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LINC00210 plays oncogenic roles in non-small cell lung cancer by sponging microRNA-328-5p

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Abstract. Long noncoding RNA (lncRNA) has an important role in regulating non-small cell lung cancer (NSCLC) progression. The present study aimed to investigate the effect of LINC00210 in NSCLC progression in order to provide a novel treatment target for patients with NSCLC. A total of 39 NSCLC patients were obtained and divided into LINC00210 high expression and low expression groups. Subsequently, the 5-year survival rate from this patient cohort was analyzed. The SK-MES-1 and A549 NSCLC and the human 16-HBE bronchial epithelial cell lines were utilized to investigate expression level of LIN00210. A549 cells were used to investigate cell proliferation, migration and invasive abilities using Cell Counting kit 8, Transwell and Matrigel assays, respectively. In addition, the luciferase reporter gene assay was performed to investigate the potential target of LINC00210. Reverse transcription-quantitative PCR was used to determine LINC00210 and microRNA (miR)-328-5p expression levels in NSCLC tissues and tumor cell lines (SK-MES-1 and A549). The results demonstrated that LINC00210 was upregulated in NSCLC tissues and cell lines compared with that in normal tissues and 16-HBE cells, and that LINC00210 expression was associated with a poor prognosis in patients with NSCLC ($P<0.05$). Furthermore, A549 cell transfection with small interfering (si)LINC00210#1 and siLINC00210#2 induced a significant decrease in cell proliferation, and migratory and invasive abilities compared with that in the control groups ($P<0.05$). In addition, miR-328-5p overexpression was stimulated by knockdown of LINC00210. Furthermore, A549 cells transfected with siLINC00210#1 and miR-328-5p inhibitor exhibited a significant increase in cell proliferation, and migratory and invasive ability compared with that in A549 cells transfected with siLINC00210#1. These findings suggest that LINC00210 may serve as an oncogenic role in NSCLC by sponging miR-328-5p.

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Introduction

Lung cancer is the leading cause of cancer-associated mortality in men and women and is responsible for 26% of all cancer-associated deaths worldwide (1). Non-small cell lung cancer (NSCLC) is the predominant type of lung cancer, accounting for ~80% of all lung cancers (1). Furthermore, >50% patients with NSCLC present with locally advanced or distantly metastatic cancer at the time of diagnosis (1,2). Although targeted therapy has improved the management and outcome of patients with NSCLC, the overall 5-year survival rates of patients with NSCLC and advanced NSCLC are only 18 and 4%, respectively (1).

Recently, long non-coding RNAs (lncRNAs) have attracted great attention in the field of cancer. They serve crucial roles in the development and progression of cancer and might therefore represent a potential therapeutic target to treat various types of cancer, including lung cancer and colon cancer (3). Selective inhibition of target lncRNA by RNA interference technology represents the most suitable approach. Small interfering (si)RNAs can significantly inhibit the expression of lncRNAs in tumor cells. For example, HOX transcript antisense RNA (HOTAIR) knockdown by siRNA can reduce lung cancer cell proliferation and invasive capacity *in vitro* (4). In addition, HOTAIR silencing by siRNA can reverse cisplatin resistance in lung adenocarcinoma cells via p21 downregulation (5). Cheng *et al* (6) reported that the expression of urothelial cancer associated 1 (UCA1) lncRNA is significantly increased in patients with NSCLC and that UCA1 knockdown can partly improve the gefitinib sensitivity of gefitinib-resistant NSCLC cell lines. *In vivo* experiments also demonstrated that high expression of UCA1 might present a novel mechanism for the acquired resistance of gefitinib-resistant NSCLC cell lines (6). A previous study also demonstrated that metastasis associated lung adenocarcinoma transcript 1 (MALAT1) inhibition using a siRNA can reduce NSCLC cell migratory and invasive ability (7). In addition, it was demonstrated *in vitro* that Pvt1 oncogene knockdown can inhibit the proliferation and

induce apoptosis of NSCLC cells by sponge-like adsorption of microRNA-195 therefore, increasing sensitivity to radiotherapy of these cells (8). LncRNAs may therefore be used to develop novel targeted therapy to treat NSCLC.

The present study determined the expression of LINC00210 in NSCLC tumor tissues and cells and investigated its effects on NSCLC progression. Moreover, a previous study identified that LINC00210 sponges miR-328-5p (9). Thus, this present study explored whether LINC00210 also sponges miR-328-5p in NSCLC. Through luciferase reporter assays, it was demonstrated that LINC00210 targeted miR-328-5p to promote NSCLC progression. The findings from the present study may provide a novel therapeutic target for the diagnosis and treatment of NSCLC.

Materials and methods

Ethical statement. The present study was approved by the Ethics Committee of China-Japan Union Hospital of Jilin University and patients provided written informed consent.

Patients and samples. A total of 39 patients who were diagnosed for the first time with NSCLC, according to the grading system of the American Joint Committee on Cancer (10), at the China-Japan Union Hospital of Jilin University, between March 2010 and July 2012, were included in the present study. Patients treated with radiotherapy or chemotherapy before surgery were excluded. The 39 patients with NSCLC included 29 men and 10 females, and their mean age was 58.7±9.2 years. The clinicopathological characteristics of all patients are presented in Table I. Patients' carcinoma tissues and corresponding adjacent normal tissues (at least 3 cm from tumor tissues) were collected during resection and immediately stored at -20˚C. Following surgery, all patients were followed-up every 3 months for 5 years through telephone consultations, to analyze their 5-year survival rate using Kaplan-Meier survival analysis.

Cell culture. The SK-MES-1 and A549 NSCLC, and the human 16-HBE bronchial epithelial cell line were all provided by The Cell Bank of Type Culture Collection of the Chinese Academy of Sciences. All cells were maintained separately in Dulbecco's Modified Eagle medium (DMEM) supplemented with 10% FBS (Invitrogen; Thermo Fisher Scientific, Inc.), 100 μg/ml streptomycin and 100 U/ml penicillin and cultured at 37˚C in a humidified incubator containing 5% CO_2.

Transfection. A total of two siRNAs against LINC00210 (siRNA#1: 5'-GGUUCUCAUUCUCAUUUAAUU-3' and siRNA#2: 5'-CGGUAUUAUGACCACUACUUU-3') as well as LINC00210 scramble siRNA negative control (NC) (5'-AAUUCUCCGAACGUGUCACGU-3'), miR-328-5p mimics (5'-GGGGGGGCAGGAGGGGCUCAGGG-3'), miR-328-5p NC (scrambled; 5'-UCACAACCUCCUAGA AAGAGUAGA-3') and miR-328-5p inhibitors (5'-CCC TGAGCCCCTCCTGCCCCCCC-3') were all synthesized by Shanghai GenePharma Co., Ltd. A549 cells cultured in serum free DMEM were transfected with 100 nM of the siRNA/miRNA/miRNA inhibitor, using Lipofectamine® 2000 (Thermo Fisher Scientific, Inc.). After transfection, A549 cells were grouped as follows: siLINC00210#1 group (transfected with LINC00210 siRNA#1), siLINC00210#2 group (transfected with LINC00210 siRNA#2), scramble group (transfected with LINC00210 siRNA NC), miR-NC group (transfected with miR-328-5p NC), miR-328-5p group (transfected with miR-328-5p mimics), siLINC00210#1 + miR-328-5p inhibitor group (co-transfected with LINC00210 siRNA#1 and miR-328-5p inhibitors). The transfection efficiency was confirmed using reverse transcription-quantitative PCR (RT-qPCR), 24 h after transfection and transfected cells were used for following experiments.

Cell proliferation assay. Cell proliferation was measured using Cell Counting Kit-8 (CCK8) assay (Sigma-Aldrich; Merck KGaA). Transfected A549 cells were seeded into 96-well plates ($2x10^3$ cells/well) and cultured for 24, 48, 72 and 96 h. CCK8 solution (20 μl; 0.5 mg/ml; Sigma-Aldrich; Merck KGaA) was added into each well and incubated for 2 h at 37˚C. Absorbance was measured at 450 nm using a microplate reader (Thermo Labsystems).

Transwell assay. To investigate cell invasion, 24-well Transwell chambers were pre-coated with Matrigel (BD Biosciences) for 30 min at 37˚C and were inserted into 24-well plates containing 1 ml DMEM supplemented with 10% FBS. A total of 100 μl of A549 cells in serum free-suspension ($1x10^5$ cells/ml) was added to 24-well Transwell chambers and incubated in a humidified incubator at 37˚C with 5% CO_2. After 2 days cells that had not penetrated the membrane were removed with a cotton swab. Cells that penetrated and adhered to the lower side of the membrane were fixed with 4% paraformaldehyde for 30 min at room temperature and stained with crystal violet (0.1%) for 10 min at room temperature. The number of invading cells was counted in five random fields under the light microscope at 100x magnification. Cell number was counted using ImageJ (version 1.41; National Institutes of Health). For the assessment of cell migration, the same method was performed, although the 24-well Transwell chambers, were not pre-coated with Matrigel.

Luciferase reporter gene assay. To determine if LINC0020 is a target of miR-328-5p the target gene prediction software miRcode 11 (http://www.mircode.org/) was used. Mutant (Mut) and wild-type (WT) sequences of the LINC00210 containing the 3'untranslated region were designed and amplified using PCR. A549 cells from the miR-NC and miR-328-5p groups were seeded in 24-well plates ($1x10^4$ cells per well), and transfected with pGL3-LINC00210-Mut and pGL3-LINC00210-WT plasmids (Promega Corporation), respectively using Lipofectamine® 2000, and cultured for 24 h at 37˚C. Cells were collected and firefly and *Renilla* luciferase activity was detected using a Dual-Luciferase Reporter Assay System kit (Promega Corporation) according to the manufacturer's protocol. Firefly luciferase activity was normalized to *Renilla* luciferase activity.

RT-qPCR. Total RNA was extracted from tissue samples and cells using TRIzol® reagent (Invitrogen; Thermo Fisher Scientific, Inc.) according to the manufacturer's protocol. Total RNA was reverse transcribed into cDNA at 37˚C for 15 min

Table I. Association between LINC00210 expression and the clinicopathological characteristics of patients with non-small cell lung cancer.

Characteristics	LINC00210 low expression (n=18)	LINC00210 high expression (n=21)	P-value
Sex			0.6508
Male	14	15	
Female	4	6	
Age, years			0.5189
≤60	13	17	
>60	5	4	
Tumor size, cm			0.0071[a]
≤3	12	5	
>3	6	16	
TNM stage			0.0160[a]
I and II	9	3	
III and IV	9	18	
Lymph node metastasis			0.0174[a]
Negative	12	6	
Positive	6	15	

[a]P<0.05 (χ^2 test). TNM, Tumor-Node-Metastasis.

using a Reverse Transcription kit (Applied Biosystems; Thermo Fisher Scientific, Inc.) according to the manufacturer's protocol. A similar amount of cDNA product in each sample was subjected to PCR amplification using the Applied Biosystems 7300 Real Time PCR system (Applied Biosystems; Thermo Fisher Scientific, Inc.). The qPCR were performed using a SYBR Green I Master Mix kit (Thermo Fisher Scientific, Inc.) according to the manufacturer's protocol. as follows: Initial denaturation for 5 min at 95˚C, followed by 39 cycles of denaturation at 95˚C for 30 sec and annealing at 60˚C for 45 sec. The sequences of the primers (Sangon Biotech Co., Ltd.) were as follows: LINC00210 forward, 5'-AACACGTTAGCGGGTTCTCA-3' and reverse, 5'-TCAAAAACCACCGAGGGAGG-3'; miR-328-5p forward, 5'-AACGAGACGACGACAGAC-3' and reverse, 5'-GGGGGG GCAGGAGGGGCTCAGGG-3'; GAPDH forward, 5'-ATG TTGCAACCGGGAAGGAA-3', reverse 5'-AGGAAAAGC ATCACCCGGAG-3'. and U6 forward, 5'-AACGAGACGACG ACAGAC-3' and reverse 5'-GCAAATTCGTGAAGCGTT CCATA-3'. The relative expressions levels of LINC00210 and miR-328-5p were normalized to the endogenous control U6 and expressed as $2^{-\Delta\Delta Cq}$ (11).

LINC00210 distribution detection. For A549 cells, nuclear and cytoplasmic RNA were separated using a Cytoplasmic and Nuclear RNA Purification kit (Norgen Biotek Corp.) according to the manufacturer's instructions. The LINC00210 expression level in the nucleus and cytoplasm was determined using RT-qPCR. U6 was used as endogenous control for LINC00210 expression level in nucleus, whereas GAPDH used as the endogenous control for cytoplasmic LINC00210 expression level.

Statistical analysis. All data are presented as the mean ± standard deviation and were analyzed using SPSS v19.0 software (IBM Corp.) or GraphPad Prism v6 (GraphPad Software, Inc.). The correlation between LINC00210 and miR-328-5p expression level was determined using Pearson's correlation analysis. Comparison between two groups was performed using unpaired two tailed Student's t-test. One-way analysis of variance followed by Tukey's post hoc test was used for comparison of multiple groups. The Kaplan-Meier method was used to create survival curves and the log-rank test was used to determine statistical significance. χ^2 test was used to determine the association between LINC00210 expression level and the clinicopathological characteristics of the 39 patients with NSCLC. P<0.05 was considered to indicate a statistically significant difference. Each experiment was repeated at least three times.

Results

LINC00210 is overexpressed in NSCLC. The results from the RT-qPCR revealed that LINC00210 expression level was significantly increased in NSCLC tissues compared with that in adjacent tissues (P<0.05; Fig. 1A). Similarly, LINC00210 expression level was significantly increased in the SK-MES-1 and A549 NSCLC cell lines compared with that in the human 16-HBE bronchial epithelial cell line (P<0.05; Fig. 1B). These results suggested that LINC00210 may by overexpressed in NSCLC.

According to the follow-up study, patients with high LINC00210 expression levels, using the median value as the cut-off (n=21), had significantly lower overall 60-month survival compared with patients with low LINC00210 expression level (n=18; P<0.05; Fig. 1C; Log Rank χ^2 value, 4.6). Analysis of the association between LINC00210 expression level and the clinicopathological characteristics of patients with NSCLC revealed that LINC00210 expression level was significantly associated with tumor size, TNM stage and lymph node metastasis (Table I; P<0.05). High LINC00210 expression level may therefore be used to predict larger tumor size, advanced tumor stage and positive lymph node metastasis. The upregulation of LINC00210 in patients with NSCLC indicated a poor prognosis.

LINC00210 knockdown inhibits A549 cell proliferation, and migratory and invasive abilities. A549 cells in the siLINC00210#1 and siLINC00210#2 groups exhibited a significantly decreased LINC00210 expression level compared with that in A549 cells from the scramble NC group (P<0.05; Fig. 2A), indicating that LINC00210 was successfully downregulated in A549 cells following transfection. The results from the CCK8 assay revealed that the absorbance was significantly reduced in A549 cells in the siLINC00210#1 and siLINC00210#2 groups compared with that in A549 cells from the scramble NC group (P<0.05; Fig. 2B). In addition, transfection with siLINC00210#1 and siLINC00210#2 induced a significant decrease in the migratory and invasive ability of A549 cells compared with that in A549 cells from the scramble NC group (P<0.05; Fig. 2C and D).

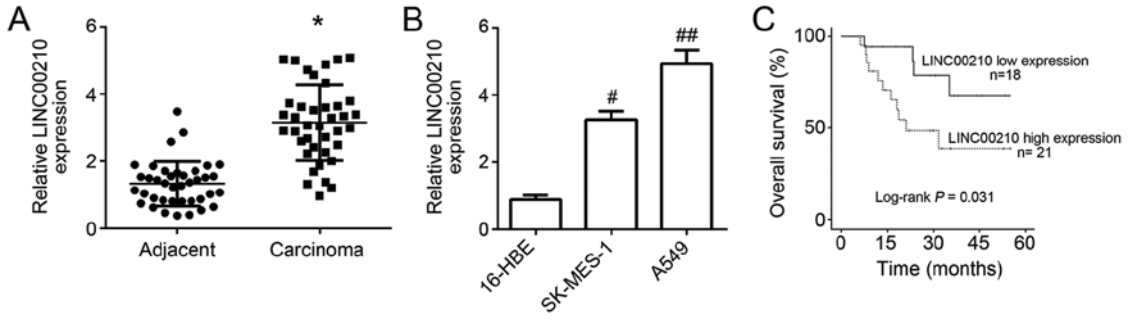

Figure 1. LINC00210 is upregulated in NSCLC. (A) Carcinoma tissues (n=39) exhibited higher LINC00210 expression level compared with that in adjacent normal tissues (n=39). (B) SK-MES-1 and A549 NSCLC cell lines had a significantly higher LINC00210 expression level compared with that in 16-HBE cells. (C) Patients with high LINC00210 expression level had a significantly lower overall 60-month survival time compared with patients with low LINC00210 expression level (Log Rank χ^2 value, 4.6). *P<0.05 vs. adjacent tissues. #P<0.05, ##P<0.01 vs. 16-HBE cells. NSCLC, non-small cell lung cancer.

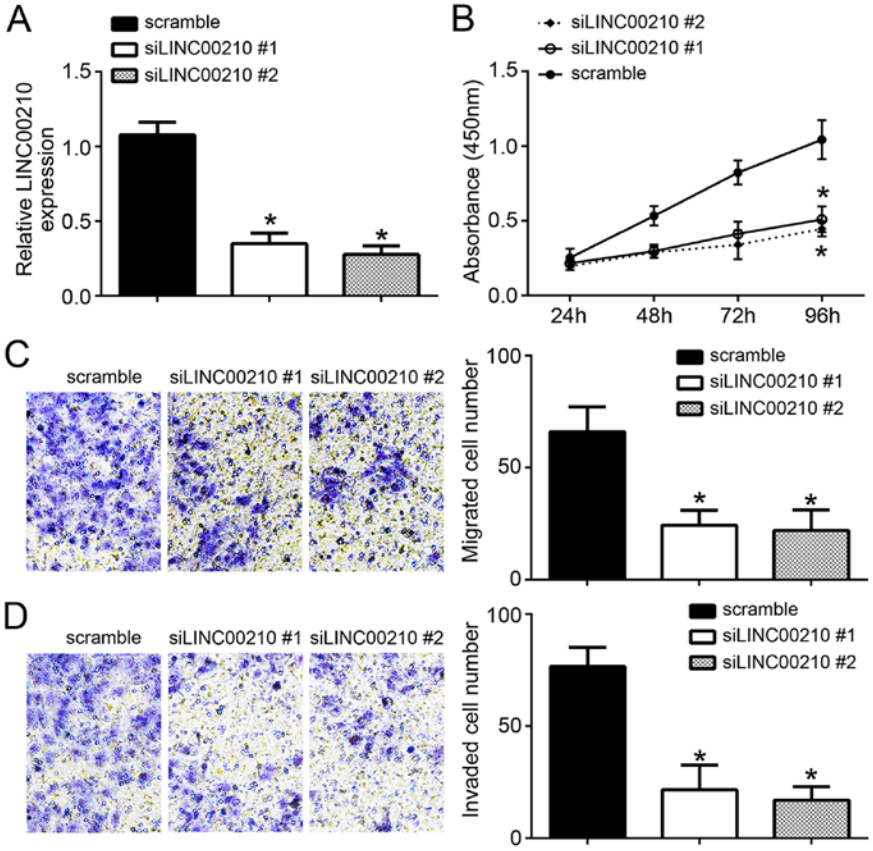

Figure 2. LINC00210 knockdown inhibits the proliferation, and migratory and invasive abilities of A549 cells. (A) LINC00210 was successfully down-regulated in A549 cells following transfection with siLINC00210#1 and #2. Expression of LINC00210 was measured 24 h after transfection. LINC00210 knockdown inhibited the (B) proliferation, and (C) migratory and (D) invasive abilities of A549 cells. *P<0.05 vs. the scrambled siRNA. si, small interfering.

LINC00210 knockdown directly promotes miR-328-5p expression. The distribution of LINC00210 expression level in A549 cells was investigated. As presented in Fig. 3A, LINC00210 was mainly distributed in the cytoplasm. A previous study reported that LINC00210 can interact with miR-328-5p (9). Through bioinformatics analysis, miR-328-5p was also identified as a potential target of LINC00210, therefore miR-328-5p was investigated in the present study. The WT and Mut sequences of LINC00210 were designed separately, and their binding sites to miR-328-5p were presented in Fig. 3B. A luciferase reporter assay was performed following A549 cell transfection with miR-328-5p mimics and WT- or Mut-LINC00210 reporter plasmid. The results revealed that miR-328-5p mimics significantly inhibited the relative luciferase activity of WT-LINC00210, but not Mut-LINC00210 (Fig. 3C; P<0.05) compared with that in miR-NC group. Furthermore, A549 cells from the siLINC00210#1 and siLINC00210#2 groups exhibited a significant increase in miR-328-5p expression level compared with that in A549 cells from the scramble NC group (P<0.05; Fig. 3D). The results from the Pearson's correlation analysis revealed a negative correlation between LINC00210 and miR-328-5p expression levels in NSCLC tissues

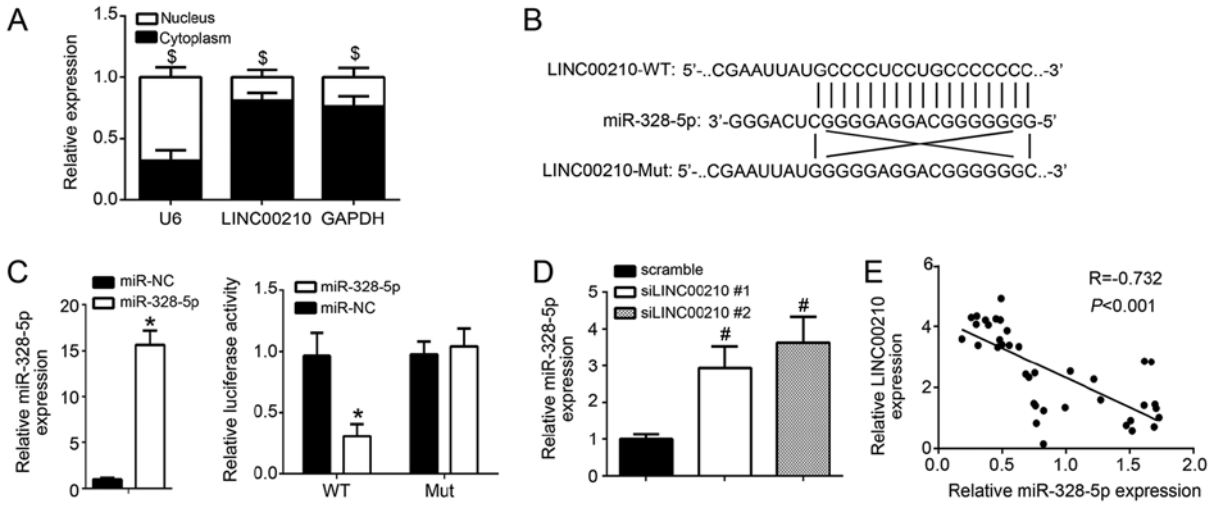

Figure 3. LINC00210 knockdown directly promotes miR-328-5p expression. (A) LINC00210 was mainly distributed in the cytoplasm. (B) Binding sites of WT or Mut LINC00210 sequences to miR-328-5p. (C) Luciferase reporter assay was conducted following A549 cell transfection with miR-328-5p mimics and WT- or Mut-LINC00210 reporter plasmids. Transfection efficiency with miR-328-5p mimics was confirmed by reverse transcription-quantitative PCR. (D) LINC00210 knockdown promoted miR-328-5p expression in A549 cells. (E) Pearson's correlation analysis revealed a negative correlation between LINC00210 and miR-328-5p expression levels in NSCLC tissues. *P<0.05 vs. miR-NC, #P<0.05 vs. scramble and $P<0.05 vs. nucleus. miR, microRNA; Mut, mutant; NC, negative control; si, small interfering; WT, wild-type; NSCLC, non-small cell lung cancer.

(Fig. 3E). These findings suggest that LINC00210 knockdown may directly promote miR-328-5p expression.

LINC00210 knockdown inhibits A549 cell proliferation, and migratory and invasive ability by promoting miR-328-5p expression. miR-328-5p inhibitor significantly inhibited the expression of miR-328-5p (Fig. 4A). For further experiments, siLINC00210#1 was chosen because it produced the largest knockdown. A549 cells from the siLINC00210#1 group exhibited a significant decrease in cell proliferation, and in the migratory and invasive ability compared with that in A549 cells from the scramble NC group and the siLINC00210#1 + miR-328-5p inhibitor group (P<0.05; Fig. 4B-D). However, there was no significant difference in the proliferation, and migratory and invasive ability of A549 cells between the scramble NC group and the siLINC00210#1 + miR-328-5p inhibitor group (Fig. 4B-D). These results suggest that LINC00210 knockdown may inhibit A549 cell proliferation, and migratory and invasive ability by promoting miR-328-5p expression.

Discussion

The present study hypothesized that LINC00210 may be considered as a novel diagnostic tool and treatment target for patients with NSCLC. A high LINC00210 expression level was also found in patients with NSCLC and was associated with poor prognosis. In addition, the results from the present study revealed that LINC00210 was primarily expressed in the cytoplasm, and that knockdown of LINC00210 could inhibit A549 cell proliferation, and migratory and invasive abilities by promoting miR-328-5p expression. LINC00210 may therefore serve an oncogenic role in NSCLC cells by sponging miR-328-5p.

lncRNAs can directly interact with DNA, mRNA or protein to regulate chromatin modification or structure, transcription and translation, therefore regulating numerous physiological and pathological processes, including cell proliferation or differentiation, stem cell reprogramming, tumorigenesis or drug resistance (12). lncRNAs are divided into carcinogenic lncRNAs and tumor suppressor lncRNAs. As with other tumor types, such as colon cancer and liver cancer (6), the upregulation of carcinogenic lncRNAs in NSCLC can enhance cell proliferation as well as the migratory and invasive capacity, and reduce tumor cell apoptosis and tumor drug sensitivity, including MALAT1, HOTAIR and AFAP1 (13-17). Recently, numerous carcinogenic lncRNAs have been discovered in NSCLC, including HOTAIR, MALAT1, colon cancer associated transcript 2, H19 imprinted maternally expressed transcript and AFAP1 antisense RNA 1 (13-17), whereas SPRY4 intronic transcript 1, maternally expressed 3, GAS6 antisense RNA 1, growth arrest specific 5 and promoter of CDKN1A antisense DNA damage activated RNA PANDAR have been reported as tumor suppressor lncRNAs in NSCLC (18-22). LINC00210 has rarely been investigated in human diseases. A previous study investigated the effect of LINC00210 in liver cancer, and reported that LINC00210 is overexpressed in liver cancer, promoting liver tumor initiating cell self-renewal and propagation via the activation of the Wnt/β-catenin signaling pathway (23). Furthermore, another previous study reported that LINC00210 regulates nasopharyngeal carcinoma progression via the miR-328-5p/NOTCH3 axis (11). However, the role of LINC00210 in lung cancer remains unknown. The results from the present study demonstrate that LINC00210 was significantly upregulated in NSCLC tissues, and that high LINC00210 expression level was significantly associated with larger tumor size, advanced TNM stage and lymph node metastasis in patients with NSCLC. In addition, reducing LINC00210 expression using siRNA inhibited NSCLC cell proliferation, and migratory and invasive abilities. As the primary cause of cancer-associated mortality worldwide, the underlying mechanisms of NSCLC have not yet been

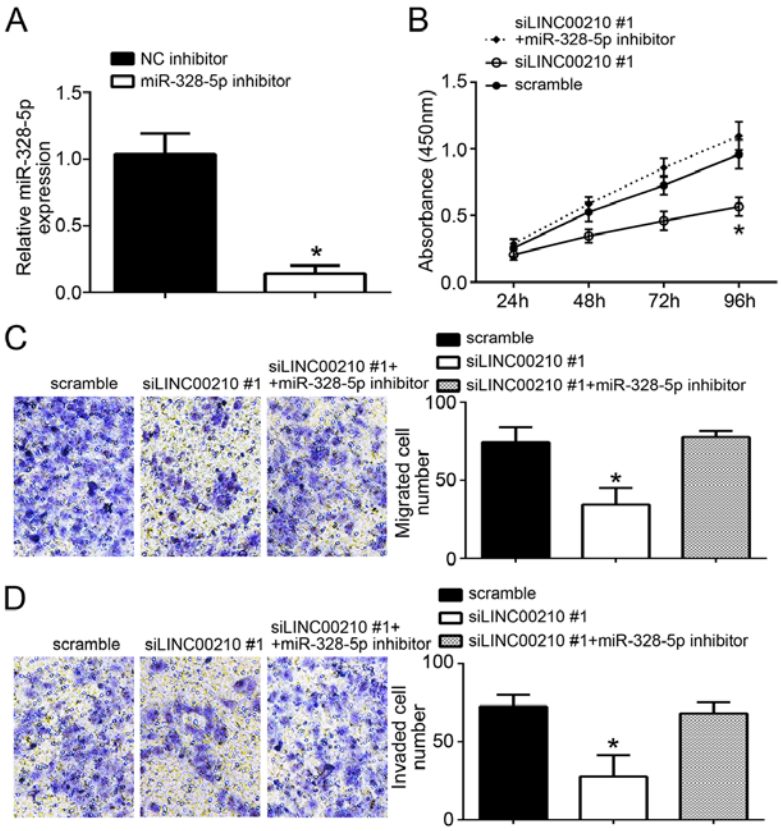

Figure 4. LINC00210 knockdown inhibits A549 cell proliferation, and migratory and invasive ability by promoting miR-328-5p expression. (A) Cell transfection with miR-328-5p inhibitor significantly reduced miR-328-5p expression level. Cell transfection with LINC00210 reduced the (B) proliferation, and (C) migratory and (D) invasive abilities compared with that in cells in the scramble NC group and the siLINC00210#1 + miR-328-5p inhibitor group. *P<0.05.

fully elucidated and the actual therapeutic options remain unsatisfactory. The discovery of lncRNAs has improved the clinical understanding of NSCLC tumorigenesis and progression, suggesting that these aforementioned lncRNAs may be considered as promising biomarkers for the early diagnosis and treatment of NSCLC. The results from this paper suggest that LINC00210 may be considered as a novel target for the diagnosis and treatment of patients with NSCLC.

miRNAs represent a class of endogenous, non-coding small RNAs found in eukaryotes. They participate in the regulation of various types of human tumor, such as liver cancer and ovarian cancer (24,25). In the present study, LINC00210 knockdown inhibited A549 cell proliferation, and migratory and invasive ability via the promotion of miR-328-5p expression. miR-328 is a type of microRNA that was been reported to be associated with the progression of numerous tumors. For example, Santasusagna *et al* (25) reported that miR-328 expression level is reduced in colon cancer tissues compared with that in adjacent normal tissues, and that miR-328 can affect colon cancer progression via solute carrier family 2 member 1/solute carrier family 2 member 1 targeting. In nasopharyngeal carcinoma, miR-328 is considered as a potential prognostic and therapeutic marker due to its inhibiting effect on the epithelial-mesenchymal transition of nasopharyngeal carcinoma cells (26). Liu *et al* (27) demonstrated that low miR-328 expression can predict a poor prognosis in patients with acute myeloid leukemia. Previous studies also reported that low miR-328 expression level is associated with poor survival in patients with high-grade glioma, and that miR-328 can impair glioma cell proliferation and invasive ability. miR-328 was therefore considered as a favorable prognostic marker in glioma (28,29). Furthermore, a previous study reported that miR-328 is reduced in NSCLC, and that miR-328 upregulation can increase NSCLC cell radiosensitvity via the DNA damage and repair signaling pathway (30). In the present study, miR-328-5p expression level was directly reduced following transfection with LINC00210, which may therefore act as a tumor suppressor in NSCLC.

In conclusion, the results from the present study suggest that LINC00210 may be considered as a novel target for the diagnosis and treatment of NSCLC. In addition, high LINC00210 expression predicted poor prognosis in patients with NSCLC. Following LINC00210 knockdown, NSCLC cell proliferation, and migratory and invasive abilities were reduced. Furthermore, the current study demonstrated that LINC00210 may serve oncogenic role in NSCLC by sponging miR-328-5p. At present, research on LINC00210 is still at an early stage, and further investigation on LINC00210 is required to determine the underlying mechanism of NSCLC and to assist with the development of novel treatment options. The present study did not analyze the effect of LINC00210 on cell cycle; however this will be performed in future studies.

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Availability of data and materials

All data generated or analyzed during this study are included in this published article.

Authors' contributions

ZL and LX contributed to the conception and design of the present study. NG analyzed and interpreted the results, and wrote the manuscript. KZ, BG and ZC performed the experiments and analyzed the data. All authors read and approved the final version of the manuscript.

Ethics approval and consent to participate

The present study was approved by The Ethics Committee of China-Japan Union Hospital of Jilin University. Written informed consent was provided from all recruited patients.

Patient consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

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