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Original Article

miR-128 suppresses bleomycin-induced pulmonary fibrosis and epithelial to mesenchymal transition by targeting Cadherin-11

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Abstract: Idiopathic pulmonary fibrosis (IPF) is a progressive and fatal lung disease characterized by the replacement of normal lung parenchyma with fibrotic tissue. Epithelial to mesenchymal transition (EMT), due to its close relationship with the process of tissue fibrosis, has been reported to play a crucial role in regulation of pulmonary fibrosis. Recently, miR-128, a brain-enriched microRNA, has been reported to be involved in EMT and tumorigenesis. However, the roles of miR-128 in pulmonary fibrosis have not yet been studied. The current study reports, for the first time, that miR-128 is downregulated in bleomycin (BLM)-induced pulmonary fibrosis and overexpression of it could significantly inhibit the progression of pulmonary fibrosis and EMT. Furthermore, this study shows that CDH11, a crucial regulator in progression of pulmonary fibrosis, is a direct target of miR-128. This direct interaction is crucial for their function in BLM-induced EMT and pulmonary fibrosis. Moreover, present results reveal that upregulation of miR-128 could prevent BLM-induced pulmonary fibrosis and EMT *in vitro* and *in vivo*. Taken together, the current study indicates that miR-128 may be a potential therapeutic target for treatment of IPF.

Keywords: Idiopathic pulmonary fibrosis, microRNA, miR-128, epithelial to mesenchymal transition, Cadherin-11

Introduction

Idiopathic pulmonary fibrosis (IPF) is a chronic, devastating, and irreversible lung disease. It is characterized by the replacement of normal lung parenchyma with fibrotic tissue [1, 2]. The crude incidence rate of IPF has been estimated between 4.3 to 4.9 per 100,000 person-years. It is more prevalent in women and older people, based on a UK analysis of general practice database [3]. IPF was once thought to be a developing inflammatory process involved with fibroblast proliferation and excessive collagen deposition [4-6]. However, accumulating evidence has demonstrated that pulmonary fibrosis is driven by abnormal alveolar epithelial cells, which produce effective mediators that induce pathological stimulation of epithelial to mesenchymal transition (EMT) [7-9]. EMT, in which the acquisition of a mesenchymal phenotype [N-cadherin and α-smooth muscle actin (α-SMA)] and the loss of epithelial characteristics (E-cadherin) are involved, occurs in both tissue development during embryogenesis and pathological processes, including wound healing, organ fibrosis, and tumorigenesis [10-13].

MicroRNAs (miRNAs) are endogenous small non-protein-coding RNA molecules that consist of approximate 22 nucleotides. They play an important role in genetic regulation via directly binding to targeted mRNA [14-16]. Numerous studies have revealed that miRNAs are involved in various biological processes, including cell proliferation, tumorigenesis, and tissue fibrosis [17-20]. For example, in non-small-cell lung cancer cells, miR-149 inhibit the invasive capability and EMT via targeting FOXM1, a potential metastasis promoter [21]. miR-192, a microRNA highly expressed in fibrotic kidneys, may mediate renal fibrosis in a TGF-β/Smad3-dependent manner [22].

miR-128, a brain-enriched microRNA, could regulate apoptosis, proliferation, and chemosensitivity in glioblastomas and ovarian cancer [23-

25]. Moreover, Liu et al. demonstrated that overexpression of miR-128 suppressed EMT development, migration, and invasion by directly targeting integrin α2 in osteosarcoma cells [26]. However, the roles of miR-128 in pulmonary fibrosis have not been elucidated.

The present study determined the expression patterns of miR-128 in bleomycin-induced pulmonary fibrosis and EMT of alveolar epithelial A549 cells and its function in bleomycin (BLM)-induced invasion and EMT of A549 cells *in vitro*. Moreover, this study found that Cadherin-11 (CDH11), a crucial regulator in progression of pulmonary fibrosis, is a direct target of miR-128, revealing the significance of miR-128/CDH11 interaction in BLM-induced pulmonary fibrosis *in vivo*.

Materials and methods

Animals and models of BLM-induced pulmonary fibrosis

Male Wistar rats (age, 8 weeks; weight, 200-240 g) were provided by Shanghai Laboratory Animal Center (SLAC), Chinese Academy of Sciences (Shanghai, China), and maintained in pathogen-free conditions. Before BLM treatment, the rats were acclimated to the environment for 1 week. They were injected intratracheally with BLM (Nippon Kayaku; Tokyo, Japan) at a dose of 5 mg/kg, dissolved in a total of 2 mL of sterile saline or 2 mL of sterile saline for control groups. The rats were killed at days 0, 7, 14, and 21 (n = 6 for each time point). All animal experiments were reviewed and permitted by the Animal Care and Use Committee of Central South University.

Cell culture and treatment

The alveolar epithelial A549 cell line was provided by Auragene Bioscience (Changsha, China). The cell line was maintained in Dulbecco's modified Eagle's medium (Hyclone; Logan, UT) supplemented with 10% fetal Calf serum (Gibco; Grand Island, NY), 100 mg/mL streptomycin (Gibco; Gaithersburg, MD), and 100 U/mL penicillin G at 37°C with 5% CO_2. Cells at a density of 1×10^5 cells per well were seeded into 6-well plates and treated with serum-free medium with or without 20 μg/mL BLM for 48 hours.

Histological analysis

For histological study, rat lungs were fixed in 4% paraformaldehyde and embedded in paraffin. They were cut into sections and stained with hematoxylin and eosin (HE), according to standard protocol.

RNA extraction and qRT-PCR analyses

TRIZOL Reagent (Invitrogen; Carlsbad, CA) was used to extract total RNA. The Reverse Transcription Kit (Takara; Dalian, China) was used to synthesize the cDNA. Quantitative RT-PCR (qRT-PCR) analyses were performed using SYBR Green Master (Roche; Indianapolis, IN), according to manufacturer recommendations. Transcription levels of β-actin and U6 were used to normalize expression levels of target genes and miR-128, respectively. Data were analyzed using an ABI 7300 instrument (Thermo Fisher Scientific; Waltham, MA). Primers for qRT-PCR were synthesized by Invitrogen (Shanghai, China). Sequences were: *CDH11* sense, 5'-ATTGTGATTAGCGATGGTGGCA-3' and antisense, 5'-TCAGACCGGCATTCAGGATGTA-3'; *α-SMA* sense, 5'-CCCAGATTATGTTTGAGACCTTCA3' and antisense, 5'-TCAGACCGGCATTCAGGATGTA-3'; *E-cadherin* sense, 5'-TGCCCAGAAAATGAAAAAGG-3' and antisense, 5'-GTGTATGTGGCAATGCGTTC-3'; *N-cadherin* sense, 5'-ATGACAATCCTCCAGAGTTTA-3' and antisense, 5'-ATCCTTATCGGTCACAGTTAG-3'; β-actin sense, 5'-AGGGGCCGGACTCGTCATACT-3' and antisense, 5'-GGCGGCACCACCATGTACCCT-3'.

Wound healing assay

A549 cells were grown to confluent monolayer in 6-well plates for 48 hours. They were scratched with a 200 μl pipette tip. Subsequently, plates were washed twice with PBS and incubated with the complete growth medium without FBS. After 0 and 24 hours of incubation, the wounded area was photographed by microscopy (magnification, ×40) for wound closure analysis.

Matrigel invasion assay

Matrigel invasion assay was performed using 24-well Transwell plates (Corning; Lowell, MA). Briefly, the treated 2×10^5 cells in DMEM media were added to the upper chamber containing 8-μm pore size filters. This was coated with 1 mg/mL matrigel. The invasion process lasted for 24 hours at 37°C in a CO_2 incubator. Subsequently, non-invasive cells on the upper surface of the membranes were scrubbed with a cotton-tipped swab. Invading cells were on the lower surface of filters. They were fixed and stained with 0.5% crystal violet for 10 minutes

at room temperature. Images of the random fields (five per membrane) were photographed by microscopy (magnification, ×100) for calculation of the cell number.

Dual luciferase reporter assay

Fragments from CDH11 containing the predicted miR-128 binding site or mutant sequence of miR-128 binding site were synthesized and cloned into the luciferase construct psi-CHECK2. The resulting vector CDH11-3'UTR-psi-CHECK2 and corresponding mutant were called WT 3'UTR and Mut 3'UTR, respectively. Transfection reagent (Invitrogen; Carlsbad, CA) was used for transfection. Forty-eight hours after transfection, luciferase activities in cell lysates were measured using the Dual-Luciferase Reporter Assay Kit (Promega, Madison, Wisconsin, USA).

Statistical analysis

Statistical analyses were performed using SPSS 20.0 (SPSS; Chicago, IL). Figures were processed with GraphPad Prism 5 (GraphPad Software; La Jolla, CA). Paired Student's *t*-test was used for statistical analyses between two groups. Data are expressed as mean ± standard deviation (SD) from at least three independent experiments. $P < 0.05$ indicates statistical significance.

Results

miR-128 was downregulated and CDH11 was upregulated in BLM-induced pulmonary fibrosis and EMT of alveolar epithelial A549 cells

The current study established a rat model of BLM-induced pulmonary fibrosis. After 7, 14, and 21 days of treatment, the structures of the alveolar and air-blood barriers were severely damaged (**Figure 1A**). These histological changes were also supported by increased levels of mesenchymal cell markers (α-SMA, N-cadherin), along with decreased levels of epithelial cell markers (E-cadherin) (**Figure 1A**). Moreover, qRT-PCR analysis revealed that miR-128 levels on days 7, 14, and 21 after BLM treatment were decreased, compared with the control group (**Figure 1B**). Additionally, CDH11 expression was significantly upregulated (**Figure 1C**). Furthermore, it was observed that BLM could induce EMT of alveolar epithelial A549 cells. miR-128 was downregulated and CDH11 was upregulated in BLM-induced EMT (**Figure 1D**). Taken together, present data suggests a potential link between dysregulation of miR-128 and CDH11 and BLM-induced pulmonary fibrosis and EMT.

miR-128 restoration and CDH11 silencing attenuated BLM-induced invasion and EMT of A549 cells in vitro

To investigate the roles of miR-128 and CDH11 in BLM-induced EMT of A549 cells, this study restored miR-128 by confecting miR-128 mimics and inhibited CDH11 by specific small interfering RNAs. Wound healing and Matrigel invasion assays showed that miR-128 restoration (**Figure 2A**, **2B**) and knockdown of CDH11 (**Figure 2C**, **2D**) significantly inhibited cell invasion and migration ability of A549 cells, respectively. In addition, miR-128 restoration and CDH11 silencing attenuated EMT related markers (**Figure 2E**, **2F**). Results indicate that miR-128 and CDH11 played an opposite role in the regulation of BLM-induced cell invasion and EMT of A549 cells *in vitro*.

miR-128 directly inhibited CDH11 expression in A549 cells

A reverse relation of miR-128 and CDH11 expression was observed in BLM-induced pulmonary fibrosis and EMT of alveolar epithelial A549 cells. Moreover, *in silico* prediction for microRNA target (**Figure 3A**) suggests that CDH11 might be a target of miR-128. Previously, CDH11 has been reported to contribute to TGF-β-induced EMT and bleomycin-induced pulmonary fibrosis [27]. To validate targeting of CDH11 by miR-128, luciferase reporter assays were conducted. The co-transfection of miR-128 mimics with CDH11 Wt-3'UTR showed significant decreased luciferase activity, compared with the control group. However, luciferase activity of CDH11 Mut-3'UTR was not changed (**Figure 3B**). In addition, qRT-PCR analysis revealed that miR-128 mimic significantly reduced expression of CDH11, while miR-128 inhibitor increased CDH11 expression in A549 cells (**Figure 3C**). Together, results reveal that miR-128 directly inhibited CDH11 expression in lung cells.

miR-128 prevented BLM-induced pulmonary fibrosis by suppressing CDH11-mediated EMT in vivo

To explore the potential roles of miR-128 in BLM-induced pulmonary fibrosis, this study co-injected the miR-128 agomir at day 0 and via the tail veins of rat at days 7 and 14 after BLM

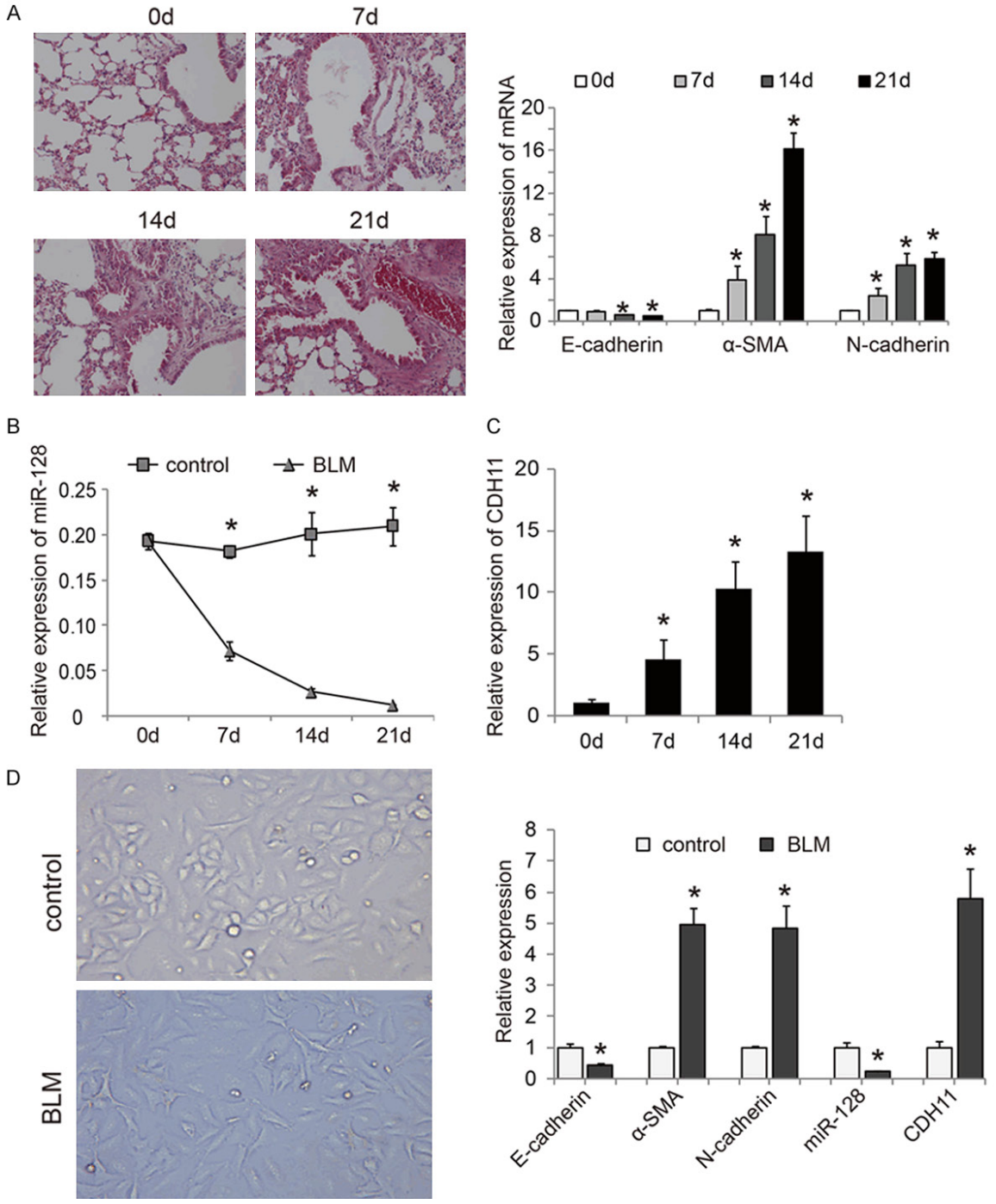

Figure 1. miR-128 was downregulated and CDH11 was upregulated in BLM-induced pulmonary fibrosis and EMT of alveolar epithelial A549 cells. A. H&E staining was applied to show pulmonary fibrosis in lung tissues after 7, 14, and 21 days of BLM treatment. qRT-PCR was used to indicate expressional changes of EMT-related markers (E-cadherin, α-SMA, and N-cadherin). B and C. Relative expression levels of miR-128 and CDH11 were examined in lung tissues of rat models of bleomycin induced pulmonary fibrosis and control groups. D. BLM was used to induce the EMT of alveolar epithelial A549 cells and relative expression of EMT-related markers, miR-128, and CDH11 was examined in these cells with qRT-PCR. (*P value < 0.05).

treatment. At day 21, the lungs were harvested. Histological examination via H&E staining showed that alveolar and air-blood barriers were severely damaged in the BLM treatment group, but slightly damaged in the BLM plus miR-128 treatment group (**Figure 4A**), suggest-

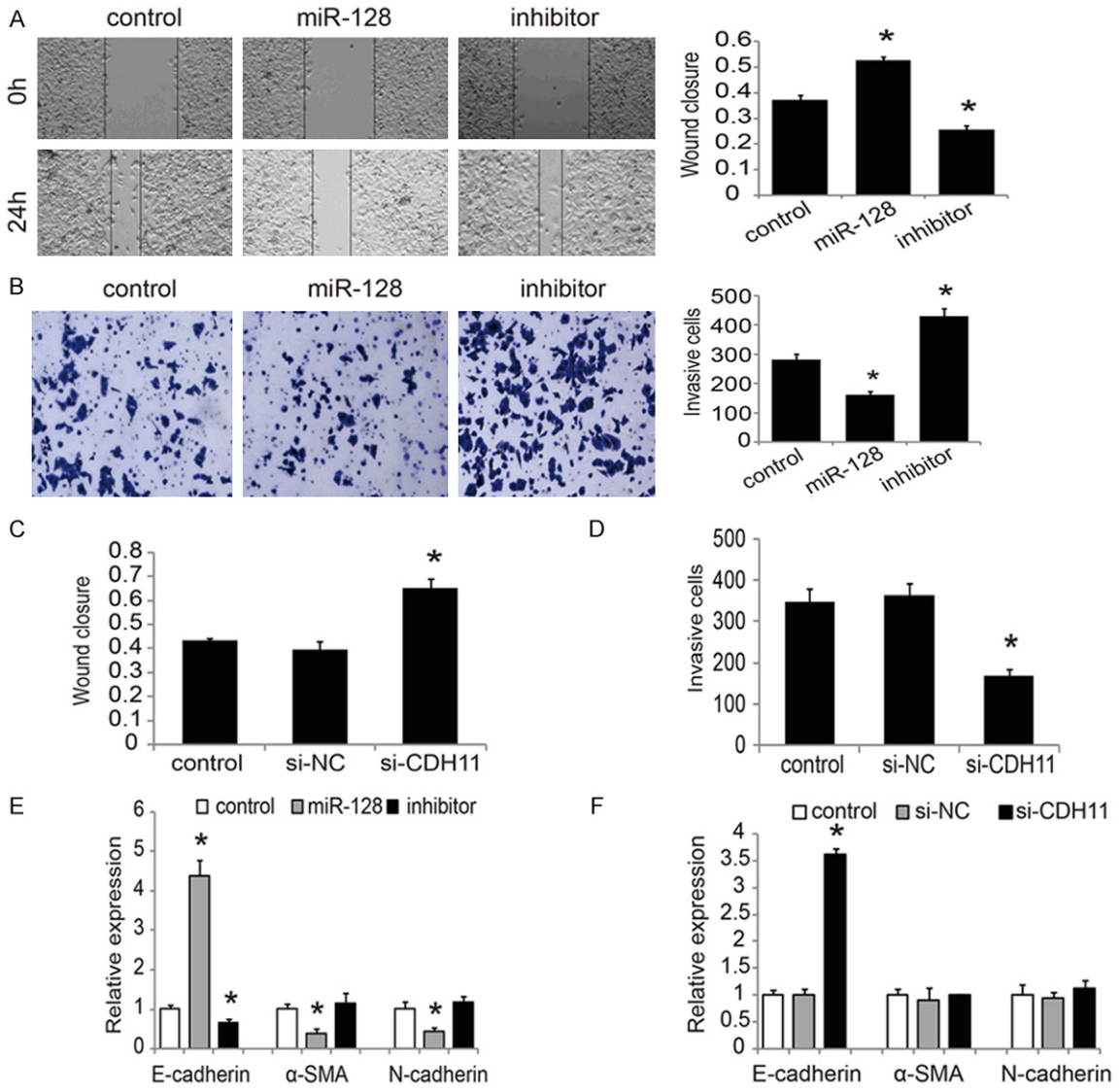

Figure 2. miR-128 restoration and CDH11 silencing attenuated BLM-induced invasion and EMT of A549 cells in vitro. A. Monolayers of treated A549 cells were scraped with a 200 μl pipette tips and the number of cells in the denuded zone was photographed and quantified after 0 and 24 hours of incubation. B. Matrigel invasion assay was performed using Transwell plates. Representative photomicrographs of the invading cells were assayed by 0.5% crystal violet stain. C and D. Wound healing assays and Matrigel invasion assays were also conducted to CDH11 silencing A549 cells and the control groups. E and F. Expression of EMT-related markers (E-cadherin, α-SMA and N-cadherin) in each group was examined by qRT-PCR. (*P value < 0.05).

ing that miR-128 may prevent BLM-induced pulmonary fibrosis *in vivo*. Twenty-one days after BLM treatment, miR-128 expression levels were decreased, while miR-128 agomir treatment increased expression of miR-128 (**Figure 4B**). Additionally, the presence of miR-128 agomir attenuated BLM-induced inhibition on E-cadherin and CDH11 (**Figure 4C**). Together, results indicate that miR-128 might prevent BLM-induced pulmonary fibrosis by suppressing CDH11-mediated EMT *in vivo*.

Discussion

Alveolar epithelial cell injuries, damage to air-blood barriers, and subsequent uncontrolled accumulation of fibroblasts and collagen are the major pathological processes in IPF patients [28, 29]. EMT, due to its relationship with process of tissue fibrosis, has been reported to play an important role in regulation of pulmonary fibrosis [30-32]. It has been demonstrated that approximately 33% of pulmonary fibrosis

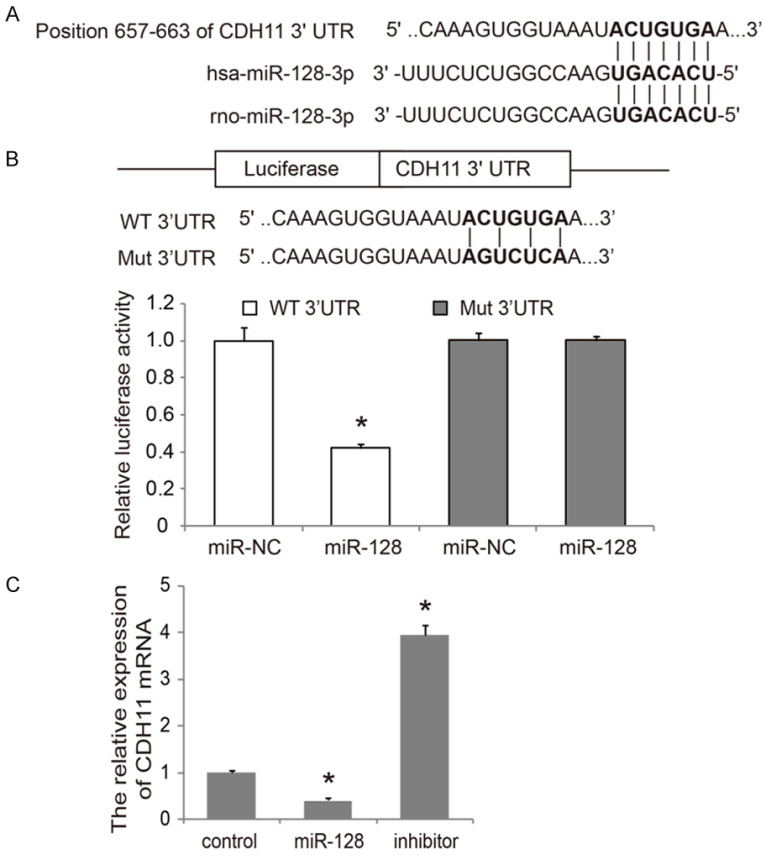

Figure 3. miR-128 directly inhibited CDH11 expression in A549 cells. A. In silico prediction the binding sites between miR-128 and CDH11. B. Relative luciferase activities were measured using the Dual-Luciferase Reporter Assay Kit in indicated cells, which were transfected with WT or Mut CDH11 3'UTR luciferase reporter plasmid with or without miR-128. C. mRNA expression of CDH11 was detected in A549 cells transfected with miR-128 mimics or inhibitors using qRT-PCR. (*P value < 0.05).

of an experimental IPF model originated from cells undergoing EMT [33, 34]. In this study, increased levels of mesenchymal cell markers (α-SMA, N-cadherin), along with decreased levels of epithelial cell markers (E-cadherin), were observed in the rat model of BLM-induced pulmonary fibrosis. Results suggest that induction of EMT was involved in BLM-induced pulmonary fibrosis.

MicroRNAs have been identified to be differentially expressed in pulmonary fibrosis. These miRNAs are involved in regulations of apoptosis and TGF-β-mediated EMT [35]. A study on human osteosarcomas demonstrated that miR-128 overexpression could suppress cell migration and invasion and EMT of human osteosarcoma cells by inhibiting its target gene integrin α2 [26]. However, the roles of miR-128 in pulmonary fibrosis have not been reported yet. Present data revealed, for the first time, that miR-128 was downregulated in bleomycin induced pulmonary fibrosis and EMT of alveolar epithelial A549 cells and that miR-128 restoration could attenuate BLM-induced invasion and EMT of A549 cells. Present results suggest that miR-128 may play a similar role in the regulation of BLM-induced pulmonary fibrosis and EMT.

Previously, many target genes of miR-128 have been identified in different types of human tumors. These genes are widely involved in cancer-related biological processes, including cell proliferation, differentiation, cellular motility, and adhesion [36-40]. CDH11 is a cell-to-cell adhesion molecule functioning in many biological functions, including cytoskeletal organization, extracellular matrix-mediated tissue remodeling, and cellular migration and invasion [41-43]. Moreover, CDH11 has been found to be upregulated in lung tissues from IPF patients. It may promote the process of fibrosis by inducing the differentiation of resident tissue fibroblasts into myofibroblasts [27, 44, 45]. Present results also indicate that expression of CDH11 was significantly upregulated in BLM-induced pulmonary fibrosis and EMT. Since this *in silico* study predicted that CDH11 might be a target of miR-128, further exploration on the roles of miR-128 in pulmonary fibrosis is critical to the understanding of IPF initiation and progression. In the present study, according to the dual luciferase reporter assay, it was found that CDH11 was a target of miR-128 in alveolar epithelial cells. Further transgenic analysis validated the direct interaction.

Since upregulation of CDH11 could contribute to EMT progression and pulmonary fibrosis, it was hypothesized that upregulation of miR-128

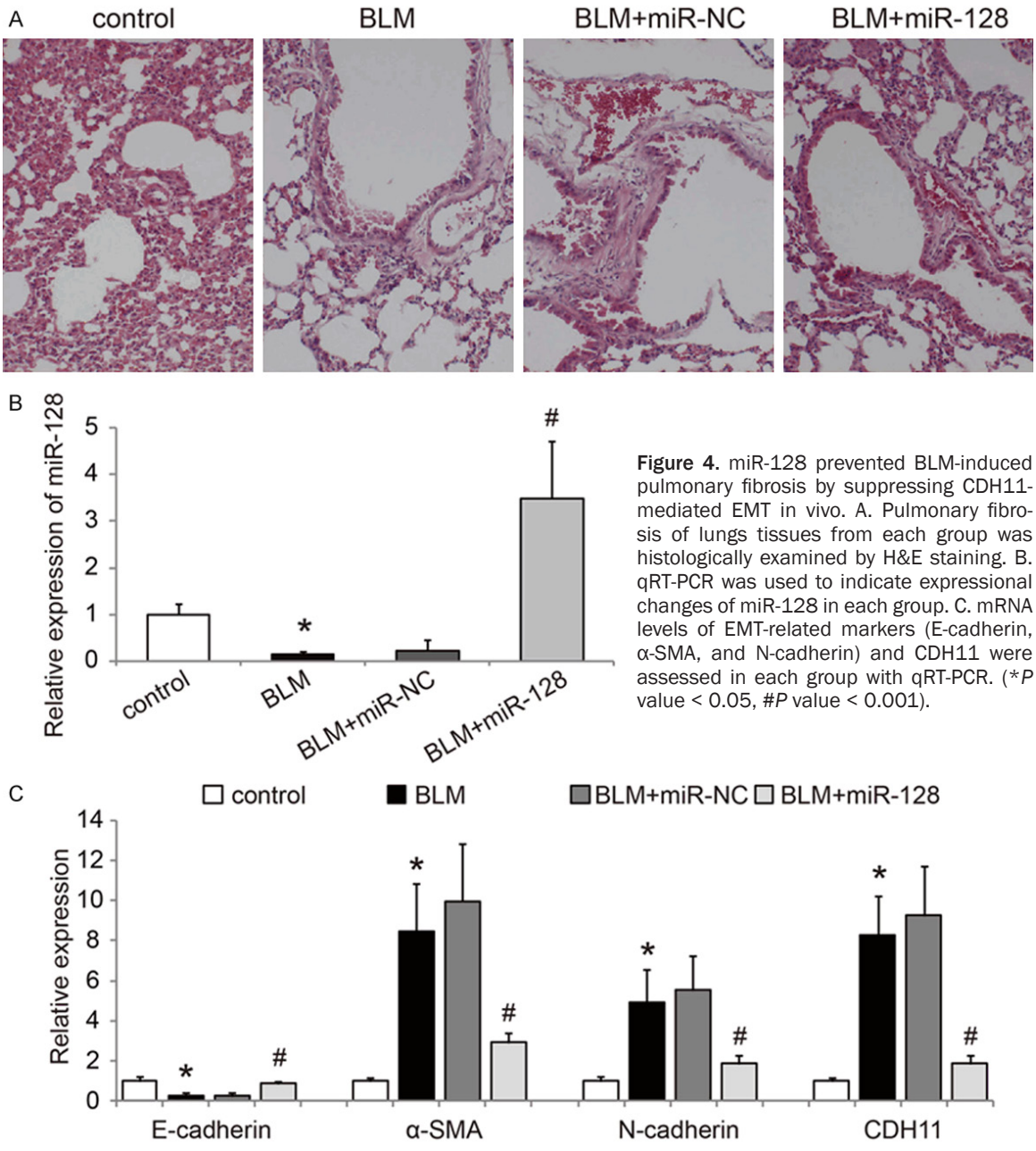

Figure 4. miR-128 prevented BLM-induced pulmonary fibrosis by suppressing CDH11-mediated EMT in vivo. A. Pulmonary fibrosis of lungs tissues from each group was histologically examined by H&E staining. B. qRT-PCR was used to indicate expressional changes of miR-128 in each group. C. mRNA levels of EMT-related markers (E-cadherin, α-SMA, and N-cadherin) and CDH11 were assessed in each group with qRT-PCR. (*P value < 0.05, #P value < 0.001).

might prevent EMT progression and BLM-induced pulmonary fibrosis via suppressing CDH11 expression [43]. To validate this hypothesis, this study co-injected miR-128 agomir into BLM-treated rats via intratracheal instillation. Results indicated that miR-128 could prevent both of bleomycin-induced pulmonary fibrosis and expression of CDH11 and EMT-related marker genes *in vivo*.

Conclusion

Taken together, the current study demonstrates, for the first time, that miR-128 is down-regulated in BLM-induced pulmonary fibrosis and EMT and that miR-128 restoration could significantly inhibit the progression of pulmonary fibrosis and EMT *in vitro* and *in vivo*, at least in part by regulating CHD11. Therefore, present findings indicate that miR-128/CDH11 interaction might be a potential therapeutic target for treatment of IPF in the future.

Disclosure of conflict of interest

None.

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References

[1] Armanios MY, Chen JJ, Cogan JD, Alder JK, Ingersoll RG, Markin C, Lawson WE, Xie M, Vulto I, Phillips JA 3rd, Lansdorp PM, Greider CW, Loyd JE. Telomerase mutations in families with idiopathic pulmonary fibrosis. N Engl J Med 2007; 29; 1317-1326.

[2] Yount SE, Beaumont JL, Chen SY, Kaiser K, Wortman K, Van Brunt DL, Swigris J, Cella D. Health-related quality of life in patients with idiopathic pulmonary fibrosis. Lung 2016; 194: 227-234.

[3] Gribbin J, Hubbard RB, Le Jeune I, Smith CJ, West J and Tata LJ. Incidence and mortality of idiopathic pulmonary fibrosis and sarcoidosis in the UK. Thorax 2006; 61: 980-985.

[4] Vittal R, Mickler EA, Fisher AJ, Zhang C, Rothhaar K, Gu H, Brown KM, Emtiazdjoo A, Lott JM, Frye SB, Smith GN, Sandusky GE, Cummings OW, Wilkes DS. Type V collagen induced tolerance suppresses collagen deposition, tgf-β and associated transcripts in pulmonary fibrosis. PLoS One 2013; 8: e76451.

[5] Low RB, Giancola MS, King TE Jr, Chapitis J, Vacek P and Davis GS. Serum and bronchoalveolar lavage of N-terminal type III procollagen peptides in idiopathic pulmonary fibrosis. Am Rev Respir Dis 1992; 146: 701-706.

[6] Ruiz V, Ordóñez RM, Berumen J, Ramírez R, Uhal B, Becerril C, Pardo A, Selman M. Unbalanced collagenases/TIMP-1 expression and epithelial apoptosis in experimental lung fibrosis. Am J Physiol Lung Cell Mol Physiol 2003; 285: 1026-1036.

[7] Selman M, Thannickal VJ, Pardo A, Zisman DA, Martinez FJ and Lynch JP. Idiopathic pulmonary fibrosis. Drugs 2011; 378: 1949.

[8] Tian K, Chen P, Liu Z, Si S, Zhang Q, Mou Y, Han L, Wang Q, Zhou X. Sirtuin 6 inhibits epithelial to mesenchymal transition during idiopathic pulmonary fibrosis via inactivating TGF-β1/Smad3 signaling. Oncotarget 2017; 9: 61011-61024.

[9] Milara J, Navarro R, Juan G, Peiró T, Serrano A, Ramón M, Morcillo E, Cortijo J. Sphingosine-1-phosphate is increased in patients with idiopathic pulmonary fibrosis and mediates epithelial to mesenchymal transition. Thorax 2012; 67: 147-156.

[10] Thuault S, Valcourt U, Petersen M, Manfioletti G, Heldin CH and Moustakas A. Transforming growth factor-β employs HMGA2 to elicit epithelial-mesenchymal transition. J Cell Biol 2006; 174: 175-183.

[11] Voulgari A and Pintzas A. Epithelial-mesenchymal transition in cancer metastasis: mechanisms, markers and strategies to overcome drug resistance in the clinic. Biochim Biophys Acta 2009; 1796: 75-90.

[12] Zeisberg M and Kalluri R. The role of epithelial-to-mesenchymal transition in renal fibrosis. J Mol Med (Berl) 2004; 82: 175-181.

[13] Yan C, Grimm WA, Garner WL, Qin L, Travis T, Tan N, Han YP. Epithelial to mesenchymal transition in human skin wound healing is induced by tumor necrosis factor-α through bone morphogenic protein-2. Am J Pathol 2010; 176: 2247-2258.

[14] Bartel DP. MicroRNAs: target recognition and regulatory functions. Cell 2009; 136: 215-233.

[15] Weiss GJ, Bemis LT, Nakajima E, Sugita M, Birks DK, Robinson WA, Varella-Garcia M, Bunn PA Jr, Haney J, Helfrich BA, Kato H, Hirsch FR, Franklin WA. EGFR regulation by microRNA in lung cancer: correlation with clinical response and survival to gefitinib and EGFR expression in cell lines. Ann Oncol 2008; 19: 1053.

[16] Yin R, Zhang S, Wu Y, Fan X, Jiang F, Zhang Z, Feng D, Guo X, Xu L. microRNA-145 suppresses lung adenocarcinoma-initiating cell proliferation by targeting OCT4. Oncol Rep 2011; 25: 1747-1754.

[17] Bueno MJ, Castro IPD and Malumbres M. Control of cell proliferation pathways by microRNAs. Cell Cycle 2008; 7: 3143.

[18] Shivdasani RA. MicroRNAs: regulators of gene expression and cell differentiation. Blood 2006; 108: 3646.

[19] Kumar MS, Lu J, Mercer KL, Golub TR and Jacks T. Impaired microRNA processing enhances cellular transformation and tumorigenesis. Nature Genetics 2007; 39: 673-677.

[20] Angelini A, Li Z, Mericskay M and Decaux JF. Regulation of connective tissue growth factor and cardiac fibrosis by an SRF/MicroRNA-133a axis. PLoS One 2015; 10: e0139858.

[21] Ke Y, Zhao W, Xiong J, Cao R. miR-149 inhibits non-small-cell lung cancer cells EMT by targeting FOXM1. Biochem Res Int 2013; 2013: 506731.

[22] Chung AC, Huang XR, Meng X, Lan HY. miR-192 mediates TGF-beta/Smad3-driven renal fibrosis.J Am Soc Nephrol 2010; 21: 1317-25.

[23] Shang C, Hong Y, Guo Y, Liu YH and Xue YX. miR-128 regulates the apoptosis and proliferation of glioma cells by targeting RhoE. Oncology Letters 2016; 11: 904.

[24] She X, Yu Z, Cui Y, Lei Q, Wang Z, Xu G, Xiang J, Wu M, Li G. miR-128 and miR-149 enhance the chemosensitivity of temozolomide by Rap1B-mediated cytoskeletal remodeling in glioblastoma. Oncol Rep 2014; 32: 957-964.

[25] Li B, Chen H, Wu N, Zhang WJ and Shang LX. Deregulation of miR-128 in ovarian cancer pro-

motes cisplatin resistance. Int J Gynecol Cancer 2014; 24: 1381-1388.
[26] Liu X, Liang Z, Gao K, Li H, Zhao G, Wang S, Fang J. MicroRNA-128 inhibits EMT of human osteosarcoma cells by directly targeting integrin α2. Tumour Biol 2016; 37: 7951.
[27] Schneider DJ, Wu M, Le TT, Cho SH, Brenner MB, Blackburn MR, Agarwal SK. Cadherin-11 contributes to pulmonary fibrosis: potential role in TGF-β production and epithelial to mesenchymal transition. FASEB J 2012; 26: 503-512.
[28] Colombi D, Dinkel J, Weinheimer O, Obermayer B, Buzan T, Nabers D, Bauer C, Oltmanns U, Palmowski K, Herth F, Kauczor HU, Sverzellati N, Kreuter M, Heussel CP. Visual vs fully automatic histogram-based assessment of idiopathic pulmonary fibrosis (ipf) progression using sequential multidetector computed tomography (MDCT). PLoS One 2015; 10: e0130653.
[29] Sime PJ. The antifibrogenic potential of PPARgamma ligands in pulmonary fibrosis. J Investig Med 2008; 56: 534-538.
[30] López-Novoa JM and Nieto MA. Inflammation and EMT: an alliance towards organ fibrosis and cancer progression. EMBO Mol Med 2009; 1: 303.
[31] Chua KN, Ma J and Thiery JP. Targeted therapies in control of EMT in carcinoma and fibrosis. Drug Discovery Today Disease Mechanisms 2008; 4: 261-267.
[32] Namba T, Tanaka KI, Ito Y, Hoshino T, Matoyama M, Yamakawa N, Isohama Y, Azuma A, Mizushima T. Induction of EMT-like phenotypes by an active metabolite of leflunomide and its contribution to pulmonary fibrosis. Cell Death Differ 2010; 17: 1882.
[33] Tanjore H, Xu XC, Polosukhin VV, Degryse AL, Li B, Han W, Sherrill TP, Plieth D, Neilson EG, Blackwell TS, Lawson WE. Contribution of epithelial-derived fibroblasts to bleomycin-induced lung fibrosis. Am J Respir Crit Care Med 2009; 180: 657-665.
[34] Ji Y, Dou YN, Zhao QW, Zhang JZ, Yang Y, Wang T, Xia YF, Dai Y, Wei ZF. Paeoniflorin suppresses TGF-β mediated epithelial-mesenchymal transition in pulmonary fibrosis through a Smad-dependent pathway. Acta Pharmacol Sin 2016; 37: 794-804.
[35] Xie T, Liang J, Guo R, Liu N, Noble PW and Jiang D. Comprehensive microRNA analysis in bleomycin-induced pulmonary fibrosis identifies multiple sites of molecular regulation. Physiol Genomics 2011; 43: 479-487.
[36] Li M, Fu W, Wo L, Shu X, Liu F and Li C. miR-128 and its target genes in tumorigenesis and metastasis. Exp Cell Res 2013; 319: 3059.
[37] Shan ZN, Tian R, Zhang M, Gui ZH, Wu J, Ding M, Zhou XF, He J. miR128-1 inhibits the growth of glioblastoma multiforme and glioma stem-like cells via targeting BMI1 and E2F3. Oncotarget 2016; 7: 78813.
[38] Shi ZM, Wang J, Yan Z, You YP, Li CY, Qian X, Yin Y, Zhao P, Wang YY, Wang XF, Li MN, Liu LZ, Liu N, Jiang BH. MiR-128 inhibits tumor growth and angiogenesis by targeting p70S6K1. PLoS One 2012; 7: e32709.
[39] Guidi M, Muiños-Gimeno M, Kagerbauer B, Martí E, Estivill X, Espinosa-Parrilla Y. Overexpression of miR-128 specifically inhibits the truncated isoform of NTRK3 and upregulates BCL2 in SH-SY5Y neuroblastoma cells. BMC Mol Biol 2010; 11: 95.
[40] Woo HH, László CF, Greco S and Chambers SK. Regulation of colony stimulating factor-1 expression and ovarian cancer cell behaviorin vitroby miR-128 and miR-152. Mol Cancer 2012; 11: 58.
[41] Birtolo C, Pham H, Morvaridi S, Chheda C, Go VL, Ptasznik A, Edderkaoui M, Weisman MH, Noss E, Brenner MB, Larson B, Guindi M, Wang Q, Pandol SJ. Cadherin-11 is a cell surface marker up-regulated in activated pancreatic stellate cells and is involved in pancreatic cancer cell migration. Am J Pathol 2017; 187: 146-155.
[42] Marchong MN, Yurkowski C, Ma C, Spencer C, Pajovic S and Gallie BL. Cdh11 acts as a tumor suppressor in a murine retinoblastoma model by facilitating tumor cell death. PLoS Genet 2010; 6: e1000923.
[43] Yao J, Deng B, Zheng L, Dou L, Guo Y and Guo K. miR-27b is upregulated in cervical carcinogenesis and promotes cell growth and invasion by regulating CDH11 and epithelial-mesenchymal transition. Oncol Rep 2016; 35: 1645.
[44] Pedroza M. Cadherin-11 regulates pulmonary fibrosis in bleomycin-induced lung injury. Journal 2014; 398.
[45] Tuch D, Charlton T, Martic M. Cadherin-11 as a target for molecular imaging of the epithelial-mesenchymal transition and early fibrosis (P687, proceedings of the 2011 World Molecular Imaging Congress). Molecular Imaging & Biology 2012; 14: S629.