miR-192 is upregulated in T1DM, regulates pancreatic β-cell development and inhibits insulin secretion through suppressing GLP-1 expression

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Abstract. MicroRNAs (miRs) post-translationally regulate gene expression by specifically binding to the mRNA of their target genes. The aim of the present study was to determine the effect of miR-192 on pancreatic β-cell development. The serum levels of miR-192 in type 1 diabetes mellitus (T1DM) and streptozotocin-induced rats were determined, and were revealed to be elevated compared with those in healthy patients and normal rats, respectively. Western blot and reverse transcription-quantitative polymerase chain reaction analysis indicated that miR-192 suppressed the expression of glucagon-like peptide-1 (GLP-1), a potent insulin secretagogue. Ectopic expression of miR-192 inhibited cell proliferation and promoted apoptosis of NIT-1 cells, while miR-192 inhibitor had the opposite effect. Collectively, the present results revealed that miR-192 was elevated in T1DM, and is implicated in pancreatic β-cell development through regulation of cell proliferation and apoptosis, thereby suppressing insulin secretion. Furthermore, miR-192 suppressed GLP-1 expression, thereby further promoting T1DM. The present study suggested that miR-192 is a novel molecular target for the management or prevention of T1DM.

Introduction

Type 1 diabetes mellitus (T1DM) is an insulin-dependent type of diabetes (1). One of the key mechanisms of T1DM development is autoimmune-mediated destruction of the pancreatic β cells with consequent severe insulin deficiency (2-5). Patients with T1DM rely on daily insulin injections, while excess insulin injections may put patients at risk of hypoglycemia. Therefore, it is necessary to investigate the detailed mechanisms of T1DM.

MicroRNAs (miRs) are a class of non-coding, single-stranded small RNAs of 21-25 nucleotides in length, which regulate the expression of target genes through binding with their mRNA. Certain miRs are involved in pathological processes, including kidney diseases (6). Plasma miRs have been reported to be stable even under harsh conditions, including boiling, long-term storage at room temperature, high or low pH and multiple freeze-thaw cycles (7,8). Various studies have suggested that certain miRs have key roles in diabetes, including miR-23 and miR-144 (9,10). miR-192 has a key function in the formation and progression of kidney diseases (11,12). Krupa *et al* (13) indicated that the reduced expression of serum miR-192 deteriorated kidney diseases through facilitating the progression of kidney fibrosis. In addition, a previous study has indicated that miR-192 is aberrantly expressed in prediabetes (14), suggesting that miR-192 takes part in the development of diabetes. miR-192 has been reported to have a key role in DM and cardiovascular diseases (15). However, the detailed function of miR-192 in T1DM has remained to be elucidated.

Glucagon-like peptide-1 (GLP-1) is a potent insulin secretagogue and promotes glucose-induced insulin secretion (16,17). Furthermore, recent studies have indicated that GLP-1 reduces food intake by interacting with the hypothalamus (18,19). Several studies using cellular models of animal origin have investigated the regulation of GLP-1 secretion, including the STC-1 murine enteroendocrine cell line (20), fetal rat intestinal cell cultures (21), isolated canine L cells (22) and GLUTag (23).

The present study indicated that miR-192 was upregulated in T1DM patients and streptozotocin (STZ)-induced rats, and the expression of miR-192 was associated with the age and glucose concentration. miR-192 suppressed the GLP-1 expression in NCI-H716 cells. In addition, miR-192 inhibited the proliferation of pancreatic β-cell lines and insulin secretion. Taken together, the present study indicated that miR-192 has a

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key function in T1DM and suggested that miR-192 may serve as a novel molecular therapeutic target of T1DM.

Materials and methods

Cell line and culture conditions. Human NCI-H716 cells and NIT-1 mouse insulinoma β cells were obtained from the American Type Culture Collection (Manassas, VA, USA). NCI-H716 cells were grown in high-glucose Dulbecco's modified Eagle's medium (HyClone; GE Healthcare Life Sciences, Logan, UT, USA) supplemented with 100 IU/ml penicillin, 100 g/ml streptomycin, 2 mm L-glutamine and 10% fetal bovine serum (FBS; HyClone; GE Healthcare Life Sciences). NIT-1 cells were cultured in Ham's F12K medium with 100 IU/ml penicillin, 100 g/ml streptomycin, 2 mM L-glutamine and 10% FBS.

Animals. A total of 24 male Sprague-Dawley rats (weight, 250-300 g; age, 7-8 weeks) were obtained from the Experimental Animal Center of The Third Affiliated Hospital of Sun Yat-sen University (Guangzhou, China). The animal experiments were approved by the Animal Care Committee of The Third Affiliated Hospital of Sun Yat-sen University. Mice were housed under a 12 h light/dark cycle in a temperature (22.0±1.0˚C; 0.05% CO_2) and humidity (40-60%) controlled room and were provided with *ad libitum* access to rodent chow (Teklad 7001; 4.4%; Envigo Teklad Global Diets, Huntington, UK) and water. STZ (60 mg/kg; Sigma-Aldrich; Merck KGaA, Darmstadt, Germany) in 0.1 mol/l citrate buffer was used to induce Type 1 diabetes mellitus through single intraperitoneal (i.p) injection as reported previously (24). Control animals received buffer only. The blood glucose concentration was assessed using a commercial glucometer (Accu-Chek Sensor; Roche Inc., Mannheim, Germany) at four days after STZ injection. The animals with blood glucose levels of >16 mmol/l were considered diabetic and were selected for the experiments. The STZ+insulin group received 1.5 UI Novolin®N (Novo Nordisk, Gatwick, West Sussex, UK) insulin twice a day.

Patients and samples. All patients and healthy volunteers provided written informed consent. The present study was approved by the Ethics Committee of The Third Affiliated Hospital of Sun Yat-sen University. Patients were diagnosed according to the criteria of the American Diabetes Association (25). A total of 73 patients with T1DM (30 male, 43 female; age range, 9-29 years; average age, 16±3.12 years). Patients were excluded from the present study if that had clinical cardiovascular disease. Healthy control subjects were recruited from age- and gender-matched healthy blood donors. Blood samples were collected from The Third Affiliated Hospital of Sun Yat-sen University between April 2014 and October 2016 and were stored at -80˚C prior to use.

Cell transfection. The mimics negative control (pre-NC), miR-192 mimics (pre-miR-192), inhibitor negative control (NC-inh) and miR-192 inhibitors (miR-192 inh) were purchased from Ribobio, Inc (Guangzhou, China). Prior to transfection, the cells were grown to 70-80% confluence in 6-well plates. The mimics and inhibitor were transfected using Lipofectamine® 2000 according to the manufacturer's protocol (Invitrogen; Thermo Fisher Scientific, Inc., Waltham, MA, USA). After transfection for 48 h, the expression of miR-192 was determined by reverse transcription-quantitative polymerase chain reaction (RT-qPCR).

RT-qPCR. Total RNA was extracted from cells or tissues using TRIzol (Invitrogen; Thermo Fisher Scientific, Inc.) according to the manufacturer's protocol. The RNA concentration was measured using a NanoDrop® ND-1000 spectrophotometer (Thermo Fisher Scientific, Inc.). The complementary (c)DNA was synthesized using the High Capacity cDNA Reverse Transcription kit (Thermo Fisher Scientific, Inc.) according to the manufacturer's protocol. RT-qPCR was performed using SYBR-Green PCR Master Mix (Takara, Tokyo, Japan) on an ABI 7500 system (Applied Biosystems; Thermo Fisher Scientific, Inc.). Duplicate samples were normalized to GAPDH expression. The relative expression of indicated genes was presented as the mean value and calculated with the formula $2^{-\Delta\Delta Cq}$ (26). The primers used were as follows: GLP-1 forward, 5'-CATCAAATGCAGACTTGCCA-3' and reverse, 5'-ACCTCATTGTTGACAAAGCAG-3'; cyclin D1 forward, 5'-CCTCTAAGATGAAGGAGACCA-3' and reverse, 5'-AAT GAACTTCACATCTGTGGC-3'; cyclin E1 forward, 5'-ATG TTGACTGCCTTGAATTTCC-3' and reverse, 5'-ACCACT GATACCCTGAAACC-3'; caspase-9 forward, 5'-CTCTTC CTTTGTTCATCTCCT-3' and reverse, 5'-CAGGATGTAAGC CAAATCTG-3'; GAPDH forward, 5'-ATGATGACATCA AGAAGGTGGT-3' and reverse, 5'-TTGTCATACCAGGAA ATGAGCT-3'. For detecting miR-192 expression, miR-192 and U6 small nuclear (sn)RNA PCR primers were purchased from RiboBio (cat. no. miRQ0000517-1-2) and U6 snRNA was used as an internal control. The cycling conditions for all reactions were as follows: 98˚C for 1 min, 57˚C for 90 sec, and 72˚C for 90 sec over 32 cycles, followed by a 5-min extension step at 72˚C.

Western blot analysis. Following treatment, the protein was extracted from the cells or tissues using radioimmunoprecipitation assay lysis buffer (Sangon Biotech, Shanghai, China) for 45 min at 4˚C, followed by centrifugation at 13,000 x g for 10 min at 4˚C. Subsequently, the protein concentration was measured using a bicinchoninic acid kit (Pierce; Thermo Fisher Scientific, Inc.). Equal amounts of protein (45 μg) were separated by 10% SDS-PAGE and then transferred onto polyvinylidene fluoride membranes (Bio-Rad Laboratories, Hercules, CA, USA). The membranes were blocked with 5% non-fat milk in Tris-buffered saline supplemented with 0.1% Tween-20 (TBST) for 60 min at room temperature and were subsequently incubated with the primary antibodies, anti-GLP-1 (cat. no. ab23468; 1:2,000 dilution; Abcam, Cambridge, UK) and β-actin (cat. no. ab8226; 1:5,000 dilution; Abcam) at 4˚C overnight. The membranes were washed with TBST three times and then incubated with horseradish peroxidase-conjugated secondary immunoglobulin G (cat. no. ab97023; 1:5,000 dilution; Abcam) at room temperature for 60 min. The specific protein bands were visualized using an enhanced chemiluminescence kit (EMD Millipore, Billerica, MA, USA) according to the manufacturer's protocol. The results were quantified using

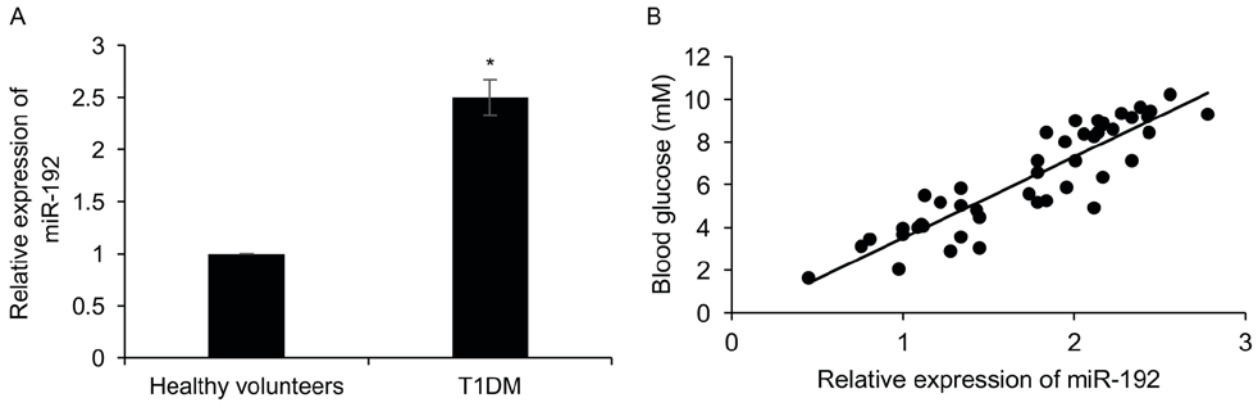

Figure 1. Expression of miR-192 is obviously upregulated in T1DM patients. (A) The expression of miR-192 in diabetic patients and healthy volunteers was determined by reverse transcription-quantitative polymerase chain reaction. *P<0.05, healthy volunteers vs. T1DM. (B) Correlation between the miR-192 expression and blood glucose concentration in T1DM analyzed by Pearson's correlation analysis (P=0.036; r=0.78). T1DM, type 1 diabetes; miR, microRNA.

Image J software (version 1.48; National Institutes of Health, Bethesda, MD, USA).

Assay of extracellular GLP-1. After transfection with miR-192 mimics or inhibitors, $3x10^5$ NCI-H716 cells were seeded in 6-well plates with 2 ml medium. After 48 h, the supernatants were collected and used to analyze the extracellular GLP-1 levels using an ELISA kit (cat. no. EZGLP1T-36K; EMD Millipore) according to the protocol of a previous study (27). Each experiment was repeated three times.

Cell proliferation assay. Following treatment, ~3,000 NIT-1 cells were placed into 96-well plates and incubated at 37˚C with 5% CO_2. Every 24 h, the cell proliferation was detected using a Cell Counting Kit-8 (CCK-8) kit (Beyotime Institute of Biotechnology, Haimen, China) according to the manufacturer's instructions. The absorbance at 450 nm was measured using a Varioskan Flash (Thermo Fisher Scientific, Inc.). Each experiment was repeated three times.

Colony formation assay. To assess the effects of miR-192 on the growth of pancreatic β cells, ~5,000 NIT-1 cells per well were seeded into 6-well plates and incubated for 14 days. The medium was replaced every two days. After the incubation, the colonies were fixed with methanol at room temperature for 15 min and then stained with 0.1% crystal violet (Sigma-Aldrich; Merck KGaA) at room temperature for 15 min. The number of colonies was counted under a light microscope. Each experiment was repeated three times.

Analysis of cell apoptosis. Annexin V-propidium iodide (PI) staining was used to detect cell apoptosis. In brief, after transfection of NIT-1 cells with miR-192 mimics or inhibitors for 48 h, the cells were collected and washed with PBS for three times. The cells were stained with 5 μl annexin V-fluorescein isothiocyanate at room temperature for 20 min in the dark in binding buffer (Sigma-Aldrich; Merck KGaA), 10 μl propidium iodide was then added and the cells were incubated at room temperature for 10 min. Finally, cell apoptosis was assessed using a FACSVerse flow cytometer (BD Biosciences, San Jose, CA, USA). Each experiment was repeated three times.

Statistical data analysis. Data analysis was performed with Prism 6 (GraphPad Inc., La Jolla, CA, USA). Values are expressed as the mean ± standard deviation. The correlation between the clinicopathological features of type 1 diabetic patients and miR-192 expression was assessed using the χ^2 test. A two-tailed Student's t-test was used to analyze differences between two groups. The correlation between blood glucose levels and miR-192 expression was determined by Pearson's correlation coefficient. P<0.05 was considered to indicate a statistically significant difference.

Results

Expression of miR-192 is obviously upregulated in type 1 diabetic patients. To investigate the role of miR-192 in diabetes, RT-qPCR analysis was performed to determine the expression of miR-192 in the plasma of 73 healthy volunteers and 46 T1DM patients, revealing that compared with the healthy volunteers group, miR-192 was obviously increased 2.5-fold in T1DM patients (Fig. 1A). In addition, the patients with high expression of miR-192 had a higher glucose concentration than those with low expression of miR-192 (r=0.78; Fig. 1B). Subsequently, the correlation between the expression of miR-192 and patients' clinicopathological characteristics was analyzed. As presented in Table I, high expression miR-192 was closely associated with age, but not with gender (Table I).

miR-192 is obviously upregulated in an STZ-induced rat model of T1DM. To determine the role of miR-192 in T1DM, STZ-induced model rats of T1DM were established. The diabetic features of the model rats, including plasma blood glucose levels and body weight, were detected. Blood samples were collected from the rats at 4 weeks post-STZ injection. Results demonstrated that body weight decreased and blood glucose levels were improved following STZ injection; however, an insulin injection only partially offset the effect of STZ, compared with control group (Table II). RT-qPCR analysis was performed to detect the expression of miR-192 compared with that in normal control rats, which revealed that miR-192 was increased 2.2-fold in

Table I. Clinicopathological parameters in 73 type 1 diabetic patients stratified by miR-192 expression status.

Parameter	No. (%)	miR-192 expression		P-value
		Low (n=25) (%)	High (n=48) (%)	
Age (years)				
<20	31 (42.5)	15 (20.5)	16 (20)	0.029
≥20	42 (57.5)	10 (13.7)	32 (43.8)	
Sex				
Male	30 (41.1)	12 (16.4)	18 (24.7)	0.387
Female	43 (58.9)	13 (17.8)	30 (41.1)	
Glucose concentration (mM)				
<7.0	24 (32.9)	16 (21.9)	8 (11.0)	<0.001
>7.0	49 (67.1)	9 (12.3)	40 (54.8)	

miR, microRNA.

Table II. Body weight and plasma glucose in the experimental rats (4 weeks).

Group	No.	Body weight (g)	Blood glucose (mmol/l)
Control	8	377.1±18.62	8.35±0.19
Diabetic (4 weeks)	8	264.3±31.46[a]	23.3±3.31[a]
Diabetic + insulin (4 weeks)	8	348.3±19.30	8.36±2.72

[a]P<0.05 vs. control group (4 weeks).

STZ-induced rats, but insulin had no effect on miR-192 expression (Fig. 2A). Subsequently, the expression of GLP-1, a potent insulin secretagogue, was detected in STZ-induced rats, which suggested that GLP-1 was downregulated in STZ-induced rats compared with that in normal rats (Fig. 2B). Of note, insulin treatment had little effect on GLP-1 expression (Fig. 2B). Taken together, miR-192 was upregulated in STZ-induced rats, and negatively correlated with the expression of GLP-1.

miR-192 suppresses GLP-1 expression in NCI-H716 cells. To identify the downstream target genes of miR-192, potential targets of miR-192 were searched with a public algorithm, TargetScan (http://www.targetscan.org). It was indicated that miR-192 binds to a sequence in the 3'-UTR of GLP-1. It is known that miRs bind to sequences in the 3'-UTRs of their target genes, resulting in either inhibition of target mRNA translation or mRNA degradation. To verify whether miR-192 suppresses GLP-1 expression, possibly by directly binding to the putative binding site in the 3'-UTR of its mRNA, the effect of miR-192 on the expression of GLP-1 mRNA and protein was first detected. For this purpose, human NCI-H716 cells were used (28), which are widely used to assess the mechanisms underlying the secretion of GLP-1 (29). The NCI-H716 cell line, derived from a poorly differentiated adenocarcinoma of human cecum (30), has been described to have certain endocrine features, in particular the formation of secretory granules and chromogranin A expression (31). Furthermore, this cell line expresses several neurohormonal receptors, including receptors for gastrin, serotonin and somatostatin (32). NCI-H716 cells were transfected with miR-192 mimics or miR-192 inhibitors, and the expression of miR-192 was determined by RT-qPCR analysis. As presented in Fig. 3A and B, miR-192 was overexpressed or knocked down in NCI-H716 cells transfected with pre-miR-192 or miR-192-inh, respectively (Fig. 3A and B). In addition, the mRNA and protein levels of GLP-1 were determined by RT-qPCR and western blot analysis, respectively. The results demonstrated that the mRNA and protein levels of GLP-1 were downregulated when miR-192 was overexpressed. Conversely, miR-192 inhibition obviously increased the expression of GLP-1 (Fig. 3C and D). The extracellular GLP-1 levels were also assessed, which demonstrated that ectopic expression of miR-192 decreased the extracellular GLP-1 levels; however, miR-192 inhibition significantly increased the extracellular GLP-1 levels (Fig. 3E). The present results therefore reveal that miR-192 suppresses GLP-1 expression in NCI-H716 cells.

miR-192 suppresses the proliferation of pancreatic β-cell lines and insulin secretion. To further investigate the functions of miR-192 in the pancreatic β-cell development process, miR-192 was first overexpressed or knocked down in NIT-1 pancreatic β cells. After transfection for 48 h,

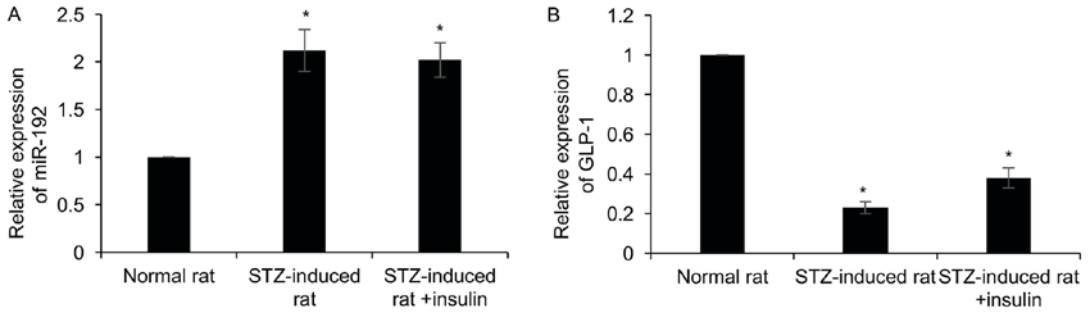

Figure 2. miR-192 is obviously upregulated in STZ-induced type 1 diabetic rats. (A) The expression of miR-192 in normal rats, STZ-induced rats and insulin-treated rats was determined by reverse transcription-quantitative polymerase chain reaction. (B) The expression of GLP-1 in the rats' blood was determined with a GLP-1 ELISA kit. *P<0.05, STZ-induced group and STZ-induced + insulin-treated group vs. normal rats. miR, microRNA; STZ, streptozotocin; GLP-1, glucagon-like peptide-1.

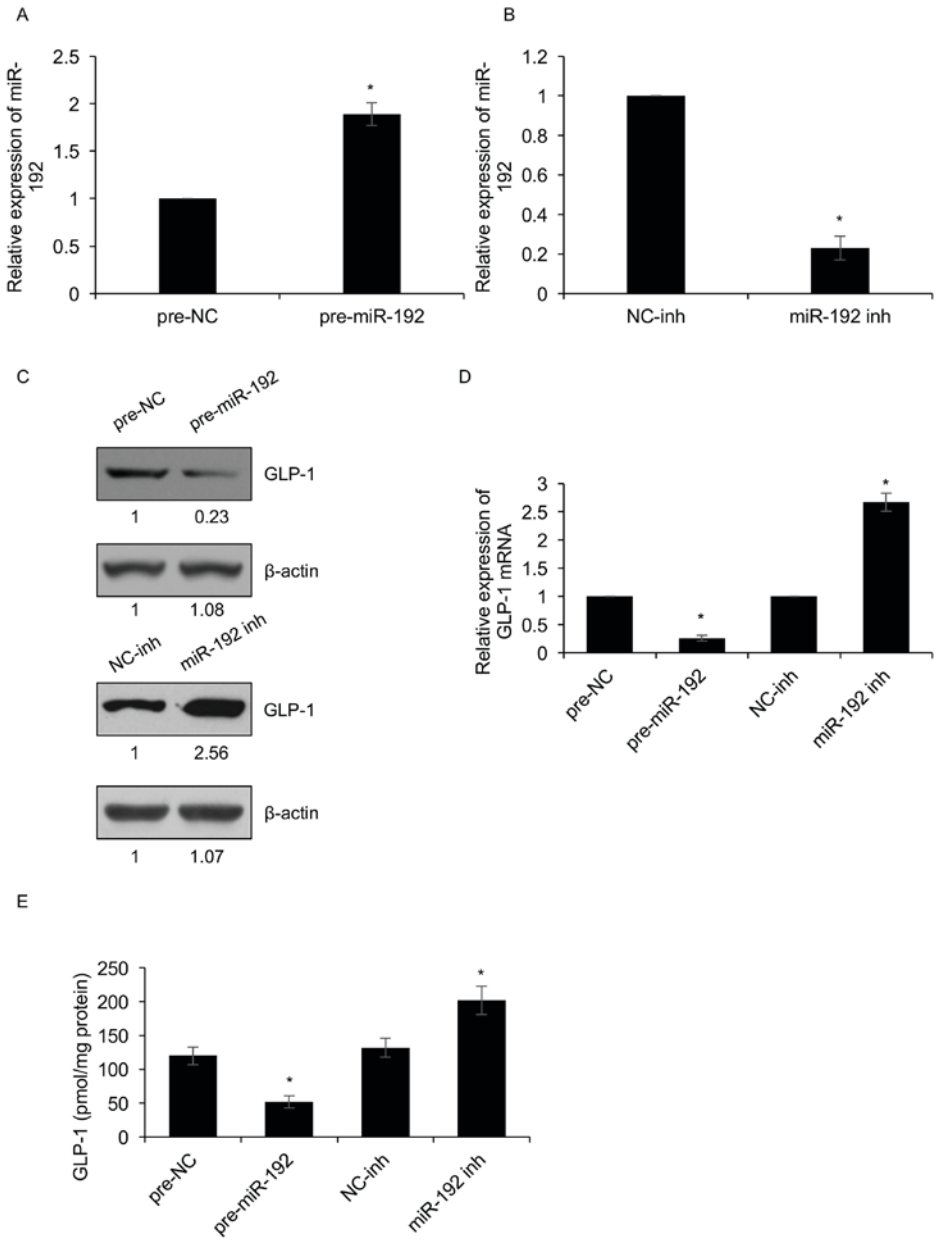

Figure 3. miR-192 suppresses GLP-1 expression in NCI-H716 cells. (A and B) Expression of miR-192 in NCI-H716 cells (A) following transfection with pre-NC or pre-miR-192 and (B) NC-inh or miR-192 inh for 48 h as determined by RT-qPCR. (C and D) Protein and mRNA levels of GLP-1 in NCI-H716 cells after transfection with miR-192 overexpression vector, inhibitor or the respective controls for 48 h as determined by (C) western blot analysis and (D) RT-qPCR, respectively. (E) GLP-1 secretion in NCI-H716 cells treated as above was determined with a GLP-1 ELISA kit. *P<0.05, pre-miR-192 vs. pre-NC, miR-192 inh vs. NC-inh. NC, negative control; pre-miR-192, miR-192 mimics; inh, inhibitor; miR, microRNA; GLP-1, glucagon-like peptide-1; RT-qPCR, reverse transcription-quantitative polymerase chain reaction.

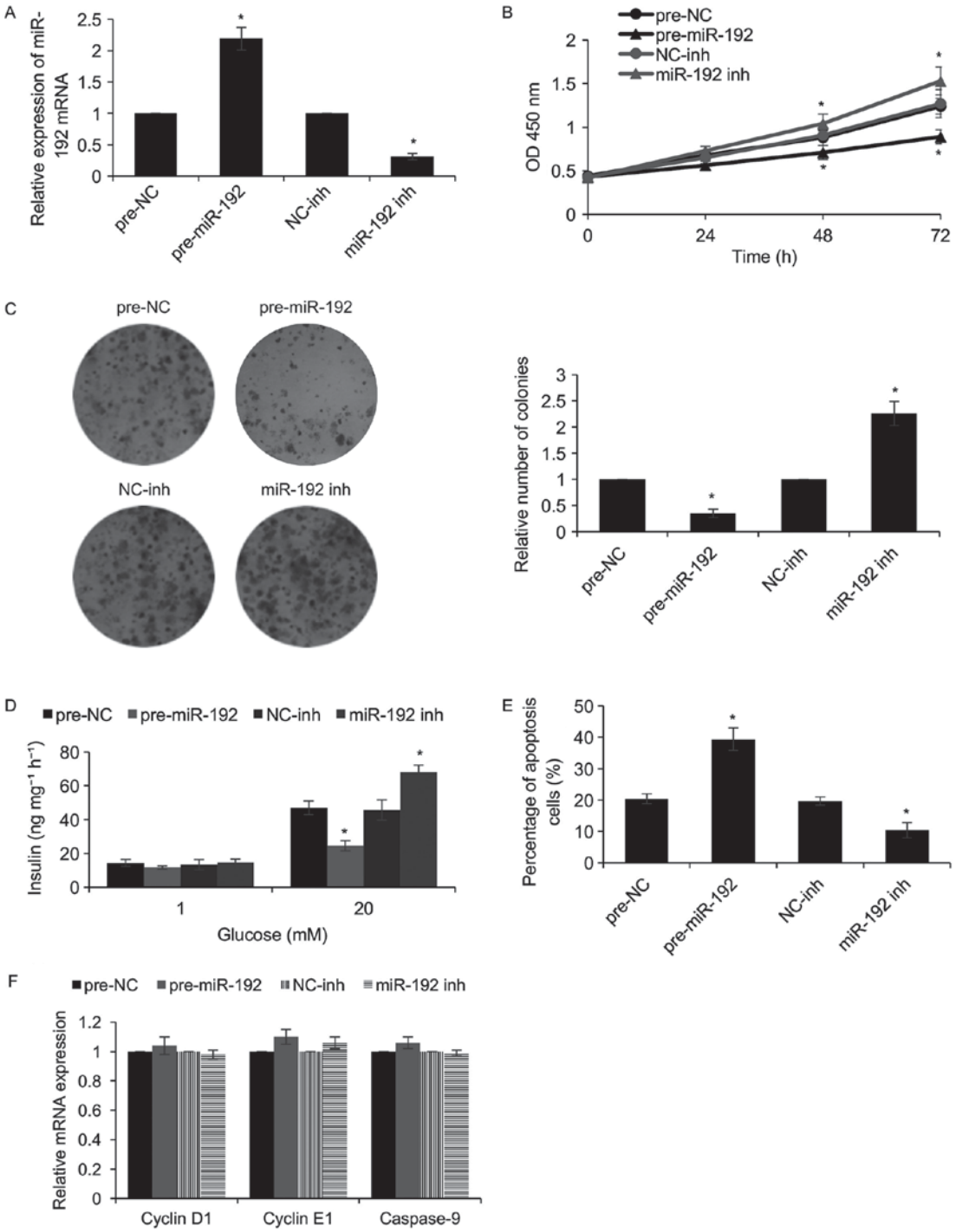

Figure 4. miR-192 suppresses the proliferation of a pancreatic β-cell line and the secretion of insulin. miR-192 was overexpressed or knocked down in NIT-1 cells by transfection for 48 h. (A) The levels of miR-192 were determined by RT-qPCR. (B) A Cell Counting Kit-8 assay was used to determine the effect of miR-192 on cell proliferation. (C) A colony formation assay was used to determine the effect of miR-192 on cell proliferation (magnification, x20). (D) The cells were incubated with 1 or 20 mM glucose for 100 min, and glucose-stimulated insulin secretion was determined in each group. Each experiment was repeated for three times. (E) The apoptotic rate in the different groups was assessed to determine the effect of miR-192 on cell apoptosis. (F) The expression of cyclin D1, cyclin E1 and caspase-9 was determined by RT-qPCR. *P<0.05, pre-miR-192 vs. pre-NC, miR-192 inh vs. NC-inh. NC, negative control; pre-miR-192, miR-192 mimics; inh, inhibitor; miR, microRNA; GLP-1, glucagon-like peptide-1; RT-qPCR, reverse transcription-quantitative polymerase chain reaction; OD, optical density.

the expression of miR-192 was detected by RT-qPCR. As presented in Fig. 4A, miR-192 was markedly increased when NIT-1 cells were transfected with miR-192 mimics when compared with that in the control groups, while miR-192 was significantly downregulated when NIT-1 cells were transfected with miR-192 inhibitors. Subsequently, a CCK-8 assay was used to determine the effect of miR-192 on cell proliferation, which revealed that overexpression of miR-192 suppressed cell proliferation, while inhibition of miR-192 promoted cell proliferation (Fig. 4B). Furthermore, a colony formation assay confirmed that ectopic expression of miR-192 suppressed the cologenicity

of NIT-1 cells, whereas inhibition of miR-192 facilitated it (Fig. 4C). In addition, in response to glucose stimulation, ectopic expression of miR-192 obviously decreased the insulin secretion, while inhibition of miR-192 markedly increased it (Fig. 4D). T1DM is an autoimmune disease, and to detect whether miR-192 regulates apoptosis of pancreatic β-cells, a flow cytometric assay was performed, which revealed that ectopic expression of miR-192 increased the percentage of apoptotic NIT-1 cells compared with that in the control groups, whereas miR-192 inhibition decreased the percentage of apoptotic cells (Fig. 4E). To further investigate the mechanisms by which miR-192 inhibits cell proliferation and induces apoptosis, it was assessed whether miR-192 regulated the expression of cell cycle- and apoptosis-associated genes, including cyclin D1, cyclin E1 and caspase-9. As presented in Fig. 4F, miR-192 had no effect on the mRNA expression of cyclin D1, cyclin E1 and caspase-9 (Fig. 4F). Taken together, the present results reveal that miR-192 suppresses the proliferation of pancreatic β cells through facilitating cell apoptosis, thereby reducing insulin secretion.

Discussion

T1DM is an insulin-dependent type of diabetes (1). One of the key pathophysiological mechanisms of T1DM is the autoimmune-mediated destruction of pancreatic β-cells with consequent severe insulin deficiency (2-5).

miR-192 has been identified to have several key roles in diabetic kidney disease (33); however, the detailed functions of miR-192 in T1DM have remained to be elucidated.

The present study revealed that miR-192 was upregulated in T1DM patients and STZ-induced rats. Furthermore, insulin treatment had no effect on the expression of miR-192 in STZ-induced rats. In addition, the expression of miR-192 was closely associated with age and high glucose levels. To further assess the effect of miR-192 in T1DM, the pancreatic β cell line NIT-1 was used. A colony formation assay and a CCK-8 assay were performed to determine the effect of miR-192 on cell proliferation, which revealed that miR-192 suppressed the proliferation of pancreatic β cell lines and insulin secretion. Furthermore, miR-192 was indicated to promote β-cell apoptosis. A recent study has demonstrated that miR-192 suppresses cell proliferation and induces apoptosis in human rheumatoid arthritis fibroblast-like synoviocytes and lung cancer (34,35). The present results were consistent with the previous ones, as it was indicated that miR-192 promoted T1DM through suppressing pancreatic β-cell proliferation and facilitating cell apoptosis. However, the detailed mechanisms by which miR-192 affects cell proliferation and apoptosis have remained elusive. It was then detected whether miR-192 regulates certain cell cycle- and apoptosis-associated proteins, including cyclin D1, cyclin E1 and caspase-9, but as it appeared to have no effect the mRNA expression of these genes, it was assumed that miR-192 regulates the proliferation and apoptosis of pancreatic β cells through other genes/proteins.

GLP-1 is a potent insulin secretagogue and promotes glucose-induced insulin secretion (16,17). As NCI-H716 cells were previously used to investigate GLP-1 (29), they were also used in the present study. Furthermore, NIT-1 cells were employed to investigate the effect of miR-192 on pancreatic β cells, including their proliferation and apoptosis. The results indicated that miR-192 suppresses GLP-1 expression, indicating miR-192 promoted T1DM also through negative regulation of GLP-1. However, the direct binding of miR-192 to the 3'UTR of GLP-1 mRNA should be assessed using a luciferase assay in future studies.

In conclusion, to the best of our knowledge, the present study indicates that miR-192 is upregulated in T1DM and determined the role of miR-192 in T1DM. miR-192 was identified as a driver in T1DM. Mechanisms via which miR-192 suppresses pancreatic β-cell proliferation and insulin secretion were provided, and it was demonstrated that miR-192 suppresses GLP-1 expression in NCI-H716 cells. Additionally, miR-192 also suppresses cellular proliferation and promotes apoptosis in NIT-1 cells. It was suggested that miR-192 is a novel diagnostic biomarker for T1DM.

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Availability of data and materials

The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

Authors' contributions

WP and JW conceived and designed the study. WP, YZ, FX JY and CZ performed the experiments. WP and JY wrote the paper. WP and JW reviewed and edited the manuscript. All authors read and approved the manuscript and agree to be accountable for all aspects of the research in ensuring that the accuracy or integrity of any part of the work are appropriately investigated and resolved.

Ethical approval and consent to participate

The animal experiments were approved by the Animal Care Committee of The Third Affiliated Hospital of Sun Yat-sen University (Guangzhou, China). All patients and healthy volunteers provided written informed consent. The patient study has been approved by the Ethics Committee of The Third Affiliated Hospital of Sun Yat-sen University.

Patient consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

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