Research Article

Resveratrol Suppresses T0901317-Induced Hepatic Fat Accumulation in Mice

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Abstract. Liver X receptor (LXR) has been identified as a potential target for treatment of atherosclerosis and diabetes. Activation of LXR, however, is associated with increased lipogenesis and fat accumulation in the liver. The objective of the current study was to examine the effect of resveratrol on LXR activator-induced fat accumulation in liver using mice as an animal model. Three groups of C57BL/6 mice were studied. Animals in group 1 were treated with T0901317, a potent activator of LXR in mice. Animals in group 2 served as the control and were treated with carrier solution and those in group 3 were treated with T0901317/resveratrol combination. Using histochemical and biochemical methods, we demonstrate that resveratrol treatment significantly suppressed fat accumulation in the liver induced by T0901317. In addition, resveratrol completely blocked elevation of blood levels of triglyceride and cholesterol and reduced blood glucose level. Quantitative PCR analysis revealed that resveratrol treatment did not change the mRNA levels of *abca1*, *abcg1*, *cyp7a1*, *srebp-1c*, *chrebp*, and *acc* genes compared to that of animals treated with T0901317 alone but reduced *pepck* and *g6p* gene expressions. Immunohistochemistry and Western blot analyses show resveratrol treatment activated AMP-activated protein kinase (AMPK) and increased phosphorylation of acetyl-CoA carboxylase. Treatment with T0901317 on hepatocytes increased intracellular fat accumulation and this increase was suppressed by resveratrol; the suppressive effect of resveratrol was greatly repressed by Compound C which is an inhibitor of AMPK. Collectively, these data suggest that resveratrol blocks T0901317-induced lipid accumulation in the liver and can be considered for inclusion into the treatment of diseases involving activation of liver X receptor.

KEY WORDS: AMPK; fatty liver; gluconeogenesis; lipogenesis; liver X receptor; resveratrol; T0901317.

INTRODUCTION

Liver X receptor (LXR) is a nuclear receptor vital for cholesterol metabolism (1,2). It binds to cholesterol derivatives, its physiological ligands, and activates expression of its target genes including *abca1*, *abcg1*, and *cyp7a1* that are directly involved in cholesterol metabolism (2,3). Due to its pivotal role in cholesterol metabolism, LXR has been studied as a potential target for treatment of atherosclerosis in which cholesterol metabolism is deregulated (4). In addition, activation of LXR has also been considered as a strategy for treatment of diabetes because of its involvement in glucose metabolism (5,6). For example, treatment of insulin-resistant Zucker (*fa/fa*) rats with T0901317, an agonist of LXR, inhibited expression of phosphoenolpyruvate carboxykinase gene (PEPCK) and thereby significantly improved insulin sensitivity (7). It was also shown using C57BL/KsJ-obese *db/db* mice as a model that T0901317 suppresses expression of glucocorticoid receptor gene and improved the phenotype of type 2 diabetes (8). These results raise the possibility for

beneficial effects of LXR activation on glucose homeostasis and diabetes. However, a major problem observed from these studies is that LXR agonists increase lipogenesis and induce hepatic steatosis (fatty liver) (9).

Although the detailed mechanism for LXR agonist-induced fat accumulation in the liver is still not clear, it is believed that activation of LXR increases expression of *srebp-1c* and *chrebp*, two master genes responsible for lipogenesis, blood hyperlipidemia, and liver fat accumulation (10,11). It is also suggested that LXR, SREBP-1c, and chREBP synergistically regulate expression of *acc* encoding acetyl-CoA carboxylase (ACC) that converts acetyl-CoA into malonyl-CoA and provides substrates for fatty acid synthesis (12). The activity of ACC can be controlled at the transcriptional level as well as by AMP-activated protein kinase (AMPK)-mediated phosphorylation (13). Phosphorylation of ACC potently suppresses its carboxylase activity and therefore reduces lipogenesis (13). As a natural activator of AMPK, resveratrol (3,5,4'-trihydroxy-trans-stilbene) has been shown to possess an anti-lipogenic effect in HepG2 cells (14).

Resveratrol is a type of natural phenol found in the skin of red grapes and in other fruits (15). Resveratrol has also been produced by chemical and biotechnological synthesis (16,17). The anticancer, anti-inflammatory, blood glucose-lowering, and beneficial cardiovascular effects of resveratrol have been reported in mice and rats (15,18,19). In one human

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trial, extremely high doses (3–5 g) of resveratrol significantly lowered blood glucose (20). Although remaining controversial, resveratrol has been shown to be able to extend life span of elderly mice (21). More recent work has shown that resveratrol is capable of competitively inhibiting various phosphodiesterases, which results in an increase in cytosolic concentration of cAMP, which acts as a second messenger for the activation of the pathway Epac1/CaMKK β /AMPK/SIRT1/PGC-1 α (22). This rise of cAMP concentration allows an increase in oxidation of fatty acids, mitochondrial biogenesis, and mitochondrial respiration (22).

In this study, we examined the activity of resveratrol on suppression of LXR activator-induced fat accumulation in the liver of C57BL/6 mice. Using T0901317, the most potent activator of LXR, we treated animals with or without resveratrol and performed series of histochemical and biochemical analyses on treated and control animals. We found that resveratrol treatment greatly suppressed liver fat accumulation while conserving the elevated expression of genes for cholesterol metabolism and decreasing the expression of genes for gluconeogenesis. These results suggest that combination of LXR activator with resveratrol could avoid the fatty liver problem for the treatment of atherosclerosis and diabetes using LXR as the target.

MATERIALS AND METHODS

Animals and Treatments

Male C57BL/6 mice were purchased from Charles River (Wilmington, MA) and housed under standard conditions. The use of mice in this study was compliant with relevant federal guidelines and institutional policies, and the animal protocol (Protocol Number, A2011 07-Y2-A3) was approved on September 12, 2011 by the Institutional Animal Use and Care Committee of the University of Georgia, Athens, Georgia. The mice were adapted for 3 days before the experiment. T0901317 (Cayman Chemical) was dissolved in DMSO, and resveratrol (Sigma-Aldrich) was suspended in phosphate buffered saline (PBS) before administration. Three groups of mice ($n=5$ for each group) were used, including the carrier solution-treated control group, T0901317-treated group (5 mg/kg, *i.p.*), and the group with combined treatment with T0901317 (5 mg/kg, *i.p.*) and resveratrol (200 mg/kg, *p.o.*). The mice were treated daily for 5 days.

Cell Culture

Murine Hepa 1–6 (CRL-1830, ATCC, Manassas, VA) hepatocytes were maintained in Dulbecco's minimum essential medium (DMEM) supplemented with 10% fetal bovine serum, penicillin (100 U/ml), and streptomycin (100 μ g/ml) in 5% CO₂ atmosphere at 37°C. Cells were incubated with T0901317 (1 μ M) with or without resveratrol (40 μ M) and compound C (10 μ M) for 24 h and fixed by 4% neutral buffered formalin for 1 h. Neutral lipids were stained using Nile Red (19123, Sigma-Aldrich) for 5 min and nuclei were counterstained with DAPI (D8417, Sigma-Aldrich). Pictures were taken using a fluorescent microscope (ECLIPSE Ti, Nikon). For quantitative analysis of cellular triglycerides, fixed cells were stained with Oil-red O (Electron Microscopy

Sciences). The incorporated dye was extracted by adding isopropanol and determined by using a spectrophotometer at 510 nm.

Histochemistry Analysis

All animals were fasted for 4 h before being sacrificed. For haematoxylin and eosin (H&E) staining, liver samples were dissected and stored in 10% neutral buffered formalin. After fixation overnight, the samples were dehydrated and embedded into paraffin. Tissue sections were cut at 6 μ m and H&E staining was performed. For frozen section and Oil-red O staining, liver samples were frozen in liquid nitrogen and sectioned at 8 μ m using a Cryostat. The tissue sections were washed with 60% isopropanol and stained with Oil-red O for 30 min before counterstained with haematoxylin. The tissue slides were examined and photographed under an optical microscope.

Liver Triglyceride Assay

Triglyceride analysis in the liver was performed as previously described (23,24). Briefly, frozen liver samples (200–400 mg per sample) were homogenized in chloroform and methanol (2:1) and incubated overnight at 4°C. Tissue homogenates were centrifuged at 12,000 rpm for 20 min and the supernatants were collected and dried. The dried samples were then dissolved in 5% Triton-X100 and triglyceride concentration was determined using a commercial kit (TR22203, Thermo-Scientific).

Blood Chemistry Analysis

Blood samples were collected and centrifuged at 4,000 rpm for 10 min to isolate plasma. The concentrations of aspartate aminotransferase (AST), alanine aminotransferase (ALT), glucose, and triglyceride were determined using commercial kits from Thermo-Scientific. The levels of cholesterol and free fatty acids were determined using commercial kits purchased from Genzyme Diagnostics and BioAssay Systems, respectively. All assays were performed according to the protocols provided by the manufacturers.

Quantitative PCR Analysis

Total RNA was isolated using TRIZOL reagent purchased from Invitrogen (Grand Island, NY). RT-PCR was performed using a Superscript RT III enzyme kit from Invitrogen. SYBR Green was used as the detection reagent for quantification using the $\Delta\Delta$ Ct method and GAPDH mRNA level as an internal control. All primer sequences employed are listed in Table I. Melting curve analysis of all real-time PCR products was performed and shown a single DNA duplex.

Immunohistochemistry and Western Blot Analysis

For immunohistochemistry, the tissue sections were immersed in citrate buffer (pH 6.0) and processed through heat-mediated antigen retrieval. The sections were incubated in 3% hydrogen peroxide to inactivate the endogenous

Table I. Primer Sequences

Sequences of real-time PCR primers
<i>Abca1</i> -F: GCTTGTGGCCTCAGTTAAGG
<i>Abca1</i> -R: GTAGCTCAGGCGTACAGAGAT
<i>Abcg1</i> -F: CTTTCCTACTCTGTACCCGAGG
<i>Abcg1</i> -R: CGGGGCATTCCATTGATAAGG
<i>Acc</i> -F: ATGGGCGGAATGGTCTCTTTC
<i>Acc</i> -R: TGGGGACCTTGTCTTCATCAT
<i>Chrebp</i> -F: AGATGGAGAACCGACGTATCA
<i>Chrebp</i> -R: ACTGAGCGTGCTGACAAGTC
<i>Cyp7a1</i> -F: GGGATTGCTGTGGTAGTGAGC
<i>Cyp7a1</i> -R: GGTATGGAATC AACCCGTTGTC
<i>Fas</i> -F: GGAGGTGGTG ATAGCCGGTAT
<i>Fas</i> -R: TGGGTAATCCATAGAGCCCAG
<i>G6p</i> -F: CGACTCGCTATCTCCAAGTGA
<i>G6p</i> -R: GTTGAACCACTCTCCGACCA
<i>Pepck</i> -F: CTGCATAACGGTCTGGACTTC
<i>Pepck</i> -R: CAGCAACTGCCCCGACTCC
<i>Scd1</i> -F: TTCTTGCGATACACTCTGGTGC
<i>Scd1</i> -R: CGGGATTGAATGTTCTTGTCGT
<i>Srebp-1c</i> -F: GCAGCCACCATCTA GCCTG
<i>Srebp-1c</i> -R: CAGCAGTGAGTCTGCCTTGAT

PCR polymerase chain reaction

hydrogen peroxidase. Antibodies against phospho-AMPK α (Thr172) and phospho-acetyl-coA carboxylase (Ser79) were purchased from Cell Signaling Technology (Boston, MA; 2535 and 3661). For Western blot, liver samples were homogenized, centrifuged at 12,000 rpm for 20 min and protein concentrations of the supernatants were measured using Bradford assay. Proteins (50 μ g in each lane) were separated using 10% SDS-PAGE and transferred to PVDF membrane (DAB150) from Millipore (Billerica, MA). Membrane blocking was performed using 5% nonfat milk (in 20 mmol/L PBST) at room temperature for 1 h. Antibody kit (9957) was purchased from Cell Signaling Technology and used as suggested by the manufacturer. Protein bands were visualized using an ECL kit (Pierce, Rockford, IL). The films were scanned and their protein bands were quantified using Bio-Rad Quantity One 1-D analysis software.

Statistical Analysis

Statistical analysis was performed using one-way analysis of variance. The results were expressed as the mean $\pm$ SD. A p value below 0.05 ($p < 0.05$) was considered significantly different.

RESULTS

Resveratrol Suppresses LXR Activator-Induced Fat Accumulation in Mouse Liver

The effect of resveratrol on LXR activator-induced fat accumulation in the liver was examined by histochemistry and measurement of liver weight and triglyceride content in control and T0901317-treated animals with or without resveratrol administration. Results in Fig. 1a show that liver becomes bigger in T0901317-treated mice with higher lipid content as indicated by the amount of vacuoles (H&E staining) and number of bright red spots in tissue sections (Oil-red-O staining). Inclusion of

resveratrol into daily injection of T0901317 suppressed liver size increase and reduced the amount of vacuoles in liver section and the bright red structures revealed by Oil-red O staining. Quantitative analysis shows that liver weight in T0901317-treated animals increased from 4.6% in control to 6.3% body weight (Fig. 1b). Fat accumulation score is 1.3 ± 0.3 in control liver and 3.4 ± 0.4 in that of T0901317-treated animals (Fig. 1c). The triglyceride content also increased from approximately 13.7 ± 2.2 mg/g (control) to 21.3 ± 1.7 mg/g of the liver with T0901317 treatment (Fig. 1d). As evidenced by the same set of analysis, resveratrol treatment suppressed the fat accumulation induced by T0901317 with a liver weight equals to about 5% body weight, maintained the score of fat accumulation at 1.9 ± 0.4 and liver content of triglyceride at approximately 17.3 ± 1.0 mg/g of liver tissue. Collectively, these data clearly show that resveratrol suppressed T0901317-induced fat accumulation in the liver.

The Effect of Resveratrol Treatment on Blood Chemistry

Blood chemistry was analyzed to assess the effect of treatment of T0901317 with or without injection of resveratrol. Results in Fig. 2a, b show that AST and ALT level remained the same between the control and treated animals, suggesting neither T0901317 nor resveratrol caused liver damage or dysfunction. The fasting glucose concentration was 10.5 ± 1.0 , 8.4 ± 0.4 , and 6.5 ± 0.5 mM for the control group, T0901317-treated group, and group treated with T0901317/resveratrol combination, respectively (Fig. 2c). T0901317 treatment increased blood concentrations of triglyceride ($p < 0.01$) and cholesterol ($p < 0.01$) while resveratrol completely blocked the elevation (Fig. 2d, e). Neither T0901317 nor resveratrol caused significant influence on free fatty acid level in blood (Fig. 2f).

The Effect of Resveratrol Treatment on mRNA Level of the LXR Target Genes and Genes That Are Involved in Regulation of Lipogenesis in the Liver

Gene expression analysis was performed to assess the effect of different treatments on expression of the LXR target genes including *abca1*, *abcg1*, and *cyp7a1* (2,25). As shown in Fig. 3, T0901317 treatment significantly increased the expression of *abca1*, *abcg1*, and *cyp7a1* by 2.5 ± 0.3 -, 3.8 ± 0.2 - and 4.9 ± 0.5 -fold, respectively (Fig. 3a–c). Inclusion of resveratrol into the T0901317 treatment did not alter expression pattern of these three genes, indicating that resveratrol does not interfere with LXR-mediated transcription activation.

Similar analysis was performed on mRNA levels of *srebp-1c* and *chrebp*, the two master genes for lipogenesis (26). As expected, T0901317 markedly increased expression of both genes (Fig. 3d, e). Interestingly, resveratrol treatment did not have any impact on the T0901317-induced elevation of *srebp-1c* and *chrebp* expression. Compared to control animals, T0901317 treatment increased expression of *acc* by 2.7 ± 0.3 -fold, and again inclusion of resveratrol in the treatment did not suppress the elevation (Fig. 3f). The mRNA levels of *pepck* and *g6p* whose expression is tightly associated with gluconeogenesis and negatively correlated with blood glucose (27,28) were also determined. T0901317 reduced expression of *pepck* to $\sim 30\%$ and inclusion of

Fig. 1. Suppression of T0901317-induced fat accumulation in mouse liver by resveratrol

resveratrol in the treatment further decreased its expression to about 10% (Fig. 3g). T0901317 did not exert significant influence on *g6p* expression but inclusion of resveratrol decreased its transcription by 50% (Fig. 3h). Resveratrol treatment alone did not significantly change the mRNA levels of *abca1*, *abcg1*, and *cyp7a1* but slightly decreased the expression levels of *srebp-1c*, *chrebp*, *acc*, *pepck*, and *g6p* (Fig. 3i)

Fig. 2. Effect of T0901317 and combination of T0901317 and resveratrol on blood chemistry

Fig. 3. Effect of T0901317 and combination of T0901317 and resveratrol on mRNA levels of genes involving in cholesterol metabolism

Resveratrol Treatment Activates AMPK and Enhances Phosphorylation of ACC

To further investigate the effect of resveratrol on T0901317-induced elevation of lipogenesis and liver fat accumulation, we performed immunohistochemistry using antibodies against phospho-AMPK α (pAMPK, Thr172) and phospho-acetyl-CoA carboxylase (pACC, Ser79). Compared to control animals, T0901317 treatment did not change the pAMPK and pACC level. However, intense positive responses are shown in the liver sections from the mice that received combined treatment of T0901317/resveratrol (Fig. 4a), suggesting that resveratrol has significantly elevated the phosphorylation of AMPK and ACC in mouse liver. To verify such an effect, we performed Western blot and analyzed the ratio of pAMPK/AMPK and pACC/ACC. Consistent with the results of immunohistochemistry, combined treatment with T0901317 and resveratrol markedly increased the ratio of pAMPK/AMPK ($p < 0.01$) and pACC/ACC ($p < 0.01$) compared to T0901317 treatment alone (Fig. 4b, c).

Resveratrol Suppresses T0901317-Induced Fat Accumulation in Hepa 1–6 Hepatocytes and the Suppressive Effect Is Reversed by Compound C

To further explore if activation of AMPK is directly involved in the suppressive effect of resveratrol on T0901317-induced fat accumulation in the liver, we performed *in vitro* studies using Hepa 1–6 hepatocytes. Results of quantitative PCR analyses showed that T0901317 with or without resveratrol increased the mRNA levels of LXR target genes including *abca1*, *abcg1*, and *cyp7a1* (Fig. 5a). Moreover, these treatments significantly increased the expression levels of genes involving in lipogenesis including *srebp-1c*, *chrebp*, *acc*, and *fas* (Fig. 5b, c). Results of Nile Red staining showed that T0901317 increased the amount of lipid droplets in hepatocytes and this increase was markedly suppressed by the combined treatment with resveratrol (Fig. 5d). Quantitative determinations showed similar results (Fig. 5e). Importantly, the suppressive effect of resveratrol on T0901317-induced intracellular fat accumulation was greatly reversed by Compound C which is an inhibitor of AMPK (Fig. 6a), and quantitative determinations showed similar trends (Fig. 6b).

Fig. 4. Effect of T0901317 and combination of T0901317 and resveratrol on activation of ACC and AMPK in the liver

Fig. 5. Effect of resveratrol on T0901317-induced fat accumulation in Hepa 1-6 hepatocytes

Fig. 6. Suppression of resveratrol effect on T0901317-induced fat accumulation in hepatocytes by Compound C

Fig. 7. Depicted pathway for resveratrol-mediated alleviation of T0901317-induced fat accumulation in the liver. T0901317 activates LXR and regulates the expression of a set of genes involved in lipid and glucose metabolism. Resveratrol activates AMPK by increasing its phosphorylation at the post-translational level, and consequently reducing carboxylase activity of ACC and suppressing lipogenesis and fat accumulation in the liver

DISCUSSION

LXR plays an important role in cholesterol metabolism, and has been studied as a potential target for treatment of atherosclerosis in which cholesterol metabolism is deregulated (4). Activation of LXR has also been considered as a strategy for treatment of diabetes because of its involvement in glucose metabolism. We have recently shown that T0901317, a LXR activator, suppresses high-fat diet-induced obesity but induces liver steatosis (29). In the current study, we demonstrate that resveratrol suppresses T0901317-induced fat accumulation in the liver (Fig. 1), blocks the T0901317-induced elevation of blood concentration of triglyceride and cholesterol, and reduces blood glucose level (Fig. 2). Gene expression analysis shows T0901317/resveratrol treatment has no effect on expression of LXR target genes and genes involved in lipogenesis, but decreases the expression of genes involved in gluconeogenesis (Fig. 3). Results from immunohistochemistry and Western blot suggest that T0901317/resveratrol treatment activates AMPK and thereby increases phosphorylation of ACC (Fig. 4).

Due to its crucial role in regulating glucose metabolism, LXR has been recognized as a potential target for the development anti-diabetic drugs (5). Previous study by Cao *et al.* demonstrated that activation of LXR by T0901317 improved glucose tolerance in insulin-resistant Zucker rats through suppressing the expression of PEPCK, a key enzyme involved in hepatic gluconeogenesis (7). Additionally, Laffitte *et al.* also demonstrated that activation of LXR improved glucose tolerance in a murine model of diet-induced obesity and insulin resistance, mainly *via* coordinately modulating the expression of a set of genes involved in glucose metabolism (30). Consistent with these studies, our previous work demonstrated that activation of LXR by T0901317 protected mice from high-fat diet-induced insulin resistance and glucose intolerance but induced liver steatosis (29). Results summarized in this report showed that combined treatment with T0901317 and resveratrol synergistically decreased blood glucose in mice (Fig. 2c). This synergistic effect may be mediated by the suppressed hepatic gluconeogenesis because combined treatment synergistically decreased the mRNA levels of *pepck* and *g6p* (Fig. 3g, h), two key genes for gluconeogenesis in the liver. Considering this synergistic effect in lowering blood glucose, it can be speculated that combined treatment by activator of LXR and resveratrol may achieve a better effect in management of diabetes.

Resveratrol-mediated attenuation of fat accumulation in hepatocytes has been reported before (14). It was demonstrated that resveratrol attenuates fat deposition mainly by inhibiting *srebp-1c* expression. Because of this earlier study, we have included not only *srebp-1c* but also *chrebp* and *acc* genes into our gene expression analysis. Data in Fig. 3d–f clearly show that resveratrol treatment has no effect on T0901317-induced elevation of these three genes. The discrepancy between these two studies is likely resulted from the two different systems employed, one employing mice as a model and the other using established cell line under non-physiological conditions. Our data suggest that alleviation effect of resveratrol on T0901317-induced fat accumulation in the liver is achieved, at least partly, by activating AMPK. Additionally, it has been reported that resveratrol is a potent activator of SIRT1 and SIRT1 activation improves hepatic lipid metabolism (31–33). Interestingly, growing evidence suggests a tight association between SIRT1 and

AMPK signaling (34–36). The possible pathway has been depicted in Fig. 7. Upon activation by T0901317, the activated LXR enters nucleus to initiate the expression of a set of genes, including its target genes *abca1*, *abcg1*, and *cyp7a1*, thereby accelerating cholesterol transport and oxidation. Meanwhile, activated LXR suppressed expression of *pepck* and *g6p*, thus reducing gluconeogenesis and decreasing blood glucose level. In addition, LXR can also enhance the expression of *srebp-1c*, *chrebp*, and *acc* genes, three vital genes for fatty acid synthesis and lipogenesis. At enzyme level, the product of ACC enzyme, malonyl-CoA, is a potent inhibitor for CPT which is a key transporter for fatty acid in β oxidation. Therefore, activation of LXR induces blood hyperlipidemia and liver fat accumulation. Resveratrol treatment has no significant impact on the elevated expression of *srebp-1c*, *chrebp*, or *acc*, but it can activate AMPK *via* increasing its phosphorylation, thereby decreasing the carboxylase activity of ACC and suppressing lipogenesis and liver fat accumulation. Unfortunately and based on data available, we cannot exclude the possibility that SIRT1 is also involved in resveratrol-mediated alleviation of T0901317-induced hepatic fat accumulation. Apparently, more mechanistic studies are needed.

Since resveratrol treatment conserved the elevated expression of genes for cholesterol metabolism while reducing the expression of genes for gluconeogenesis, resveratrol may be considered as additional drug included for treatment of metabolic diseases such as atherosclerosis and/or diabetes in which the metabolism of cholesterol and/or glucose is deregulated. The anticipated benefits would include total preservation of the effect of activation of LXR and blockade of fat accumulation in the liver. Evidently, due to species differences between rodent models and humans, additional studies are still required to further evaluate the application of the strategy of the combined treatment in managing metabolic diseases.

In summary, we demonstrate in this study that resveratrol is able to block LXR activator-induced fat accumulation in the liver. The mechanism involves activation of AMPK through phosphorylation. Consequently, pAMPK phosphorylates ACC and suppresses lipogenesis in the liver. These results strongly suggest that inclusion of resveratrol into disease treatment using LXR activation as a target may be valuable strategy to avoid development of fatty liver and its related problems.

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