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ORIGINAL RESEARCH

LINC00665 Promotes the Progression of Multiple Myeloma by Adsorbing miR-214-3p and Positively Regulating the Expression of PSMD10 and ASF1B

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Background: Although assertion that long non-coding RNA (lncRNA) exerts crucial functions in the progression of multiple myeloma (MM) is well documented, few studies investigate function and underlying mechanism of long intergenic non-protein coding RNA 665 (LINC00665) in MM.

Patients and Methods: A total of 25 MM patient samples and 15 healthy volunteer samples were collected, and quantitative real-time polymerase chain reaction (qRT-PCR) was employed to detect the expressions of LINC00665. PSMD10 and ASF1B expressions were determined by qRT-PCR and Western blot assays. U266 cell and H929 cell were used in functional experiments. Besides, CCK-8 assay and flow cytometry analysis were utilized to determine cell proliferation and apoptosis. Bioinformatics analysis and dual-luciferase reporter assays were used to predict and verify the targeting relationships between LINC00665 and miR-214-3p, PSMD10 and miR-214-3p, as well as ASF1B and miR-214-3p. Moreover, the regulatory function of LINC00665 on the expression of PSMD10 and ASF1B was detected by Western blot.

Results: The expression of LINC00665 was up-regulated in MM samples and cell lines. In vitro functional assays indicated that LINC00665 enhanced MM cell proliferation and inhibited its apoptosis. PSMD10 and ASF1B were identified as target genes of miR-214-3p. Additionally, LINC00665 negatively regulated miR-214-3p expression through sponging miR-214-3p and positively regulated PSMD10 and ASF1B.

Conclusion: LINC00665 can promote the expression of PSMD10 and ASF1B by inhibiting the expression of miR-214-3p, thus facilitating the proliferation and inhibiting apoptosis of MM cells.

Keywords: MM, LINC00665, miR-214-3p, PSMD10, ASF1B

Introduction

Multiple myeloma (MM) is characterized as a complicated hematological malignancy featured with abnormal clonal proliferation of plasma cells in the bone marrow.[1] In clinical treatment, though emerging therapies like immunomodulatory factors (lenalidomide, pomalidomide) or proteasome inhibitors (bortezomib, carfilzomib) have remarkably elevated the response rate and overall survival rate,[2,3] MM still cannot be cured completely. Therefore, it is essential to further study the molecular mechanism of MM pathogenesis in order to provide new molecular targets for drug design.

Less than 2% of the DNA sequences in the human genome can encode proteins, while at least 90% of DNA sequences do not have the function of protein coding.[4]

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Non-coding RNAs (ncRNAs) are a class of RNAs that have no capacity for protein synthesis, including ribosomal RNA (rRNA), transit RNA (tRNA), microRNA (miRNA), circular RNA (circRNA) and long non-coding RNA (lncRNA), etc. LncRNAs are transcripts with over 200 nucleotides. An increasing number of researches demonstrated that lncRNAs are involved in multiple biological processes, and some of them blend in regulating the malignant biological behaviors of cancer cells.[5,6] Accumulating experiments display that lncRNAs are closely related to MM. For example, the expression of lncRNA CRNDE in MM cell lines and clinical samples is significantly increased, and its up-regulation was related to poor overall survival (OS);[7] lncRNA PRAL is down-regulated in MM, especially in the cases of chromosome del (17p); this change is related to MM's international staging system (ISS) staging and Durie-Salmon (D-S) staging.[8] LINC00665 is a member of LncRNAs, and little is known about its molecular mechanisms in MM progression.

miRNAs, a kind of short ncRNAs embracing 20 to 24 nucleotides in length, can inhibit translation or degrade mRNA.[9] Numerous experiments indicate that many miRNAs participate in the progression of MM. For example, research finds that miR-92a expression is remarkably down-regulated in MM patients compared to healthy people; in patients with MM who achieve complete remission, miR-92a expression returns to normal levels, but in MM patients with partially remission, miR-92a expression is still low, normal levels, suggesting that miR-92a expression is related to MM disease status and may be directly involved in the pathogenesis of MM.[10] miR-214-3p is reported to take part in regulating the occurrence and progression of various cancers like esophageal squamous cancer, hepatocellular carcinoma, gastric cancer and MM.[11–14]

In this study, we investigated the regulatory effects of LINC00665, miR-214-3p, proteasome 26S subunit, non-ATPase 10 (PSMD10), and anti-silencing function 1B histone chaperone (ASF1B) on the pathogenesis of MM, the emphasis of which lied in providing a new theoretical basis for the prevention and treatment of MM.

Patients and Methods

Specimen Collection

The collection of human samples was conducted in accordance with the Declaration of Helsinki. Bone marrow samples of 25 MM patients and 15 healthy volunteers from the First Affiliated Hospital of Zhengzhou University were available in this study. Equipped with written informed consents signed by all subjects, this work was endorsed by Ethics Committee of the First Affiliated Hospital of Zhengzhou University.

Cell Culture and Transfection

Chinese Academy of Sciences (Shanghai, China) was the provider of the five MM cell lines (MM.1S, U266, RPMI-8226, KM3 and H929) and normal plasma cells (nPCs). Cells were cultured in RPMI-1640 medium (Thermo Fisher Scientific, MA, USA) with 10% fetal bovine serum (FBS), 100 U/mL penicillin and 100 mg/mL streptomycin (Gibco, Carlsbad, CA, USA) at 37 °C in 5% CO_2 with 100% humidity. miR-214-3p mimics, miR-214-3p inhibitors, small hairpin RNA (shRNA) targeting LINC00665 (sh-LINC00665), LINC00665 overexpressing plasmid (pcDNA-LINC00665) and negative controls (miR-NC, inhibitor-NC, shRNA-NC and pcDNA-vector) were obtained from RiboBio (Guangzhou, China). U266, H929 and MM.1S cells were inoculated in a 6-well cell plate with a density of 1×10^5/mL, cultured at 37 °C in 5% CO_2 for 16 h and then transfected. Lipofectamine 2000 reagent (Invitrogen, Thermo Fisher Scientific, Inc.) was adopted to transfect miR-214-3p mimics, miR-214-3p inhibitors, LINC00665 shRNA or pcDNA-LINC00665 into the cells in accordance to the supplier's instructions.

Quantitative Real-Time Polymerase Chain Reaction (qRT-PCR)

TRIzol reagent (Invitrogen, Carlsbad, CA, USA) was adopted to extract total RNA from tissues or cells. To quantify the expression level of miR-214-3p, reverse transcription was performed with TaqMan Advanced miRNA cDNA Synthesis Kit (Thermo Fisher, Shanghai, China). And TaqMan® Universal PCR Master Mix (Applied Biosystems, San Francisco, CA, USA) was used to detect the relative expression of miR-214-3p levels normalized to the U6 endogenous control. To detect the expression of LINC00665, PSMD10 and ASF1B, total RNA was reversely transcribed to cDNA using PrimeScript™ RT Kit (TaKaRa, Dalian, China) consistent with the instructions. RT-PCR reactions were performed employing SYBR Premix Ex Taq (TaKaRa, Dalian, China) on ABI7500 real-time PCR system (Applied Biosystems, San Francisco, CA, USA). β-actin was treated as an endogenous control. The relative expression levels of the target genes were measured by $2^{-\Delta\Delta CT}$ method. The primers

were designed and provided by Beijing Genomics Institute (BGI, Shenzhen, China).

Western Blot

Cells were collected and rinsed 3 times with cold PBS, and RIPA lysate (Beyotime Biotechnology, Shanghai, China) with protease inhibitors (Roche, Basel, Switzerland) was loaded for total protein extraction. The protein concentration was detected by the BCA assay. After denaturation, the total protein was separated by 10% SDS-PAGE. The protein was transferred to PVDF membrane prior to the block with 5% skim milk for 2 h, and then incubated with primary antibodies (Anti-Gankyrin/PSMD10 antibody (1: 500, ab154676, Abcam, Cambridge, UK), Anti-ASF1B (1: 500, ab235358, Abcam, Cambridge, UK), and anti-GAPDH (1: 2000, ab181602, Abcam, Cambridge, UK)) at 4 °C for 12 h. Rinsed with TBST, the membrane was incubated employing horseradish peroxidase-labeled secondary antibody at room temperature for 1 h. After the membrane was washed again, chemiluminescence was performed using hypersensitive ECL (Amersham Pharmacia Biotech, Little Chalfont, UK). The membrane was then scanned on a Tanon-5200 imaging system (Shanghai, China).

Cell Viability Assay

Cell viability was measured by Cell Counting Kit-8 (CCK-8, Dojindo Molecular Technologies, Kumamoto, Japan) assay. Transfected cells were harvested and inoculated into 96-well plates at 1×10^3 cells/well. Cell proliferation was examined at 24, 48, 72 and 96 h after inoculation. 10 μL of CCK-8 reagent was dripped into each well, with which the cells were incubated in a 37 °C for 1 h and then the value of OD_{450nm} was measured on a microplate reader (Bio-Rad, CA, USA) in according to the manufacturer's instructions.

Apoptosis Determination

Apoptosis was determined by Annexin V-FITC/PI Apoptosis Detection Kit (Shanghai Yeasen BioTechnologies co., Ltd.). In brief, after transfection, cells were trypsinized with trypsin, centrifuged at 1000 r/min for 4 min, and washed with buffer 3 times, and the pellet was resuspended to reach a cell density of 3×10^6 cells/mL. FITC-Annexin V and PI solution were loaded before the cells were sequentially incubated in the dark for 15 min. Then apoptosis was detected by flow cytometry (BD Biosciences, New Jersey, USA).

Luciferase Reporter Gene Assay

Wild type LINC00665 sequence (LINC00665-wt) and mutant LINC00665 sequence (LINC00665-mut), wild type PSMD10 3′UTR sequence (PSMD10-wt) and mutant PSMD10 3′UTR sequence (PSMD10-mut) and wile type ASF1B 3′UTR sequence (ASF1B-wt) and mutant ASF1B 3′UTR sequence (ASF1B-mut) containing miR-214-3p binding site were constructed and integrated into pmirGLO dual-luciferase vector (Promega, Madison, WI, USA). HEK293 cells were seeded into 24-well plates and co-transfected with the reporter plasmid and miR-214-3p mimics or miRNA negative control using Lipofectamine 2000 (Invitrogen, Thermo Fisher Scientific, Inc.). After 48 h, the luciferase activity was measured on a dual-luciferase reporter assay system (Promega, Madison, WI, USA) in compliance with the manufacturer's protocols.

Statistical Analysis

Data were expressed as the mean ± standard deviation (x ± s) of three independent experiments. Statistical analysis was performed utilizing SPSS 20.0 software (Chicago, IL, USA). Differences between the two groups were analyzed using independent sample *t*-test or one-way ANOVA analysis. $P < 0.05$ was regarded statistically significant.

Results

LINC00665 Was Up-Regulated in MM Samples and Cell Lines

First of all, we employed qRT-PCR to determine LINC00665 expressions in MM patients (n = 25) and normal healthy controls (n = 15), the findings of which showed that compared with the normal group, LINC00665 expression in MM patients was significantly up-regulated (Figure 1A). Furthermore, we evaluated LINC00665 expressions in MM cell lines (MM.1S, U266, RPMI-8226, KM3, and H929) and normal plasma cells (nPCs). The results manifested that compared with nPCs cells, the expression of LINC00665 in each MM cell line was increased, and the changes were the most significant in U266 and H929 cells, while lnc00665 expression was at a lowest level in MM.1S (Figure 1B).

Knockdown of LINC00665 Inhibited MM Cell Proliferation and Promoted Its Apoptosis

To further explore the role of LINC00665 in the progression of MM, we transfected shRNA targeting LINC00665

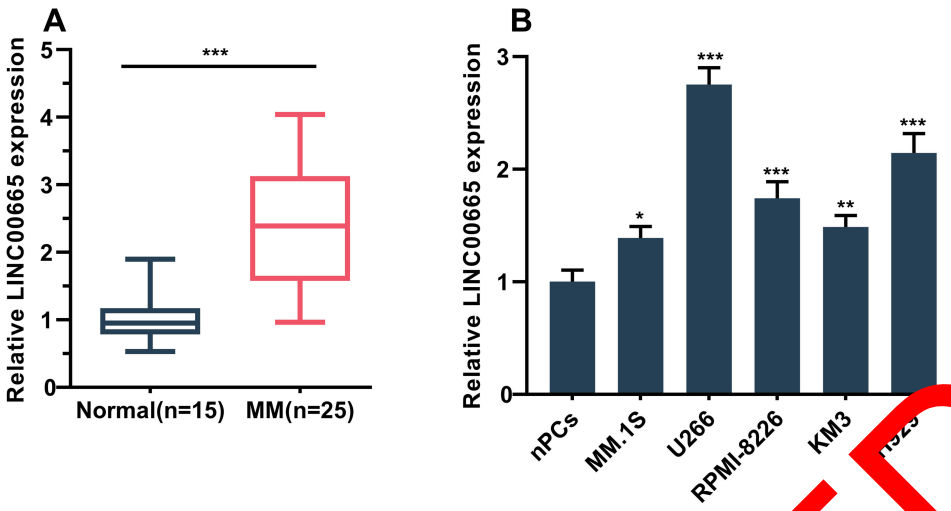

Figure 1 LINC00665 expression was up-regulated in MM samples and cell lines. (**A**) The relative expression level of LINC00665 in bone marrow samples of healthy control group (n = 15) and MM patients (n = 25) was detected by qRT-PCR. (**B**) Relative expressions of LINC00665 in normal plasma cells (nPCs) and MM cell lines (MM.1S, U266, RPMI-8226, KM3, and H929) were detected by qRT-PCR. *$P < 0.05$, **$P < 0.01$, and ***$P < 0.001$.

(sh-LINC00665) into U66 and H929 cell lines. Besides, we transfected pcDNA-LINC00665 into MM.1S cells. qRT-PCR results displayed that LINC00665 expression was dramatically reduced in MM cells transfected with sh-LINC00665 and significantly increased in MM.1S transfected with pcDNA-LINC00665 (Figure 2A). CCK-8 assay showed that knockdown of LINC00665 observably restrained the cell viability of U66 and H929 cells compared to the sh-NC group (Figure 2B and 2C). Additionally, LINC00665 overexpression markedly promoted cell viability of MM.1S (Figure 2D). Besides, flow cytometry analysis showed that knocking down LINC00665 observably facilitated MM cell apoptosis and LINC00665 overexpression significantly inhibited cell apoptosis of MM.1S (Figure 2E–G).

LINC00665 Directly Interacted with miR-214-3p

To investigate the regulatory role of LINC00665 on downstream molecules in MM, we used bioinformatics tool StarBase v2.0 to predict the potential lncRNA-miRNA interactions. We noticed that LINC00665 contained a conserved binding site for miR-214-3p, suggesting that miR-214-3p could be a potential target for LINC00665 (Figure 3A). To validate this prediction, we conducted a dual-luciferase reporter gene assay with HEK293T cells. As shown, miR-214-3p mimics remarkably reduced the relative luciferase activity of the wild-type LINC00665 luciferase reporter vector (LINC00665-wt); instead, it had no influence on the relative luciferase activity of empty vector or mutated LINC00665 luciferase reporter vector (LINC00665-mut) (Figure 3B). In addition, we examined miR-214-3p expressions in clinical samples by qRT-PCR analysis. The results proved that miR-214-3p was markedly down-regulated in MM patients (n = 25) compared to the normal group (n = 15) (Figure 3C). The interrelation between the expression of LINC00665 and miR-214-3p in MM tissue was further evaluated by Pearson's correlation analysis, and the results proved a significant negative correlation between the expressions of LINC00665 and miR-214-3p (Figure 3D). Next, we investigated whether changes in LINC00665 in U66, H929 and MM.1S cell lines affected the expression of miR-214-3p. The results showed that overexpression of LINC00665 significantly inhibited miR-214-3p expressions, while knockdown of LINC00665 markedly increased miR-214-3p expressions (Figure 3E). In summary, LINC00665 could directly bind with miR-214-3p and repress its expressions.

miR-214-3p Inhibitors Reversed the Effects of LINC00665 Knockdown on MM Cells

To further investigate whether LINC00665 exerted biological functions by targeting miR-214-3p, we co-transfected U66 and H929 cells with sh-LINC00665 and miR-214-3p inhibitors and co-transfected MM.1S cells with pcDNA-LINC00665 and miR-214-3p mimics. CCK-8 assay indicated that LINC00665 knockdown dramatically inhibited the proliferation of MM cells, and co-transfection with miR-214-3p inhibitors significantly reversed the above inhibitory effect (Figure 4A and B). Moreover, miR-214-3p inhibitors could also boost the proliferation of U266 and H929 cells

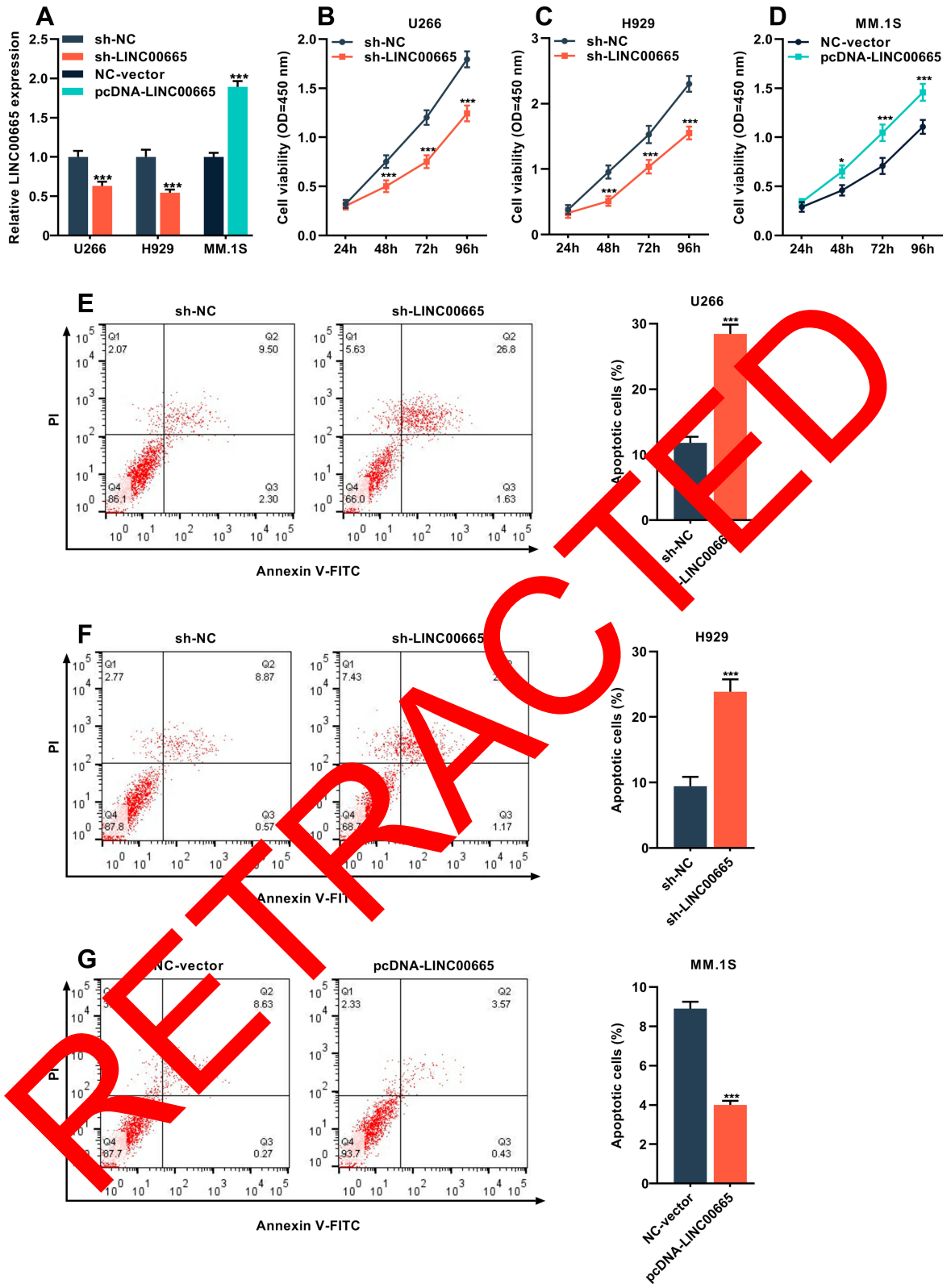

Figure 2 Knockdown of LINC00665 inhibited MM cell proliferation and promotes apoptosis. (**A**) LINC00665 knockdown cell models were successfully constructed in U266 and H929 cell lines. Besides, pcDNA-LINC00665 was successfully transfected into MM.1S. The transfection efficiency was validated by qRT-PCR. (**B–D**) Cell viability of U266, H929 and MM.1S cell lines was measured employing CCK-8 method. (**E–G**) MM cell apoptosis ratio was detected by flow cytometry. *$P < 0.05$, ***$P < 0.001$.

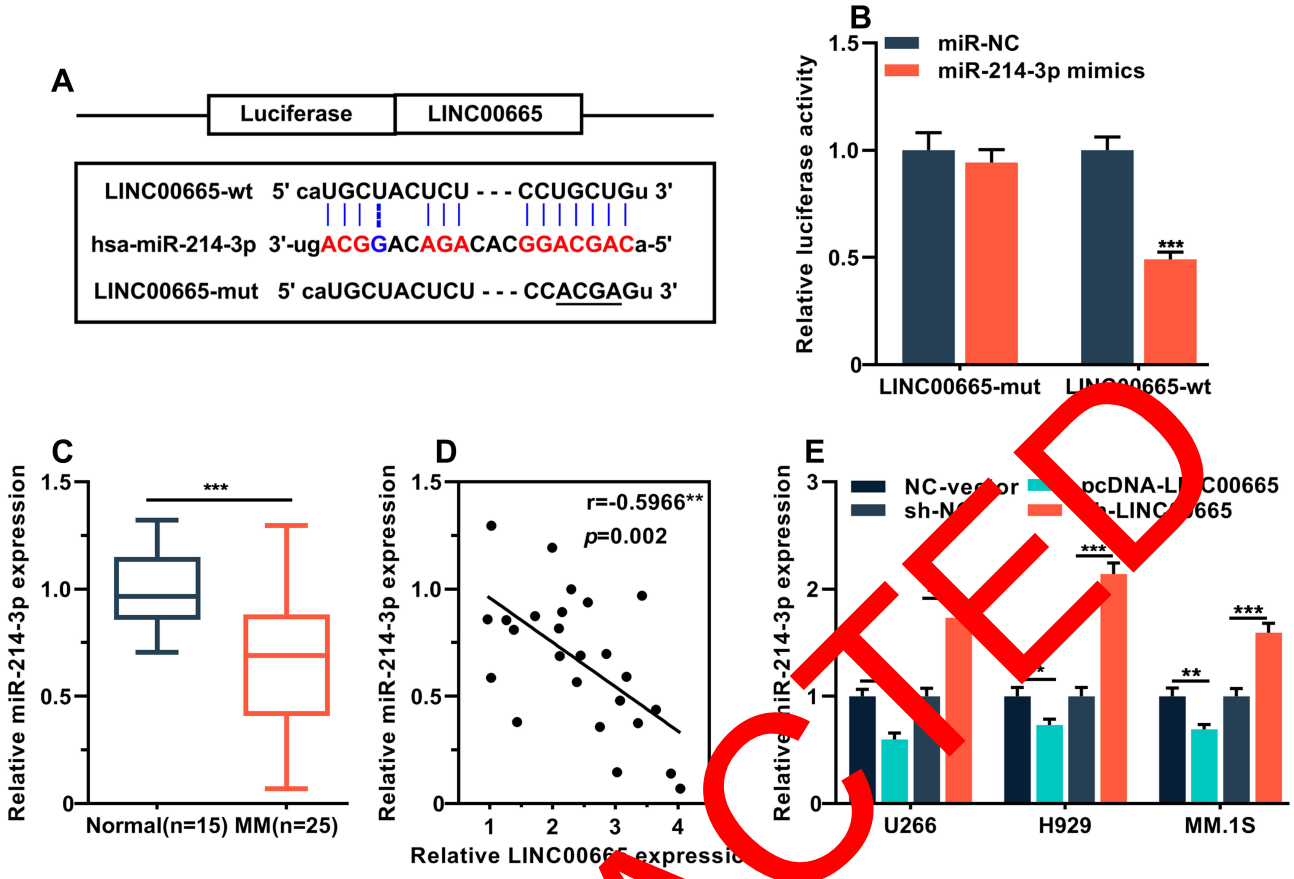

Figure 3 LINC00665 directly interacted with miR-214-3p. (**A**) StarBase database was used to predict the binding sites between LINC00665 and miR-214-3p. (**B**) The luciferase reporter assay confirmed the targeting relationship between LINC00665 and miR-214-3p. (**C**) qRT-PCR was used to detect the relative expression of miR-214-3p in bone marrow tissues of healthy control group (n = 15) and MM patients (n = 25). (**D**) Pearson's correlation analysis was used to evaluate the relationship between LINC00665 expression and miR-214-3p expressions in MM samples. (**E**) qRT-PCR was used to detect the effects of overexpression of LINC00665 or knockdown of LINC00665 on miR-214-3p expression in U266, H929 and MM.1S cell lines. **$P < 0.01$ and ***$P < 0.001$.

(Figure 4A and B). Oppositely, LINC00665 overexpression remarkably promoted the proliferation of MM.1S cells, and miR-214-3p mimics significantly reduced the cell viability of MM.1S cells transfected alone or co-transfected with pcDNA-LINC00665 (Figure 4C). Additionally, miR-214-3p inhibitors could observably reduce the apoptosis of U266 and H929 cells, and reverse the effects of LINC00665 knockdown on the apoptosis (Figure 4D and E), whereas miR-214-3p mimics significantly facilitated the rate of apoptosis and reversed the inhibitory effects of LINC00665 overexpression on the apoptosis of MM.1S cells (Figure 4F). These results further validated that LINC00665 could play a tumor-promoting role in MM cells through targeting regulation of miR-214-3p.

PSMD10 and ASF1B Were the Targets of miR-214-3p, and Both Were Regulated by LINC00665

To explore the molecular regulatory mechanism of miR-214-3p in MM, we used TargetScan to predict its downstream targets. It was found that both the 3′UTR of PSMD10 and the 3′UTR of ASF1B contained potential binding sites for miR-214-3p (Figure 5A). The luciferase reporter assay demonstrated that miR-214-3p mimics reduced the luciferase activity of PSMD10-wt and ASF1B-wt, but had no significant impact on the luciferase activity of PSMD10-mut and ASF1B-mut (Figure 5B). Besides, qRT-PCR proved that miR-214-3p mimics could inhibit the expression of PSMD10 and ASF1B in U266, H929 and MM.1S cell lines (Figure 5C); whereas, miR-214-3p inhibitors could promote the expression of PSMD10 and ASF1B in U266, H929 and MM.1S cell lines (Figure 5D). To determine whether LINC00665 functioned through PSMD10, we detected the expression of PSMD10 in MM samples by qRT-PCR. It was found that PSMD10 expression was increased in samples from patients with MM compared to normal healthy controls (Figure 6A). Pearson's correlation analysis indicated that PSMD10 expression was negatively related with miR-214-3p expression in MM samples and positively correlated with

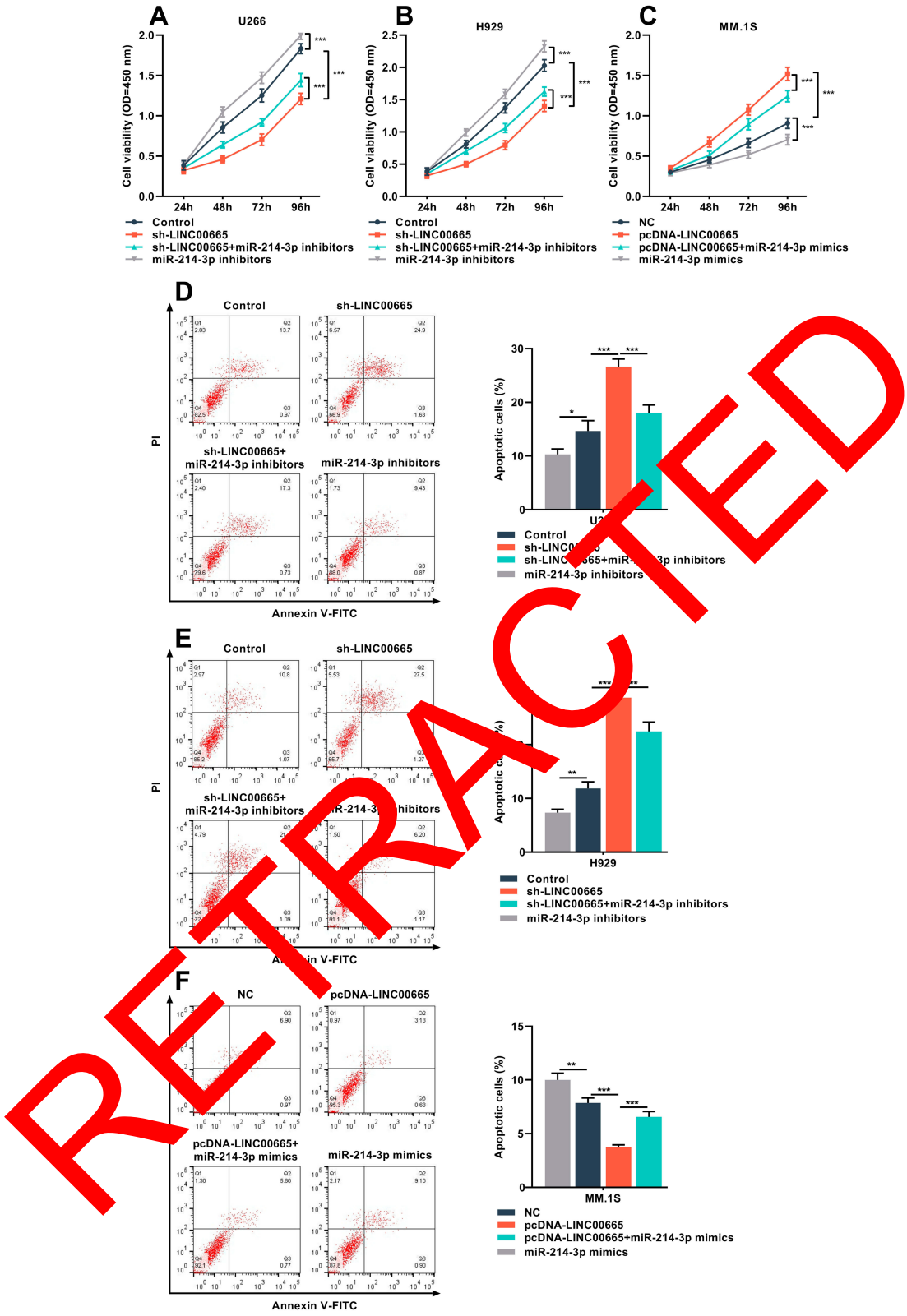

Figure 4 MiR-214-3p inhibitors reversed the effects of LINC00665 knockdown on MM cells. U266 and H929 cells were transfected with sh-LINC00665 or miR-214-3p inhibitors, or co-transfected with sh-LINC00665 and miR-214-3p inhibitors. MM.1S cells were transfected with pcDNA-LINC00665 or miR-214-3p mimics, or co-transfected with pcDNA-LINC00665 and miR-214-3p mimics. (**A–C**) CCK-8 was used to detect the cell viability of U266, H929 and MM.1S cell lines. (**D–F**) Flow cytometry was used to analyze the apoptosis of U266, H929 and MM.1S cell lines. *$P < 0.05$, **$P < 0.01$, and ***$P < 0.001$.

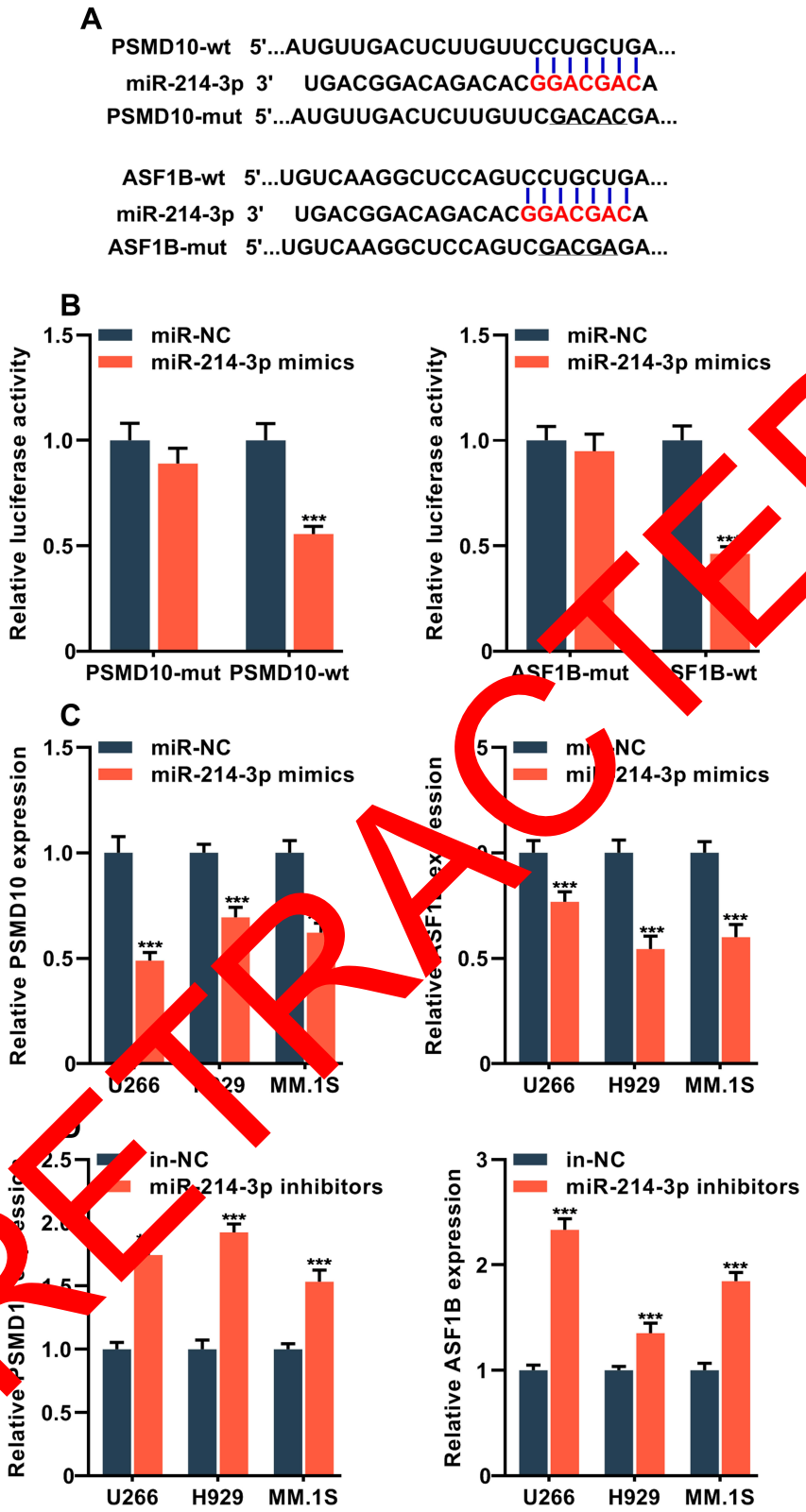

Figure 5 PSMD10 and ASF1B were targets of miR-214-3p, and both were regulated by LINC00665. (**A**) The binding sites between PSMD10 and miR-214-3p, and between ASF1B and miR-214-3p were predicted by TargetScan database. (**B**) The luciferase reporter assay confirmed the targeting relationships between PSMD10 and miR-214-3p, and ASF1B and miR-214-3p. (**C**) qRT-PCR was used to detect the expression of PSMD10 and ASF1B in U266, H929 and MM.1S cells transfected with miR-214-3p mimics. (**D**) qRT-PCR was used to detect the expression of PSMD10 and ASF1B in U266, H929 and MM.1S cells transfected with miR-214-3p inhibitors. ***$P < 0.001$.

LINC00665 expression (Figure 6B and C). Similarly, ASF1B was also found to be significantly up-regulated in MM patient samples, negatively correlated with miR-214-3p expression, and positively correlated with LINC00665 expression (Figure 6D–F). In addition, we detected the expression of PSMD10 and ASF1B at mRNA and protein levels after transfection of sh-LINC00665 and miR-214-3p inhibitors into U266 and H929 cells and of pcDNA-LINC00665 and miR-214-3p mimics into MM.1S cells. The results proved that knocking down LINC00665 significantly inhibited the expressions of PSMD10 and ASF1B but overexpression of LINC00665 significantly promoted these expressions; Moreover, co-transfection of miR-214-3p inhibitors or miR-214-3p mimics significantly reversed the these effects (Figure 6G–J). Collectively, LINC00665 could regulate the expression of PSMD10 and ASF1B by targeting miR-214-3p.

Discussion

Accounts for 10% of hematological malignancies,[15] MM is tough to be cured due to the lack of effective drug, which contributes to the necessity to further study the molecular mechanism of MM to explore potential therapy targets. An enormous number of researches suggest that lncRNA is closely related to MM pathogenesis and development. For example, lncRNA FEZF1-AS1 is significantly increased both in MM clinical samples and cell lines, and its high expression is responsible for the proliferation of MM cells;[16] lncRNA PVT1 is up-regulated in bone marrow samples from MM patients and MM cell lines, and it enhances the proliferation of MM cells and inhibits apoptosis.[17] LINC00665 is proved to be up-regulated in various tumors such as hepatocellular carcinoma and lung adenocarcinoma, which is involved in the development of these diseases as an oncogenic lncRNA.[18] In this work, for the first time, we found that the expression of LINC00665 was significantly increased in bone marrow tissues of patients with MM. Functional experiments indicated that knocking down LINC00665 could significantly repress viability and promote apoptosis of MM cells, suggesting that LINC00665 might be involved in MM progression as a tumor-promoting factor.

In recent years, dysregulated expression of miRNA has been reported to participate in the pathogenesis and progression of many cancers, including MM.[20,21] For example, compared to normal plasma cells, expressions of miR-32, miR-21, miR-17-92 cluster, miR-106b-25 and miR-181a/b are up-regulated in MM cells, and these miRNAs are associated with the dysregulation of MM-related genes such as SOCS-1 and PCAF.[22] It is reported that miR-214-3p is lowly expressed in MM, and overexpression of miR-214-3p reduces the proliferation of MM cells through negative regulation of PSMD10 and ASF1B, suggesting it is a tumor suppressor in MM.[23] However, the mechanism of miR-214-3p dysregulation in MM is obscure. Emerging researches suggest that lncRNA participates in tumor progression by functioning as competitive endogenous RNA (ceRNA), which can block the binding between miRNA and their target mRNAs. For example, lncRNA OIP5-AS1 is down-regulated in MM cells, and deficit of OIP5-AS1 will mediate the accumulation of miR-410, and in turn reduce the expression level of KLF10 to promote MM progression;[24] lncRNA MALAT1 acts as an oncogenic lnRNA in MM through sponging miR-509-5p to up-regulate FOXP1 expression;[25] ceRNA CCAT1, as a ceRNA, promotes MM progression by repressing miR-181a-5p and up-regulating HOXA9.[26] In this work, it was demonstrated that LINC00665 was negatively correlated with miR-214-3p expressions in MM samples, and LINC00665 could negatively regulate the expression of miR-214-3p, but positively modulate the expressions of PSMD10 and ASF1B. PSMD10 (p28 gankyrin) negatively regulates p53 and acts as an oncoprotein.[27] It is reported that knockdown of PSMD10 induces the apoptosis of RPMI-8226 MM cells.[28] ASF1B is a histone H3-H4 chaperone, participating in DNA replication, DNA damage repair, and transcriptional regulation; overexpression of ASF1B leads to distinct transcriptional signatures consistent with increased cellular proliferation and reduced cellular apoptosis.[29,30] As is mentioned above, miR-214-3p suppressed the expressions of PSMD10 and ASF1B to block the progression of MM.[23] In the present work, we proved that LINC00665 could probably function as a ceRNA to up-regulate oncogenes PSMD10 and ASF1B via repressing miR-214-3p in MM.

There are still several limitations in this work. Firstly, animal studies are necessary to further verify our conclusion in the future; moreover, other mechanism by which LINC00665 promotes MM progression remains to be clarified, and other potential downstream miRNAs remain to be screened and validated; additionally, whether other malignant phenotypes of MM cells, such as invasion and chemoresistance regulated by LINC00665, are also worth investigating; last but not least, to evaluate the potential of LINC00665 as a biomarker, more patients from multiple centers shall be enrolled in, and the survival analysis of MM patients based on the expression of LINC00665 is desirable.

In summary, LINC00665 expression is remarkably up-regulated in MM samples and cell lines. In addition, it is

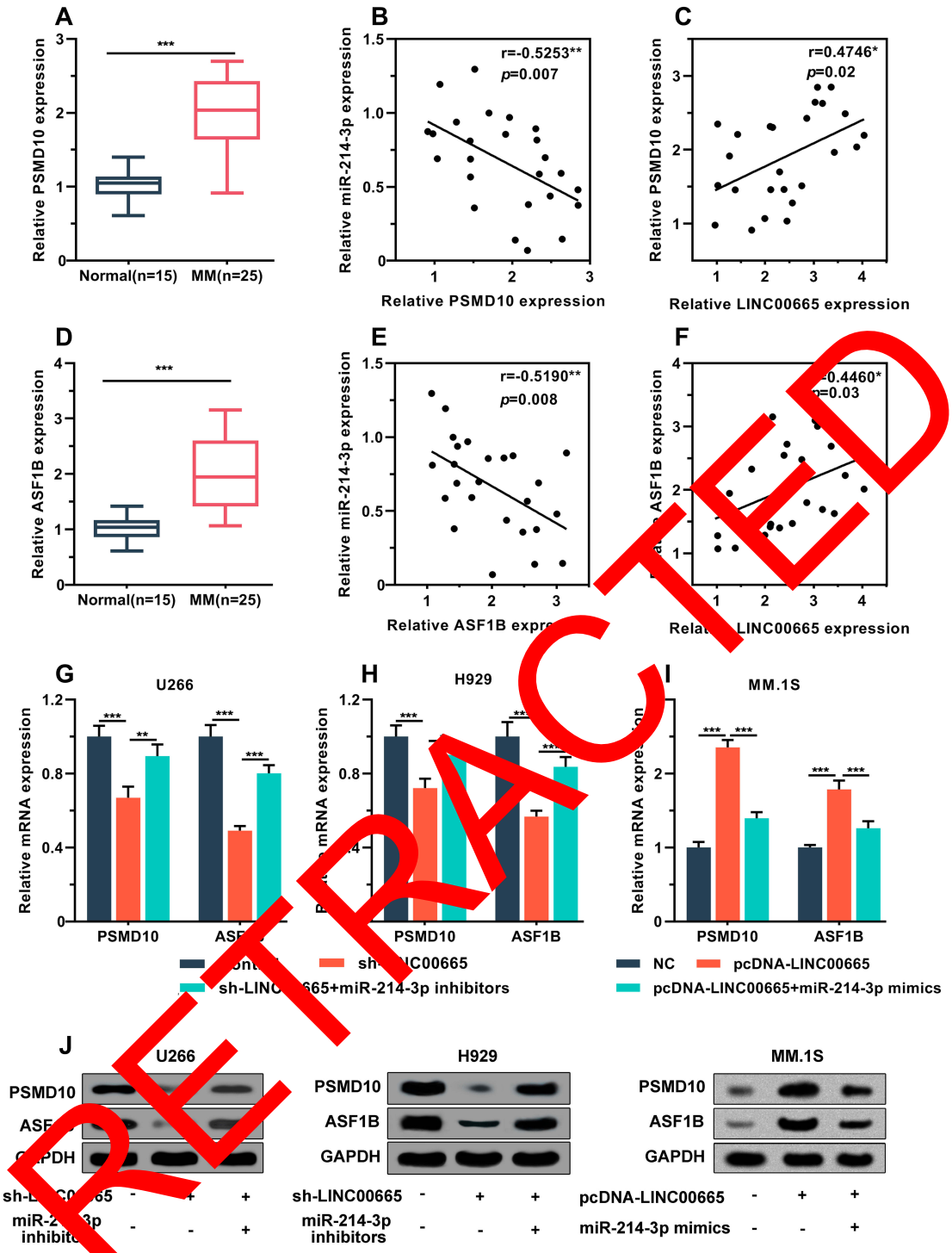

Figure 6 LINC00665 regulated PSMD10 and ASF1B expressions by inhibiting miR-214-3p. (**A**) qRT-PCR was used to detect the relative expression of PSMD10 in bone marrow tissues of healthy control group (n = 15) and MM patients (n = 25). (**B**) Pearson's correlation analysis was used to determine the relationship between PSMD10 and miR-214-3p expressions in MM tissues. (**C**) Pearson's correlation analysis was used to detect the relationship between LINC00665 and PSMD10 expressions in MM tissues. (**D**) qRT-PCR was used to detect the relative expression of ASF1B in bone marrow tissues of healthy control group (n = 15) and MM patients (n = 25). (**E**) Pearson's correlation analysis was used to detect the relationship between ASF1B and miR-214-3p expressions in MM tissue. (**F**) Pearson's correlation analysis was used to detect the relationship between LINC00665 and ASF1B expressions in MM tissues. (**G** and **H**) qRT-PCR was used to detect the expressions of PSMD10 and ASF1B in U266 and H929 cells transfected with sh-LINC00665, or co-transfected with sh-LINC00665 and miR-214-3p inhibitors. (**I**) qRT-PCR was used to detect the expression of PSMD10 and ASF1B in MM.1S cells transfected with pcDNA-LINC00665, or co-transfected with pcDNA-LINC00665 and miR-214-3p mimics. (**J**) Western blot was used to detect the expression of PSMD10 and ASF1B in U266, H929 and MM.1S cells $^{*}P < 0.05$, $^{**}P < 0.01$, and $^{***}P < 0.001$.

demonstrated that LINC00665 can promote the expression of PSMD10 and ASF1B through targetedly binding with miR-214-3p, thereby facilitating cancer cell proliferation and inhibiting apoptosis. The above results indicate that LINC00665 is promising to be an effective diagnostic marker and a therapeutic target for MM.

Data Sharing Statement

The data used to confirm the findings of this study are available from the corresponding author upon request.

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Disclosure

The authors report no conflicts of interest in this work.

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