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JAZF1 can regulate the expression of lipid metabolic genes and inhibit lipid accumulation in adipocytes

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ABSTRACT

JAZF1 is a newly identified gene with unknown functions. A recent genome-wide association study showed that *JAZF1* is associated with type 2 diabetes and is highly expressed in liver and adipose tissue. Studies have demonstrated that JAZF1 is the co-repressor for nuclear orphan receptor TAK1, whereas most nuclear orphan receptor family members are involved in the regulation of lipid metabolism. Therefore, JAZF1 could be closely related to glycolipid metabolism. In this study, JAZF1 was significantly upregulated during the induced differentiation process of 3T3-L1 preadipocytes. The overexpression of JAZF1 inhibited lipid accumulation in differentiated mature 3T3-L1 adipocytes and significantly inhibited the expression of SREBPl, ACC, and FAS, which were important in lipid synthesis, while upregulating the expression of key enzyme hormone-sensitive lipase in lipoclasis. Moreover, SREBPl exhibited an inhibitory function on the expression of JAZF1. SREBP1 reversed the inhibitory action on lipid accumulation of JAZF1. SREBP1 and JAZF1 were observed to regulate each other in adipocytes. Therefore, JAZF1 could regulate the expression of particular genes related to lipid metabolism and inhibit lipid accumulation in adipocytes. This result suggests that JAZF1 may be a potential target for the treatment of diseases, such as obesity and lipid metabolism disorders.

1. Introduction

A recent genome-wide association study showed that different single-nucleotide polymorphisms (SNPs) in *JAZF1* (juxtaposed with another zinc finger gene 1) are associated with the risk of type 2 diabetes mellitus (T2DM) [1]. A study on the correlation between diabetes risk and insulin sensitivity, obesity, and insulin release performed in a Danish population showed that *JAZF1* rs86474 variant has a 0.21 kg/m^2 decreased body mass index (BMI), a 0.47 cm decreased waist circumference, and a 2.6% decreased insulin release as assessed by the BIGTT-acute insulin response (AIR) index, with statistically significant differences [2]. Another study conducted in Finland showed that risk variant in the *JAZF1* genes can increase the risk of T2DM, and the variant of *JAZF1* is associated with lower birth weight, which is one of the high risk factors for T2DM [3].

JAZF1, a newly identified gene with unknown function, is also referred to as Tip27 and ZNF802 (molecular weight of coding protein is 27 KDa and its isoelectric point is 8.83) [4]. TAK1, which is also known as nuclear receptor subfamily 2, Group C, member 2 (NR2C2), or testicular orphan nuclear receptor-4 (TR4), is a member of the nuclear orphan receptor family [5]. Studies have demonstrated that the nuclear orphan receptor is involved in metabolic regulation, embryonic development, cell differentiation, gene expression in animals, and regulation of lipid metabolism [6,7]. Mice lacking TAK1 have relatively low serum insulin-like growth factor 1 and early postnatal or perinatal hypoglycemia, which can prevent obesity, fatty liver, low-grade inflammation, insulin resistance (IR), and impaired glucose tolerance (IGT) in adipose tissue induced by age and high-fat diet (HFD) [8,9]. JAZF1 is a transcriptional repressor of TAK1, which indirectly suggests that JAZF1 is related to gluconeogenesis and insulin sensitivity. Therefore, JAZF1 may be involved in the genesis and progression of IR and T2DM. A recent study has shown that the expression level of JAZF1 in the pancreatic gland of a patient with T2DM is significantly lower than that in healthy people [10], which indicated that JAZF1 possibly has an important function in the pathogenesis of IR and T2DM. Other studies have shown that TAK1 and peroxisome proliferator-activated receptor alpha (PPARα) are co-expressed in the liver, but TAK1 has higher expression levels; TAK1 can inhibit

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transcriptional activation mediated by PPARα, and this repression appears to be mediated by competition binding with PPAR-responsive elements (PPRE) and co-activators [11]. PPARα regulates gene expression in multiple links during lipid metabolism in the liver [12,13], whereas JAZF1 is an inhibitor of TAK1, which indirectly suggests that JAZF1 may regulate lipid metabolism.

A recent study has shown that JAZFl is related to prostate cancer, endometrial stromal tumor, and T2DM [14–16], but its function and the specific pathogenic mechanisms of these diseases remain unclear. No report on its function in metabolomics is available. The 3T3-L1 adipocytes are currently recognized as an important cell model for in vitro study on glycolipid metabolism. In this study, the effect of JAZFl on the expression of genes related to glycolipid metabolism was investigated to elucidate the physiological function of JAZFl gene and its possible action mechanism on the regulation of glycolipid metabolism. This study could provide a molecular theory for the treatment of metabolic diseases, such as obesity and diabetes.

2. Materials and methods

2.1. Culture and differentiation of 3T3-L1 preadipocytes

3T3-L1 preadipocytes were obtained from American Type Culture Collection (Rockville, MD), and maintained in Dulbecco's modified Eagle's medium (DMEM; Cellgro Mediatech, Inc. Manassas, VA) that was supplemented with 10% inactivated fetal bovine serum (FBS; Cambrex, East Rutherford, NJ), 100 U/ml penicillin, and 0.1 μg/ml streptomycin at 37 °C and 5% CO_2. Before differentiation, 3T3-L1 preadipocytes (after five generations, within 10 generations) were inoculated in a six-well culture plate until cell confluence. 2 days after confluence (referred to as day 0), the induced differentiation solution (containing 1.7 mM insulin, 1 mM dexamethasone, and 0.5 mM IBMX in DMEM with 10% FBS) was changed for culture for 48 h (day 2). After that, the medium was changed to DMEM containing 1.7 mM insulin (Sigma, St. Louis, MO, USA) and 10% FBS and cultured for 48 h (day 4). The medium was then changed to DMEM containing 10% FBS for cell culture, and was changed once every 2–3 days. The differentiated mature adipocytes exhibited the accumulation of triglyceride (TG) on day 7 or 8.

2.2. Construction of JAZF1 overexpression vector and synthesis of siRNA

RNA was extracted from differentiated mature 3T3-L1 adipocytes with Trizol assay kit (Invitrogen, Auckland, New Zealand) according to the instructions of the manufacturer, followed by reverse transcription PCR to amplify the coding regions of JAZF1 and SREBP1. The primers were shown in Table 1. Then the products were digested with KpnI and EocRI (TaKaRa), cloned into pcDNA3.1 vectors, sequenced and verified. An Invitrogen Block-iT RNAi Designer was used to design and synthesize three siRNAs for targeting JAZF1 (Table 1). Meanwhile, the negative control siRNA were synthesized.

2.3. Cell transfection

The mature 3T3-L1 adipocytes were seeded into 6-well plate at 1×10^5 cells/well and incubated. Transfection was performed the next day when cell confluence reached approximately 70%. The concentration of the transfection plasmid was 2 μg/mL, and the final concentration of siRNA was 50 nM. Lipofectamine 2000 (Invitrogen) was used according to the manufacturer's instructions. The medium was changed at 4–6 h after transfection and cultured for 48 h to perform subsequent tests.

2.4. Fluorescence quantitative PCR

The cells were collected, and the total RNA was extracted using Trizol. Reverse transcription was performed with 1 μg of RNA after quantitation. Quantitative PCR was conducted using the SYBR-Green dye method (SYBR Green PCR Master Mix purchased from TOYOBO) with 100 ng of cDNA in a 20 μL system. The primers were shown in Table 2. The reaction conditions were as follows: 95 °C for 5 min; followed by 40 cycles of 95 °C for 30 s, 55 °C for 30 s, and 72 °C for 30 s. Each sample was repeated thrice, and quantitative analysis was performed by comparing the $2^{-\Delta\Delta Ct}$ values.

2.5. Western blot

Total cellular proteins were extracted by incubating cells in lysis buffer with protease inhibitors. The protein concentrations in the cell lysates were determined by BCA Protein Assay Reagent Kit (Pierce). Equal amount of proteins was loaded per lane in 8% tricine gels of SDS–PAGE. Then, separated proteins were transferred to PVDF membrane after electrophoresis. 5% nonfat milk in TBST as blocking buffer was used to block with the membranes for1 h. After that, the membranes were incubated with JAZF1 (Santa Cruz Biotech, 1:200 dilution),GLUT1 (Santa Cruz Biotech, 1:500 dilution), GLUT4 (Cell Signaling Technology, 1:1000 dilution),FAS (Santa Cruz Biotech, 1:800 dilution),ACC (Santa Cruz Biotech, 1:800 dilution),SREBP1 (Santa Cruz Biotech, 1:800 dilution),ATGL (Cell Signaling Technology, 1:1000 dilution),HSL (Cell Signaling Technology, 1:1000 dilution) and GAPDH (Novus Biologicals, 1:1000 dilution) antibodies in 5% non-fat milk overnight at 4 °C, and then anti-rabbit IgG monoclonal antibody conjugated with horseradish peroxidase (Southern Biotech) at 1:2000 dilution for 1 h at room temperature. Protein bands were detected using the West Femto system (PIERCE).

Table 1
Sequences of primers and siRNAs.

Name	Sense (5′-3′)	Antisense (5′-3′)
Primer set for JAZF1 CDS amplification	GGGGTACCGCCACC ATGACAGGCATCGCCGCC	CGGAATTC CTACTGCTGCATCTTCCTGATCATC
Primer set for SREBP1 mature peptide region amplification	GGGGTACCGCCACC ATGGACGAGCTGGCCTTCGG	CGGAATTC CTACAGGGCCAGGCGGGAGCG
siRNAs		
JAZF1-si1	AUAGCAGUGGCAGCCUUACTT	GUAAGGCUGCCACUGCUAUTT
JAZF1-si2	GAAUGGAGGGGAAGAGAAGTT	CUUCUCUUCCCCUCCAUUCTT
JAZF1-si3	GUCCGCAAACCAUUCAAAUTT	AUUUGAAUGGUUUGCGGACTT
Negative control	UUCUCCGAACGUGUCACGUTT	ACGUGACACGUUCGGAGAATT

Table 2
Primers for quantitative PCR.

Genes	Primer sequences	Product sizes (bp)	Accession No.
JAZF1	F: 5′-CACTCCACCTCGACATAG-3′ R: 5′-ACCTCTTCCTCATCATACTC-3′	117	NM_173406
GLUT1	F: 5′-GGAGAAGAAGGTCACCATC-3′ R: 5′-GAGTAGTAGAACACAGCATTG-3′	123	NM_011400
GLUT4	F: 5′-GAGAGAGCGTCCAATGTC-3′ R: 5′-CGAAGATGCTGGTTGAATAG-3′	140	NM_009204
FAS	F: 5′-GTGACCGCCATCTATATCG-3′ R: 5′-CTGTCGTCTGTAGTCTTGAG-3′	148	NM_007988
ACC	F: 5′-GCACTGACTGTAACCACATCTTC-3′ R: 5′-TTCCATAGCCGACTTCCATAGC-3′	115	NM_133360
SREBP1	F: 5′-CAGCACAGCAACCAGAAGC-3′ R: 5′-CCTCCTCCACTGCCACAAG-3′	104	NM_011480
ATGL	F: 5′-ACCAGCATCCAGTTCAAC-3′ R: 5′-CGAAGTCCATCTCTGTAGC-3′	113	NM_001163689
HSL	F: 5′-CTGAGATTGAGGTGCTGTC-3′ R: 5′-GGTGAGATGGTAACTGTGAG-3′	136	NM_010719
GAPDH	F: 5′-GCCTTCCGTGTTCCTACC-3′ R: 5′-CTTCACCACCTTCTTGATGTC-3′	96	NM_008084

2.6. Oil red O staining

For Oil red O staining, cells were washed with PBS buffer thrice and fixed with 10% paraformaldehyde at room temperature for 30 min. After washing with PBS, the cells were stained with Oil red O solution (0.6% Oil red O in isopropanol:water, 3:2) for 1 h at room temperature. The Oil-Red O staining solution was then removed, and the plates were rinsed with water and dried. The stained lipid droplets were viewed on an Olympus microscope (Tokyo, Japan). Lipids were extracted from the cells using isopropanol for 1 h and quantified by measuring absorbance at 510 nm. The results are expressed relative to control differentiated cells.

2.7. Measurement of triglyceride content in 3T3-L1 cells

To analyze the content of cellular triglycerides, the cells were washed with PBS, scraped into 200 ml PBS and sonicated for 1 min. After cells were sonicated, triglyceride contents were measured with a Triglyceride E-test kit (Wako) [17]. Normalized protein contents were measured using a Bio-Rad protein assay kit.

2.8. Statistical process

All data are represented by mean ± SD ($\bar{x} \pm s$). Group results were compared by an analysis of variance (ANOVA), followed by Duncan's test using SPSS 18.0 software. $P < 0.05$ or $P < 0.01$ was considered significant.

3. Results

3.1. JAZFl expression during the differentiation of 3T3-L1 preadipocytes

The study by Li et al. [18] found that JAZFl was relatively highly expressed in adipose tissue and liver. Therefore, JAZFl is possibly involved in regulation of glycolipid metabolism in vivo. 3T3-L1 adipocytes are currently generally accepted as an important cell model for the in vitro study on glycolipid metabolism, so 3T3-L1 was chosen as the model for the in vitro study of JAZF1 function. The expression of JAZF1 was detected in the differentiation process of 3T3-L1 preadipocytes first. The 3T3-L1 cells were collected at various typical phases during the differentiation (preadipocytes, day 0, day 2, day 5, and day 8). Fluorescent quantitative PCR and Western blot results show that the expression of JAZF1 was significantly upregulated during the induced differentiation of 3T3-L1 preadipocytes (Fig. 1A–C). However, the expression in day 5 suddenly decreased, but still higher than the 3T3-L1 preadipocytes. This result indicates that JAZF1 may have an important function in adipocytes.

3.2. JAZF1 can inhibit lipid accumulation in adipocytes

To investigate the specific function of JAZFl in adipocytes, we constructed JAZFl overexpression vector, designed and synthesized siRNAs targeting JAZFl, and then transfected them into differentiated mature 3T3-L1 adipocytes. After 72 h, the cells were collected and subjected to fluorescence quantitative PCR and Western blotting analysis. For instance, the expression of JAZFl was significantly increased after JAZFl overexpression vector transfection (Fig. 1E–G). Fluorescence quantitative PCR was used to evaluate the ability of different JAZFl siRNAs to silence JAZFl expression in vitro. The results showed that 50 nM JAZF1-si3 was the most effective silencer with suppression rate of 84.65% (Fig. 1D). So, JAZF1-si3 was used in the subsequent experiments for gene knockdown. Our results showed that the JAZFl expression was significantly suppressed after transfection with JAZF1-si3 (Fig. 1E–G).

Oil red O staining was used to test the changes in intracellular lipid accumulation in each group. The result shows that the amount of red lipid droplets significantly decreased in the cytoplasm when JAZF1 was overexpressed (Fig. 2). By contrast, the amount of red lipid droplets significantly increased in the cytoplasm when JAZF1 expression was downregulated (Fig. 2). Further detection was performed on intracellular triglycerides, and the results were consistent with those of oil red O staining test. The triglyceride content in adipose cells significantly decreased when JAZFl was overexpressed, whereas the triglyceride content in adipocytes increased when JAZFl expression was downregulated (Fig. 2I). This result shows that JAZFl could inhibit lipid accumulation in adipocytes.

3.3. Regulation of glucose transport and lipid metabolic genes by JAZF1

Given that JAZFl can affect lipid accumulation in adipocytes, it should be able to regulate the expression of specific genes related to glycolipid metabolism. GLUT1 and GLUT4 are glucose transporters that are extensively distributed and highly active [19]. When JAZFl was overexpressed, no significant changes were observed in GLUT1 and GLUT4 mRNA (Fig. 1E–G). However, the mRNA levels of GLUT1 and GLUT4 significantly decreased when the JAZF1 expression was downregulated (Fig. 1E–G). Regardless if the JAZFl expression was up- or downregulated, the difference in protein

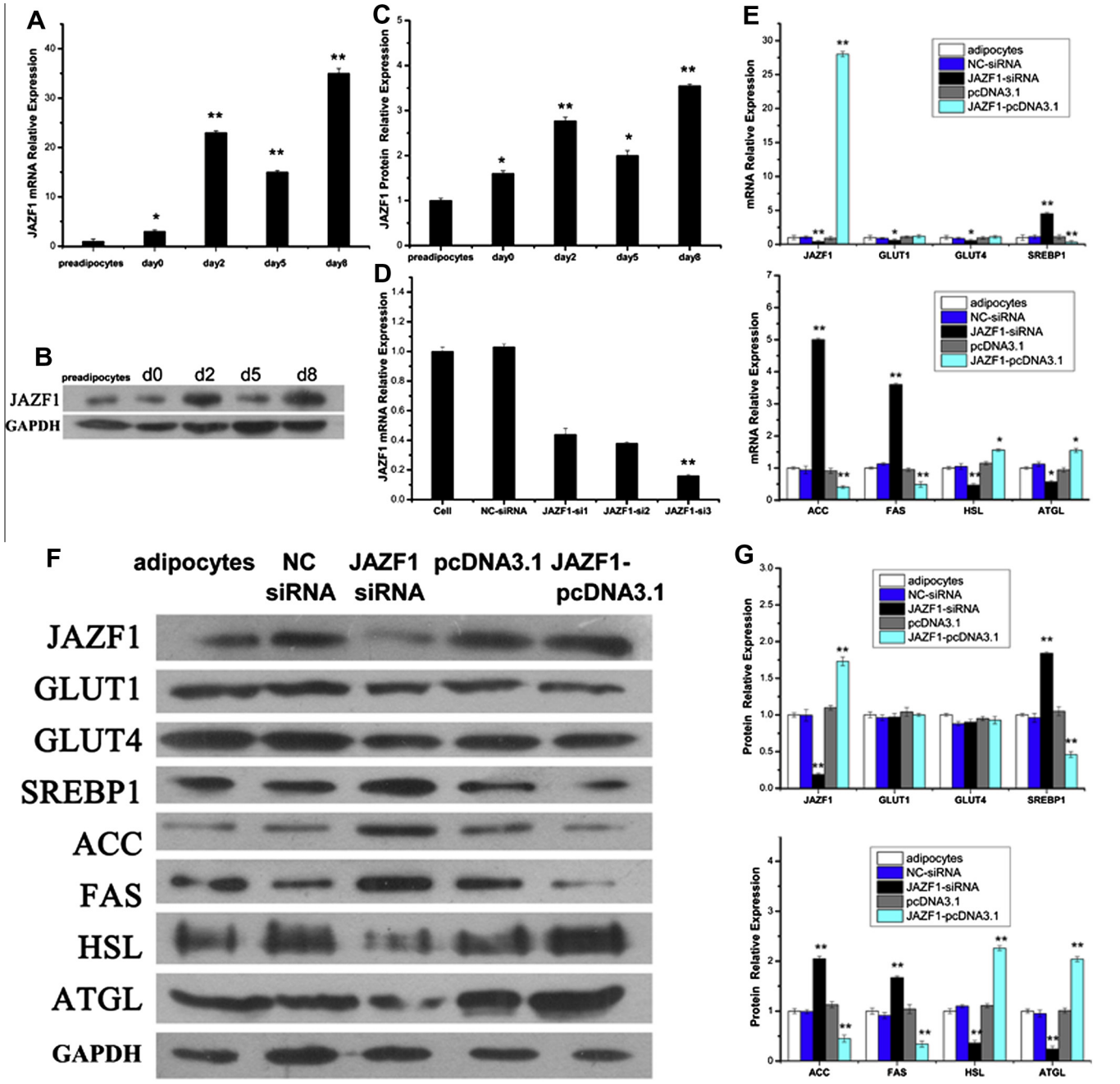

Fig. 1. Expression of JAZF1 in 3T3-L1 cells and regulation of JAZF1 on lipid metabolic genes. (A–C) Expression of JAZF1 in differentiated 3T3-L1 cells as determined by fluorescence quantitative PCR and Western blot analysis. (D) JAZF1 expression in differentiated mature 3T3-L1 adipocytes after transfection with JAZF1 siRNAs as measured by fluorescence quantitative PCR. (E–G) The differentiated mature 3T3-L1 adipocytes were transfected with pcDNA3.1, JAZF1-pcDNA3.1, JAZF1 siRNA and NC siRNA separately, and then JAZF1, GLUT1, GLUT4, FAS, ACC, mature SREBP1, ATGL and HSL expression as measured by fluorescence quantitative PCR and Western blot analysis. Experiments were carried out at least in triplicate and the results were expressed as mean values, $^{*}P < 0.05$ vs. the control, $^{**}P < 0.01$ vs. the control. GAPDH serves as a loading control.

levels between GLUT1 and GLUT4 was insignificant (Fig. 1E–G). Therefore, the regulation of JAZFl on GLUT1 and GLUT4 in adipocytes was not significant. When JAZFl was overexpressed, the mRNA and protein levels of sterol regulatory element-binding protein (SREBPl), acetyl coenzyme A carboxylase (ACC), and fatty acid synthetase (FAS) were significantly downregulated ($P < 0.01$), whereas the mRNA and protein levels of hormone-sensitive lipase (HSL) and adipocyte triglyceride lipase (ATGL) were significantly upregulated ($P < 0.05$) (Fig. 1E–G). When the expression of JAZFl was downregulated, the mRNA and protein levels of SREBPl, ACC, and FAS were significantly upregulated, whereas HSL and ATGL were significantly downregulated (Fig. 1E–G). Therefore, in adipocytes, JAZFl could regulate the expression of particular genes related to lipid metabolism, thereby affecting lipid accumulation in adipocytes.

3.4. SREBP1 can reverse JAZF1 induced lipid accumulation in adipocytes

In this study, JAZF1 could inhibit the expression of SREBP1, which is an important nuclear transcription factor that regulates fat synthesis in animals by regulating the gene expression of

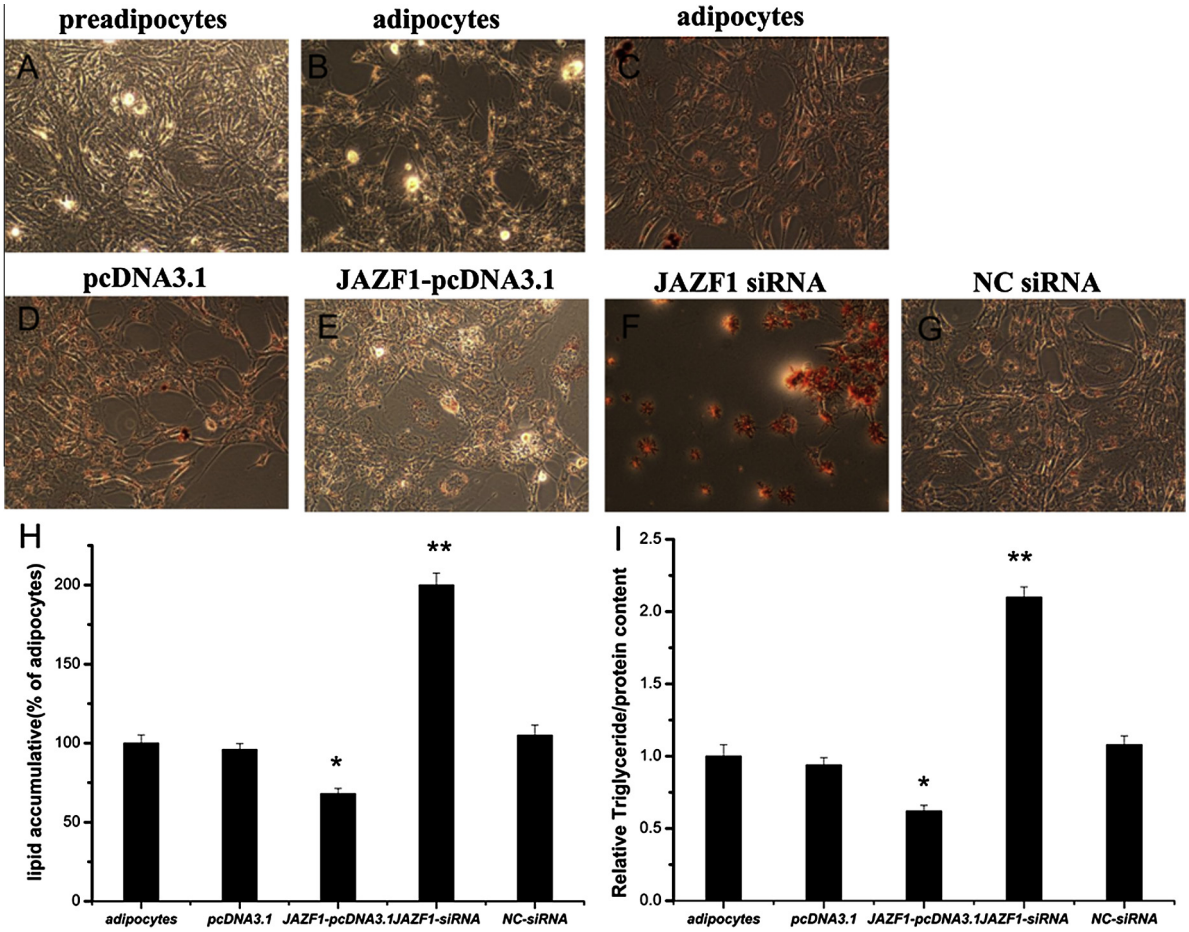

Fig. 2. Effect of JAZF1 on the lipid accumulation of 3T3-L1 adipocytes. (A) 3T3-L1 preadipocytes (100× magnification); (B) Confluent 3T3-L1 cells were exposed to differentiation cocktail for 9 days (100× magnification); (C–G) The differentiated mature 3T3-L1 adipocytes were transfected with pcDNA3.1, JAZF1-pcDNA3.1, JAZF1 siRNA and NC siRNA separately or not, and then stained with Oil Red O and examined by optical microscopy (100× magnification); (H) Data are expressed as percent lipid accumulation rate of cells transfected with vector or siRNA compared with differentiated untransfected control cells (100%); (I) Stimulation of triglyceride accumulation in 3T3-L1 cells. $^{*}P < 0.05$ versus untransfected control, $^{**}P < 0.01$ versus untransfected control.

enzymes related to lipid metabolism and directly regulate ACC and FAS [20]. Therefore, inhibition of JAZF1 on lipid accumulation in adipocytes may be realized by regulating SREBP1. When the overexpression vectors of JAZFl and SREBP1 were simultaneously transfected into adipocytes, We found that the overexpression of SREBP1 significantly increased the amount red lipid droplets in the cytoplasm of adipocytes and reversed the inhibition of JAZF1 on lipid accumulation in adipocytes (Fig. 3). Moreover, SREBP1 also exhibited a regulatory function in JAZF1, and they mutually inhibited each other (Fig. 4).

4. Discussion

Nakajima et al. [4] analyzed the expression of JAZF1 gene in human tissues via Northern blot and found that JAZF1 is expressed in multiple human tissues, but they did not detect the fat, muscle, and kidney tissues. Li et al. [18] observed that JAZFl is relatively highly expressed in adipose tissue and liver. In the present study, the expression of JAZF1 was dramatically upregulated during the induced differentiation of 3T3-L1 preadipocytes. Thus, JAZF1 may be involved in the regulation lipid metabolism in vivo. Surprisingly, the expression of JAZF1 decreased in day 5 but was still significantly higher than the 3T3-L1 preadipocytes. This result may be due to other factors that regulate the expression of JAZF1 during the differentiation process of preadipocytes.

After transfecting the JAZFl overexpression vector into differentiated mature 3T3-L1 adipocytes, JAZF1 significantly reduced the amount of red lipid droplets in the cytoplasm of adipocytes, and the triglyceride content was reduced significantly. When the expression of JAZFl was downregulated, the contents of red lipid droplets and triglyceride in the cytoplasm significantly increased. This result demonstrates that JAZFl inhibited lipid accumulation and regulated lipid metabolism in adipocytes. JAZF1 is a gene with unknown functions, and its mutation is related to diseases, such as T2DM, endometrial stromal sarcoma, and prostate cancer [14–16]. In T2DM, the expression of JAZF1 gene is downregulated [10]. JAZF1 and TAK1 are expressed in the nucleus in a very similar punctated pattern and are part of the same protein complex, which demonstrates that JAZF1 is a coregulator for TAK1 and inhibits the activation of TAK1-mediated transcription by repressing the aggregation of coactivators [4]. TAK1 is one of the nuclear orphan receptor family members, most of which are involved in regulating lipid metabolism. Studies have shown that mice lacking TAK1 can successfully prevent obesity, obesity-associated fatty liver, inflammation related to white adipose tissue, IR, and IGT induced by age and HFD [9]. Kim et al. [21] showed that the suppression of SCD1 via loss of TR4 resulted in reduced fat mass and increased insulin sensitivity with increased β-oxidation and decreased lipogenic

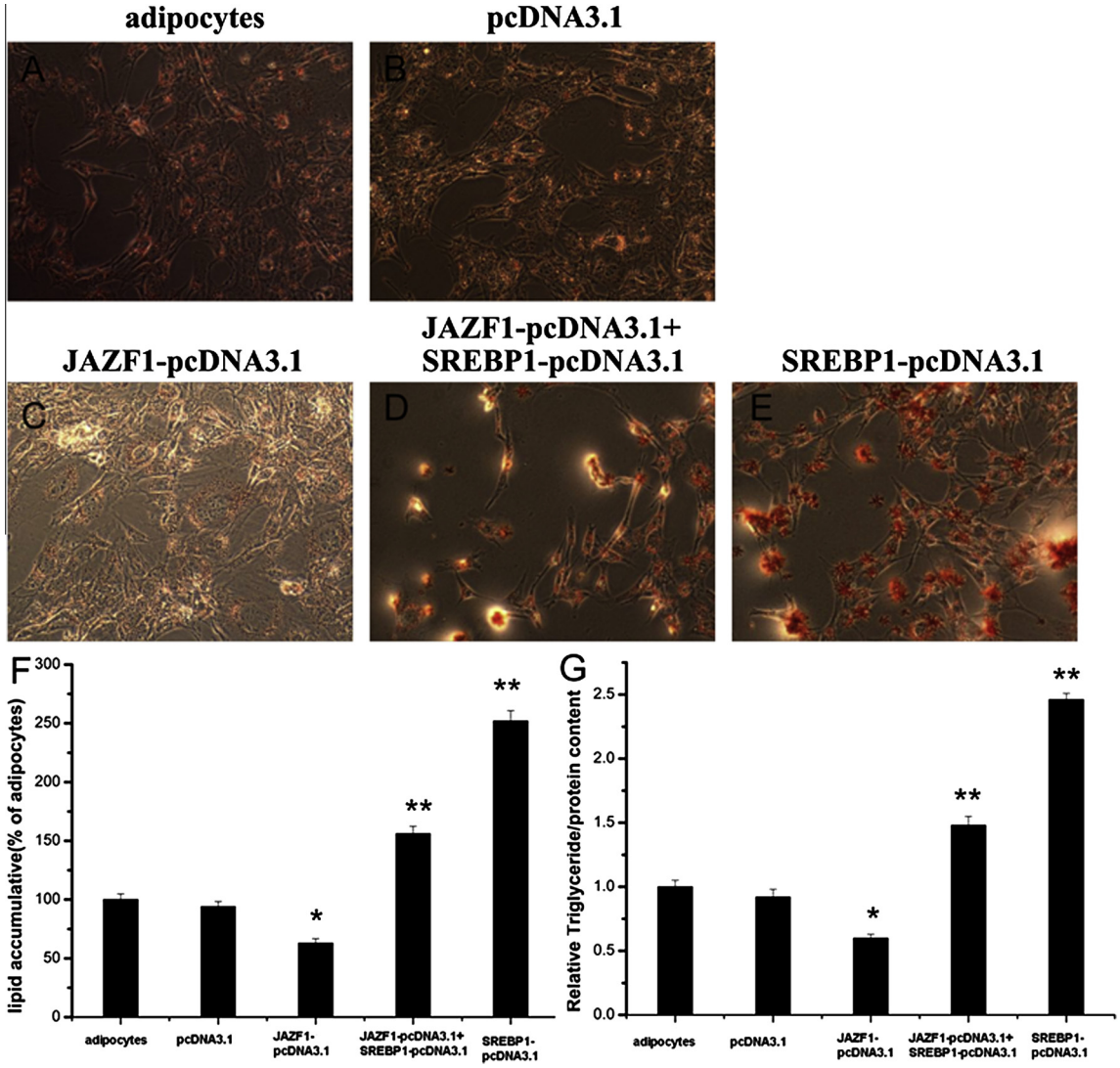

Fig. 3. Effect of JAZF1 and SREBP1 on the lipid accumulation of 3T3-L1 adipocytes. (A–E) The differentiated mature 3T3-L1 adipocytes were transfected with pcDNA3.1, JAZF1-pcDNA3.1, JAZF1-pcDNA3.1 + SREBP1-pcDNA3.1, SREBP1-pcDNA3.1 separately, and then stained with Oil Red O and examined by optical microscopy (100× magnification); (F) Data are expressed as percent lipid accumulation rate of cells transfected with vector compared with differentiated untransfected control cells (100%); (G) Stimulation of triglyceride accumulation in 3T3-L1 cells. $^{*}P < 0.05$ versus untransfected control, $^{**}P < 0.01$ versus untransfected control.

gene expression. Other studies have reported that TAK1 is involved in the regulation of gluconeogenesis and regulates the activation of PPARα-mediated transcription [7,11]. All these studies indirectly demonstrate the association of JAZF1 with lipid metabolism.

Further analysis showed that JAZFl significantly inhibited the expression of SREBPl, ACC, and FAS. SREBPs are regulators of transcription for lipid homeostasis in animal cells and directly activate the expression of multiple genes involved in the synthesis and uptake of cholesterol, fatty acid, triglyceride, and phospholipid to regulate metabolism for lipids, such as cholesterol and fatty acids [22–24]. SREBP1, which is also called adipocyte determination and differentiation factor-1, preferentially activates enzymes related to fatty acid synthesis and directly regulates ACC and FAS [20]. FAS is a basic metabolic enzyme, in which the primary product is palmitic acid, which stores energy in the form of triglycerides, and the quantity and activity of FAS are important in animal fat deposition [25]. ACC, a rate-limiting enzyme for fatty acid synthesis, catalyzes the first reaction step in fatty acid synthesis, promotes synthesis of long-chain fatty acids, and reduces its oxidation [26]. The downregulation of expression for these genes may lead to the reduction of the amount of synthesized fatty acid, thereby resulting in a decrease in the amount of synthesized triglyceride. Lipolytic enzymes are needed in the decomposition of triglycerides that are stored in fatty tissues or cells. If triglycerides do not decompose, the dynamic balance of body energy may be destroyed, which may cause obesity and IR. Both HSL and ATGL are two important rate-limiting enzymes for fat decomposition reactions [27,28]. In this study, JAZF1 could promote the expression of HSL and ATGL. Therefore, JAZFl may interfere with the signal transduction of SREBPs by downregulating the expression of SREBP1 in adipocytes. This downregulation could lead to SREBPl downstream target genes ACC and FAS synthesis decrease, and result in decreased amount of synthesized lipid. Meanwhile, lipolysis was enhanced by increasing HSL and ATGL expression. Li et al. [18] showed that JAZF1 regulates the expression of several genes of lipid metabolism, which was consistent with our findings. However, only the changes in mRNA levels caused by the overexpression of JAZF1 were tested in this study. The changes in protein levels and the effect caused by downregulation of JAZF1 expression were not investigated.

JAZFl not only regulates the expression of SREBPl, but the latter could also inhibit the expression of JAZFl. The overexpression of

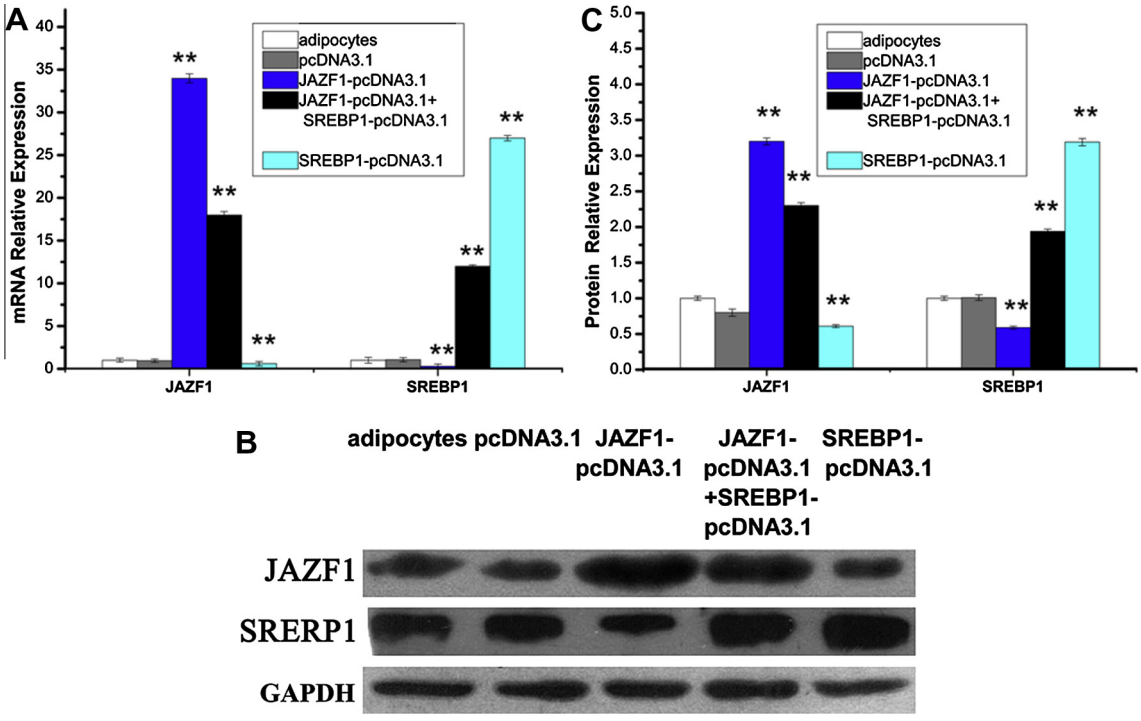

Fig. 4. Expression of JAZF1 and SREBP1 in 3T3-L1 cells. The differentiated mature 3T3-L1 adipocytes were transfected with pcDNA3.1, JAZF1-pcDNA3.1, JAZF1-pcDNA3.1 + SREBP1-pcDNA3.1, SREBP1-pcDNA3.1 separately, and then JAZF1 and mature SREBP1 expression as measured by fluorescence quantitative PCR and Western blot analysis. The results are presented as mean ($n = 3$) ± SD, $^{*}P < 0.05$ vs. the control, $^{**}P < 0.01$ vs. the control. GAPDH serves as a loading control.

SREBPl could reverse the inhibition on lipid accumulation in adipocytes caused by the upregulation of JAZFl expression. This result indicates that JAZFl and SREBPl regulated each other in adipocytes and mutually regulated the differentiation of adipocytes. Adipose tissue is not only a simple energy-storing organ but is also a tissue and organ with important endocrine and immune functions, which can secrete various adipocytokines, including adiponectin, leptin, visfatin, and resistin, whereas adipocyte factors have an important function in regulating the balance in energy metabolism for the entire body [29,30]. JAZFl was highly expressed in fatty tissue, but its effects of reducing lipid synthesis, enhancing lipolysis, and improving lipid accumulation suggest that it may provide a potential target for the treatment of obesity and lipid metabolism disorders.

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