Roles of vitamin A in the regulation of fatty acid synthesis

Fu-Chen Yang, Feng Xu, Tian-Nan Wang, Guo-Xun Chen

ORCID number: Fu-Chen Yang 0000-0001-6203-4220; Feng Xu 0000-0001-8888-0142; Tian-Nan Wang 0000-0001-5584-3628; Guo-Xun Chen 0000-0001-6226-4050.

Author contributions: All authors contributed to literature search and manuscript writing; Yang FC and Chen GX initiated the design and outline of the manuscript.

Supported by: the Financial Support of the Overseas Training Program for Outstanding Young and Middle-Aged Teachers in Universities in Jiangsu Province, China (to Yang FC).

Conflict-of-interest statement: The authors declare no conflict of interest related to this manuscript.

Open-Access: This article is an open-access article that was selected by an in-house editor and fully peer-reviewed by external reviewers. It is distributed in accordance with the Creative Commons Attribution NonCommercial (CC BY-NC 4.0) license, which permits others to distribute, remix, adapt, build upon this work non-commercially, and license their derivative works on different terms, provided the original work is properly cited and the use is non-commercial. See: http://creativecommons.org/Licenses/by-nc/4.0/

Manuscript source: Invited

Abstract

Dietary macronutrients and micronutrients play important roles in human health. On the other hand, the excessive energy derived from food is stored in the form of triacylglycerol. A variety of dietary and hormonal factors affect this process through the regulation of the activities and expression levels of those key player enzymes involved in fatty acid biosynthesis such as acetyl-CoA carboxylase, fatty acid synthase, fatty acid elongases, and desaturases. As a micronutrient, vitamin A is essential for the health of humans. Recently, vitamin A has been shown to play a role in the regulation of glucose and lipid metabolism. This review summarizes recent research progresses about the roles of vitamin A in fatty acid synthesis. It focuses on the effects of vitamin A on the activities and expression levels of mRNA and proteins of key enzymes for fatty acid synthesis in vitro and in vivo. It appears that vitamin A status and its signaling pathway regulate the expression levels of enzymes involved in fatty acid synthesis. Future research directions are also discussed.

Key Words: Vitamin A; Acetyl-CoA carboxylase; Fatty acid synthase; Fatty acid elongase; Stearoyl-CoA desaturase; Fatty acid synthesis

©The Author(s) 2021. Published by Baishideng Publishing Group Inc. All rights reserved.

Core Tip: Recent studies have shown that vitamin A plays a role in the regulation of glucose and fatty acid metabolism. Vitamin A status, its supplementation, and the treatment with its metabolite, retinoic acid, have been shown to regulate the activities,
and the expression levels of protein and mRNA of acetyl-CoA carboxylase, fatty acid synthase, fatty acid elongases, and fatty acid desaturases in the animal tissues and cells. Systematic evaluations of the roles of vitamin A in the fatty acid metabolism are needed for the treatment and prevention of metabolic diseases such as obesity and type 2 diabetes.

Citation: Yang FC, Xu F, Wang TN, Chen GX. Roles of vitamin A in the regulation of fatty acid synthesis. World J Clin Cases 2021; 9(18): 4506-4519
URL: https://www.wjgnet.com/2307-8960/full/v9/i18/4506.htm
DOI: https://dx.doi.org/10.12998/wjcc.v9.i18.4506

INTRODUCTION
Excessive accumulation of fat leads to obesity. Currently, human obesity has become a global concern of public health[1]. It is one of the main risk factors affecting human health and causes many chronic diseases such as diabetes and cardiovascular diseases [2]. Both dietary and de novo synthesized fatty acids (either saturated or unsaturated) are esterified to a glycerol to make a triacylglycerol (TAG)[3], which is stored in adipocytes that increase in sizes and number with the obesity development. In addition to a place for TAG deposition, the adipose tissue also acts as an endocrine organ and secretes adipokines with a variety of physiological functions[4]. Alterations of adipose tissue functions occur with the development of obesity and other chronic metabolic diseases[5].

Fatty acids are also components of other molecules such as phospholipids, sphingolipids, and esters. Furthermore, they also participate in mediating signal transduction in cells[6]. Dietary linoleic acid and alpha-linolenic acid are the two essential fatty acids for human health. Intracellular fatty acids synthesized can be further elongated and desaturated through multiple enzymes responsible for desaturation and elongation reactions[7,8].

Vitamin A (retinol), a micronutrient, regulates a variety of physiological functions [9]. Retinol molecule contains a β-ionone ring with a polyunsaturated chain and an alcohol group[10]. Its derivatives function in the vision cycle, and regulate cell growth and differentiation, etc. Dietary molecules with vitamin A activities are preformed vitamin A retinyl esters and provitamin A carotenoids, which are from animal and plant sources, respectively. Provitamin A molecules can be converted into vitamin A [9].

Recently, it has become clear that vitamin A plays a role in the regulation of glucose and fatty acid metabolism[11,12]. This is achieved through the regulation of gene expression by retinoic acid (RA), a product of retinol metabolism[13-15]. How RA regulates the expression of genes involved in lipid metabolism and their signaling pathways is something worth being investigated. Here, we try to summarize the effects of vitamin A status and its metabolites on the regulation of genes involved in fatty acid synthesis, desaturation, and elongation pathways. In July 2020, key words such as vitamin A, retinol, acetyl-CoA carboxylase (ACC), fatty acid synthase (FAS), fatty acid elongases, and desaturases were used to search PubMed and retrieve the relevant articles for further reading.

OVERVIEW OF FATTY ACID SYNTHESIS IN MAMMALIAN CELLS
Understanding the regulation of the hepatic fatty acid metabolism pathways has both theoretical and clinical significance for health. The liver plays a major role in the de novo lipogenesis[16,17]. After the formation of TAG, hepatocytes secrete very low-density lipoprotein containing the newly synthesized TAG to be stored or used in other tissues[18-20]. The synthesis of fatty acids occurs in the cytosol and uses acetyl-CoA as the building block. Acetyl-CoA comes from three sources, product of pyruvate dehydrogenase, β-oxidation of fatty acids, and catabolism of amino acids. It is mainly produced in the mitochondrion, and is first converted into citrate, which enters into cytosol using the citric acid transport system. After that, ATP citrate lyase in the cytosol hydrolyzes citrate into oxaloacetate and acetyl-CoA which is used in
lipogenesis[16].

The steps of mammalian fatty acid synthesis are shown in Figure 1. Acetyl-CoA is first converted to malonyl-CoA by ACC. Both acetyl-CoA and malonyl-CoA are loaded onto acyl carrier protein (ACP) domains of FAS to form acetyl-ACP and malonyl-ACP, respectively[21,22]. Mammalian FAS is a polypeptide containing multi-functional subdomains with seven enzymatic activities, which are acetyl-CoA-ACP transacylase, malonyl-CoA-ACP transacylase, β-ketoacyl-ACP condensase, β-ketoacyl-ACP reductase, β-hydroxyacyl-ACP dehydratase, enoyl-ACP reductase, and palmitoyl-ACP thioesterase. FAS repetitively catalyzes condensation, reduction, dehydrogenation, and reduction reactions to add two carbons each time until a 16-carbon acyl chain is formed. The final product of FAS is palmitic acid, a saturated fatty acid.

Fatty acids with longer chain length and double bonds can be produced from palmitic acid via other enzymes[23,24]. Elongases (ELOVLs) add two carbons each time to create a longer chain fatty acid. Desaturases introduce double bonds to the saturated and unsaturated fatty acids, which results in the production of monounsaturated fatty acids (MUFAs) and polyunsaturated fatty acids (PUFAs), respectively. Desaturases are specific for double bond formation in specific positions on a fatty acid molecule. The formation of each double bond requires an oxygen molecule and two electrons to participate. The first desaturation reaction is catalyzed by stearoyl-CoA desaturase 1 (SCD1), which introduces the first double bond in palmitic acid and stearic acid[25,26]. Saturated fatty acids and unsaturated fatty acids can be used to form TAG, phospholipids, and other lipids. The impacts of nutritional factors on the expression and activities of these lipogenic enzymes have been the interests of nutritional sciences.

OVERVIEW OF VITAMIN A METABOLISM

Humans and other mammals are unable to synthesize vitamin A in the body. Dietary preformed vitamin A and provitamin A are from animal and plant foods, respectively. Vitamin A is mainly stored as retinyl esters such as retinyl palmitate in the liver[27-30]. Figure 2 shows the digestion, absorption, transport, and storage of vitamin A in the body. Dietary retinyl esters and carotenoids form micelles with other lipids in the digestion and absorption processes[10,27]. The released retinol and carotenoids are absorbed into the enterocytes after crossing the unstirred water layer. Within the enterocytes, β-carotene is hydrolyzed to retinal by 15,15'-dioxygenase, and retinal is reduced to retinol by retinal reductase[28]. Retinol is re-esterified to form retinyl esters again by lecithin retinol acyltransferase or acyl-CoA retinol acyltransferase and then incorporated into chylomicrons, the lipoprotein for the transport of dietary lipids, which enter the lymph circulation and then the blood circulation[29].

The retinol released from retinyl esters in chylomicron remnants is catabolized into retinal and then RA (all trans RA unless defined otherwise) in the liver. The excessive retinol is re-esterified into retinyl esters, which are stored in the hepatic stellate cells[29]. A cellular retinol-binding protein binds to free retinol in the cells and is responsible to modulate the intracellular free retinol concentration. Retinol is secreted back from the liver in a complex containing retinol-binding protein 4, transthyretin, and thyroxine in the blood. Peripheral cells uptake retinol, and convert it into retinal and then RA to exert physiological responses. Other metabolites derived from retinol and RA can be excreted in the urine and the bile. Extrahepatic tissues, retina, adipose tissues, skeletal muscle, bone marrow, blood cells, spleen, heart, lungs, and kidneys can also uptake retinyl esters and metabolize retinol as the liver.

RA enters the nucleus, and binds to RA receptors (RARs) and retinoid X receptors (RXRs) interacting with the RA response elements (RAREs) in the promoter of the targeted genes[30-32]. Retinoids regulate gene expression through the RAREs in the promoters of their targeted genes. Some of those genes are involved in the regulation of glucose and fatty acid metabolism as reviewed[11,12]. Our lab has shown that the vitamin A metabolism participates in the regulation of hepatic lipogenic gene expressions during the cycle of fasting and refeeding in rats[33]. Therefore, understanding the role of vitamin A in fatty acid synthesis helps to clarify the regulation of lipogenesis for the prevention and treatment of chronic metabolic diseases such as obesity.
Figure 1 Fatty acid synthesis process in a mammalian cell. Dietary nutrients are metabolized into acetyl-CoA, which is converted into malonyl-CoA by acetyl-CoA carboxylase. Malonyl-CoA and acetyl-CoA are used by fatty acid synthase to generate palmitic acid, which can be either elongated into stearic acids by elongases (ELOVLs) or desaturated into palmitoleic acid, a monounsaturated fatty acid (MUFA), by stearoyl-CoA desaturase 1 (SCD1). Oleic acid (an MUFA) can be created either via elongation of palmitoleic acid by ELOVLs or desaturation of stearic acid by SCD1. Additional fatty acid with longer chain length or more double bonds can be generated from oleic acid through the activities of ELOVLs and fatty acid desaturases. ACC: Acetyl-CoA carboxylase; FAS: Fatty acid synthase; SCD1: Stearoyl-CoA desaturase 1; FADS: Fatty acid desaturases.

Figure 2 Overview of vitamin A metabolism in the body. Vitamin A (retinol) is in the forms of preformed vitamin A, retinyl esters, and provitamin A, carotenoids, in our diets. After digestion and absorption, resynthesized retinyl esters are packed as chylomicrons and released into the lymph circulation and then blood circulation to be delivered to the peripheral tissues first. The chylomicron remnants are taken up by the hepatocytes, which will hydrolyze retinyl esters to retinol, which is used for the productions of retinoic acid, stored again in the form of retinyl ester in stellate cells, or a complex containing retinol, retinol binding protein, and transthyretin, which is released into the blood circulation again. Retinol in the circulation is taken up by cells and oxidized into retinal, and then retinoic acid, which participates into the regulation of gene expression, and in turn cellular responses.

EFFECTS OF VITAMIN A STATUS AND RA TREATMENT ON ACC

ACC catalyzes the conversion of acetyl-CoA to malonyl-CoA, and plays an important role in the control of fatty acid metabolism[34,35]. Two isoforms of ACC have been identified, ACCα and ACCβ. The malonyl-CoA produced by ACC is located in the cytoplasm and can be used for fatty acid synthesis, and suppression of fatty acid oxidation through the inhibition of carnitine palmitoyl transferase I (CPT1) activity [36]. Vitamin A can regulate ACC activity and gene expression levels, thereby affecting fatty acid synthesis.

Reports have shown that vitamin A signaling system regulates the activity and expression of ACC. The effect of vitamin A deficiency (VAD) on myocardial lipid metabolism has been studied in rats. VAD causes changes of lipid synthesis and composition, and reduces the ACC activity significantly[37]. In the heart, the VAD rats have reductions of ACC activity (but not Accb mRNA), mRNA levels of Rtrs, and...
cardiolipin content, and increases in CPT1 activity and its mRNA level, and phosphatidic acid levels compared with those fed the same diet supplemented with 8 mg of retinyl palmitate/kg diet. The incorporations of [1-14C]-acetate into cholesterol and [methyl-14C]-choline into phosphatidylcholine are increased in the VAD animals. All these changes returned to their original levels after the VAD rats were fed a vitamin A sufficient (VAS) diet for 15 d, demonstrating the significant alterations of lipid metabolism in the heart of VAD rats.[37]

Changes of the hepatic ACC activity and its mRNA expression levels in VAD status are observed. Male Wistar rats at weaning (3 wk of age) fed a VAD diet for 3 mo have lower body weight gain, liver and plasma retinol levels, and plasma TAG and cholesterol levels than those fed a VAS diet or those refed the control diet for 15 d.[38] The incorporations of [1-14C]-acetate into cholesterol and [methyl-14C]-choline into phosphatidylcholine are lowered due to the VAD status. The activity of hepatic ACC in the VAD male Wistar rat is significantly lower than that in the control group.[38] Similarly, the hepatic expression of Acc mRNA in the VAD group is significantly lower than that in the control group and the refed group. When the VAD animals were refed a VAS diet, the expression of Acc mRNA returned to that of the control value.[38]

NMRI male mice at 12-wk-old age have been treated with RA at 10, 50, or 100 mg/kg body weight/d for 4 d. Levels of Pparβ, Accb, Rxsα, and Cpt1 mRNA in the skeletal muscle tissue are induced by the RA treatment. The proteins levels of PPARβ and RXRα are elevated in the group of mice treated with the 50 mg/kg body weight/d RA.[39]

The effect of RA on fatty acid synthesis in bovine mammary alveolar cells (MAC-T) have been studied.[40] MAC-T cells after being differentiated for 4 d have been treated with 0, 1.0, 1.5, and 2.0 μmol/L RA for additional 3 d. RA treatment increases the amounts of short-chain and medium-chain, saturated and monosaturated fatty acids, and reduces the amounts of long-chain and PUFAs in the cells. Interestingly, the mRNA level of Acca is reduced by 1 μmol/L RA, but induced by 2 μmol/L RA.[40]

In H9C2 myotube, a rat heart muscle cell line, RA treatment significantly induces the expression of Accb gene expression through the RXRα-mediated activation of muscle regulatory factor 4, which interacts with the Accb gene promoter.[41] In HL-60 promyelocytic leukemia cells treated with RA for 24 h and 7 d, the activity of ACC decreased by 44.9% and 99.7%, respectively.[42]

Table 1 summarizes the effects of vitamin A status and RA treatment on the ACC activity and its mRNA and protein expression levels in animals and cells. It seems that the enzymatic activity and mRNA expression levels of ACC have been studied to certain extent. However, more protein data appear to be needed.

**EFFECTS OF VITAMIN A STATUS AND RA TREATMENT ON FAS**

As one of the key enzymes of de novo lipogenesis, FAS in the cytosol uses acetyl-CoA and malonyl-CoA to produce the 16-carbon palmitic acid.[22] The expression level of Fas mRNA changes in response to nutritional states.[43]

In the male lambs, supplementation of VA at 500000 IU/animal twice per week from birth to 100 d of age does not affect body weight and lipid content during growth. However, this treatment increases the number of adipocytes in the perirenal depot, but reduces the sizes of adipocytes in the omental and perirenal depots, which is associated with the reduction of FAS activity in the perirenal fat depot.[44] On the other hand, a vitamin A supplementation study did not show any change of the marbling scores and lipogenic enzyme activities including the FAS activity in the adipose tissues of yearling beef steer.[45] Glucocorticoids can stimulate fatty acid biosynthesis and increase the activity of FAS protein and Fas mRNA levels. However, studies of the Fas gene in the lungs of rat fetuses in late pregnancy indicate that glucocorticoid-stimulated Fas gene expression is antagonized by the RA treatment.[46]

Doses of RA at 10 or 100 mg/kg body weight have been injected daily subcutaneously for 4 d to 13-wk-old NMRI male mice.[47] The 100 mg/kg body weight, but not 10 mg/kg body weight, dose reduces the epididymal white adipose tissue weight. However, both doses of RA reduce hepatic Srebp-1c and Fas mRNA levels.[47] Our lab has shown that the VAD status leads to the reduction of hepatic expression levels of FAS protein in the refeeding of a VAS diet in the VAD rat liver.[33] In addition, vitamin A status regulates the Fas mRNA levels in both Zucker lean (ZL) and Zucker fat (ZF) rats. The VAD ZL and ZF rats have lower hepatic Fas mRNA levels than their respective VAS controls.[48].
Table 1 Effects of vitamin A status and retinoic acid treatment on acetyl-CoA carboxylase enzymatic activity and its mRNA and protein expression levels in cells and tissues

| Treatment         | Tissue/cells        | ACC activity | mRNA levels                                                                 | Protein levels         | Ref.   |
|-------------------|---------------------|--------------|-----------------------------------------------------------------------------|------------------------|--------|
| Vitamin A deficiency | Rat heart            | Reduced      | No change of Accb                                                          | ND                     | Vega et al[37] |
|                   | Rat liver            | Reduced      | Reduced Acc                                                                | ND                     | Oliveros et al[38] |
| RA treatment      | Rat liver            | ND           | ND                                                                           | Reduced in ad libitum  | Li et al[33]   |
|                   | NMRI mouse muscle   | ND           | Increased Accb                                                             | ND                     | Amengual et al[39] |
|                   | MAC-T cells          | Acca reduced by 1 μmol/L and induced by 2 μmol/L | ND                                                                           | Liao et al[40]  |
|                   | H9C2 myotube         | Increased Accb| ND                                                                           | Kim et al[41]  |
|                   | HL-60 PL cells       | Reduced      | ND                                                                           | Fischkoff et al[42] |

ACC: Acetyl-CoA carboxylase; HL-60 PL: HL-60 promyelocytic leukemia cells; MAC-T: Bovine mammary alveolar cells; ND: Not determined; RA: Retinoic acid; VAD: Vitamin A deficiency.

The effect of glucose on the RA-induced lipogenesis has been investigated in 3T3L1 adipocytes cells. RA has been shown to induce or suppress lipid accumulation when medium glucose concentrations are 25 and 5.5 mmol/L, respectively. These RA effects are associated with inductions and reductions of Ap2 and Fas mRNA expression at 25 mmol/L and 5.5 mmol/L medium glucose concentrations, respectively[49]. On the other hand, 0.5, 5, or 50 μmol/L RA treatment increases the expression levels of Ap2 mRNA[49]. Other studies from the same group show that RA at 1 μmol/L inhibits the mRNA levels of Srebp-1c and Fas, which is associated with the reduction of lipid accumulation in 3T3-L1 cells[50]. Interestingly, RA at 1 μmol/L is able to inhibit the differentiation of 3T3-L1 cells and suppress the FAS activity[51]. In human AML-I preadipocytes, the treatment with 50 μmol/L RA or 9-cis RA induces the cell growth arrest and cell death[52]. The 50 μmol/L RA treatment for 4 to 5 d results in the elevation of Fas mRNA[52].

In primary rat hepatocytes, RA synergizes with insulin to induce the Srebp-1c and Fas expression, which is mediated by the two liver X responsive elements in the Srebp-1c promoter[14]. In HepG2 cells, RA treatment induces the activation of FAS promoter in a transient reporter gene assay[53]. The responsive element is attributed to an E-box region that is considered a place for multiple hormonal effects[54]. The mechanism of the RA-induced expression of FAS mRNA and protein is thought to be mediated by SREBP-1c[55]. RXR, but not RAR, is thought to be responsible for this phenomenon [55]. Interestingly, in HepG2 cells, 1 μmol/L RA treatment for 24 h induces the mRNA levels of CPT1, SREBP-1c, and FAS, which is associated with the elevation of the fatty acid oxidation based on the authors’ conclusions[56]. Farnesol treatment significantly down-regulates the mRNA level of FAS in the clone-9 cultured rat hepatocytes, which involves a 9-cis RA mediated mechanism[57]. Stimulatory proteins 1 and 3, nuclear factor Y, upstream stimulatory factor, and SREBP-1 have cognate binding sites in the FAS promoter, which may contribute to the RA-regulated FAS expression in HepG2 cells[58]. The region of the rat Fas promoter contains specific cis-elements responsible for the RA responses, which might not be RAREs. It is possible that RA induces SREBP-1c expression in hepatocytes, and in turn, SREBP-1c mediates the RA signal to activate the FAS promoter[59].

In LNCaP prostate cells, 1 μmol/L RA treatment for 24 or 72 h is sufficient to induce FAS mRNA expression, which is accompanied by the incorporation of [2-14C] acetate into lipids, especially TAG, indicating the elevation of lipid synthesis and accumulation[60]. Retinol in human glioblastoma cells affects fatty acid biosynthetic pathways. FAS protein expression is down-regulated after the treatment with retinol[61].

Table 2 summarizes the effects of vitamin A status and RA treatment on the FAS protein and mRNA expression levels in different cells and organs. It appears that vitamin A status affects the Fas mRNA levels in rat hepatocytes. In addition, RA treatments also regulate FAS protein and mRNA expression levels. The outcomes depend on the cell types and glucose content in the culture media.
Table 2 Effects of vitamin A supplementation, vitamin A status, and retinoic acid treatment on the Fas mRNA and fatty acid synthase protein levels in cells and tissues

| Treatment | Tissue/cells         | FAS activity | Fas mRNA | Protein levels | Ref.                          |
|-----------|----------------------|--------------|-----------|----------------|-------------------------------|
| Vitamin A supplementation | Perirenal fat depot of lamb | Reduced | ND | ND | Arana et al[44] |
|           | Adipose tissue of yearling beef steers | No change | ND | ND | Bryant et al[45] |
| Vitamin A deficiency | ZL rat liver | ND | Not changed in ad libitum | Reduced in the refeeding of a VAS diet | Li et al[33] |
| RA treatments | ZL and ZF rat liver | ND | Reduced in 6 h-fasting | ND | Zhang et al[48] |
|           | Rat fetus lung | ND | Reduced GC-induced activity | Reduced GC-induced Fas | ND | Xu et al[46] |
|           | Mouse EWAT | ND | Reduces Fas | ND | Amengual et al[47] |
|           | 3T3-L1 cells | ND | Induced at 25 mmol/L glucose, and reduced at 5.5 mmol/L glucose | ND | Abd Eldaim et al[49] |
|           | 3T3-L1 cells | ND | Reduced | ND | Amengual et al[50] and Murray et al[51] |
|           | Human AML-I preadipocytes | ND | Induced | ND | Morikawa et al[52] |
|           | Primary rat hepatocytes | ND | Synergized with insulin to induce Fas | ND | Li et al[14] |
|           | HepG2 cells | ND | Induced FAS | Induced FAS | Roder et al[53] and Amengual et al[56] |
|           | LNCAP prostate cells | ND | Induced FAS | ND | Duncan and Archer[60] |

GC: Glucocorticoid; EWAT: Epididymal white adipose tissue; FAS: Fatty acid synthase; ND: Not determined; RA: Retinoic acid; VAD: Vitamin A deficiency; ZL: Zucker lean; ZF: Zucker fatty; VAS: Vitamin A sufficient.

EFFECTS OF VITAMIN A STATUS AND RA ON ELOVLs

Currently, seven isoforms of ELOVLs, ELOVL1 to 7, have been identified in mammalian cells. They participate in the elongation reactions for the synthesis of very long chain fatty acids. Each ELOVL isozyme has its preferred acyl-CoA with particular carbon chain length and saturation[7,62]. Few reports have shown the relationship between vitamin A and ELOVLs.

Both male and female C57BL/6J mice at 35 d of age have been fed a stock diet with 20% total energy from ground nut oil (10% w/w) or 54% total energy from beef tallow (high-fat diet, 33% w/w) for 26 wk[63]. The hepatic retinol content in mice fed the high-fat diet is much higher (more than 5-fold) than that of mice fed the stock diet. The hepatic content of docosahexaenoic acid (C22:6n-3) and expression levels of ELOVL2 protein in male and female mice fed the high-fat diet are higher than those of mice fed the stock diet, but the mRNA was not determined[63]. VA supplementation increases the ELOVL4 in the retina of WNIN/Ob obese rats[64].

EFFECTS OF VITAMIN A STATUS AND RA ON DESATURASES

Fatty acid desaturases introduce double bonds onto saturated and unsaturated fatty acids. For example, SCD1 is responsible for the formation of the first double bond to produce MUFA s and regulation of lipogenesis[25,26]. Vitamin A restriction in beef cattle has been shown to improve the marbling scores in Japanese Black cattle[65]. Changes in the expression of genes for lipid synthesis are observed in the muscle tissues of the Japanese Black steers with the vitamin A restriction, and associated with the marbling phenotype[66]. However, semi-quantitative polymerase chain reaction did not find any significant change of Scd mRNA in the vitamin A restriction group[66]. In Angus-based steers, vitamin A restriction does not affect the marbling scores, but induces MUFA amounts and desaturase index in adipose tissues[67]. However, the SCD activity was not measured.
in the study[67].

Wistar rats fed a VAD diet for 16 wk have reduced plasma TAG and hepatic expression levels of Scd1 mRNA, but not SCD protein levels, both of which are induced by feeding of a high fructose diet[68]. The mRNA expression levels of Elovl6 and Scd1 are reduced after female mice were fed a high-fat diet for 4 wk[69]. The vitamin A supplementation does not have any impact on the Elovl6 and Scd1 mRNA expression in this experimental setting[69]. In mice, feeding of a diet containing high retinyl palmitate (0.1% w/w) for 36 h induces the hepatic Scd1 mRNA expression[70]. However, the SCD1 protein and its mRNA levels in the kidney are not affected by the vitamin A status even though VAD leads to the elevation of oleic (C18:1) and total MUFA levels in the same tissue[71]. The feeding of a VAD diet reduces the retinol content, and increases the MUFAs in the kidney probably due to the regulation of SCD1 activity[71]. In the pancreas of rats fed a VAD diet for 16 wk, the oleic acid content is reduced, which is associated with the reduction of SCD1 protein level[72].

The restriction or deficiency of vitamin A in rats induces the hepatic mRNA levels of fatty acid delta-5 desaturase[73]. Interestingly, the liver of rats fed a VAD diet for 19 wk have higher microsomal activity of SCD1, but not delta 6-desaturase, than those fed a VAS diet[74]. The replenishment of vitamin A in the VAD rats restored the SCD1 activity to the level equal to that in the VAS rats[74]. In the skin of Scd1 knockout mice, the retinoid metabolism is disturbed with the elevations of retinol and RA contents, and the expression levels of RA-induced genes such as Rhp1, Cnnp2, and Lcn2, which is also associated with the elevations of Il1b and Tnfα mRNA levels[75].

In rat primary astrocytes pretreated with 10 μmol/L retinol for 24 h, treatment with 50 μmol/L docosahexaenoic acid for 24 h induces the mRNA expression of Fads2 (delta 6-desaturase gene)[76]. In the initiation phase of 3T3-L1 adipocyte differentiation, the RA treatment prevents the lipid accumulation[77]. RA dose-dependently suppressed the Scd1 and Albp mRNA expression, which is induced during the differentiation process[77]. In human retinal pigment epithelial cells, RA dose-dependently induces the SCD mRNA, which is mediated by the activation of both RAR and RXR[78].

Table 3 summarizes the effects of vitamin A status and RA treatments on the SCD1 activity, and the expression levels of its mRNA and protein in cells and tissues. In the liver, VAD reduces the SCD1 protein and mRNA expression levels. RA treatment affects the Scd1 mRNA expression levels depending on the cells tested.

VITAMIN A AND NON-ALCOHOLIC FATTY LIVER DISEASE

Non-alcoholic fatty liver disease (NAFLD) has become a disease that challenges the public health systems in many countries[79,80]. NAFLD can develop to nonalcoholic steatohepatitis (NASH), cirrhosis, and then hepatocellular carcinoma in certain percent of the patients[81]. Elevation of fatty acid synthesis from acetyl CoA has been indicated as the major contribution to the development of NAFLD in humans[81].

The VA intakes, blood levels of retinol and RA, and the hepatic retinoid contents have been investigated in human subjects with NAFLD. The VA intakes of subjects with NAFLD have been shown to be lower than the control ones[82,83], higher than the control ones[84] or not different from the control groups[85]. The blood retinol is inversely associated with liver damages in patients with NAFLD[83,86]. Patients with a genetic variant of patatin-like phospholipase domain-containing 3 (I148M) that reduces its activity and is linked to NAFLD have reduced blood retinol levels[87]. In one study, the blood RA levels in patients with NAFLD and NASH are lower than that in the age-matched control group[88]. The hepatic levels of retinyl palmitate, RA, 13-cis-RA, and 4-oxo-atRA in NAFLD patients with simple steatosis and NASH are lower than that in the control subjects[89].

The hepatic genes involved in VA metabolism and signaling are also altered in the liver samples from patients with NAFLD. The expression level of RXRα mRNA in the liver biopsy samples of NAFLD patients is inversely correlated with hepatic steatosis[88]. The expression levels of retinaldehyde dehydrogenase 1 family, member A2 (ALDH1A2) and retinaldehyde dehydrogenase 1 family, member A3 (ALDH1A3), enzymes for RA synthesis, in NAFLD subjects are lower than those of the control subjects as well[85]. However, the mRNA levels of lecithin retinol acyltransferase (LRAT), ALDH1A1, CYP26A1, RARA, and RARB are not different between the control and NAFLD groups[89].

Animal studies have been done to determine the impacts of NAFLD on VA metabolism and VA signaling on the NAFLD development. Feeding a methionine and
choline deficient (MCD) diet, a diet that induces fatty liver in mice and rats, for 6 wk lowered plasma retinol level and increased hepatic retinol content in Wistar rats[90]. The expression levels of the hepatic Lrat, Aldh1a1, and Aldh1a2 are also elevated in the rats fed the MCD diet[90]. The increased expression level of Lrat was also observed in mice fed a high-fat/cholesterol diet for 12 or 20 wk or in ob/ob mice, which is attributed to the increase in the formation of retinyl esters in hepatocytes, not hepatic stellate cells[91]. This is associated with the decrease of liver retinol and increase in retinyl palmitate contents[91]. The MCD diet also induces the expression level of platelet-type 12S-lipoxygenase in the hepatic stellate cells of mice, cells for VA storage and the development of hepatic fibrosis[92].

VA deficiency prevents the high-fructose diet-induced TAG in the liver and plasma in Wistar rats[68], and HFD-induced steatosis in mice[93]. On the other hand, treatment with RA has been shown to reduce lipid accumulation and steatosis in NAFLD mice[90, 94-96]. Interestingly, RA improved insulin sensitivity and reduced blood glucose and liver damage in wild type, but not ob/ob mice[95]. M80 (an RARα specific agonist) treatment reduces insulin and leptin resistance in KK-Ay mice and increases leptin receptor mRNA levels[95]. However, in another study, mice fed an HFD for 3 mo were treated with vehicle, and then treated with AM80 or AC261066 (an RARβ2 specific agonist) for another month[97]. Mice in the AM80 group have higher degree of steatosis, TAG levels, inflammation, and blood glucose levels than those in the control and AC261066 groups [97]. The AC201066 group has higher hepatic TAG content than the control group as well[97].

Reduced RA production via heterozygous deletion of retinol dehydrogenase 10 (Rdh10) gene has been attributed to the increased adiposity and insulin resistance in mice fed an HFD for 16 wk, which can be corrected to certain extent by RA treatment via capsules for 3 wk[94]. On the other hand, altered location of retinyl ester formation from the stellate cells to hepatocytes is also considered a reason for fatty liver development in mice fed an HFD or ob/ob mice[91]. Furthermore, the Sirtuin pathway seems to be required for HFD-induced hyperglycemia, insulin resistance, and steatosis in mice fed an HFD, which can be attenuated if the HFD is supplemented with RA[96]. Additionally, the adenovirus-mediated overexpression of RXRα and RA treatment are shown to reduce the lipid accumulation in the liver and steatosis in mice[90]. It appears that VA metabolism is altered with the development of NAFLD based on the data of human and animal studies summarized here. Whether these alterations are the causes or consequences of the NAFLD remains to be determined. Interventional studies in rodents seem to yield conflicting results. The VA deficiency status appears to reduce fatty liver in rats[68], and HFD-induced steatosis in mice[93]. The reduced RA production led to insulin resistance, and RA supplementation rescued the phenotype in mice[94]. It is interesting that extremely low VA availability in the body and exogenously derived RA both can reduce fatty liver phenotypes in rodents. Apparently, the role of VA in the lipid metabolism is more complicated than whether

| Treatments | Tissue/cells | Activity | mRNA | Protein levels | Ref. |
|------------|-------------|---------|------|----------------|-----|
| Vitamin A supplement | Mouse liver | ND | No change | ND | Weiss et al[69] |
| Vitamin A restriction | Muscle tissues of the Japanese Black steers | ND | No change | ND | Hayashi et al[66] |
| Vitamin A deficiency | Rat liver | ND | Reduced | No change | Raja Gopal Reddy et al[68] |
| RA treatments | 3T3-L1 cells | ND | ND | Reduced | Stone and Bernlohr[77] |
| Human retinal cells | ND | Induced | ND | | Samuel et al[78] |

ND: Not determined; RA: Retinoic acid.
there is enough RA produced or not. The VA metabolism probably should be considered more dynamically in the context of spatial and temporal manners, such as the transition of the cycle of fasting and refeeding[33].

Due to the lack of clear etiology and effective biomarkers for diagnosis (liver biopsy is the gold standard), the treatment methods for NAFLD are limited[98]. Nevertheless, given the roles of VA in the regulation of glucose and lipid metabolism, cautions must be given when supplementations of micronutrients are recommended in a clinical setting for the intervention of NAFLD. Another area that deserves more attention is the changes of VA and other micronutrients metabolism and their roles in the drug-induced hepatotoxicity that involves multiple mechanisms and pathways[99]. This is especially important as drugs are often used to treat comorbidities associated with NAFLD such as obesity and type 2 diabetes.

CONCLUSION

Fatty acid synthesis is closely related to the development of chronic metabolic diseases such as obesity, diabetes, and cardiovascular diseases. The key lipogenic enzymes and their genes seem to be regulated by the vitamin A statuses and its metabolite, RA. This has become more and more obvious with the accumulation of research data. As demonstrated in this review, this area is still in the preliminary stage, and more in-depth and systematic research is anticipated. The following areas are especially important in the future: (1) Systematic studies of the effects of vitamin A on the activities of key lipogenic genes in various mammalian cells should be conducted to establish the link between these two; (2) In the meantime, their mRNA and protein levels are also worth to be determined to indicate the potential mechanisms; (3) The interactions of vitamin A with insulin and other regulatory factors of lipogenesis should receive more attention; and (4) In addition to metabolism, since RA treatments affect tumor growth and cell apoptosis, the role of RA-regulated fatty acid synthesis is also worth to be investigated. Nevertheless, understanding the role of vitamin A in the regulation of lipogenesis will benefit not only metabolic studies but also interventions of human metabolic diseases.

REFERENCES

1. Yanovski SZ, Yanovski JA. Obesity prevalence in the United States--up, down, or sideways? N Engl J Med 2011; 364: 987-989 [PMID: 21410367 DOI: 10.1056/NEJMip1009229]
2. Saeedi P, Petersohn I, Salpea P, Malanda B, Karuranga S, Unwin N, Colagiuri S, Guariguata L, Motala AA, Ogurtsova K, Shaw JE, Bright D, Williams R; IDF Diabetes Atlas Committee. Global and regional diabetes prevalence estimates for 2019 and projections for 2030 and 2040: Results from the International Diabetes Federation Diabetes Atlas, 9th edition. Diabetes Res Clin Pract 2019; 157: 107843 [PMID: 31518657 DOI: 10.1016/j.diabres.2019.107843]
3. Yan CL, Stone SJ, Koliwad S, Harris C, Farese RV Jr. Thematic review series: glycerolipids. DGAT enzymes and triacylglycerol biosynthesis. J Lipid Res 2008; 49: 2283-2301 [PMID: 18757836 DOI: 10.1194/jlr.R800018-JLR200]
4. Friedman JM. Leptin at 14 y of age: an ongoing story. Am J Clin Nutr 2009; 89: 973S-979S [PMID: 19190071 DOI: 10.3945/ajcn.2008.26788B]
5. Goossens GH. The Metabolic Phenotype in Obesity: Fat Mass, Body Fat Distribution, and Adipose Tissue Function. Obes Facts 2017; 10: 207-215 [PMID: 28564650 DOI: 10.1159/000471488]
6. Turner N, Cooney GJ, Kraegen EW, Bruce CR. Fatty acid metabolism, energy expenditure and insulin resistance in muscle. J Endocrinol 2014; 220: T61-T79 [PMID: 24323910 DOI: 10.1530/JEO-13-0357]
7. Jakobsson A, Westerberg R, Jacobsson A. Fatty acid elongases in mammals: their regulation and roles in metabolism. Prog Lipid Res 2006; 45: 237-249 [PMID: 16564093 DOI: 10.1016/j.plipres.2006.01.004]
8. Jump DB. N-3 polysaturated fatty acid regulation of hepatic gene transcription. Curr Opin Lipidol 2008; 19: 242-247 [PMID: 18460914 DOI: 10.1097/MOL.0b013e3282f1eafe]
9. Ross AC, Harrison E. Vitamin A and Carotenoids. In: McCormick D, Rucker R, Suttie J, Zempleni J. Handbook of Vitamins. 4th ed. Boca Raton: Dekker/CRC Press, 2006
10. Blomhoff R, Blomhoff RK. Overview of retinoid metabolism and function. J Neurobiol 2006; 66: 606-630 [PMID: 16688755 DOI: 10.1002/neu.20242]
11. Chen W, Chen G. The Roles of Vitamin A in the Regulation of Carbohydrate, Lipid, and Protein Metabolism. J Clin Endocrinol Metab 2014; 3: 453-479 [PMID: 26237385 DOI: 10.1016/j.jcme.2014.04.053]
12. Blaner WS. Vitamin A signaling and homeostasis in obesity, diabetes, and metabolic disorders. Pharmacol Ther 2019; 197: 153-178 [PMID: 30703416 DOI: 10.1016/j.pharmthera.2019.01.006]
Yang FC et al. Vitamin A and lipogenesis

13 Chen W, Howell ML, Li Y, Li R, Chen G. Vitamin A and feeding statuses modulate the insulin-regulated gene expression in Zucker lean and fatty primary rat hepatocytes. *PLoS One* 2014; 9: e100868 [PMID: 25105869 DOI: 10.1371/journal.pone.0100868]

14 Li R, Chen W, Li Y, Zhang Y, Chen G. Retinoids synergized with insulin to induce Srebp-1c expression and activated its promoter via the two liver X receptor binding sites that mediate insulin action. *Biochem Biophys Res Commun* 2011; 406: 268-272 [PMID: 21363446 DOI: 10.1016/j.bbrc.2011.02.031]

15 Zhang Y, Chen W, Li R, Li Y, Ge Y, Chen G. Insulin-regulated Srebp-1c and Pck1 mRNA expression in primary hepatocytes from Zucker fatty but not lean rats is affected by feeding conditions. *PLoS One* 2011; 6: e21342 [PMID: 21731709 DOI: 10.1371/journal.pone.0021342]

16 Chen G. Roles of Vitamin A Metabolism in the Development of Hepatic Insulin Resistance. *JCR* 2013; 5: 534972 [PMID: 27335827 DOI: 10.1155/2013/534972]

17 Hodson I, Frayn KN. Hepatic fatty acid partitioning. *Curr Opin Lipidol* 2011; 22: 216-224 [PMID: 21494414 DOI: 10.1097/ML0.0b013e3283462e16]

18 Cahill GF Jr. Fuel metabolism in starvation. *Ann Rev Nutr* 2006; 26: 1-22 [PMID: 16848698 DOI: 10.1146/annurev.nutr.26.061505.111258]

19 Carey MC, Small DM, Bliss CM. Lipid digestion and absorption. *Ann Rev Physiol* 1983; 45: 651-677 [PMID: 6342528 DOI: 10.1146/annurev.ph.45.030183.003251]

20 Havel RJ. Postprandial lipid metabolism: an overview. *Proc Nutr Soc* 1997; 56: 659-666 [PMID: 9264115 DOI: 10.1079/pnm19970605]

21 Van Vranken JG, Nowinski SM, Clowers KJ, Jeong MY, Ouyang Y, Berg JA, Gygi JP, Gygi SP, Winge DR, Rutter J. ACP Acylation Is an Acetyl-CoA-Dependent Modification Required for Electron Transport Chain Assembly. *Mol Cell* 2018; 71: 567-580. e4 [PMID: 30118679 DOI: 10.1016/j.molcel.2018.06.039]

22 Maier T, Leibundgut M, Boehringer D, Ban N. Structure and function of eukaryotic fatty acid synthases. *Q Rev Biophys* 2010; 43: 373-422 [PMID: 207311893 DOI: 10.1017/s0033585510000156]

23 Zhang JY, Kothapalli KS, Bremta JT. Desaturase and elongase-limiting endogenous long-chain polyunsaturated fatty acid biosynthesis. *Curr Opin Clin Nutr Metab Care* 2016; 19: 103-110 [PMID: 26825851 DOI: 10.1097/MCO.0000000000000254]

24 Kihara A. Very long-chain fatty acids: elongation, physiology and related disorders. *J Biochem* 2012; 152: 387-395 [PMID: 22984065 DOI: 10.1093/jb/mvs105]

25 AlJohani AM, Syed DN, Ntambi JM. Insights into Stearoyl-CoA Desaturase-1 Regulation of Systemic Metabolism. *Trends Endocrinol Metab* 2017; 28: 831-842 [PMID: 29089222 DOI: 10.1016/j.tem.2017.10.003]

26 Koeberle A, Löser K, Thümmer M. Stearoyl-CoA desaturase-1 and adaptive stress signaling. *Biochim Biophys Acta* 2016; 1861: 1719-1726 [PMID: 27550503 DOI: 10.1016/j.bbadis.2016.08.009]

27 Harrison EH. Mechanisms of digestion and absorption of dietary vitamin A. *Ann Rev Nutr* 2005; 25: 87-103 [PMID: 16011460 DOI: 10.1146/annurev.nutr.25.050304.092164]

28 Bohn T, Desmarchelier C, El SN, Keijer J, van Schorsthorst E, Rühl R, Borel P. B-Carotene in the human body: metabolic bioactivation pathways - from digestion to tissue distribution and excretion. *Proc Nutr Soc* 2019; 78: 68-87 [PMID: 30747092 DOI: 10.1017/S0029665119000241]

29 D’Ambrosio DN, Clugston RD, Blaner WS. Vitamin A metabolism: an update. *Nutrients* 2011; 3: 63-103 [PMID: 21350678 DOI: 10.3390/nu3010063]

30 Iskakova M, Karbyshov M, Piskunov A, Rochette-Egly C. Nuclear and extranuclear effects of vitamin A. *Can J Physiol Pharmacol* 2015; 93: 1065-1075 [PMID: 26459513 DOI: 10.1139/cjpp-2014-0522]

31 Zhang R, Wang Y, Li R, Chen G. Transcriptional Factors Mediating Retinoid Acid Signals in the Control of Energy Metabolism. *Int J Mol Sci* 2015; 16: 14210-14244 [PMID: 26110391 DOI: 10.3390/jms160614210]

32 Gutierrez-Mazariegos J, Schubert M, Laudet V. Evolution of retinoic acid receptors and retinoic acid signaling. *Subcell Biochem* 2014; 70: 55-73 [PMID: 24962881 DOI: 10.1007/978-94-017-9050-5_5]

33 Li Y, Li R, Chen W, Chen G. Vitamin A status and its metabolism contribute to the regulation of hepatic genes during the cycle of fasting and refeeding in rats. *J Nutr Biochem* 2016; 30: 33-43 [PMID: 27012619 DOI: 10.1016/j.jnba.2015.11.012]

34 Browasey RW, Boone AN, Elliott JE, Kulpa JE, Lee WM. Regulation of acetyl-CoA carboxylase. *Biochem Soc Trans* 2006; 34: 223-227 [PMID: 16545081 DOI: 10.1042/bst0600223]

35 Kusunoki J, Kanatani A, Moller DE. Modulation of fatty acid metabolism as a potential approach to the treatment of obesity and the metabolic syndrome. *Endocrine* 2006; 39: 91-100 [PMID: 16622296 DOI: 10.1385/endo:29:1:91]

36 McGarry JD. Glucose-fatty acid interactions in health and disease. *Am J Clin Nutr* 1998; 67: 5005-504S [PMID: 9497160 DOI: 10.1093/ajcn/67.3.500S]

37 Vega VA, Anzulovich AC, Varas SM, Bonomi MR, Giménez MS, Oliveros LB. Effect of nutritional vitamin A deficiency on lipid metabolism in the rat heart: Its relation to PPAR gene expression. *Nutrition* 2009; 25: 828-838 [PMID: 19342198 DOI: 10.1016/j.nut.2009.01.008]

38 Oliveros LB, Domeniconi MA, Vega VA, Gatica LV, Brigada AM, Gimenez MS. Vitamin A deficiency modifies lipid metabolism in rat liver. *Br J Nutr* 2007; 97: 263-272 [PMID: 17298694 DOI: 10.1017/s0007114507182659]

39 Amengual J, Ribot J, Bonet ML, Palo A. Retinoic acid treatment increases lipid oxidation capacity
in skeletal muscle of mice. *Obesity (Silver Spring)* 2008; 16: 585-591 [PMID: 18239600 DOI: 10.1038/oby.2007.104]

40 Liao XD, Zhou CH, Zhang J, Shen JL, Wang YJ, Jin YC, Li SL. Effect of all-trans retinoic acid on casein and fatty acid synthesis in MAC-T cells. *Asian-Australas J Anim Sci* 2020; 33: 1012-1022 [PMID: 31480153 DOI: 10.5713/ajas.19.0315]

41 Kim JY, Lee JJ, Kim KS. Acetyl-CoA carboxylase beta expression mediated by MyoD and muscle regulatory factor 4 is differentially affected by retinoic acid receptor and retinoid X receptor. *Exp Mol Med* 2003; 35: 23-29 [PMID: 12642900 DOI: 10.1038/emm.2003.4]

42 Fischhoff SA, Papachou GC, Nickols WA. Decreased activity of acetyl-CoA carboxylase during chemically induced neutrophilic differentiation of human promyelocytic leukemia cells. *J Cell Biochem* 1984; 26: 75-81 [PMID: 6151950 DOI: 10.1002/jcb.240260203]

43 Clarke SD, Abraham S. Gene expression: nutrient control of pre- and posttranscriptional events. *FASEB J* 1992; 6: 3146-3152 [PMID: 1397836 DOI: 10.1096/fasebj.6.13.1397836]

44 Arana A, Mendizabal JA, Alzón M, Soret B, Purroy A. The effect of vitamin A supplementation on postnatal adipose tissue development of lambs. *J Anim Sci* 2008; 86: 3393-3400 [PMID: 18676724 DOI: 10.2527/jas.2008-0889]

45 Bryant TC, Wagner JJ, Tatum JD, Galvez ML, Anthony RV, Engle TE. Effect of dietary supplemental vitamin A concentration on performance, carcass merit, serum metabolites, and lipogenic enzyme activity in yearling beef steers. *J Anim Sci* 2010; 88: 1463-1478 [PMID: 20023133 DOI: 10.2527/jas.2009-231]}

46 Xu ZX, Viviano CJ, Rooney SA. Glucocorticoid stimulation of fatty-acid synthase gene transcription in fetal lung: antagonism by retinoic acid. *Am J Physiol* 1995; 268: L683-L690 [PMID: 7537465 DOI: 10.1152/ajplung.1995.268.l.683]

47 Amengual J, Ribot J, Bonet ML, Palou A. Retinoic acid treatment enhances lipid oxidation and inhibits lipid biosynthesis capacities in the liver of mice. *Cell Physiol Biochem* 2010; 25: 657-666 [PMID: 20511711 DOI: 10.1159/000315085]

48 Zhang Y, Li R, Li Y, Chen W, Zhao S, Chen G. Vitamin A status affects obesity development and hepatic expression of key genes for fuel metabolism in Zucker fatty rats. *Biochem Cell Biol* 2012; 90: 548-557 [PMID: 22554462 DOI: 10.1139/o12-012]

49 Abd Eldaim MA, Matsuoka S, Okamatsu-Ogura Y, Kamikawa A, Ahmed MM, Terao A, Nakajima KI, Kimura K. Retinoic acid modulates lipid accumulation glucose concentration dependently through inverse regulation of SREBP-1 expression in 3T3L1 adipocytes. *Genes Cells* 2017; 22: 568-582 [PMID: 28488421 DOI: 10.1111/gtc.12498]

50 Abd Eldaim MA, Okamatsu-Ogura Y, Terao A, Kimura K. Effects of retinoic acid and hydrogen peroxide on sterol regulatory element-binding protein-1a activation during adipogenic differentiation of 3T3-L1 cells. *Jpn J Vet Res* 2010; 58: 149-154 [PMID: 21180254 DOI: 10.1136/imp.c6672]

51 Murray T, Russell TR. Inhibition of adipocyte conversion in 3T3-L2 cells by retinoic acid. *J Supramol Struct* 1980; 14: 255-266 [PMID: 6164877 DOI: 10.1002/jss.400140214]

52 Morikawa K, Hanada H, Hirota K, Nonaka M, Ikeda C. All-trans retinoic acid displays multiple effects on the growth, lipogenesis and adipokine gene expression of AML-1 preadipocyte cell line. *Cell Biol Int* 2013; 37: 36-46 [PMID: 23319320 DOI: 10.1002/cbi.10005]

53 Roder K, Wolf SS, Schweizer M. Regulation of the fatty acid synthase promoter by retinoic acid. *Biochem Soc Trans* 1996; 24: 233S [PMID: 8736891 DOI: 10.1042/bst24233s]

54 Roder KH, Wolf SS, Schweizer M. Regulation of the fatty acid synthase gene by retinoic acid in HepG2 cells is mediated by an E-box-containing multihormonal response element and two neighbouring upstream sequences. *Biochem Soc Trans* 1997; 25: 157S [PMID: 9191201 DOI: 10.1042/bst025157s]

55 Roder K, Zhang L, Schweizer M. SREBP-1c mediates the retinoid-dependent increase in fatty acid synthase promoter activity in HepG2. *FEBS Lett* 2007; 581: 2715-2720 [PMID: 17531980 DOI: 10.1016/j.febslet.2007.05.022]

56 Amengual J, Petro P, Bonet ML, Ribot J, Palou A. Induction of carnitine palmitoyl transferase 1 and fatty acid oxidation by retinoic acid in HepG2 cells. *Int J Biochem Cell Biol* 2012; 44: 2019-2027 [PMID: 22871568 DOI: 10.1016/j.biocel.2012.07.026]

57 Duncan RE, Archer MC. Farnesol decreases serum triglycerides in rats: identification of mechanisms including up-regulation of PPARalpha and down-regulation of fatty acid synthase in hepatocytes. *Lipids* 2008; 43: 619-627 [PMID: 18509688 DOI: 10.1007/s11745-008-3192-3]

58 Schweizer M, Roder K, Zhang L, Wolf SS. Transcription factors acting on the promoter of the fatty acid synthase gene. *Biochem Soc Trans* 2002; 30: 1070-1072 [PMID: 12440974 DOI: 10.1042/bst0301070]

59 Roder K, Schweizer M. Retinoic acid-mediated transcription and maturation of SREBP-1c regulates fatty acid synthase via cis-elements responsible for nutritional regulation. *Biochem Soc Trans* 2007; 35: 1211-1214 [PMID: 17956315 DOI: 10.1042/bst035121]

60 Esquenat M, Swinnen JV, Van Veldhoven PP, Denef C, Heyns W, Verhoeven G. Retinoids stimulate lipid synthesis and accumulation in LNCaP prostate adenocarcinoma cells. *Mol Cell Endocrinol* 1997; 136: 37-46 [PMID: 9510065 DOI: 10.1016/s0303-7207(97)00210-4]

61 Facchin I, Ignarro RS, Rodrigues-Silva E, Vieira AS, Lopes-Cendes I, Castillo RF, Rogerio F. Toxic effects of phytol and retinol on human glioblastoma cells are associated with modulation of cholesterol and fatty acid biosynthetic pathways. *J Neuroloncol* 2018; 136: 435-443 [PMID: 29159775 DOI: 10.1007/s11060-017-2672-9]
Yang FC et al. Vitamin A and lipogenesis

62 Guillou H, Zadravec D, Martin PG, Jacobsson A. The key roles of elongases and desaturases in mammalian fatty acid metabolism: Insights from transgenic mice. *Prog Lipid Res* 2010; 49: 186-199 [PMID: 20018209 DOI: 10.1016/j.plipres.2009.12.002]

63 Reddy MRG, Asha GV, Manchirayla SK, Putcha UK, Vajreswari A, Jeyakumar SM. High-Fat Diet Elevates Liver Docosahexaenoic Acid Possibly through Over-Expression of Very Long-Chain Fatty Acid Elongase 2 in C57BL/6 Mice. *Int J Vitam Nutr Res* 2019; 89: 62-72 [PMID: 30957704 DOI: 10.1024/0300-9831/a000432]

64 Tiruvalluru M, Ananthalakshmi P, Ayyalasomayajula V, Nappanventtil G, Ayyagari R, Reddy GB. Vitamin A supplementation ameliorates obesity-associated retinal degeneration in WNN1/Ob rats. *Nutrition* 2013; 29: 298-304 [PMID: 23036575 DOI: 10.1016/j.nut.2012.06.006]

65 Oka A, Maruo Y, Miki T, Yamasaki T, Saito T. Influence of vitamin A on the quality of beef from the Tajima strain of Japanese Black cattle. *Meat Sci* 1998; 48: 159-167 [PMID: 12206288 DOI: 10.1016/s0309-1740(97)00086-7]

66 Hayashi M, Kido K, Hodate K. Microarray analysis of Longissimus thoracis muscle gene expressions in vitamin A-restricted Japanese Black steers in middle fattening stage. *Anim Sci J* 2018; 89: 88-96 [PMID: 28960613 DOI: 10.1111/asj.12898]

67 Gorocica-Buenfil MA, Fluharty FL, Loerch SC. Effect of vitamin A restriction on carcass characteristics and immune status of steers. *Anim Sci J* 2008; 86: 1609-1616 [PMID: 18344289 DOI: 10.2527/jas.2007-0241]

68 Raja Gopal Reddy M, Pavan Kumar C, Mahesh M, Sravan Kumar M, Mullapudi Venkata S, Putcha UK, Vajreswari A, Jeyakumar SM. Vitamin A deficiency suppresses high fructose-induced triglyceride synthesis and elevates resolvin D1 levels. *Biochim Biophys Acta* 2016; 1861: 156-165 [PMID: 26597784 DOI: 10.1016/j.bbalip.2015.11.005]

69 Weiss K, Mihály J, Liebsch G, Marosvölgyi T, Garcia AL, Schmitz G, Decsi T, Rühl R. Effect of high versus low doses of fat and vitamin A dietary supplementation on fatty acid composition of phospholipids in mice. *Genes Nutr* 2014; 9: 368 [PMID: 24306959 DOI: 10.1007/s12263-013-0368-0]

70 Miller CW, Waters KM, Ntambi JM. Regulation of hepatic stearoyl-CoA desaturase gene 1 by vitamin A. *Biochem Biophys Res Commun* 1997; 231: 206-210 [PMID: 9070250 DOI: 10.1006/bbrc.1997.6707]

71 Gopal Reddy MR, Kumar MS, Acharya V, Venkata SM, Putcha UK, Jeyakumar SM. Vitamin A deficiency increases the oleic acid (C18:1) levels in the kidney of high fructose diet-fed rats. *Indian J Med Res* 2019; 150: 620-629 [PMID: 30488626 DOI: 10.4103/imjr.IJMIR_1574_17]

72 Raja Gopal Reddy M, Mullapudi Venkata S, Putcha UK, Jeyakumar SM. Vitamin A deficiency induces endoplasmic reticulum stress and apoptosis in pancreatic islet cells: Implications of stearoyl-CoA desaturase 1-mediated oleic acid synthesis. *Exp Cell Res* 2018; 364: 104-112 [PMID: 29409096 DOI: 10.1016/j.yexcr.2018.01.040]

73 Zolfaghari R, Cifelli CJ, Banta MD, Ross AC. Fatty acid delta(5)-desaturase mRNA is regulated by dietary vitamin A and exogenous retinolic acid in liver of adult rats. *Arch Biochem Biophys* 2001; 391: 8-15 [PMID: 11414679 DOI: 10.1006/abbi.2001.2361]

74 Alam SQ, Alam BS. Microsomal fatty acid desaturase activities in vitamin A-deficient rat liver. *Biochim Biophys Acta* 1985; 833: 175-177 [PMID: 3967039 DOI: 10.1016/0005-2765(85)90267-x]

75 Flowers MT, Paton CM, O’Byrne SM, Schiesser K, Dawson JA, Blaner WS, Kendziorski C, Ntambi JM. Metabolic changes in skin caused by Scd1 deficiency: a focus on retinol metabolism. *PLoS One* 2011; 6: e19734 [PMID: 21573029 DOI: 10.1371/journal.pone.0019734]

76 Dziedzic B, Bewicz-Binkowska D, Zgorzynska E, Stulczewski D, Wieteska L, Kaza B, Walczewska A. DHA upregulates FADS2 expression in primary cortical astrocytes exposed to vitamin A. *Physiol Res* 2018; 67: 663-668 [PMID: 29750879 DOI: 10.3354/physres.93708]

77 Stone RL, Bernholz DA. The molecular basis for inhibition of adipose conversion of murine 3T3-L1 cells by retinoic acid. *Differentiation* 1990; 45: 119-127 [PMID: 1982997 DOI: 10.1111/j.1432-0436.1990.tb00465.x]

78 Samuel W, Kutty RK, Naginini S, Gordon JS, Prouty SM, Chandraratna RA, Wiggert B. Regulation of stearoyl coenzyme A desaturase expression in human retinal pigment epithelial cells by retinolic acid. *J Biol Chem* 2001; 276: 28744-28750 [PMID: 11397803 DOI: 10.1074/jbc.M103387200]

79 Lazarus JV, Palayew A, Carrieri P, Ekstedt M, Marchesini G, Novak K, Ratziu V, Ruhl R. Effect of high versus low doses of fat and vitamin A dietary supplementation on fatty acid composition of phospholipids in mice. *Genes Nutr* 2014; 9: 368 [PMID: 24306959 DOI: 10.1007/s12263-013-0368-0]

80 Younossi ZM. Non-alcoholic fatty liver disease - A global public health perspective. *J Hepatol* 2019; 70: 531-544 [PMID: 30414863 DOI: 10.1016/j.jhep.2018.10.033]

81 Cohen JC, Horton JD, Hobbs HH. Human fatty liver disease: old questions and new insights. *Science* 2011; 322: 1519-1523 [PMID: 21700865 DOI: 10.1126/science.1204265]

82 Musso G, Gambino R, De Michieli F, Birolì G, Premoli A, Pagano G, Bo S, Durazzo M, Cassader M. Nitrosative stress predicts the presence and severity of nonalcoholic fatty liver at different stages of the development of insulin resistance and metabolic syndrome: possible role of vitamin A intake. *Am J Clin Nutr* 2007; 86: 661-671 [PMID: 17823431 DOI: 10.1093/ajcn/86.3.661]

83 Coelho JM, Cansanção K, Perez RM, Leite NC, Padilha P, Ramalho A, Peres W. Association between serum and dietary antioxidant micronutrients and advanced liver fibrosis in non-alcoholic fatty liver disease: an observational study. *PeerJ* 2020; 8: e9838 [PMID: 32995080 DOI: 10.4103/ijmr.IJMR_1574_17]
Yang FC et al. Vitamin A and lipogenesis

10.7717/peeaj.9838

84 Lim HS, Choi J, Lee B, Kim SG, Kim YS, Yoo JJ. Association between Inflammatory Biomarkers and Nutritional Status in Fatty Liver. Clin Nutr Res 2020; 9: 182-194 [PMID: 32789148 DOI: 10.7762/cnr.2020.9.3.182]

85 Pettinelli P, Arendt BM, Teterina A, McGilvray I, Comelli EM, Fung SK, Fischer SE, Allard JP. Altered hepatic genes related to retinol metabolism and plasma retinol in patients with non-alcoholic fatty liver disease. PLoS One 2018; 13: e0205747 [PMID: 30379862 DOI: 10.1371/journal.pone.0205747]

86 Botella-Carretero JI, Balsa JA, Vázquez C, Peromingo R, Díaz-Enríquez M, Escobar-Morreale HF. Retinol and alpha-tocopherol in morbid obesity and nonalcoholic fatty liver disease. Obes Surg 2010; 20: 69-76 [PMID: 18830789 DOI: 10.1007/s11695-008-9686-5]

87 Mondul A, Mancina RM, Merlo A, Dongiovanni P, Rametta R, Montalcini T, Valenti L, Albanes D, Romeo S. PNPLA3 I148M Variant Influences Circulating Retinol in Adults with Nonalcoholic Fatty Liver Disease or Obesity. J Nutr 2015; 145: 1687-1691 [PMID: 26136587 DOI: 10.3945/jn.115.210633]

88 Liu Y, Chen H, Wang J, Zhou W, Sun R, Xia M. Association of serum retinoic acid with hepatic steatosis and liver injury in nonalcoholic fatty liver disease. Am J Clin Nutr 2015; 102: 130-137 [PMID: 25948673 DOI: 10.1394/jcn.114.105153]

89 Zhong G, Kirkwood J, Won KJ, Tjota N, Jeong H, Isoherranen N. Characterization of Vitamin A Metabolome in Human Livers With and Without Nonalcoholic Fatty Liver Disease. J Pharmacol Exp Ther 2019; 370: 92-103 [PMID: 31043436 DOI: 10.1124/jpet.119.255517]

90 Kim SC, Kim CK, Axé D, Cook A, Lee M, Li T, Smallwood N, Chiang JY, Hardwick JP, Moore DD, Lee YK. All-trans-retinoic acid ameliorates hepatic steatosis in mice by a novel transcriptional cascade. Hepatology 2014; 59: 1750-1760 [PMID: 24038081 DOI: 10.1002/hep.26699]

91 Saeed A, Bartuçi P, Heegsma J, Dekker D, Kloosterhuis N, de Bruin A, Jonker JW, van de Sluis B, Faber KN. Impaired Hepatic Vitamin A Metabolism in NAFLD Mice Leading to Vitamin A Accumulation in Hepatocytes. Cell Mol Gastroenterol Hepatol 2021; 11: 309-325. e3 [PMID: 32698042 DOI: 10.1016/j.jcmgh.2020.07.006]

92 Mori Y, Kawakami Y, Kanzaki K, Otsuki A, Kimura Y, Kanji H, Tanaka R, Tsukayama I, Hojo N, Suzuki-Yamamoto T, Kawakami T, Takahashi Y. Arachidonate 12S-lipoxygenase of platelet-type in hepatic stellate cells of methionine and choline-deficient diet-fed mice. J Biochem 2020; 168: 455-463 [PMID: 32492133 DOI: 10.1093/jb/mva9062]

93 Maguire M, Bushkołsky JR, Larsen MC, Foong YH, Tanumihardjo SA, Jefcoate CR. Diet-dependent retinoid effects on liver gene expression include stellate and inflammation markers and parallel effects of the nuclear repressor Shp. J Nutr Biochem 2017; 47: 63-74 [PMID: 28570941 DOI: 10.1016/j.jnutbio.2017.04.009]

94 Yang D, Vuckovic MG, Smullin CP, Kim M, Lo CP, Deverrick S, Yoo HS, Tintcheva M, Deng Y, Napoli JL. Modest Decreases in Endogenous All-trans-Retinoic Acid Produced by a Mouse Rbh1 Heterozygote Provoke Major Abnormalities in Adipogenesis and Lipid Metabolism. Diabetes 2018; 67: 662-673 [PMID: 29321172 DOI: 10.2337/db17-0946]

95 Tsuichiya H, Ikeda Y, Ebata Y, Kojima C, Katsumas R, Tsuruyama T, Sakabe T, Shomori K, Komeda N, Oshiro S, Okamoto H, Takubo K, Hama S, Shudo K, Kogure K, Shiota G. Retinoids ameliorate insulin resistance in a leptin-dependent manner in mice. Hepatology 2012; 56: 1319-1330 [PMID: 22531980 DOI: 10.1002/hep.25798]

96 Geng C, Xu H, Zhang Y, Gao Y, Li M, Liu X, Gao M, Wang X, Fang Y, Chang Y. Retinoid acid ameliorates high-fat diet-induced liver steatosis through sirt1. Sci China Life Sci 2017; 60: 1234-1241 [PMID: 28667519 DOI: 10.1007/s11427-016-9027-6]

97 Melis M, Tang XH, Trasino SE, Patel VM, Stummer DJ, Jessurun J, Gudas LJ. Effects of AM80 compared to AC261066 in a high fat diet mouse model of liver disease. PLoS One 2019; 14: e0211071 [PMID: 30677086 DOI: 10.1371/journal.pone.0211071]

98 Sheka AC, Adeyi O, Thompson J, Hameed B, Crawford PA, Ikramuddin S. Nonalcoholic Steatohepatitis: A Review. JAMA 2020; 323: 1175-1183 [PMID: 32207804 DOI: 10.1001/jama.2020.2290]

99 Maddrey WC. Drug-induced hepatotoxicity: 2005. J Clin Gastroenterol 2005; 39: S83-S89 [PMID: 15758665 DOI: 10.1097/01.mcg.0000155548.91524.6c]
