesis and lipid uptake (*Cd36*). The expression of these genes leads to the activation of the cAMP/PKA/CREB pathway, and facilitates NAFLD steatosis.[108] NAFLD in pregnancy is a risk factor for maternal and child health.[109] In this context, PTH reduces insulin sensitivity and β-cell functions in pregnancy.[110] Various clinical trials have been documented, ensuring the effect of vitamin D supplementation on the reduction in serum PTH levels in obese populations.[111]

Role of vitamin D in hepatic inflammation

Inflammation plays an important role in the development of advanced stage NAFLD, and progression of NASH. Inflammation occurs through the liver, white adipose tissues (WATs), and the intestines (Fig. 5a). WATs functionally work to secrete bioactive substances, such as adipokines and cytokines, which participate in inflammatory pathways.[112] An *in vivo* study revealed that the surgical removal of epididymal WATs from high-fat diet-fed mice diminished the development of NASH.[113] Furthermore, the antibody treatment against proinflammatory mediators can improve the steatosis.[114] Inflammation in the progressive stage of NAFLD may be explained through the "multiple parallel hit model".[115] According to this hypothesis, lipotoxicity, the gut microbiota, lack of short-chain fatty acids, toll-like receptors (TLR), nutrients (trans fatty acids and fructose), adipokines, cytokines, Fas death receptor, and PPARγ are involved in hepatic inflammation.[115] In addition, the FFA pool would result in increased tumor necrosis factor alpha

Fig. 6. Molecular targets of vitamin D to control oxidative stress linked to liver injury and NAFLD. Vitamin D induces the reduction in lipid peroxidation, GSH, MDA, and SOD1 levels, improving the ROS-mediated cellular toxicity. In addition, vitamin D increases the cellular (Ca^{2+}) content, in order to improve mitochondrial biogenesis and membrane potential. Vitamin D interacts with hydroxyacyl-CoA dehydrogenase, and suppresses the biosynthesis of carnitine, ultimately leading to the restriction of cellular FFA trafficking. Symbols "↑" and "↓" refer to "high" and "low", respectively. FFA, free fatty acid; GSH, glutathione; MDA, malondialdehyde; NAFLD, nonalcoholic fatty liver disease; NOX, NADPH oxidase; SOD1, superoxide dismutase 1.

(TNFα)- and reactive oxygen species (ROS)-mediated hepatocyte apoptosis.[116] Excessive triglyceride accumulation and less VDR in adipocytes may regulate the gene expression of adipokines. Moreover, vitamin D deficiency is associated with low levels of adiponectin and elevated levels of leptin.[117,118] In this context, the up-regulation of hepatocyte adiponectin or serum adiponectin, and the downregulation of leptin may be promising therapeutic approaches to control hepatic inflammation in advanced stage NAFLD.[119] It was reported that vitamin D supplementation (100,000 IU bolus and 4,000 IU/day for 16 weeks) can elevate adiponectin levels in an obese adult population.[120] Another study revealed that vitamin D-fortified yogurt can significantly increase the serum adiponectin concentration in patients with T2DM.[121] It is also reported that aerobic exercise can help to promote the vitamin D-mediated elevation of adiponectin level in T2DM.[122] Moreover, a single intramuscular injection of vitamin D (300,000 IU) can improve the adiponectin level in gestational diabetes mellitus.[123] Multiple studies have revealed that vitamin D is inversely correlated with the level of proinflammatory cytokines, such as TNFα, interleukin (IL) 1 beta (IL1β), IL6, IL8, IL17A and IL33 (Fig. 5b).[124] In this context, vitamin D supplementation can improve inflammation by lowering the level of proinflammatory cytokines, especially IL6 and TNFα.[125] Vitamin D and VDR are strong regulators of T cell-mediated immunity. It has been reported that vitamin D inhibits the proliferation of CD4+ T helper cells, thereby diminishing the secretion of proinflammatory cytokines.[126] In addition, vitamin D diminishes NFκB by inhibiting the RelB protein, and elevating the production of IκBα. RelB, which is an NFκB subunit, has been reported to promote liver fibrosis by inducing inflammation.[127] It has been reported that the degradation of phosphorylated IκB kinase (α and β subunits) would result in the entry of NFκB from the cytoplasm to the nucleus. This would subsequently upregulate genes that encode different cytokines and chemokines, and activate the NFκB pathway, leading to apoptosis-mediated liver damage.[128] Hepatic macrophages are a potential source of liver inflammation through the gut-liver axis. The TLR2, TLR4, TLR5 and TLR9

expressed on hepatic macrophages interact with TLR ligands released by commensal gut microbiota, and promote the secretion of proinflammatory cytokines, such as TNFα and IL1β.[129] However, TLR-mediated hepatic inflammation may be reduced by vitamin D through the downregulation of the TLR gene expression.[130] The M1/M2 polarization of Kupffer cell exhibits more M1 than M2 in NAFLD, which in turn, promotes the increase in secretion of proinflammatory cytokines, and generates profound liver inflammation, injury and fibrosis.[131] An *in vivo* study suggested that vitamin D deficiency is correlated with increased M1 macrophages, while the supplementation of vitamin D may reduce proinflammatory cytokines, and elevate M2 macrophages.[132] It is pivotal to note that the infiltration of monocyte-derived macrophages plays a crucial role in inflammation and NAFLD progression (Fig. 5a). The monocyte chemoattractant protein 1 (MCP1) chemokine secreted by Kupffer cells or stellate cells promote the infiltration of these macrophages to adipose tissues.[133] However, the inhibition of MCP1 may diminish the NAFLD progression and liver injury.[134] Vitamin D has been shown to be effective in downregulating the MCP1 expression, which may be beneficial for controlling adipocyte differentiation, insulin resistance, T2DM, and NAFLD pathogenesis.[135] The deficiency of anti-inflammatory cytokines, such as IL10, is potentially linked with the pathogenesis of NAFLD. An *in vivo* study revealed that a lack of IL10 would result in liver inflammation and insulin resistance. Additionally, it has been reported that vitamin D increases the expression of IL10 in dendritic cells and T cells, and impairs inflammation.[136]

Role of vitamin D in oxidative stress

An excessive FFA supply beyond the capacity of hepatocytes may lead to mitochondrial dysfunction (Fig. 6). FFA successively undergoes the process of β-oxidation and Krebs cycle.[137] Briefly, the FFA passes through fatty acid-transport protein 1, and enters the cytosol. In the cytosol, the FFA is converted into acyl-CoA through fatty acyl-CoA synthase. The elevated acyl-CoA would result in the production of increased ketone bodies (acetone and D-β-

hydroxybutyrate) as terminal products. Simultaneously, the high concentration of acetyl CoA is synthesized through β-oxidation, which in turn, participates to induce an excessive Krebs cycle. In addition, plasma membrane NOX (NADPH oxidase) catalyzes the conversion of NADPH to $NADP^+$, and the conversion of oxygen to superoxide anion ($O_2^{-\bullet}$), which ultimately leads to the activation of myofibroblasts, inducing liver fibrosis.[138] NOX-induced high ROS may lead to hypoxia-induced hepatocyte apoptosis and necrosis.[139,140] Simultaneously, high rates of gluconeogenesis would take place to meet the energy demand, while the uncoupling of the electron transport chain would occur, which may lead to less ATP production.[141] Vitamin D significantly reduces oxidative stress parameters. It was reported that vitamin D supplementation (50,000 IU every 14 days for four months) can reduce the level of malondialdehyde in NAFLD patients.[142] In addition, vitamin D supplementation can increase cellular glutathione biosynthesis, and reduce ROS production.[143] A clinical trial conducted for intensive care unit patients revealed that a high dose of vitamin D supplementation (300,000 IU) can improve the total antioxidant capacity.[144] Furthermore, an *in vivo* study revealed that vitamin D supplementation can potentially upregulate superoxide dismutase type 1 (SOD1), and prevent cellular damage.[145] The overexpression of SOD has been shown to prevent alcohol-induced liver injury in a rat model.[146] In another *in vivo* study, it was shown that SOD1 acts to protect hepatocytes from type-1 interferon-mediated oxidative damage.[147] Vitamin D is an important regulator of mitochondrial function. It was reported that vitamin D is capable of enhancing the mitochondrial biogenesis and membrane potential by increasing Ca^{2+} accumulation[148] In addition, vitamin D interacts with hydroxyacyl-CoA dehydrogenase trifunctional multienzyme complex subunit alpha, and suppresses the biosynthesis of carnitine, which limits the fatty acid transport to the mitochondria, and impairs β-oxidation.[149] As mentioned earlier, the regulation of NOX is important in NAFLD pathogenesis.[150] The role of vitamin D in NOX expression has been reported in cerebral ischemia. The pre-treatment of mice with vitamin D potentially reduced the expression of NOX-2, and minimized the high ROS-induced brain injury.[151,152] However, the role of vitamin D in downregulating NOX-2 remains to be explored for NAFLD.

Pitfalls and future directions

The primary diagnosis of NAFLD generally relies on imaging techniques, such as abdominal ultrasonography and computed tomography. However, these non-invasive techniques often lead to a poor diagnosis, in the case of mild steatosis.[153–155] In this context, non-invasive biomarkers for the detection of fibrosis, combined with imaging, may be helpful to arrive at a proper diagnosis. However, a liver biopsy warrants further confirmation. In addition, patients with liver injury and comorbidities represent inconsistencies in clinical findings.[156]

The emergence of inadequate vitamin D levels has become a serious clinical issue worldwide. As mentioned earlier, extensive research has been carried out on the low vitamin D status in NAFLD. It has been reported that vitamin D supplementation can significantly improve NAFLD-related abnormalities. However, there is an urgent need to investigate the role of vitamin D in the regulation of MAFLD. Furthermore, there is a need to explore the vitamin D-associated molecular interactions, in order to obtain a better understanding of the progression of the disease. Vitamin D is actively involved in regulating cells, such as adipocytes, hepatocytes, pancreatic β-cells, liver macrophages, and their cellular processes.

Hence, the effect of vitamin D needs to be closely monitored using *in vitro* cell lines, and *in vivo* animal models. Besides, the effect of vitamin D needs to be checked on the gut microbiota, in order to explore the role of vitamin D in the regulation of the gut-liver axis in NAFLD progression.

Conclusions

The global incidence of NAFLD is increasing. Metabolic disorders and lifestyle significantly impact the NAFLD progression. The underlying molecular mechanism of NAFLD is multifactorial and complex to understand. In addition, prominent and targeted therapeutic strategies to treat NAFLD are presently being developed. After the careful analysis and consideration of relevant published data, the results suggested that WATs play an initiative role in NAFLD progression. The generation of high FFA flux and cellular trafficking may be correlated with the associated downstream events of NAFLD pathogenesis, such as inflammation, high ROS, fibrosis, and liver damage, while metabolic abnormalities and the gut microbiota aggravate the disease progression.

Recently, vitamin D deficiency and increasing incidents of NAFLD cases have been reported. Extensive research has been carried out through *in vitro* and *in vivo* studies, and clinical trials to elucidate the beneficial effect of vitamin D supplementation on NAFLD. The present review highlights the possible molecular targets of vitamin D to diminish the NAFLD pathogenesis. Vitamin D is capable of regulating molecular pathways linked to obesity, insulin resistance and diabetes, which are the major risk factors of NAFLD. Vitamin D tremendously acts on adipocytes to control the FFA trafficking, lipogenesis, and inflammation. Similarly, vitamin D acts on hepatocytes to reduce *de novo* lipogenesis and cellular FFA trafficking. This potentially acts on pancreatic β-cells to improve insulin secretion, cell survival, and cellular functions. In addition, this improves the glucose uptake and insulin sensitivity. Moreover, vitamin D decreases proinflammatory cytokines, and reduces the chance of liver injury in NAFLD. In addition, vitamin D acts on the mitochondria to control ROS-mediated cellular toxicity. Hence, there is an urgent need to diagnostically screen out vitamin D deficient NAFLD patients, and conduct extensive research at the molecular level, in order to analyze the indispensable role of vitamin D, and control NAFLD. Vitamin D supplementation therapy may be beneficial for the treatment and control of NAFLD in the future.

Conflict of interest

The author has no conflict of interest to declare.

Author contributions

SS: performed the literature survey, data compilation and interpretation; constructed the study design and diagrams; wrote the manuscript.

References

[1] Riazi K, Azhari H, Charette JH, Underwood FE, King JA, Afshar EE, *et al*. The prevalence and incidence of NAFLD worldwide: A systematic review and meta-analysis. Lancet Gastroenterol Hepatol 2022;7(9):851–861. doi:10.1016/S2468-1253(22)00165-0, PMID:35798021.

[2] Sonsuz A, Basaranoglu M, Ozbay G. Relationship between aminotransferase levels and histopathological findings in patients with nonalcoholic steatohepatitis. Am J Gastroenterol 2000;95(5):1370–1371. doi:10.1111/j.1572-0241.2000.02046.x, PMID:10811364.

[3] Brown GT, Kleiner DE. Histopathology of nonalcoholic fatty liver disease and nonalcoholic steatohepatitis. Metabolism 2016;65(8):1080–1086. doi:10.1016/j.metabol.2015.11.008, PMID:26775559.

[4] Younossi ZM, Henry L. Epidemiology of non-alcoholic fatty liver disease and hepatocellular carcinoma. JHEP Rep 2021;3(4):100305. doi:10.1016/j.jhepr.2021.100305, PMID:34189448.

[5] Eslam M, Sanyal AJ, George J, International Consensus Panel. MAFLD: A consensus-driven proposed nomenclature for metabolic associated fatty liver disease. Gastroenterology 2020;158(7):1999–2014.e1. doi:10.1053/j.gastro.2019.11.312, PMID:32044314.

[6] Lin S, Huang J, Wang M, Kumar R, Liu Y, Liu S, *et al*. Comparison of MAFLD and NAFLD diagnostic criteria in real world. Liver Int 2020;40(9):2082–2089. doi:10.1111/liv.14548, PMID:32478487.

[7] Kosmalski M, Frankowski R, Ziółkowska S, Różycka-Kosmalska M, Pietras T. What's New in the Treatment of Non-Alcoholic Fatty Liver Disease (NAFLD). J Clin Med 2023;12(5):1852. doi:10.3390/jcm12051852, PMID:36902639.

[8] Babu AF, Csader S, Männistö V, Tauriainen MM, Pentikäinen H, Savonen K, *et al*. Effects of exercise on NAFLD using non-targeted metabolomics in adipose tissue, plasma, urine, and stool. Sci Rep 2022;12(1):6485. doi:10.1038/s41598-022-10481-9, PMID:35444259.

[9] Yoshioka N, Ishigami M, Watanabe Y, Sumi H, Doisaki M, Yamaguchi T, *et al*. Effect of weight change and lifestyle modifications on the development or remission of nonalcoholic fatty liver disease: sex-specific analysis. Sci Rep 2020;10(1):481. doi:10.1038/s41598-019-57369-9, PMID:31949229.

[10] Yang KC, Hung HF, Lu CW, Chang HH, Lee LT, Huang KC. Association of non-alcoholic fatty liver disease with metabolic syndrome independently of central obesity and insulin resistance. Sci Rep 2016;6:27034. doi:10.1038/srep27034, PMID:27246655.

[11] Rau M, Geier A. An update on drug development for the treatment of nonalcoholic fatty liver disease - from ongoing clinical trials to future therapy. Expert Rev Clin Pharmacol 2021;14(3):333–340. doi:10.1080/17512433.2021.1884068, PMID:33535836.

[12] Romeo S, Kozlitina J, Xing C, Pertsemlidis A, Cox D, Pennacchio LA, *et al*. Genetic variation in PNPLA3 confers susceptibility to nonalcoholic fatty liver disease. Nat Genet 2008;40(12):1461–1465. doi:10.1038/ng.257, PMID:18820647.

[13] Yki-Järvinen H. Non-alcoholic fatty liver disease as a cause and a consequence of metabolic syndrome. Lancet Diabetes Endocrinol 2014;2(11):901–910. doi:10.1016/S2213-8587(14)70032-4, PMID:24731669.

[14] Seppälä-Lindroos A, Vehkavaara S, Häkkinen AM, Goto T, Westerbacka J, Sovijärvi A, *et al*. Fat accumulation in the liver is associated with defects in insulin suppression of glucose production and serum free fatty acids independent of obesity in normal men. J Clin Endocrinol Metab 2002;87(7):3023–3028. doi:10.1210/jcem.87.7.8638, PMID:12107194.

[15] Dalvi PS, Yang S, Swain N, Kim J, Saha S, Bourdon C, *et al*. Long-term metabolic effects of malnutrition: Liver steatosis and insulin resistance following early-life protein restriction. PLoS One 2018;13(7):e0199916. doi:10.1371/journal.pone.0199916, PMID:29965973.

[16] Pickett-Blakely O, Young K, Carr RM. Micronutrients in nonalcoholic fatty liver disease pathogenesis. Cell Mol Gastroenterol Hepatol 2018;6(4):451–462. doi:10.1016/j.jcmgh.2018.07.004, PMID:30294653.

[17] Sindhughosa DA, Wibawa IDN, Mariadi IK, Somayana G. Additional treatment of vitamin D for improvement of insulin resistance in non-alcoholic fatty liver disease patients: a systematic review and meta-analysis. Sci Rep 2022;12(1):7716. doi:10.1038/s41598-022-11950-x, PMID:35546181.

[18] Aggeletopoulou I, Thomopoulos K, Mouzaki A, Triantos C. Vitamin

[19] D-VDR novel anti-inflammatory molecules-new insights into their effects on liver diseases. Int J Mol Sci 2022;23(15):8465. doi:10.3390/ijms23158465, PMID:35955597.

[19] Zhang H, Shen Z, Lin Y, Zhang J, Zhang Y, Liu P, *et al*. Vitamin D receptor targets hepatocyte nuclear factor 4α and mediates protective effects of vitamin D in nonalcoholic fatty liver disease. J Biol Chem 2020;295(12):3891–3905. doi:10.1074/jbc.RA119.011487, PMID:32051143.

[20] Holick MF, Smith E, Pincus S. Skin as the site of vitamin D synthesis and target tissue for 1,25-dihydroxyvitamin D3. Use of calcitriol (1,25-dihydroxyvitamin D3) for treatment of psoriasis. Arch Dermatol 1987;123(12):1677–1683a. PMID:2825606.

[21] Trang HM, Cole DE, Rubin LA, Pierratos A, Siu S, Vieth R. Evidence that vitamin D3 increases serum 25-hydroxyvitamin D more efficiently than does vitamin D2. Am J Clin Nutr 1998;68(4):854–858. doi:10.1093/ajcn/68.4.854, PMID:9771862.

[22] Kalajian TA, Aldoukhi A, Veronikis AJ, Persons K, Holick MF. Ultraviolet B light emitting diodes (LEDs) are more efficient and effective in producing vitamin D(3) in human skin compared to natural sunlight. Sci Rep 2017;7(1):11489. doi:10.1038/s41598-017-11362-2, PMID:28904394.

[23] Lamberg-Allardt C. Vitamin D in foods and as supplements. Prog Biophys Mol Biol 2006;92(1):33–38. doi:10.1016/j.pbiomolbio.2006.02.017, PMID:16618499.

[24] Haddad JG, Matsuoka LY, Hollis BW, Hu YZ, Wortsman J. Human plasma transport of vitamin D after its endogenous synthesis. J Clin Invest 1993;91(6):2552–2555. doi:10.1172/JCI116492, PMID:8390483.

[25] Hollis BW, Wagner CL. New insights into the vitamin D requirements during pregnancy. Bone Res 2017;5:17030. doi:10.1038/boneres.2017.30, PMID:28868163.

[26] Huang CY, Weng YT, Li PC, Hsieh NT, Li CI, Liu HS, *et al*. Calcitriol suppresses warburg effect and cell growth in human colorectal cancer cells. Life (Basel) 2021;11(9):963. doi:10.3390/life11090963, PMID:34575112.

[27] Institute of Medicine. Dietary reference intakes for calcium and vitamin D. Washington, DC: The National Academies Press; 2011.

[28] Jablonski KL, Jovanovich A, Holmen J, Targher G, McFann K, Kendrick J, *et al*. Low 25-hydroxyvitamin D level is independently associated with non-alcoholic fatty liver disease. Nutr Metab Cardiovasc Dis 2013;23(8):792–798. doi:10.1016/j.numecd.2012.12.006, PMID:23415456.

[29] Yodoshi T, Orkin S, Arce-Clachar AC, Bramlage K, Liu C, Fei L, *et al*. Vitamin D deficiency: prevalence and association with liver disease severity in pediatric nonalcoholic fatty liver disease. Eur J Clin Nutr 2020;74(3):427–435. doi:10.1038/s41430-019-0493-y, PMID:31444465.

[30] Gad AI, Elmedames MR, Abdelhai AR, Marei AM. The association between vitamin D status and non-alcoholic fatty liver disease in adults: a hospital-based study. Egypt Liver J 2020;10:25. doi:10.1186/s43066-020-00033-z.

[31] Amrein K, Scherkl M, Hoffmann M, Neuwersch-Sommeregger S, Köstenberger M, Tmava Berisha A, *et al*. Vitamin D deficiency 2.0: an update on the current status worldwide. Eur J Clin Nutr 2020;74(11):1498–1513. doi:10.1038/s41430-020-0558-y, PMID:31959942.

[32] Ge X, Zheng L, Wang M, Du Y, Jiang J. Prevalence trends in non-alcoholic fatty liver disease at the global, regional and national levels, 1990-2017: a population-based observational study. BMJ Open 2020;10(8):e036663. doi:10.1136/bmjopen-2019-036663, PMID:32747349.

[33] Ching-Yeung Yu B, Kwok D, Wong VW. Magnitude of nonalcoholic fatty liver disease: Eastern perspective. J Clin Exp Hepatol 2019;9(4):491–496. doi:10.1016/j.jceh.2019.01.007, PMID:31516265.

[34] Le MH, Yeo YH, Li X, Li J, Zou B, Wu Y, *et al*. 2019 Global NAFLD prevalence: A systematic review and meta-analysis. Clin Gastroenterol Hepatol 2022;20(12):2809–2817.e28. doi:10.1016/j.cgh.2021.12.002, PMID:34890795.

[35] Jiang Z, Pu R, Li N, Chen C, Li J, Dai W, *et al*. High prevalence of vitamin D deficiency in Asia: A systematic review and meta-analysis. Crit Rev Food Sci Nutr 2021. doi:10.1080/10408398.2021.1990850, PMID:34783278.

[36] Mogire RM, Atkinson SH. Challenges in estimating the prevalence of vitamin D deficiency in Africa – Authors' reply. Lancet Glob Health 2022; 10(4):e474. doi:10.1016/S2214-109X(22)00040-7, PMID:35303451.

[37] Siddiqee MH, Bhattacharjee B, Siddiqi UR, Rahman MM. High prevalence of vitamin D deficiency among the South Asian adults: a systematic review and meta-analysis. BMC Public Health 2021;21:1823. doi:10.1186/s12889-021-11888-1, PMID:34627207.

[38] Nimitphong H, Holick MF. Vitamin D status and sun exposure in southeast Asia. Dermatoendocrinol 2013;5(1):34–37. doi:10.4161/derm.24054, PMID:24494040.

[39] Cho YH, Kim JW, Shim JO, Yang HR, Chang JY, Moon JS, et al. Association between vitamin D deficiency and suspected nonalcoholic fatty liver disease in an adolescent population. Pediatr Gastroenterol Hepatol Nutr 2019;22(3):233–241. doi:10.5223/pghn.2019.22.3.233, PMID:31110956.

[40] Lowe NM, Bhojani I. Special considerations for vitamin D in the south Asian population in the UK. Ther Adv Musculoskelet Dis 2017;9(6):137–144. doi:10.1177/1759720X17704430, PMID:28620422.

[41] Doycheva I, Watt KD, Alkhouri N. Nonalcoholic fatty liver disease in adolescents and young adults: The next frontier in the epidemic. Hepatology 2017;65(6):2100–2109. doi:10.1002/hep.29068, PMID:28103626.

[42] Bennouar S, Cherif AB, Kessira A, Bennouar DE, Abdi S. Association and interaction between vitamin D level and metabolic syndrome for nonalcoholic fatty liver disease. J Diabetes Metab Disord 2021;20(2):1309–1317. doi:10.1007/s40200-021-00857-5, PMID:34900782.

[43] Wang M, Wang M, Zhang R, Shen C, Zhang L, Ding Y, et al. Influences of Vitamin D Levels and Vitamin D-Binding Protein Polymorphisms on Nonalcoholic Fatty Liver Disease Risk in a Chinese Population. Ann Nutr Metab 2022;78(2):61–72. doi:10.1159/000522193, PMID:35100585.

[44] Xing Y, Cheng T, Zhou F, Ma H. The association between vitamin D and type 2 diabetes mellitus complicated with non-alcoholic fatty liver disease: An observational cross-sectional study. Diabetes Metab Syndr Obes 2022;15:269–280. doi:10.2147/DMSO.S348870, PMID:35140487.

[45] Hamzehzadeh Alamdari A, Ahrabi S, Khoshbaten M, Roustaei S, Araqchin Ahrabi S, Asghari Jafarabadi M. Effect of Oral and parenteral routes of vitamin D supplementation on serum 25(OH) vitamin D levels in patients with non-alcoholic fatty liver disease. Caspian J Intern Med 2022;13(1):23–28. doi:10.22088/cjim.13.1.23, PMID:35178204.

[46] Wang Q, Shi X, Wang J, Zhang J, Xu C. Low serum vitamin D concentrations are associated with obese but not lean NAFLD: a cross-sectional study. Nutr J 2021;20(1):30. doi:10.1186/s12937-021-00690-9, PMID:33794916.

[47] Cordeiro A, Pereira S, Saboya CJ, Ramalho A. Relationship between nonalcoholic fatty liver disease and vitamin d nutritional status in extreme obesity. Can J Gastroenterol Hepatol 2017;2017:9456897. doi:10.1155/2017/9456897, PMID:28685131.

[48] Sayed M, Abozeid EM, Amin HM, Elsayed NM, Nassar YH, Elsheimy HA. Non-alcoholic fatty liver and vitamin D levels in extremely obese individuals: A case-control study. Open Access Maced J Med Sci 2021;9:541–546. doi:10.3889/oamjms.2021.5976.

[49] Kasapoglu B, Turkay C, Yalcin KS, Carlioglu A, Sozen M, Koktener A. Low vitamin D levels are associated with increased risk for fatty liver disease among non-obese adults. Clin Med (Lond) 2013;13(6):576–579. doi:10.7861/clinmedicine.13-6-576, PMID:24298105.

[50] Ehrampoush E, Ghaemi A, Osati S, Maleki M, Shahraki HR, Honarvar B, et al. The association between serum vitamin D level and nonalcoholic fatty liver disease. Hepat Mon 2019;19:e92992. doi:10.5812/hepatmon.92992.

[51] Mohamed Ahmed A, Abdel Ghany M, Abdel Hakeem GL, Kamal A, Khattab R, Abdalla A, et al. Assessment of vitamin D status in a group of Egyptian children with non alcoholic fatty liver disease (multicenter study). Nutr Metab (Lond) 2016;13:53. doi:10.1186/s12986-016-0112-z, PMID:27547235.

[52] Dasarathy J, Varghese R, Feldman A, Khiyami A, McCullough AJ, Dasarathy S. Patients with nonalcoholic fatty liver disease have a low response rate to vitamin D supplementation. J Nutr 2017;147(10):1938–1946. doi:10.3945/jn.117.254292, PMID:28814531.

[53] Moussa MF, Ahmed S, Abdelhady S, Aboelmagd M, Farrag M, Hassan

M, et al. Pattern of serum vitamin D in individuals with non-alcoholic fatty liver and its relation to metabolic dysfunction. Afro-Egypt J Infect Endem Dis 2021;11:412–422. doi:10.21608/aeji.2021.100732.1188.

[54] Cai J, Zhang Z, Liu J, Xiao X, Wang C, Deng M, et al. Correlation between serum 25-OH vitamin D expression and non-alcoholic fatty liver disease. Exp Ther Med 2020;19(3):1681–1686. doi:10.3892/etm.2020.8411, PMID:32104220.

[55] Küçükazman M, Ata N, Dal K, Yeniova AÖ, Kefeli A, Basyigit S, et al. The association of vitamin D deficiency with non-alcoholic fatty liver disease. Clinics (Sao Paulo) 2014;69(8):542–546. doi:10.6061/clinics/2014(08)07, PMID:25141113.

[56] Kim HS, Rotundo L, Kothari N, Kim SH, Pyrsopoulos N. Vitamin D is associated with severity and mortality of non-alcoholic fatty liver disease: A US population-based study. J Clin Transl Hepatol 2017;5(3):185–192. doi:10.14218/JCTH.2017.00025, PMID:28936398.

[57] Shawky MA, Hassan AM, Mohammed AQ, Oraby MI. Vitamin D levels in Egyptian patients with non-alcoholic fatty liver disease. J Gastroenterol Hepatol Res 2018;7:2529–2533. doi:10.17554/j.issn.2224-3992.2018.07.745.

[58] Chakraborty A, Choudhury A, Saha A. Development of non-alcoholic fatty liver disease (NAFLD) in young obese tribal subjects of Tripura: Link between low 25 (OH) vitamin-d levels and immune modulators. J Assoc Physicians India 2019;67(8):52–56. PMID:31562717.

[59] Hao R, Zhao Z, Wei W, You H, Jia J. Relationship between serum 25-OH vitamin D level and nonalcoholic fatty liver disease in the elderly. Int J Clin Exp Med 2018;11:9911–9915.

[60] Mohamed WS, Alswat KA. Relation between non-alcoholic fatty liver disease and vitamin D level. Int J Med Res Prof 2017;3:104–107.

[61] Chung GE, Kim D, Kwak MS, Yang JI, Yim JY, Lim SH, et al. The serum vitamin D level is inversely correlated with nonalcoholic fatty liver disease. Clin Mol Hepatol 2016;22(1):146–151. doi:10.3350/cmh.2016.22.1.146, PMID:27044765.

[62] Çakmak G, Kaplan DS, Yıldırım C, Ulusal H, Tarakçıoğlu M, Öztürk ZA. Improvement of cognitive deficit of curcumin on scopolamine-induced Alzheimer's disease models. Caspian J Intern Med 2022;13(1):16–22. doi:10.22088/cjim.13.1.16, PMID:35178203.

[63] Gungor S, Köylü AA, Saglam S, Neselioglu S, Erel Ö, Acıpayam C. Effects of oxidant-antioxidant and vitamin D levels on clinical and laboratory data in children with fatty liver disease. Cureus 2020;12(12):e11849. doi:10.7759/cureus.11849, PMID:33282605.

[64] Sah MK, Shrestha R, Shrestha B, Khadka D. Relationship between nonalcoholic fatty liver disease and vitamin D in Nepal. NMMJ 2021;2:1–4. doi:10.3126/nmmj.v2i1.37217.

[65] Wang N, Chen C, Zhao L, Chen Y, Han B, Xia F, et al. Vitamin D and nonalcoholic fatty liver disease: Bi-directional mendelian randomization analysis. EBioMedicine 2018;28:187–193. doi:10.1016/j.ebiom.2017.12.027, PMID:29339098.

[66] Lorvand Amiri H, Agah S, Mousavi SN, Hosseini AF, Shidfar F. Regression of non-alcoholic fatty liver by vitamin D supplement: A double-blind randomized controlled clinical trial. Arch Iran Med 2016;19(9):631–638. PMID:27631178.

[67] Day CP, James OF. Steatohepatitis: a tale of two "hits"? Gastroenterology 1998;114(4):842–845. doi:10.1016/s0016-5085(98)70599-2, PMID:9547102.

[68] Paschos P, Paletas K. Non alcoholic fatty liver disease two-hit process: Multifactorial character of the second hit. Hippokratia 2009;13(2):128.

[69] Buzzetti E, Pinzani M, Tsochatzis EA. The multiple-hit pathogenesis of non-alcoholic fatty liver disease (NAFLD). Metabolism 2016;65(8):1038–1048. doi:10.1016/j.metabol.2015.12.012, PMID:26823198.

[70] Qureshi K, Abrams GA. Metabolic liver disease of obesity and role of adipose tissue in the pathogenesis of nonalcoholic fatty liver disease. World J Gastroenterol 2007;13(26):3540–3553. doi:10.3748/wjg.v13.i26.3540, PMID:17659704.

[71] Guilherme A, Virbasius JV, Puri V, Czech MP. Adipocyte dysfunctions linking obesity to insulin resistance and type 2 diabetes. Nat Rev Mol Cell Biol 2008;9(5):367–377. doi:10.1038/nrm2391, PMID:18401346.

[72] Centers for Disease Control and Prevention. Defining Adult Overweight & Obesity. Available from: https://www.cdc.gov/obesity/basics/adult-defining.html#:~:text=If%20your%20BMI%20is%20

less,falls%20within%20the%20obesity%20range. Accessed November 29, 2022.

[73] Longo M, Zatterale F, Naderi J, Parrillo L, Formisano P, Raciti GA, et al. Adipose tissue dysfunction as determinant of obesity-associated metabolic complications. Int J Mol Sci 2019;20(9):2358. doi:10.3390/ijms20092358, PMID:31085992.

[74] Ahmad B, Serpell CJ, Fong IL, Wong EH. Molecular mechanisms of adipogenesis: The anti-adipogenic role of AMP-activated protein kinase. Front Mol Biosci 2020;7:76. doi:10.3389/fmolb.2020.00076, PMID:32457917.

[75] Kitade H, Chen G, Ni Y, Ota T. Nonalcoholic fatty liver disease and insulin resistance: New insights and potential new treatments. Nutrients 2017;9(4):387. doi:10.3390/nu9040387, PMID:28420094.

[76] El Hadi H, Di Vincenzo A, Vettor R, Rossato M. Food ingredients involved in white-to-brown adipose tissue conversion and in calorie burning. Front Physiol 2018;9:1954. doi:10.3389/fphys.2018.01954, PMID:30687134.

[77] Thethi T, Kamiyama M, Kobori H. The link between the renin-angiotensin-aldosterone system and renal injury in obesity and the metabolic syndrome. Curr Hypertens Rep 2012;14(2):160–169. doi:10.1007/s11906-012-0245-z, PMID:22302531.

[78] Lim H, Lee H, Lim Y. Effect of vitamin D(3) supplementation on hepatic lipid dysregulation associated with autophagy regulatory AMPK/Akt-mTOR signaling in type 2 diabetic mice. Exp Biol Med (Maywood) 2021;246(10):1139–1147. doi:10.1177/1535370220987524, PMID:33541129.

[79] Zhang Y, Leung DY, Richers BN, Liu Y, Remigio LK, Riches DW, et al. Vitamin D inhibits monocyte/macrophage proinflammatory cytokine production by targeting MAPK phosphatase-1. J Immunol 2012;188(5):2127–2135. doi:10.4049/jimmunol.1102412, PMID:22301548.

[80] Chen Y, Zhang J, Ge X, Du J, Deb DK, Li YC. Vitamin D receptor inhibits nuclear factor κB activation by interacting with IκB kinase β protein. J Biol Chem 2013;288(27):19450–19458. doi:10.1074/jbc.M113.467670, PMID:23671281.

[81] Szymczak-Pajor I, Miazek K, Selmi A, Balcerczyk A, Śliwińska A. The action of vitamin D in adipose tissue: Is there the link between vitamin D deficiency and adipose tissue-related metabolic disorders? Int J Mol Sci 2022;23(2):956. doi:10.3390/ijms23020956, PMID:35055140.

[82] Kota SK, Kota SK, Jammula S, Meher LK, Panda S, Tripathy PR, et al. Renin-angiotensin system activity in vitamin D deficient, obese individuals with hypertension: An urban Indian study. Indian J Endocrinol Metab 2011;15(Suppl 4):S395–S401. doi:10.4103/2230-8210.86985, PMID:22145146.

[83] Kim KM, Roh JH, Lee S, Yoon JH. Clinical implications of renin-angiotensin system inhibitors for development and progression of nonalcoholic fatty liver disease. Sci Rep 2021;11(1):2884. doi:10.1038/s41598-021-81959-1, PMID:33536442.

[84] Williams JS. Vitamin D supplementation reduces renin-angiotensin system activity in obesity. ClinicalTrials.gov Identifier: NCT01068418. Available from: https://clinicaltrials.gov/ct2/show/NCT01068418. Accessed November 28, 2022.

[85] Li YC, Kong J, Wei M, Chen ZF, Liu SQ, Cao LP. 1,25-Dihydroxyvitamin D(3) is a negative endocrine regulator of the renin-angiotensin system. J Clin Invest 2002;110(2):229–238. doi:10.1172/JCI15219, PMID:12122115.

[86] Lomonaco R, Ortiz-Lopez C, Orsak B, Webb A, Hardies J, Darland C, et al. Effect of adipose tissue insulin resistance on metabolic parameters and liver histology in obese patients with nonalcoholic fatty liver disease. Hepatology 2012;55(5):1389–1397. doi:10.1002/hep.25539, PMID:22183689.

[87] Gaggini M, Morelli M, Buzzigoli E, DeFronzo RA, Bugianesi E, Gastaldelli A. Non-alcoholic fatty liver disease (NAFLD) and its connection with insulin resistance, dyslipidemia, atherosclerosis and coronary heart disease. Nutrients 2013;5(5):1544–1560. doi:10.3390/nu5051544, PMID:23666091.

[88] Bugianesi E, Moscatiello S, Ciaravella MF, Marchesini G. Insulin resistance in nonalcoholic fatty liver disease. Curr Pharm Des 2010;16(17):1941–1951. doi:10.2174/138161210791208875, PMID:20370677.

[89] Liu E, Meigs JB, Pittas AG, McKeown NM, Economos CD, Booth SL, et al. Plasma 25-hydroxyvitamin d is associated with markers of the insulin resistant phenotype in nondiabetic adults. J Nutr 2009;139(2):329–334. doi:10.3945/jn.108.093831, PMID:19106328.

[90] Hosny SS, Ali HM, Mohammed WA, El Ghannam MH. Study of relationship between total vitamin D level and NAFLD in a sample of Egyptian patients with and without T2DM. Diabetes Metab Syndr 2019;13(3):1769–1771. doi:10.1016/j.dsx.2019.04.002, PMID:31235092.

[91] Talaei A, Mohamadi M, Adgi Z. The effect of vitamin D on insulin resistance in patients with type 2 diabetes. Diabetol Metab Syndr 2013;5(1):8. doi:10.1186/1758-5996-5-8, PMID:23443033.

[92] Maestro B, Molero S, Bajo S, Dávila N, Calle C. Transcriptional activation of the human insulin receptor gene by 1,25-dihydroxyvitamin D(3). Cell Biochem Funct 2002;20(3):227–232. doi:10.1002/cbf.951, PMID:12125099.

[93] Shepherd PR, Withers DJ, Siddle K. Phosphoinositide 3-kinase: the key switch mechanism in insulin signalling. Biochem J 1998;333 (Pt 3)(Pt 3):471–490. doi:10.1042/bj3330471, PMID:9677303.

[94] Copps KD, White MF. Regulation of insulin sensitivity by serine/threonine phosphorylation of insulin receptor substrate proteins IRS1 and IRS2. Diabetologia 2012;55(10):2565–2582. doi:10.1007/s00125-012-2644-8, PMID:22869320.

[95] Nelson VL, Jiang YP, Dickman KG, Ballou LM, Lin RZ. Adipose tissue insulin resistance due to loss of PI3K p110α leads to decreased energy expenditure and obesity. Am J Physiol Endocrinol Metab 2014;306(10):E1205–E1216. doi:10.1152/ajpendo.00625.2013, PMID:24691033.

[96] Ricoult SJ, Yecies JL, Ben-Sahra I, Manning BD. Oncogenic PI3K and K-Ras stimulate de novo lipid synthesis through mTORC1 and SREBP. Oncogene 2016;35(10):1250–1260. doi:10.1038/onc.2015.179, PMID:26028026.

[97] Lemieux P, Weisnagel SJ, Caron AZ, Julien AS, Morisset AS, Carreau AM, et al. Effects of 6-month vitamin D supplementation on insulin sensitivity and secretion: a randomised, placebo-controlled trial. Eur J Endocrinol 2019;181(3):287–299. doi:10.1530/EJE-19-0156, PMID:31344685.

[98] Klec C, Ziomek G, Pichler M, Malli R, Graier WF. Calcium signaling in ß-cell physiology and pathology: A revisit. Int J Mol Sci 2019;20(24):E6110. doi:10.3390/ijms20246110, PMID:31817135.

[99] Hoseini R, Damirchi A, Babaei P. Vitamin D increases PPARγ expression and promotes beneficial effects of physical activity in metabolic syndrome. Nutrition 2017;36:54–59. doi:10.1016/j.nut.2016.06.010, PMID:28336108.

[100] Leonardini A, Laviola L, Perrini S, Natalicchio A, Giorgino F. Cross-Talk between PPARgamma and Insulin Signaling and Modulation of Insulin Sensitivity. PPAR Res 2009;2009:818945. doi:10.1155/2009/818945, PMID:20182551.

[101] Choi SS, Park J, Choi JH. Revisiting PPARγ as a target for the treatment of metabolic disorders. BMB Rep 2014;47(11):599–608. doi:10.5483/bmbrep.2014.47.11.174, PMID:25154720.

[102] Meier JJ, Bonadonna RC. Role of reduced β-cell mass versus impaired β-cell function in the pathogenesis of type 2 diabetes. Diabetes Care 2013;36(Suppl 2):S113–S119. doi:10.2337/dcS13-2008, PMID:23882035.

[103] Bornstedt ME, Gjerlaugsen N, Pepaj M, Bredahl MKL, Thorsby PM. Vitamin D increases glucose stimulated insulin secretion from insulin producing beta cells (INS1E). Int J Endocrinol Metab 2019;17(1):e74255. doi:10.5812/ijem.74255, PMID:30881469.

[104] Wang Y, He D, Ni C, Zhou H, Wu S, Xue Z, et al. Vitamin D induces autophagy of pancreatic β-cells and enhances insulin secretion. Mol Med Rep 2016;14(3):2644–2650. doi:10.3892/mmr.2016.5531, PMID:27430408.

[105] Lai X, Liu X, Cai X, Zou F. Vitamin D supplementation induces CatG-mediated CD4(+) T cell inactivation and restores pancreatic β-cell function in mice with type 1 diabetes. Am J Physiol Endocrinol Metab 2022;322(1):E74–E84. doi:10.1152/ajpendo.00066.2021, PMID:34779254.

[106] Leung PS. The Modulatory Action of Vitamin D on the Renin-Angiotensin System and the Determination of Hepatic Insulin Resistance. Molecules 2019;24(13):2479. doi:10.3390/molecules24132479, PMID:31284538.

[107] Cheng Q, Li YC, Boucher BJ, Leung PS. A novel role for vitamin D: modulation of expression and function of the local renin-angiotensin

system in mouse pancreatic islets. Diabetologia 2011;54(8):2077–2081. doi:10.1007/s00125-011-2100-1, PMID:21424540.

[108] Feng X, Xiao Y, Guo Q, Peng H, Zhou HY, Wang JP, *et al*. Parathyroid hormone alleviates non-alcoholic liver steatosis via activating the hepatic cAMP/PKA/CREB pathway. Front Endocrinol (Lausanne) 2022;13:899731. doi:10.3389/fendo.2022.899731, PMID:36060945.

[109] Sarkar M, Grab J, Dodge JL, Gunderson EP, Rubin J, Irani RA, *et al*. Non-alcoholic fatty liver disease in pregnancy is associated with adverse maternal and perinatal outcomes. J Hepatol 2020;73(3):516–522. doi:10.1016/j.jhep.2020.03.049, PMID:32531415.

[110] Nachankar A, Kotwal N, Upreti V, Verma V, Hari Kumar KVS. Association of vitamin D and parathyroid hormone with insulin sensitivity, beta cell function and gestational diabetes in pregnancy: A cross-sectional, observational study. Diabetes Ther 2018;9(5):2081–2090. doi:10.1007/s13300-018-0508-z, PMID:30206904.

[111] Lotito A, Teramoto M, Cheung M, Becker K, Sukumar D. Serum parathyroid hormone responses to vitamin D supplementation in overweight/obese adults: A systematic review and meta-analysis of randomized clinical trials. Nutrients 2017;9(3):241. doi:10.3390/nu9030241, PMID:28272298.

[112] Stojsavljević S, Gomerčić Palčić M, Virović Jukić L, Smirčić Duvnjak L, Duvnjak M. Adipokines and proinflammatory cytokines, the key mediators in the pathogenesis of nonalcoholic fatty liver disease. World J Gastroenterol 2014;20(48):18070–18091. doi:10.3748/wjg.v20.i48.18070, PMID:25561778.

[113] Mulder P, Morrison MC, Wielinga PY, van Duyvenvoorde W, Kooistra T, Kleemann R. Surgical removal of inflamed epididymal white adipose tissue attenuates the development of non-alcoholic steatohepatitis in obesity. Int J Obes (Lond) 2016;40(4):675–684. doi:10.1038/ijo.2015.226, PMID:26499443.

[114] Li Z, Yang S, Lin H, Huang J, Watkins PA, Moser AB, *et al*. Probiotics and antibodies to TNF inhibit inflammatory activity and improve nonalcoholic fatty liver disease. Hepatology 2003;37(2):343–350. doi:10.1053/jhep.2003.50048, PMID:12540784.

[115] Tilg H, Moschen AR. Evolution of inflammation in nonalcoholic fatty liver disease: the multiple parallel hits hypothesis. Hepatology 2010;52(5):1836–1846. doi:10.1002/hep.24001, PMID:21038418.

[116] Feldstein AE, Werneburg NW, Canbay A, Guicciardi ME, Bronk SF, Rydzewski R, *et al*. Free fatty acids promote hepatic lipotoxicity by stimulating TNF-alpha expression via a lysosomal pathway. Hepatology 2004;40(1):185–194. doi:10.1002/hep.20283, PMID:15239102.

[117] Naini AE, Vahdat S, Hedaiati ZP, Shahzeidi S, Pezeshki AH, Nasri H. The effect of vitamin D administration on serum leptin and adiponectin levels in end-stage renal disease patients on hemodialysis with vitamin D deficiency: A placebo-controlled double-blind clinical trial. J Res Med Sci 2016;21:1. doi:10.4103/1735-1995.175144, PMID:27904547.

[118] Chen LW, Chien CH, Kuo SF, Yu CY, Lin CL, Chien RN. Low vitamin D level was associated with metabolic syndrome and high leptin level in subjects with nonalcoholic fatty liver disease: a community-based study. BMC Gastroenterol 2019;19(1):126. doi:10.1186/s12876-019-1040-y, PMID:31311491.

[119] Shabalala SC, Dludla PV, Mabasa L, Kappo AP, Basson AK, Pheiffer C, *et al*. The effect of adiponectin in the pathogenesis of non-alcoholic fatty liver disease (NAFLD) and the potential role of polyphenols in the modulation of adiponectin signaling. Biomed Pharmacother 2020;131:110785. doi:10.1016/j.biopha.2020.110785, PMID:33152943.

[120] Mousa A, Naderpoor N, Wilson K, Plebanski M, de Courten MPJ, Scragg R, *et al*. Vitamin D supplementation increases adipokine concentrations in overweight or obese adults. Eur J Nutr 2020;59(1):195–204. doi:10.1007/s00394-019-01899-5, PMID:30649593.

[121] Nikooyeh B, Hollis BW, Neyestani TR. The effect of daily intake of vitamin D-fortified yogurt drink, with and without added calcium, on serum adiponectin and sirtuins 1 and 6 in adult subjects with type 2 diabetes. Nutr Diabetes 2021;11(1):26. doi:10.1038/s41387-021-00168-x, PMID:34389701.

[122] Mudjanarko SW, Irawati A, Tinduh D. Effects of aerobic exercise on adiponectin levels potentially mediated by vitamin D in type 2 diabetic patients. Endocr Regul 2022;56(3):201–208. doi:10.2478/enr-2022-0021, PMID:35843714.

[123] Hosseinzadeha M, Razmpoosh E, Shareghfarid E, Hosseinzadeh E, Hadinedoushan H, Salamie M, *et al*. The effect of a single mega dose injection of vitamin D on serum adiponectin concentration at first gestational diabetes mellitus: A randomized controlled clinical trial. Clin Nutr Exp 2020;33:39–48. doi:10.1016/j.yclnex.2020.08.001.

[124] Roffe-Vazquez DN, Huerta-Delgado AS, Castillo EC, Villarreal-Calderón JR, Gonzalez-Gil AM, Enriquez C, *et al*. Correlation of vitamin D with inflammatory cytokines, atherosclerotic parameters, and lifestyle factors in the setting of heart failure: A 12-month follow-up study. Int J Mol Sci 2019;20(22):5811. doi:10.3390/ijms20225811, PMID:31752330.

[125] AlGhamdi SA, Enaibsi NN, Alsufiani HM, Alshaibi HF, Khoja SO, Carlberg C. A single oral vitamin D(3) bolus reduces inflammatory markers in healthy saudi males. Int J Mol Sci 2022;23(19):11992. doi:10.3390/ijms231911992, PMID:36233290.

[126] Sheikh V, Kasapoglu P, Zamani A, Basiri Z, Tahamoli-Roudsari A, Alahgholi-Hajibehzad M. Vitamin D3 inhibits the proliferation of T helper cells, downregulate CD4(+) T cell cytokines and upregulate inhibitory markers. Hum Immunol 2018;79(6):439–445. doi:10.1016/j.humimm.2018.03.001, PMID:29522785.

[127] Cohen-Lahav M, Shany S, Tobvin D, Chaimovitz C, Douvdevani A. Vitamin D decreases NFkappaB activity by increasing IkappaBalpha levels. Nephrol Dial Transplant 2006;21(4):889–897. doi:10.1093/ndt/gfi254, PMID:16455676.

[128] Yamamoto Y, Gaynor RB. Therapeutic potential of inhibition of the NF-kappaB pathway in the treatment of inflammation and cancer. J Clin Invest 2001;107(2):135–142. doi:10.1172/JCI11914, PMID:11160126.

[129] Miura K, Ohnishi H. Role of gut microbiota and Toll-like receptors in non-alcoholic fatty liver disease. World J Gastroenterol 2014;20(23):7381–7391. doi:10.3748/wjg.v20.i23.7381, PMID:24966608.

[130] Do JE, Kwon SY, Park S, Lee ES. Effects of vitamin D on expression of Toll-like receptors of monocytes from patients with Behcet's disease. Rheumatology (Oxford) 2008;47(6):840–848. doi:10.1093/rheumatology/ken109, PMID:18411217.

[131] Chen J, Deng X, Liu Y, Tan Q, Huang G, Che Q, *et al*. Kupffer Cells in Non-alcoholic Fatty Liver Disease: Friend or Foe? Int J Biol Sci 2020;16(13):2367–2378. doi:10.7150/ijbs.47143, PMID:32760204.

[132] Gunasekar P, Swier VJ, Fleegel JP, Boosani CS, Radwan MM, Agrawal DK. Vitamin D and macrophage polarization in epicardial adipose tissue of atherosclerotic swine. PLoS One 2018;13(10):e0199411. doi:10.1371/journal.pone.0199411, PMID:30296271.

[133] Lefere S, Tacke F. Macrophages in obesity and non-alcoholic fatty liver disease: Crosstalk with metabolism. JHEP Rep 2019;1(1):30–43. doi:10.1016/j.jhepr.2019.02.004, PMID:32149275.

[134] Baeck C, Wehr A, Karlmark KR, Heymann F, Vucur M, Gassler N, *et al*. Pharmacological inhibition of the chemokine CCL2 (MCP-1) diminishes liver macrophage infiltration and steatohepatitis in chronic hepatic injury. Gut 2012;61(3):416–426. doi:10.1136/gutjnl-2011-300304, PMID:21813474.

[135] Wang YC, Hsieh CC, Kuo HF, Tsai MK, Yang SN, Kuo CH, *et al*. Effect of vitamin D3 on monocyte chemoattractant protein 1 production in monocytes and macrophages. Acta Cardiol Sin 2014;30(2):144–150. PMID:27122781.

[136] Heine G, Niesner U, Chang HD, Steinmeyer A, Zügel U, Zuberbier T, *et al*. 1,25-dihydroxyvitamin D(3) promotes IL-10 production in human B cells. Eur J Immunol 2008;38(8):2210–2218. doi:10.1002/eji.200838216, PMID:18651709.

[137] Delarue J, Magnan C. Free fatty acids and insulin resistance. Curr Opin Clin Nutr Metab Care 2007;10(2):142–148. doi:10.1097/MCO.0b013e328042ba90, PMID:17285001.

[138] Liang S, Kisseleva T, Brenner DA. The Role of NADPH Oxidases (NOXs) in Liver Fibrosis and the Activation of Myofibroblasts. Front Physiol 2016;7:17. doi:10.3389/fphys.2016.00017, PMID:26869935.

[139] Bhogal RH, Curbishley SM, Weston CJ, Adams DH, Afford SC. Reactive oxygen species mediate human hepatocyte injury during hypoxia/reoxygenation. Liver Transpl 2010;16(11):1303–1313. doi:10.1002/lt.22157, PMID:21031546.

[140] Loffredo L, Del Ben M, Perri L, Carnevale R, Nocella C, Catasca E, *et al*. Effects of dark chocolate on NOX-2-generated oxidative stress in

patients with non-alcoholic steatohepatitis. Aliment Pharmacol Ther 2016;44(3):279–86. doi:10.1111/apt.13687, PMID:27265388.

[141] García-Ruiz I, Solís-Muñoz P, Fernández-Moreira D, Grau M, Colina F, Muñoz-Yagüe T, et al. High-fat diet decreases activity of the oxidative phosphorylation complexes and causes nonalcoholic steatohepatitis in mice. Dis Model Mech 2014;7(11):1287–1296. doi:10.1242/dmm.016766, PMID:25261569.

[142] Sharifi N, Amani R, Hajiani E, Cheraghian B. Does vitamin D improve liver enzymes, oxidative stress, and inflammatory biomarkers in adults with non-alcoholic fatty liver disease? A randomized clinical trial. Endocrine 2014;47(1):70–80. doi:10.1007/s12020-014-0336-5, PMID:24968737.

[143] Jain SK, Micinski D. Vitamin D upregulates glutamate cysteine ligase and glutathione reductase, and GSH formation, and decreases ROS and MCP-1 and IL-8 secretion in high-glucose exposed U937 monocytes. Biochem Biophys Res Commun 2013;437(1):7–11. doi:10.1016/j.bbrc.2013.06.004, PMID:23770363.

[144] Sistanizadab M, Kouchekc M, Miri M, Salarian S, Shojaei S, Vasegh FM, et al. High dose vitamin D improves total serum antioxidant capacity and ICU outcome in critically ill patients - A randomized, double-blind clinical trial. Eur J Integr Med 2021;42:101271. doi:10.1016/j.eujim.2020.101271.

[145] Romi MM, Arefian N, Wahyu Setyaningsih WA, Perdana Putri RG, Juffrie M, Ratna Sari DC. Calcitriol treatment attenuates uric acid-induced kidney injury via super oxide dismutase-1 (SOD-1) upregulation and fibrosis reduction. Iran Biomed J 2021;25(6):417–425. doi:10.52547/ibj.25.6.417, PMID:34641645.

[146] Wheeler MD, Nakagami M, Bradford BU, Uesugi T, Mason RP, Connor HD, et al. Overexpression of manganese superoxide dismutase prevents alcohol-induced liver injury in the rat. J Biol Chem 2001;276(39):36664–36672. doi:10.1074/jbc.M105352200, PMID:11477087.

[147] Bhattacharya A, Hegazy AN, Deigendesch N, Kosack L, Cupovic J, Kandasamy RK, et al. Superoxide dismutase 1 protects hepatocytes from type i interferon-driven oxidative damage. Immunity 2015;43(5):974–986. doi:10.1016/j.immuni.2015.10.013, PMID:26588782.

[148] Safaei Z, Bakhshalizadeh SH, Nasr Esfahani MH, Akbari Sene A, Najafzadeh V, Soleimani M, et al. Effect of vitamin D3 on mitochondrial biogenesis in granulosa cells derived from polycystic ovary syndrome. Int J Fertil Steril 2020;14(2):143–149. doi:10.22074/ijfs.2020.6077, PMID:32681627.

[149] Mendoza A, Takemoto Y, Cruzado KT, Masoud SS, Nagata A, Tantipanjaporn A, et al. Controlled lipid β-oxidation and carnitine biosynthesis by a vitamin D metabolite. Cell Chem Biol 2022;29(4):660–669. e12. doi:10.1016/j.chembiol.2021.08.008, PMID:34506728.

[150] Gabbia D, Cannella L, De Martin S. The Role of Oxidative Stress in NAFLD-NASH-HCC Transition-Focus on NADPH Oxidases. Biomedicines 2021;9(6):687. doi:10.3390/biomedicines9060687, PMID:34204571.

[151] Velimirović M, Jevtić Dožudić G, Selaković V, Stojković T, Puškaš N, Zaletel I, et al. Effects of vitamin D3 on the NADPH oxidase and matrix metalloproteinase 9 in an animal model of global cerebral ischemia. Oxid Med Cell Longev 2018;2018:3273654. doi:10.1155/2018/3273654, PMID:29849881.

[152] Cui C, Song S, Cui J, Feng Y, Gao J, Jiang P. Vitamin D receptor activation influences NADPH oxidase (NOX(2)) activity and protects against neurological deficits and apoptosis in a rat model of traumatic brain injury. Oxid Med Cell Longev 2017;2017:9245702. doi:10.1155/2017/9245702, PMID:29410737.

[153] Golabi P, Sayiner M, Fazel Y, Koenig A, Henry L, Younossi ZM. Current complications and challenges in nonalcoholic steatohepatitis screening and diagnosis. Expert Rev Gastroenterol Hepatol 2016;10(1):63–71. doi:10.1586/17474124.2016.1099433, PMID:26469309.

[154] Barchetta I, Cimini FA, Cavallo MG. Vitamin D and metabolic dysfunction-associated fatty liver disease (MAFLD): An update. Nutrients 2020;12(11):3302. doi:10.3390/nu12113302, PMID:33126575.

[155] Zhang Z, Thorne JL, Moore JB. Vitamin D and nonalcoholic fatty liver disease. Curr Opin Clin Nutr Metab Care 2019;22(6):449–458. doi:10.1097/MCO.0000000000000605, PMID:31589177.

[156] Iqbal U, Perumpail BJ, Akhtar D, Kim D, Ahmed A. The epidemiology, risk profiling and diagnostic challenges of nonalcoholic fatty liver disease. Medicines (Basel) 2019;6(1):41. doi:10.3390/medicines6010041, PMID:30889791.