Life Sciences 224 (2019) 197–203

Contents lists available at ScienceDirect

Life Sciences

journal homepage: www.elsevier.com/locate/lifescie

AMPK/FOXO1 signaling pathway is indispensable in visfatin-regulated myosin heavy chain expression in C2C12 myotubes

Li Na Zhou[a,b,1], Ying Ni Lin[a,1], Chen Juan Gu[a], Jian Ping Zhou[a], Xian Wen Sun[a], Xiao Ting Cai[b], Juan Du[b], Qing Yun Li[a,*,2]

[a] Department of Respiratory and Critical Care Medicine, Ruijin Hospital, Shanghai Jiao Tong University School of Medicine, Shanghai 200025, China
[b] Department of Respiratory Medicine, Ruijin Hospital North, Shanghai Jiao Tong University School of Medicine, Shanghai 281000, China

ARTICLE INFO

Keywords:
Obesity
Visfatin
Myosin heavy chain
AMPK/FOXO1 signaling pathway

ABSTRACT

Objective: Few studies have addressed the effects of visfatin on skeletal muscle remodeling. The aim of the study was to investigate the effects of visfatin on the expressions of myosin heavy chain (MHC) and its isoforms, the major indicator of fiber types and contractile properties of skeletal muscle.

Materials and methods: Levels of MHC, MHC I, MHC IIa, MHC IIb, adenosine 5′-monophosphate (AMP)-activated protein kinase (AMPK), p-AMPK and forkhead box protein O1 (FOXO1) were tested in visfatin-treated C2C12 myotubes. C2C12 myotubes were treated with visfatin combined with AMPK inhibitor or AMPK activator to investigate the role of AMPK in visfatin-mediated MHC expression. FOXO1 was overexpressed or knocked down in C2C12 myotubes to explore the role of FOXO1 in visfatin-mediated MHC expression.

Results: Compared with the vehicle group, treatment with 5 μg/ml visfatin increased the levels of total MHC and its isoforms, MHC I, MHC IIa and MHC IIb, by 1.93, 1.84, 1.80, and 1.92 folds, respectively (all p = 0,001). Visfatin suppressed AMPK phosphorylation and decreased FOXO1 expression in C2C12 myotubes. The effects of visfatin on MHC I and MHC IIa expression were canceled by AMPK activator AICAR. FOXO1 overexpression minimized the visfatin-induced upregulation of MHC I, MHC IIa and MHC IIb. The effect of AMPK activator AICAR on MHC and its isoforms expression was minimized by knockdown of FOXO1.

Conclusions: The findings revealed that visfatin promoted expressions of MHC and its isoforms in C2C12 myotubes via suppressing AMPK/FOXO1 signaling pathway.

1. Introduction

Adipose tissue has been identified as an active endocrine organ that secretes a wide spectrum of cytokines, namely adipokines. Visfatin was originally recognized as a cytokine-like molecule termed pre-B cell enhancing factor [1], and was firstly reported by Fukuhara et al. in 2005 as a novel adipokine [2], which is abundantly expressed in visceral fat and regulates glucose metabolism. Since then, studies on biological functions of visfatin have emerged. Plasma visfatin level is elevated in patients with type 2 diabetes mellitus, polycystic ovary syndrome, or cardiovascular diseases [3,4]. We previously showed that plasma visfatin levels positively correlated with apnea-hyponea index (AHI) in patients with obstructive sleep apnea (OSA) [5].

Given that obesity is a major public health problem and responsible for many diseases, secretion of adipokines may be altered in dysfunctional adipose tissues and account for some obesity-associated diseases. It is well-established that obesity could induce skeletal muscle remodeling and insulin resistance [6,7]. Obesity is related to the switch from slow to fast muscle fiber type, which ultimately affects skeletal muscle contraction and relaxation [8]. Adipokines, including leptin, resistin, visfatin and adiponectin have been reported to affect skeletal muscle insulin sensitivity [9]. Leptin and adiponectin could prevent skeletal muscle wasting via anti-muscle inflammation and modulation of skeletal muscle myogenesis, which indicate a key role of adipokine in muscle remodeling [10,11].

However, the effects of visfatin on skeletal muscle remodeling are still unknown. Thus, in this study, we aimed to elucidate the effects of visfatin on expressions of myosin heavy chain (MHC), which is a major indicator of skeletal muscle fiber types and functional property, and to explore the potential mechanism.

* Corresponding author at: Department of Respiratory Medicine, Ruijin Hospital, Shanghai Jiao Tong University School of Medicine, Shanghai 200025, China.
E-mail address: liqingyun68@hotmail.com (Q.Y. Li).
[1] Co-first author.
[2] Address: No. 197 Rui Jin Er Road, Shanghai, China.

https://doi.org/10.1016/j.lfs.2019.03.060
Received 12 January 2019; Received in revised form 9 March 2019; Accepted 25 March 2019
Available online 26 March 2019

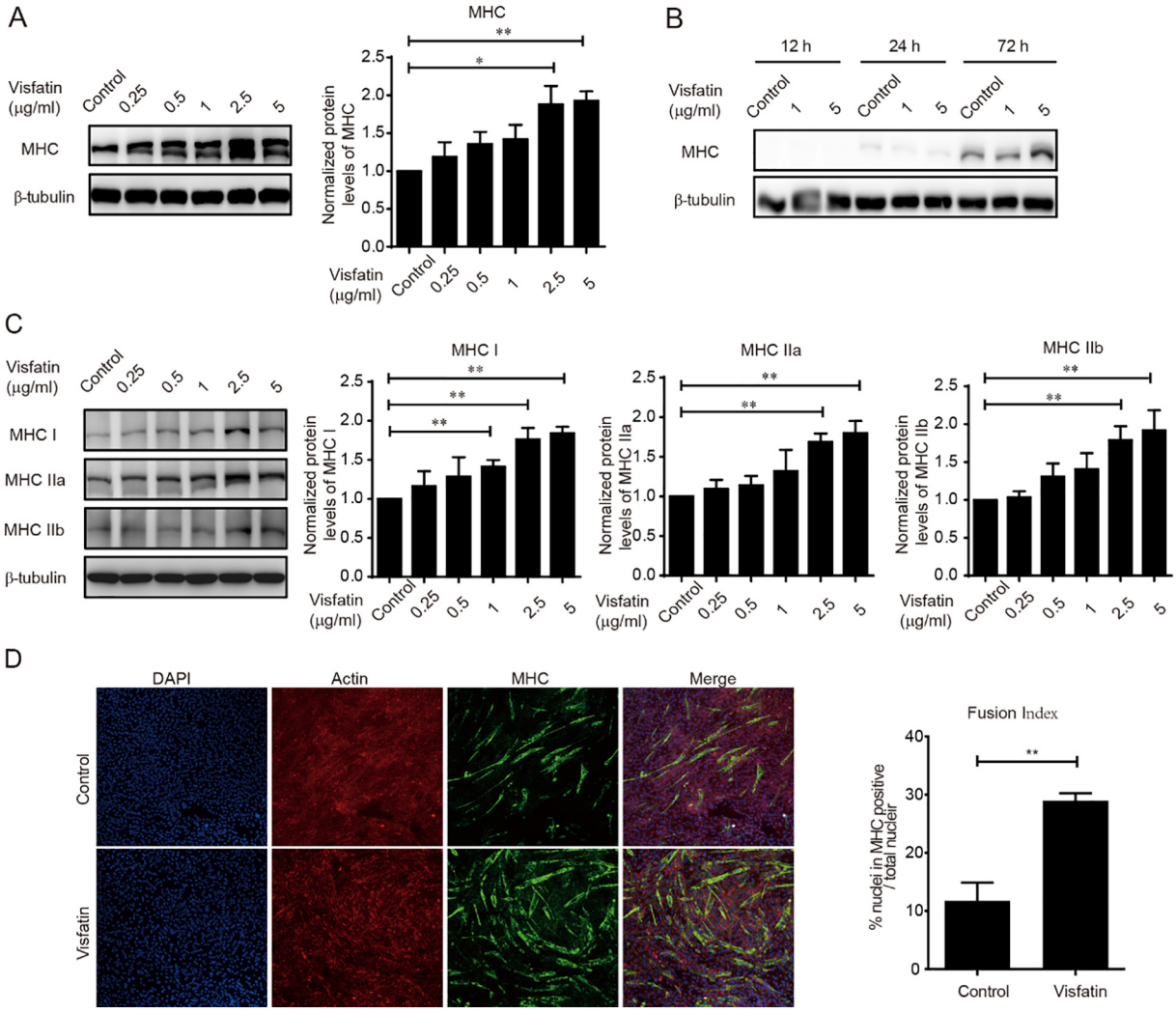

Fig. 1. Visfatin promotes expressions of MHC and its three isoforms.
(A) Visfatin upregulated the protein level of MHC in a dose-dependent manner in C2C12 myotubes. Representative Western blots are shown with densitometry of MHC/β-tubulin ($n = 3$).
(B) Upregulation of MHC protein level by visfatin was observed for 72 h. Representative Western blots are shown.
(C) Immunofluorescence staining with antibody against actin and MHC showed that visfatin (control, 5 μg/ml) increased MHC level in C2C12 myotubes. Representative images in the left panels are shown the ratio of MHC-positive cells to DAPI-positive cells in the right panel;
(D) Visfatin increased MHC I, MHC IIa and MHC IIb protein levels in C2C12 myotubes. Representative Western blots are shown with densitometry of MHC I/β-tubulin, MHC IIa/β-tubulin, MHC IIb/β-tubulin ($n = 3$).
Mean and SEM are plotted. *$P < 0.05$ and **$P < 0.01$ by ANOVA or Student's *t*-test.

2. Method

2.1. Reagents

C2C12 cells were obtained from Cell Bank of the Chinese Academy of Sciences (Shanghai, China). Antibodies against the following proteins were used in the present study: β-tubulin, AMPKa2, phospho-AMPKa2 (Thr172), acetyl-CoA carboxylase (ACC), phospho-ACC (Ser79), FOXO1 (Cell Signaling Technology, USA); MyoG, MHC, MHC I, MHC IIa and MHC IIb (Developmental Studies Hybridoma Bank, USA); β-actin (Shanghai Harmonious One Biotechnology, China). Compound C and AICAR were obtained from Selleck Chemicals (USA). Visfatin was purchased from Biovision (USA). The DAPI, Alexa Fluor® 488 goat anti-mouse IgG (H + L) and Alexa Fluor® 647 goat anti-rabbit IgG (H + L) were obtained from Invitrogen (USA).

2.2. Cell culture

C2C12 mouse myoblasts were cultured in Dulbecco's modified eagle medium (DMEM) supplemented with 10% fetal bovine serum (FBS) (growth medium, GM) (37 °C, 5% CO_2). For differentiation, when myoblasts were > 70–80% confluence, the medium was switched to DMEM with 2% horse serum (HS) (differentiation medium, DM) treated with visfatin, compound C, or AICAR.

2.3. Western blot analysis

C2C12 cells were differentiated in 6-well plates in the presence of indicated treatments. Myotubes were washed twice with ice-cold PBS and lysed with 110 μl per well of SDS lysis buffer [1 M Tris–HCl (pH 7.5), 10% SDS]. The samples were incubated at 100 °C for 10 min, and the resulting supernatants were collected for western blot analysis. The protein concentrations of lysates were measured using Byotime Protein Assay Reagent (Beyotime Institute of Biotechnology, China).

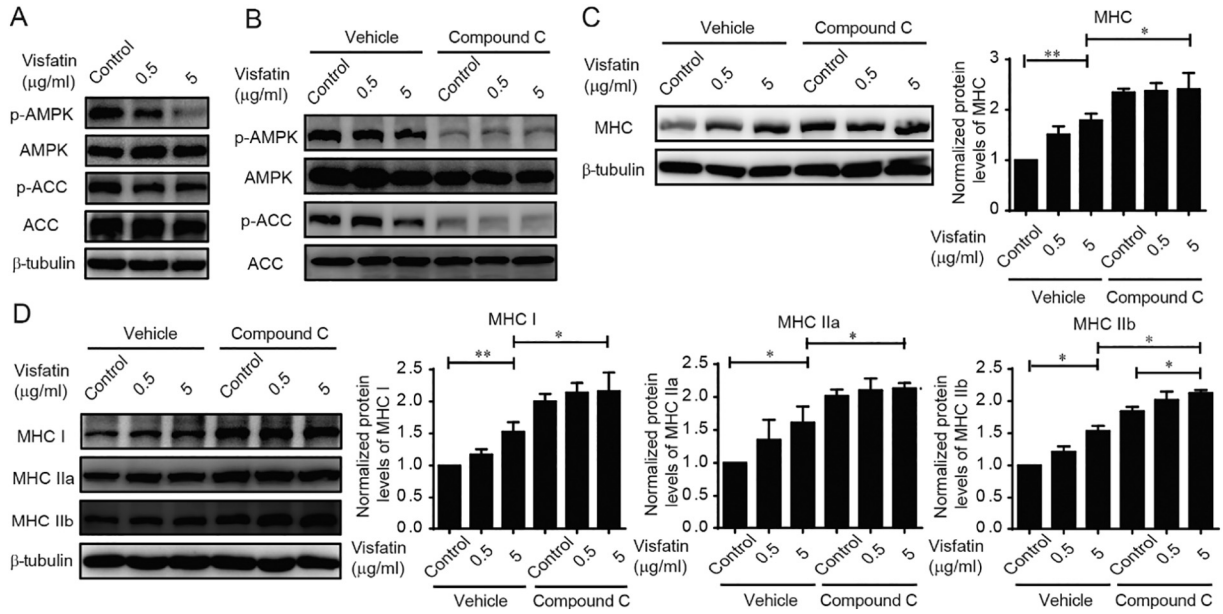

Fig. 2. The effect of visfatin on the expressions of total MHC and its isoforms in AMPK-inhibited C2C12 myotubes.
(A) Visfatin decreased the phosphorylation of AMPK and ACC in C2C12 myotubes. Representative Western blots are shown.
(B) Compound C (1 μM), which is an inhibitor for AMPK, suppressed the phosphorylation of AMPK and ACC, and addition of visfatin did not show additive effects in C2C12 myotubes. Representative Western blots are shown.
(C) Compound C (1 μM) increased MHC expression, and visfatin did not show additive effects in C2C12 myotubes. Representative western blots are shown with densitometry of MHC/β-tubulin ($n = 3$).
(D) Combination of visfatin and compound C, compared to compound C alone, significantly upregulated protein level of MHC IIb but not that of MHC I and MHC IIa. Representative western blots are shown with densitometry of MHC I/β-tubulin, MHC IIa/β-tubulin, MHC IIb/β-tubulin ($n = 3$).
Mean and SEM are plotted. $^{*}P < 0.05$ and $^{**}P < 0.01$ by ANOVA.

The supernatants were separated by 8% SDS–PAGE gels and then transferred onto PVDF membranes. The membranes were incubated with primary antibodies overnight at 4 °C. Membranes were washed with TBS/0.1% Tween-20 three times and incubated with secondary antibodies for 1 h at room temperature. Proteins were visualized with chemiluminescence using the LAS-4000 Western Blotting Detection System (FUJIFILM, Japan). Densitometric analysis was performed with ImageJ Software (National Institutes of Health, Bethesda, MD, USA).

2.4. Immunofluorescence analysis

Myotubes were fixed in 4% paraformaldehyde for 30 min, washed with PBS for 3 times, and incubated with 0.2% TritonX for 30 min at room temperature. Then the cells were incubated with anti-MHC antibody at 4 °C overnight. After washing with PBS and following incubation with Alexa Fluor® 488 goat anti-mouse IgG (H + L) and Alexa Fluor® 647 goat anti-rabbit IgG (H + L), cells were incubated with DAPI for 10 min and then observed under confocal laser scanning fluorescence microscope. Fusion index was determined by calculating the ratio of MHC-positive cells to DAPI-positive cells [12].

2.5. Construction of retrovirus vectors and viral packaging

Mouse FOXO1 cloned in MigR1 vector (MigR1-FOXO1 vector) was provided from Shanghai Institution of Hematology. To produce retroviral particles, MigR1-FOXO1 vector or empty MigR1 vector (as control) with ECOR1 packaging plasmid were co-transfected into 293T cells using Lipofectamine® (Thermo Fisher Scientific, USA) according to the manufacturer's instructions. The minimum infection dose (MID) of the purified virus was determined in 293T cells using serial dilution method. Viral concentrate was added to C2C12 myoblast in 6-well plates for 24 h. Then the culture medium was changed to DM for subsequent experiments.

2.6. Construction of shRNA lentivirus vectors and viral packaging

Recombinant lentivirus expressing short hairpin RNA (shRNA) were constructed using pLVX-shRNA2 cloning vector. The expression of shRNA is controlled by the human U6 promoter. To produce viral particles, FOXO1 shRNA vector or scramble shRNA vector (as control) with psPAX2 packaging plasmid and pMD2.G envelope expressing vector were co-transfected into 293T cells using Lipofectamine® (Thermo Fisher Scientific, USA) according to the manufacturer's instructions. After minimum infection dose (MID) of the purified virus was determined in 293T cells using serial dilution method, viral concentrate was added to C2C12 myoblast in 6-well plates for 24 h. Then the culture medium was changed to DM for subsequent experiments.

2.7. Statistical analysis

All results were derived from at least three independent experiments. The data were expressed as the mean ± SEM and were analyzed using analysis of variance (ANOVA), or a standard two-tailed Student's *t*-test. *P*-values < 0.05 were considered significant.

3. Results

3.1. Visfatin promotes the expressions of MHC and its isoforms

We treated C2C12 skeletal muscle cells with visfatin at different concentrations of 0, 0.25 μg/ml, 0.5 μg/ml, 1 μg/ml, 2.5 μg/ml, 5 μg/ml. Total MHC protein was significantly upregulated by visfatin at the concentrations of 2.5 μg/ml and 5.0 μg/ml (Fig. 1A). We also observed a time-dependent effect of visfatin on the upregulation of MHC protein (Fig. 1B). After 72-hour treatment, MHC isoforms, MHC I, MHC IIa and MHC IIb, were induced by visfatin in a dose-dependent manner (Fig. 1C). The upregulation of MHC by visfatin in C2C12 myotubes was further confirmed by immunofluorescence staining (Fig. 1D).

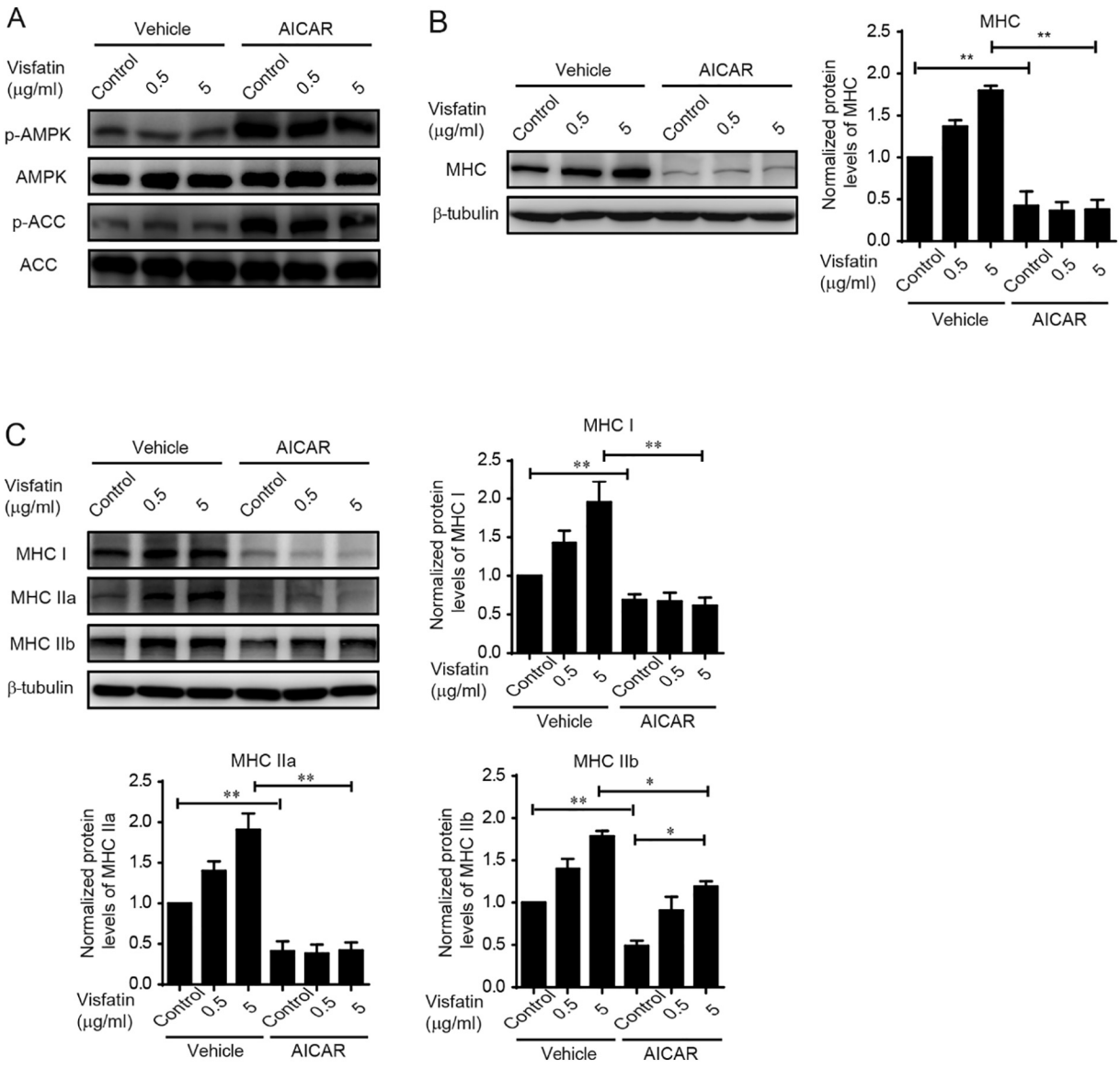

Fig. 3. The effect of visfatin on the expressions of total MHC and its isoforms in AMPK-activated C2C12 myotubes.
(A) AICAR (0.5 μM), which is an activator for AMPK, promoted the phosphorylation of AMPK and ACC in C2C12 myotubes. Representative Western blots are shown; (B) AICAR (0.5 μM) downregulated the protein level of MHC in C2C12 myotubes and minimized the effect of visfatin on increasing MHC level. Representative Western blots are shown with densitometry of MHC/β-tubulin ($n = 3$);
(C) AICAR (0.5 μM) reduced the protein levels of MHC I, MHC IIa and MHC IIb in C2C12 myotubes. The effects of visfatin on MHC I and MHC IIa but not on MHC IIb were canceled by addition of AICAR. Representative Western blots are shown with densitometry of MHC I/β-tubulin, MHC IIa/β-tubulin, MHC IIb/β-tubulin ($n = 3$); Mean and SEM are plotted. $^*P < 0.05$ and $^{**}P < 0.01$ by ANOVA.

3.2. Visfatin increases MHC expressions via suppressing AMPK activity

AMP-activated protein kinase (AMPK) is reported to be involved in the regulation of MHC expression. Upon glucose restriction, C2C12 myotubes failed to differentiate properly and expression of MHC was repressed via AMPK activation [12]. Thus, we investigated whether AMPK mediates the effect of visfatin on inducing MHC expression. We observed that phosphorylation of AMPK and acetyl-CoA-carboxylase (ACC) were suppressed by visfatin in C2C12 myotubes (Fig. 2A). We then treated C2C12 myotubes with compound C, which is an inhibitor of AMPK. Compound C inhibited the phosphorylation of AMPK and ACC (Fig. 2B), but promoted the expressions of total MHC (Fig. 2C) and its isoforms in C2C12 myotubes (Fig. 2D). Addition of visfatin further increased MHC IIb expression, but not MHC I and MHC IIa expressions (Fig. 2C and D).

We further treated C2C12 myotubes with the AMP mimetic 5-aminoimidazole-4-carboxamide-1-beta-D-ribofuranoside (AICAR), which is a specific activator of AMPK. As expected, AICAR increased the phosphorylation of AMPK and ACC (Fig. 3A), but decreased expressions of total MHC (Fig. 3B) and its isoforms (Fig. 3C). In C2C12 myotubes co-treated with AICAR and visfatin, the effect of visfatin on increasing the expressions of MHC I and MHC IIa was minimized by AICAR (Fig. 3B and C), but still existed on MHC IIb expression (Fig. 3C).

3.3. FOXO1 is required in AMPK-mediated regulation of MHC and its isoforms expression

Transcriptional factor forkhead box protein O1 (FOXO1) plays a pivotal role in controlling the activation of ubiquitin-proteasomal system and autophagic/lysosomal pathways, which can decrease MHC protein level and cause muscle atrophy [13,14]. We observed that FOXO1 expression was significantly downregulated by visfatin

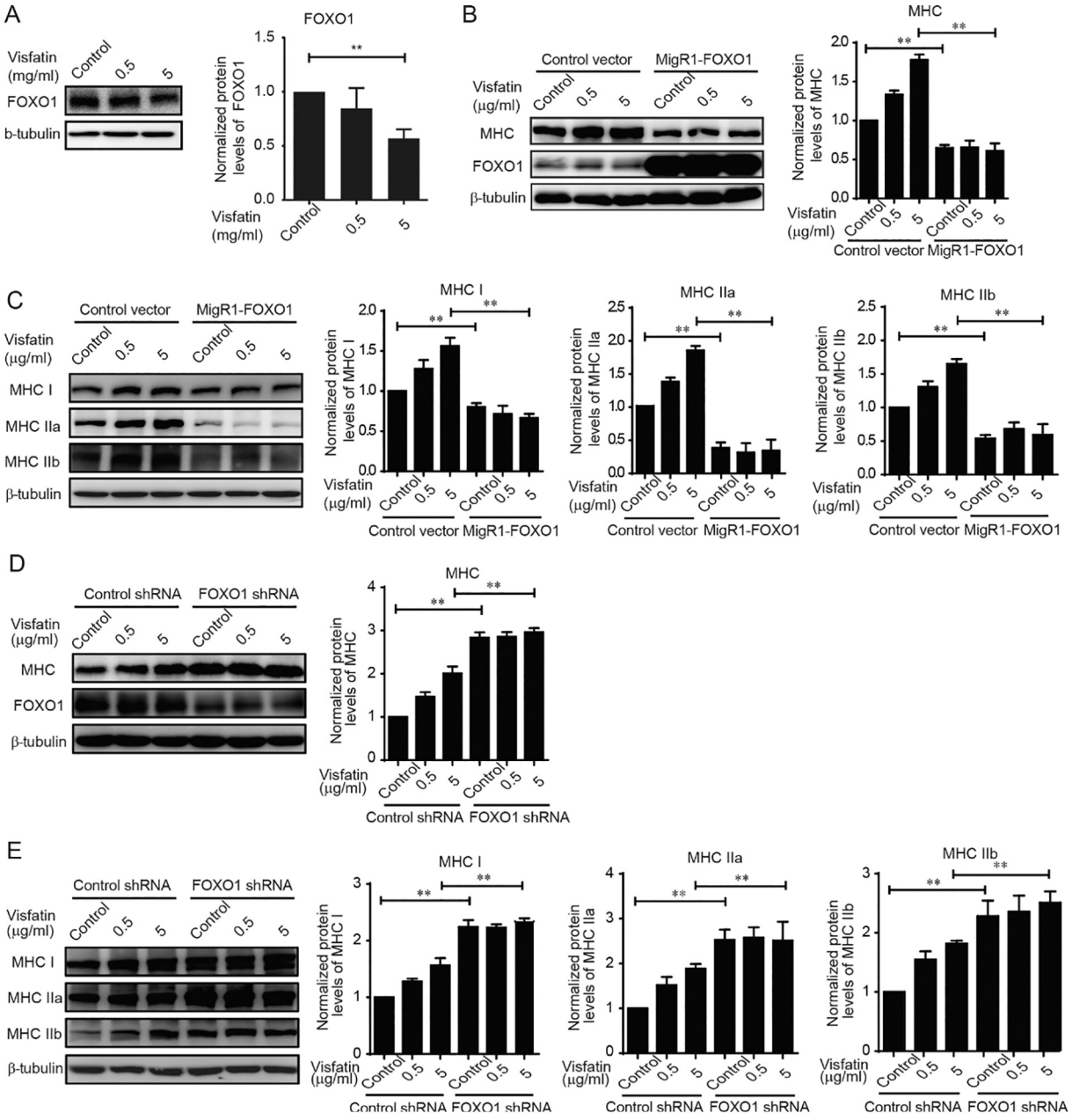

Fig. 4. Visfatin increases MHC expressions via decreasing FOXO1 level.

(A) Visfatin decreased FOXO1 protein level in C2C12 myotubes. Representative Western blots are shown with densitometry of FOXO1/β-tubulin ($n = 3$);

(B) Overexpression of FOXO1 downregulated the protein level of MHC in C2C12 myotubes and abolished the effects of visfatin on increasing MHC level. Representative Western blots are shown with densitometry of MHC/β-tubulin ($n = 3$);

(C) Visfatin could not further increase protein expressions of MHC I, MHC IIa and MHC IIb in FOXO1-overexpressed C2C12 myotubes. Representative Western blots are shown with densitometry of MHC I/β-tubulin, MHC IIa/β-tubulin, MHC IIb/β-tubulin ($n = 3$);

(D) Knockdown of FOXO1 upregulated the protein level of MHC in C2C12 myotubes, and addition of visfatin did not show additive effects in C2C12 myotubes. Representative Western blots are shown with densitometry of MHC/β-tubulin ($n = 3$);

(E) Visfatin failed to promote protein expressions of MHC I, MHC IIa and MHC IIb in FOXO1-knockdown C2C12 myotubes. Representative Western blots are shown with densitometry of MHC I/β-tubulin, MHC IIa/β-tubulin, MHC IIb/β-tubulin ($n = 3$);

Mean and SEM are plotted. $^{*}P < 0.05$ and $^{**}P < 0.01$ by ANOVA.

(Fig. 4A). We then overexpressed FOXO1 in C2C12 myotubes by retrovirally transfecting C2C12 myotubes with MigR1-FOXO1 vector, and the transfection efficiency of FOXO1 was confirmed by Western Blotting (Fig. 4B). In FOXO1-overexpressed C2C12 myotubes, total MHC and its isoforms expressions were reduced, which could not be reversed by visfatin treatment (Fig. 4B and C). Then, we knocked down FOXO1 by a lentivirus vector expressing a short hairpin RNA (shRNA) specifically targeting the FOXO1 mRNA. We confirmed the knockdown efficiency of FOXO1 and upregulation of MHC in FOXO1-knockdown myotubes (Fig. 4D). Knockdown of FOXO1 increased MHC isoforms expressions, and addition of visfatin treatment did not show an additive effect (Fig. 4E). The results indicated that the effects of visfatin on MHC expression required FOXO1.

Given that AMPK and FOXO1 were both involved in visfatin-induced expressions of MHC and its isoforms, and FOXO1 could be a downstream target of AMPK, we then investigated whether AMPK

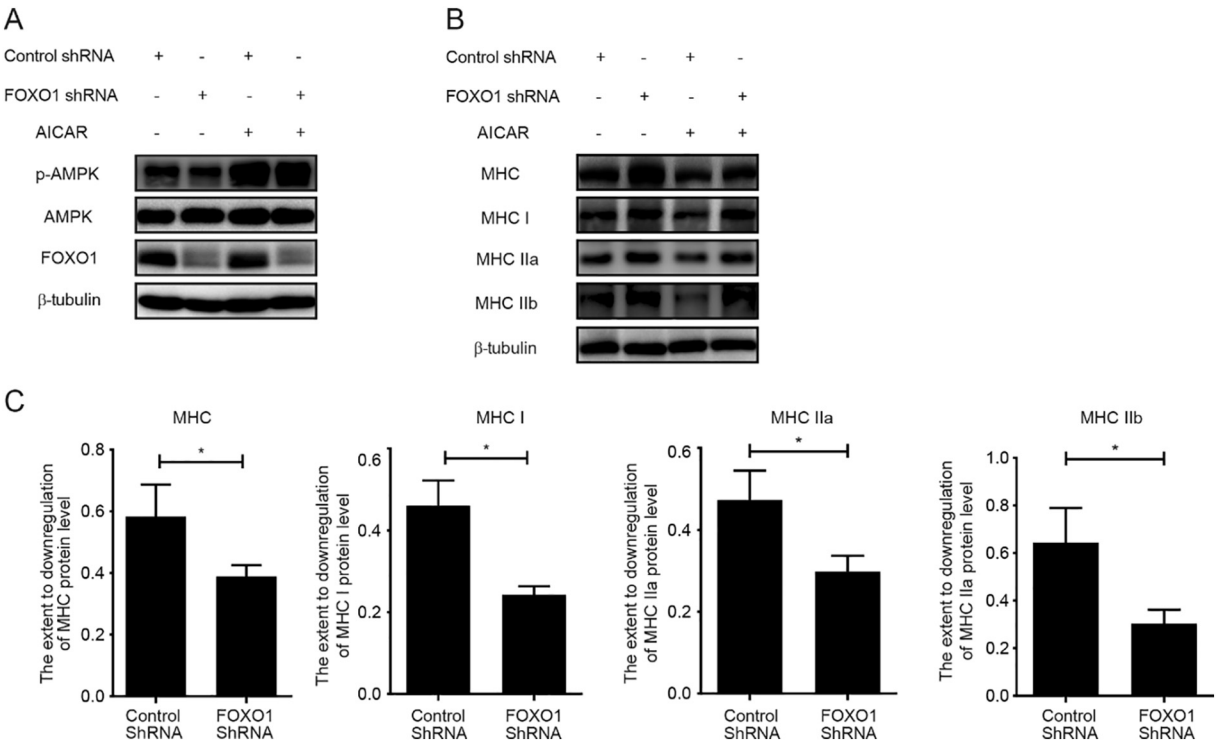

Fig. 5. FOXO1 is required in AMPK-mediated regulation of MHC and its isoforms expression.
(A) The knockdown efficiency of FOXO1 were confirmed by Western blotting. Representative Western blots are shown.
(B, C) In FOXO1-knockdown C2C12 myotubes, the downregulation of MHC, MHC I, MHC IIa and MHC IIb expression in response to AICAR, was weaker compared to C2C12 myotubes transfected with control shRNA. Representative Western blots are shown in (B). Densitometry of MHC/β-tubulin, MHC I/β-tubulin, MHC IIa/β-tubulin, MHC IIb/β-tubulin ($n = 3$) were calculated and used to analysis the extent that AICAR vs vehicle downregulates MHC, MHC I, MHC IIa and MHC IIb in control shRNA group or in FOXO1 shRNA group in (C);
Mean and SEM are plotted. *$P < 0.05$ by two-tailed Student's *t*-test.

works together with FOXO1 in regulation of MHC expression. We treated FOXO1-knockdown C2C12 myotubes with AICAR (Fig. 5A), and observed that the effects of AICAR on downregulation of MHC and its isoforms were dampened by knockdown of FOXO1 (Fig. 5B, C). The results indicated that FOXO1 was required in AMPK-mediated regulation of MHC and its isoforms expression.

4. Discussion

In this study, we identified the role of visfatin in promoting the expressions of MHC and its isoforms. Repressed AMPK activity was involved in visfatin-mediated upregulation of MHC I and MHC IIa, whereas visfatin increased MHC IIb expression partially independent of AMPK activity. FOXO1 was also required in the effects of visfatin on MHC isoforms expressions. Furthermore, FOXO1 participated in AMPK-mediated regulation of MHC expression. To the best of our knowledge, we reported for the first time the roles of visfatin and AMPK/FOXO1 signaling pathway in the regulation of MHC expression in C2C12 myotubes.

Previous studies showed that leptin and adiponectin increased MHC expression [15,16]. In the study, we observed the upregulation of total MHC by visfatin. MHC isoforms are vital indicators of muscle fiber types. MHC I is related to slow-twitch (type I) muscle fibers, while MHC IIa and MHC IIb are mainly expressed in fast-twitch ones (type IIa and type IIb) [17]. The switch of MHC isoforms is observed in obesity. In aged obese male rats, sternohyoid muscle tended to be fiber hypertrophy and had increased fast-twitch fiber density [18]. Clinically, the upper airway muscle fiber types were concerned mostly in the obstructive sleep apnea patients. It was reported that MHC IIa was predominated in uvula and genioglossus muscles of OSA patients [19,20]. We also observed an increase in plasma visfatin levels in severe OSA patients compared with the control and mild OSA group [5]. Herein, we found that visfatin upregulated the expressions of MHC I, MHC IIa and MHC IIb protein in C2C12 myotubes, which implied that visfatin may contribute to the dysfunction of upper airway muscles in OSA patients via alteration of contractile properties of skeleton muscles.

Then, we tried to find out the potential mechanisms of visfatin-mediated upregulation of MHC and its isoforms. AMPK is an essential energy sensor. AMPK activity is repressed in obesity and is associated with fiber type transition [21–23]. AMPK has also been reported to regulate MHC expression. MHC expression is repressed via AMPK activation upon glucose restriction in C2C12 myotubes [12]. Consistently, we found that inhibition of AMPK by compound C promoted MHC expression, and activation of AMPK by AICAR suppressed MHC expression. Moreover, this was the first time to find that visfatin repressed AMPK activity and AMPK was involved in visfatin-induced upregulation of MHC and its isoforms. We also observed that visfatin increased MHC IIb expression partially independent of AMPK, which implied that other unknown mechanisms were involved. Specific pathways responsible for MHC IIb expression have not yet been reported. Pathway including Ras/mitogen-activated protein kinase (MAPK), calcium/calmodulin-dependent protein kinase IV, and the peroxisome proliferator γ coactivator 1 are reported to regulate MHC expression [24–26]. Whether these pathways mediate the effect of visfatin on MHC IIb expression will need further investigation.

It is well-stablished that FOXO1 regulates skeletal muscle mass, but few studies investigated the role of FOXO1 in skeletal muscle fiber type expression. Kamei et al. [13] showed that mice with FOXO1 over-expression in skeletal muscle had less expression of MHC I fiber type. We firstly found that FOXO1 was repressed by visfatin. Visfatin-mediated upregulation of MHC I, MHC IIa, and MHC IIb was disrupted by FOXO1 overexpression. More importantly, we found reduced FOXO1 activity

was required in AMPK-mediated expressions of MHC in skeletal muscle cells, which is accordance to the results of previous studies. AMPK increased FOXO1 activity to inhibit MHC protein expression in cardiomyocytes [27]. In animal models, AMPK activation increased the activity of FOXO1 and MuRF1, a downstream atrogene which stimulated skeletal myofibrillar protein degradation [14]. All those finding showed that AMPK/FOXO1 signaling pathway was involved in the effects of visfatin on MHC expression.

5. Conclusion

In summary, we found that visfatin, an adipokine, promoted MHC expression, and showed that depressed AMPK/FOXO1 signaling pathway was involved in the effect of visfatin on MHC expression. Our study may provide new insight into the role of visfatin on skeletal muscle myogenesis and possible mechanisms of obesity-related skeletal muscle diseases.

Acknowledgements

The authors would additionally like to thank Jiang Zhu and Jian Lin (Shanghai Institution of hematology, Ruijin Hospital, Shanghai Jiao Tong University School of Medicine, Shanghai 200025, China.) for their technical assistance. The study was supported by the National Natural Science Foundation of China (No. 81270148, 81570082, 81770084, 81700084).

Author contribution to study

L.N.Z, and Q.Y.L designed the study, made plans, and collected the data. L.N.Z, Y.N.L and Q.Y.L completed the data analysis and interpretation. L.N.Z, Y.N.L C.J.G, J.P.Z and X.W.S, X.T.C, J.D, Q.Y.L drafted and revised the manuscript for important intellectual content. All authors approved the manuscript.

Declarations of interest

All authors declare that there are no conflicts of interest.

References

[1] B. Samal, Y. Sun, G. Stearns, C. Xie, S. Suggs, I. McNiece, Cloning and characterization of the cDNA encoding a novel human pre-B-cell colony-enhancing factor, Mol. Cell. Biol. 14 (2) (1994) 1431–1437.
[2] A. Fukuhara, M. Matsuda, M. Nishizawa, K. Segawa, M. Tanaka, K. Kishimoto, et al., Visfatin: a protein secreted by visceral fat that mimics the effects of insulin, Science 307 (5708) (2005) 426–430.
[3] Y.H. Chang, D.M. Chang, K.C. Lin, S.J. Shin, Y.J. Lee, Visfatin in overweight/obesity, type 2 diabetes mellitus, insulin resistance, metabolic syndrome and cardiovascular diseases: a meta-analysis and systemic review, Diabetes Metab. Res. Rev. 27 (2011) 515–527.
[4] Y. Sun, Z. Wu, L. Wei, C. Liu, S. Zhu, S. Tang, High-visfatin levels in women with polycystic ovary syndrome: evidence from a meta-analysis, Gynecol. Endocrinol. 31 (10) (2015) 808–814.
[5] Y. Wang, Q. Li, Y. Lin, X. Zhang, H. Xu, Serum visfatin levels in adult male patients with obstructive sleep apnea-hypopnea syndrome, J Shanghai Jiaotong University Medical Science 32 (2012) (2012) 503–507.
[6] H. Wu, C.M. Ballantyne, Skeletal muscle inflammation and insulin resistance in obesity, J. Clin. Invest. 127 (1) (2017) 43–54.
[7] X. Wang, D. Zhao, Y. Cui, S. Lu, D. Gao, J. Liu, Proinflammatory macrophages impair skeletal muscle differentiation in obesity through secretion of tumor necrosis factor-α via sustained activation of p38 mitogen-activated protein kinase, J. Cell. Physiol. 234 (3) (2019) 2566–2580.
[8] J. Tallis, R.S. James, F. Seebacher, The effects of obesity on skeletal muscle contractile function, J. Exp. Biol. 221 (Pt 13) (2018).
[9] F. Li, Y. Li, Y. Duan, C.A. Hu, et al., Myokines and adipokines: involvement in the crosstalk between skeletal muscle and adipose tissue, Cytokine Growth Factor Rev. 33 (2017) 73–82.
[10] A. Rodríguez, S. Becerril, L. Méndez-Giménez, B. Ramírez, N. Sáinz, V. Catalán, J. Gómez-Ambrosi, G. Frühbeck, Leptin administration activates irisin-induced myogenesis via nitric oxide-dependent mechanisms, but reduces its effect on subcutaneous fat browning in mice, Int. J. Obes. 39 (3) (2015) 397–407.
[11] Y. Ren, Y. Li, J. Yan, M. Ma, D. Zhou, Z. Xue, Z. Zhang, H. Liu, H. Yang, L. Jia, L. Zhang, Q. Zhang, S. Mu, R. Zhang, Y. Da, Adiponectin modulates oxidative stress-induced mitophagy and protects C2C12 myoblasts against apoptosis, Sci. Rep. 7 (1) (2017) 3209.
[12] M. Fulco, Y. Cen, P. Zhao, et al., Glucose restriction inhibits skeletal myoblast differentiation by activating SIRT1 through AMPK-mediated regulation of Nampt, Dev. Cell 14 (2008) 661–673.
[13] G. Milan, V. Romanello, F. Pescatore, et al., Regulation of autophagy and the ubiquitin-proteasome system by the FoxO transcriptional network during muscle atrophy, Nat. Commun. 6 (2015) 6670–6683.
[14] Y. Kamei, S. Miura, M. Suzuki, et al., Skeletal muscle FOXO1 (FKHR) transgenic mice have less skeletal muscle mass, down-regulated Type I (slow twitch/red muscle) fiber genes, and impaired glycemic control, J. Biol. Chem. 279 (2004) 41114–41123.
[15] S. Masuda, T. Tanaka, H. Masuzaki, K. Nakao, S. Taguchi, Overexpression of leptin reduces the ratio of glycolytic to oxidative enzymatic activities without changing muscle fiber types in mouse skeletal muscle, Biol. Pharm. Bull. 37 (2014) 169–173.
[16] T. Fiaschi, D. Cirelli, G. Comito, et al., Globular adiponectin induces differentiation and fusion of skeletal muscle cells, Cell Res. 19 (2009) 584–597.
[17] D. Pette, The adaptive potential of skelstal muscle fibers, Can. J. Appl. Physiol. 27 (2002) 423–448.
[18] J.R. Skelly, R.A. O'Connell, J.F. Jones, K.D. O'Halloran, Structural and functional properties of an upper airway dilator muscle in aged obese male rats, Respiration 82 (2011) 539–549.
[19] D. Friberg, T. Ansved, K. Borg, B. Carlsson-Nordlander, H. Larsson, E. Svanborg, Histological indications of a progressive snorer disease in an upper airway muscle, Am. J. Respir. Crit. Care Med. 157 (1998) 586–593.
[20] F.J. Sériès, S.A. Simoneau, S. St Pierre, I. Marc, Characteristics of the genioglossus and musculus uvulae in sleep apnea hypopnea syndrome and in snorers, Am. J. Respir. Crit. Care Med. 153 (1996) 1870–1874.
[21] G.R. Steinberg, B.J. Michell, B.J. van Denderen, M.J. Watt, A.L. Carey, B.C. Fam, S. Andrikopoulos, J. Proietto, C.Z. Görgün, D. Carling, G.S. Hotamisligil, M.A. Febbraio, T.W. Kay, B.E. Kemp, Tumor necrosis factor alpha-induced skeletal muscle insulin resistance involves suppression of AMP-kinase signaling, Cell Metab. 4 (6) (2006) 465–474.
[22] N.A. Vilchinskaya, E.P. Mochalova, T.L. Nemirovskaya, T.M. Mirzoev, O.V. Turtikova, B.S. Shenkman, Rapid decline in MyHC I(β) mRNA expression in rat soleus during hindlimb unloading is associated with AMPK dephosphorylation, J. Physiol. Lond. 595 (2017) 7123–7134.
[23] R. Bassel-Duby, E.N. Olson, Signaling pathways in skeletal muscle remodeling, Annu. Rev. Biochem. 75 (2006) 19–37.
[24] S.J. Xie, J.H. Li, H.F. Chen, Y.Y. Tan, S.R. Liu, Y. Zhang, H. Xu, J.H. Yang, S. Liu, L.L. Zheng, M.B. Huang, Y.H. Guo, Q. Zhang, H. Zhou, L.H. Qu, Inhibition of the JNK/MAPK signaling pathway by myogenesis-associated miRNAs is required for skeletal muscle development, Cell Death Differ. 25 (9) (2018) 1581–1597.
[25] L. Sun, X. Cao, B. Liu, H. Huang, X. Wang, L. Sui, W. Yin, K. Ma, CaMK IV phosphorylates prohibitin 2 and regulates prohibitin 2-mediated repression of MEF2 transcription, Cell. Signal. 23 (10) (2011) 1686–1690.
[26] N.L. Reyes, G.B. Banks, M. Tsang, D. Margineantu, H. Gu, D. Djukovic, J. Chan, M. Torres, H.D. Liggitt, D.K. Hirenallur-S, D.M. Hockenbery, D. Raftery, B.M. Iritani, Fnip1 regulates skeletal muscle fiber type specification, fatigue resistance, and susceptibility to muscular dystrophy, Proc. Natl. Acad. Sci. U. S. A. 112 (2) (2015) 424–429 Jan 13.
[27] K. Nakashima, Y. Yakabe, AMPK activation stimulates myofibrillar protein degradation and expression of atrophy-related ubiquitin ligases by increasing FOXO transcription factors in C2C12 myotubes, Biosci. Biotechnol. Biochem. 71 (7) (2007) 1650–1656.