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Research article

LINC00115 aggravates thyroid cancer progression by targeting *miR-489-3p,* which downregulates *EVA1A* to regulate the Hippo signaling pathway

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ABSTRACT

LINC00115 has been documented to regulate many different cancers; however, its function in thyroid cancer (THCA) remains unexplored. Therefore, we examined the effects of *LINC00115* on THCA and the associated molecular mechanisms. In THCA cell lines and tumor samples, the expression levels of *LINC00115*, *miR-489-3p*, and *EVA1A* were analyzed by qRT-PCR along with respective controls. Cell viability, migration, and apoptosis were analyzed by employing CCK-8, transwell, and western blotting assays, respectively. Xenograft experiments were done to assess *in vivo* tumor growth. The interaction among *LINC00115*, miR-489-3p, and *EVA1A* was tested using RNA-binding protein immunoprecipitation and luciferase assays. Key proteins of the Hippo signaling pathway were ascertained by western blotting. The outcomes elucidated that *LINC00115* was overexpressed in THCA cell lines and tumor tissues. *LIN00115* knockdown reduced *in vitro* proliferation and migration but facilitated apoptosis in THCA cells and inhibited *in vivo* tumor growth. The target of *LINC00115* was *miR-489-3p*, which binds to *EVA1A* in THCA. Functional assays revealed that *miR-489-3p* inhibition boosted THCA cell proliferation and migration, but hindered apoptosis. However, *EVA1A* knockdown resulted in the opposite effects via the Hippo signaling pathway. Additionally, *miR-489-3p* inhibition partially negated the effects of *LINC00115* knockdown in THCA cells, and EVA1A knockdown remarkably impeded the effects of *miR-489-3p* inhibition in THCA cells. Thus, *LINC00115* knockdown suppressed THCA carcinogenesis via targeting *miR-489-3p*, which regulates *EVA1A* expression and affects the Hippo signaling pathway.

1. Introduction

Thyroid cancer (THCA) is recognized as an extremely widespread cancer of the endocrine system, contributing to 3.4 % of all annual cases of cancer globally [1]. With an annual global number of 550,000 cases, its morbidity has persistently increased over the last three decades [2]. In addition to the favorable prognosis of patients diagnosed with THCA at an early stage, when treated with

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thyroidectomy and radioactive iodine, the likelihood of its occurrence increases with the existence of metastasis, which occurs in 10 % of patients with THCA [3–5]. Therefore, pursuing necessary investigations to illustrate the molecular mechanisms linked with the occurrence and advancement of THCA is of great importance.

In many cancers, long non-coding RNAs (lncRNAs) of >200 nt are pivotal regulators [6]. This is supported by evidence that an increase in the expression levels of lncRNAs, including *RUNDC3A-AS1* [7], *CCDC26* [8], *XIST* [9], and *TUG1* [10] leads to an unfavorable THCA prognosis. *LINC00115*, an lncRNA, was originally identified as a possible biomarker for bladder [11] and lung cancers [12]. High expression levels of *LINC00115* contribute to progression of glioma, and its higher levels indicate a poor prognosis [13]. Significant overexpression of *LINC00115* has been observed in colorectal cancer (CRC) and predicts poor patient outcomes [14]. Nonetheless, the significance of *LINC00115* in THCA has not been studied.

MicroRNAs (miRNAs) are short (approximately 22 nt long) endogenous single-stranded RNA molecules that bind to the 3′ untranslated regions (UTRs) of targeted genes to modulate their expression [15]. These miRNAs have significant roles in the pathogenesis of various diseases, particularly cancer [16]. Previous reports suggest that lncRNAs mediate their effects by functioning as miRNA sponges that inhibit miRNA activity in various cancers [17]. For instance, *LINC00115* is instrumental in the progression of CRC by sponging *miR-489-3p* [14]. However, further research is needed to determine whether the *LINC00115–miR-489-3p* axis regulates THCA progression.

In this work, the effects of *LINC00115* on THCA and its potential downstream targets during THCA development were investigated by using cell function experiments. Our results provide additional information regarding the molecular mechanisms underlying the progression of THCA.

2. Materials and methods

2.1. Tissue sample collection

Thirty-four THCA patients who undertook surgical treatments in our hospital (between February 2019 and September 2021) provided written informed consent. THCA tissues and corresponding adjacent normal tissues were obtained and cryopreserved (liquid nitrogen) for future usage. The study was approved by the Ethics Committee of our hospital.

2.2. Cell culture

The normal human thyroid follicular epithelial cell line (Nthy-ori-3-1: Cat# 90011609) was acquired from Millipore (Burlington, MA, USA), and the three THCA cell lines (TPC-1: Cat# BNCC337912; BCPAP: Cat# BNCC358025; SW579: Cat# BNCC100182) were bought from BeNa Culture Collection (Kunshan, China). The culturing of BCPAP and Nthy-ori-3-1 cells was done in RPMI 1640 medium, TPC-1 cells in DMEM-H, and SW579 cells in L-15 medium, all of which contained 10 % FBS. All the media were purchased from Gibco (Carlsbad, CA, USA). For culturing, these cells were maintained in a humid environment with 5 % CO_2 concentration and 37 °C temperature.

2.3. Quantitative reverse transcription-polymerase chain reaction (qRT-PCR)

The extraction of total RNA from THCA cells and tissue samples was done by TRIzol reagent (Invitrogen, Carlsbad, CA, USA) as per the instructions given by the Invitrogen. Reverse transcription was carried out on 5 μg of RNA to generate cDNA using a TaqMan one-step reverse transcription kit (Applied Biosystems, Waltham, MA, USA). Then, qRT-PCR was done utilizing the TB Green Fast qPCR Mix (Takara Biotechnology, Kusatsu, Japan). Data were assessed using the $2^{-\Delta\Delta Ct}$ method following normalization to *GAPDH* or *U6* mRNA levels. The primer sequences are presented in Table 1.

2.4. Cell transfection

siRNAs targeting *LINC00115* and *EVA1A* and non-targeting siRNAs (negative control [NC]), together with oligonucleotides,

Table 1
Sequence of the primers used in this study.

Gene	Primer type	Sequence
LINC00115	Forward	5′-AGCGGTGACTGTTGGGTG-3′
	Reverse	5′-CATCCACAGCGAGGCAAT-3′
miR-489-3p	Forward	5′-CTGACATGTGAGAGGCACTCAA-3
	Reverse	5′-GCTGCCGTATATGTGATGTCACT-3′
EVA1A	Forward	5′-CGTGGAGATGGCTTTGCTCA-3′
	Reverse	5′-AGCTGCTCGCTCAGGATTTT-3′
GADPH	Forward	5′-GGTCGGAGTCAACGGATTTG-3′
	Reverse	5′-ATGAGCCCCAGCCTTCTCCAT-3′
U6	Forward	5′-CTCGCTTCGGCAGCACA-3′
	Reverse	5′-CGCTTCACGAATTTGCGT-3′

including inhibitor/mimic-NC, *miR-489-3p* inhibitor/mimic oligonucleotides, were manufactured by GenePharma (Suzhou, China). Transfection of 50 nM plasmids into THCA cells was accomplished by Lipofectamine 2000 (Thermo Fisher Scientific, Waltham, MA, USA), only after the cells attained 80 % confluence in 96-well plates. The efficiency of transfections was confirmed 48 h post-transfection by qRT-PCR before the following experiments.

2.5. Cell counting Kit-8 (CCK-8) assay

The CCK-8 kit (Beyotime Biotechnology, Nantong, China) was utilized to assess the cell viability. Briefly, 5×10^3 cells/well were seeded into 96-well plates and cultured for 0, 24, 48, or 72 h, after which, 10 μL of CCK-8 reagent was added. Following an incubation at 37 °C for 1.5 h, absorbance (@450 nm) was recorded.

2.6. Western blotting

Total protein from THCA cells was extracted with the help of RIPA lysis buffer (Thermo Fisher Scientific) having protease inhibitor cocktail (Roche, Basel, Switzerland). The quantity of individual protein samples was evaluated availing a BCA test kit (Thermo Fisher Scientific). Through a 12 % SDS-PAGE, protein samples (20 μg) were separated at 150 V for 1.5 h and then transferred onto PVFD membranes (Millipore). Following blocking these with 5 % non-fat milk, the membranes were treated with primary antibodies at 4 °C overnight. The following antibodies were used in this study at a dilution of 1:1000 and were acquired from abcam, Cambridge, UK - Bcl-2 (ab32124), Bax (ab32503), *EVA1A* (ab216043), and GAPDH (ab9485). The membranes were then treated with hrp-conjugated goat anti-rabbit IgG secondary antibody (abcam, 1:10,000). Finally, each membrane was visualized using a chemical immunogenicity system (GE Healthcare, Chicago, IL, USA), and ImageJ software (NIH, Bethesda, MD, USA) was employed for quantitation of the intensities of the protein bands.

2.7. Cell migration assay

Cell migration was investigated by employing a transwell assay. Transfected cells (2×10^4 cells/well) were seeded into the upper chamber of transwell plates in medium without FBS, whereas the lower chamber was loaded with medium comprising 20 % FBS. After 24 h incubation, the cells migrating to the lower chamber were fixed with 4 % PFA and stained with 0.5 % crystal violet for 15 and 10 min, respectively, at 37 °C. These cells were photographed under a microscope (250 × magnification) for counting the number of migrated cells after washing with water.

2.8. Xenograft assay

Six BALB/c nude mice (male, approximately 4-week-old) provided by Wuhan Myhalic Biotechnological Co., Ltd. (Wuhan, China) were used to perform xenograft assays. The *LINC00115*-knockdown lentiviral vector (sh-lnc) and its corresponding NC (sh-NC), constructed by GenePharma, were introduced into TPC-1 cells to stably silence *LINC00115*. Next, transfected TPC-1 cells (5×10^6) were injected into the nude mice subcutaneously, and the tumor volume was determined by recording the length and width of the resulting tumors weekly for five weeks, following which these mice were euthanized, and the tumors were extracted to obtain the weight. This study was approved by the Ethics Committee of our hospital.

2.9. Luciferase assay

The ENCORI database was used to predict the binding sites of *miR-489-3p* in the 3′ UTR of *LINC00115* and *EVA1A*. Following amplification of the fragments with or without binding sites by PCR, they were cloned into the pmirGLO vector (Promega, Madison, WI, USA) and are designated as *LINC00115/EVA1A*-WT or *LINC00115/EVA1A*-MUT, respectively. The transfection was done with the constructed vectors along with *miR-489-3p* mimic or mimic-NC in the THCA cells. Finally, relative luciferase activity was calculated by a dual-luciferase reporter system (Promega).

2.10. RNA-binding protein immunoprecipitation (RIP)

RIP was conducted to determine if there was an endogenous interaction among *LINC00115* and *miR-489-3p* by means of a Magna RIP Kit (Millipore). Briefly, 2×10^7 cells were lysed with RIPA lysis buffer (Beyotime Biotechnology), and then incubation of these lysates was done with magnetic beads conjugated with an Ago2 antibody (Millipore) or IgG at 4 °C for 24 h. Finally, the immunoprecipitated RNA was extracted and evaluated via qRT-PCR.

2.11. Statistics analysis

Analyses were performed in GraphPad Prism 8 software (GraphPad, San Diego, CA, USA). The data are presented as the mean ± standard deviation of three replicate experiments. The statistical differences were checked using the student's t-test for two groups or analysis of variance followed by Dunnett's multiple comparisons test for more than two groups. The Pearson's correlation analysis was employed to verify any correlation between the expressions of the two genes in THCA tissues. A *P*-value <0.05 was considered

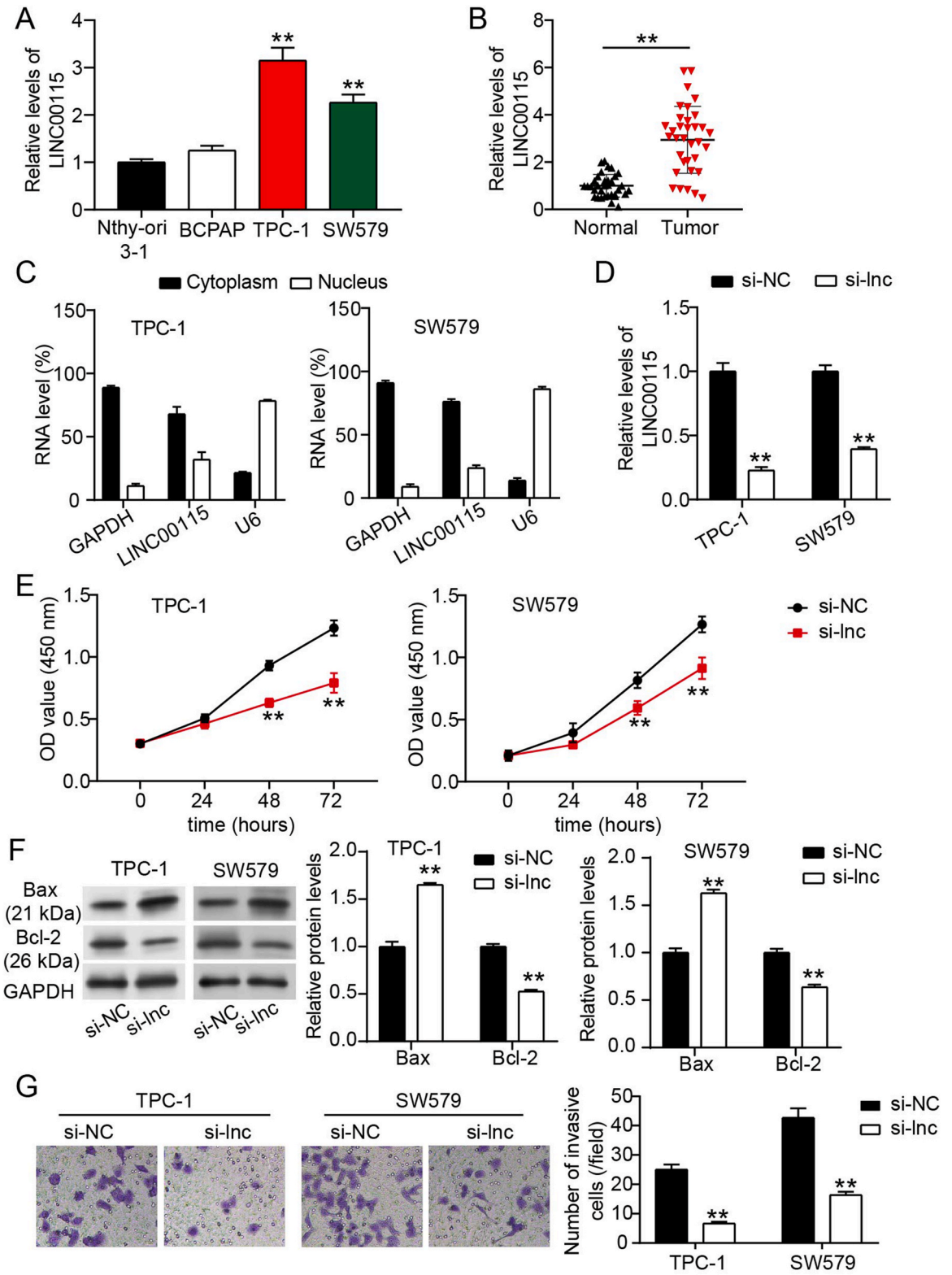

Fig. 1. Effects of LINC00115 on THCA cells *in vitro*. A. The relative level of LINC00115 in Nthy-ori-3-1 and BCPAP, TPC-1 and SW579. Nthy-ori-3-1, normal human thyroid cell line. Other cell lines, THCA cell lines. **P < 0.001 vs Nthy-ori 3-1. B. The relative level of LINC00115 in THCA tumor and corresponding paracarcinoma normal tissues (n = 34). **P < 0.001. C. The location of LINC00115 in TPC-1 and SW579 cells. D. The relative level of LINC00115 in TPC-1 and SW579 cells transfected with si-NC or LINC00115 siRNA (si-lnc). E. The proliferation of TPC-1 and SW579 cells

transfected with si-lnc or si-NC. F. Bax and Bcl-2 protein expression in TPC-1 and SW579 cells transfected with si-lnc or si-NC. G. The representative images and quantitative analysis for Transwell assay of transfected cells (TPC-1 and SW579) with si-NC or si-lnc. (D–G) *P < 0.05, and **P < 0.001 vs si-NC.

statistically significant.

3. Results

3.1. LINC00115 was upregulated in THCA and its knockdown lowered THCA progression in vitro

The expression of *LINC00115* was assessed in Nthy-ori-3-1 and THCA (TPC-1 and SW579) cell lines and it was observed that its mRNA levels were markedly elevated in the THCA cells in contrast to Nthy-ori-3-1 cells (Fig. 1A). Similarly, *LINC00115* mRNA levels were consistently upregulated in THCA tumor samples as compared to control samples (Fig. 1B). The localization of *LINC0015* was mainly seen in the cytoplasm of the THCA cells (Fig. 1C). We knocked down *LINC00115* expression in THCA cells using siRNA (Fig. 1D). The cell proliferation notably lowered after *LIN00115* knockdown, as disclosed by the outcomes of CCK-8 assay (Fig. 1E). *LINC00115* knockdown increased Bax expression levels, whereas decreased Bcl-2 expression levels in both THCA cell lines, indicating an upsurge in apoptosis after *LINC00115* knockdown (Fig. 1F). In addition, THCA cells transfected with siRNA- *LINC00115* displayed fewer migrated cells in contrast to those with si-NC (Fig. 1G). These results elucidate that *LINC00115* knockdown significantly inhibits THCA cell proliferation and migration but boosts apoptosis.

3.2. LINC00115 knockdown attenuated THCA tumor growth in vivo

After developing a xenograft mouse model, we found that the tumors were smaller in the *LINC00115*-knockdown group than the sh-NC control group (Fig. 2A). Additionally, *LINC00115* knockdown reduced tumor volume and weight (Fig. 2B and C). Taken together, *LINC00115* knockdown lowers the *in vivo* tumor growth of THCA.

LINC00115 targeted *miR-489-3p* and negatively regulated its expression in THCA.

We identified an association among the expressions of *LINC00115* and *miR-489-3p* in THCA. The ENCORI database was used to envisage the binding sites between these two, which are presented in Fig. 3A. RIP and luciferase assays were done to corroborate their interactions. The results illustrated enrichment of *LINC00115* and *miR-489-3p* in anti-Ago2 antibody precipitates, and the *miR-489-3p* mimic induced a decrease in luciferase activity in the *LINC*00115-WT group (Fig. 3B and C). Moreover, *miR-489-3p* expression was distinctly reduced in THCA tumor samples and THCA cells (Fig. 3D and E). There was an inverse correlation between *LINC00115* and *miR-489-3p* expression levels in THCA samples (Fig. 3F). These data indicate that *LINC00115* targets *miR-489-3p* in THCA.

Inhibition of *miR-489-3p* expression aggravated the progression of THCA and reversed the effect of *LINC00115 silencing.*

To evaluate the impact of *miR-489-3p* on THCA, si-*LINC00115* and *miR-489-3p* inhibitor were transfected into THCA cells (TPC-1 and SW579), resulting in an upregulation and downregulation of *miR-489-3p*, respectively (Fig. 4A). Interestingly, the *miR-489-3p* suppression culminated in an increase in cell proliferation, and the decrease in cell proliferation induced by si-*LINC00115* was blocked by *miR-489-3p* inhibitor (Fig. 4B). Additionally, the *miR-489-3p* inhibitor markedly reduced apoptosis in THCA cells and the enhanced apoptosis by si-*LINC00115* was also significantly blocked by the *miR-489-3p* inhibitor (Fig. 4C). Moreover, the presence of the *miR-489-3p* inhibitor promoted cell migration, and increased the count of migrated cells that were lowered by si-*LINC00115* (Fig. 4D). These findings imply that *miR-489-3p* inhibition may aggravate THCA progression and reverse the suppressive effects of si-*LINC00115* on THCA progression.

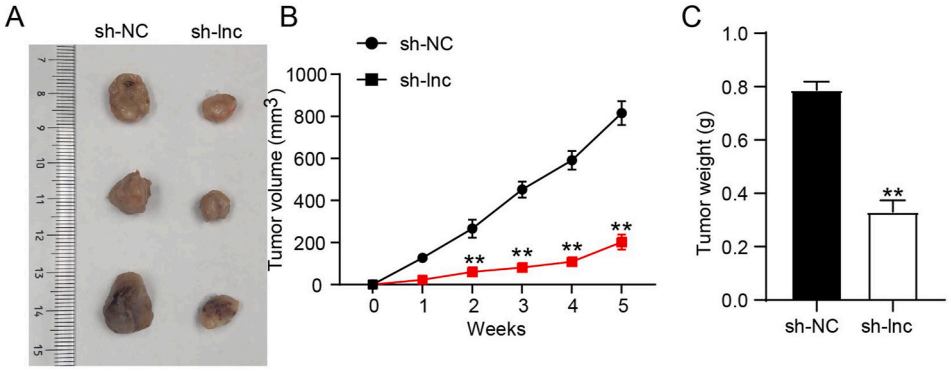

Fig. 2. Effects of LINC00115 knockdown on THCA cells *in vivo*. A. The tumor images from nude mice injected with sh-NC or sh-LINC00115 (sh-lnc). B–C. The tumor volume (B) and weight (C) from nude mice injected with sh-NC or sh-LINC00115 (sh-lnc). **P < 0.001 vs sh-NC.

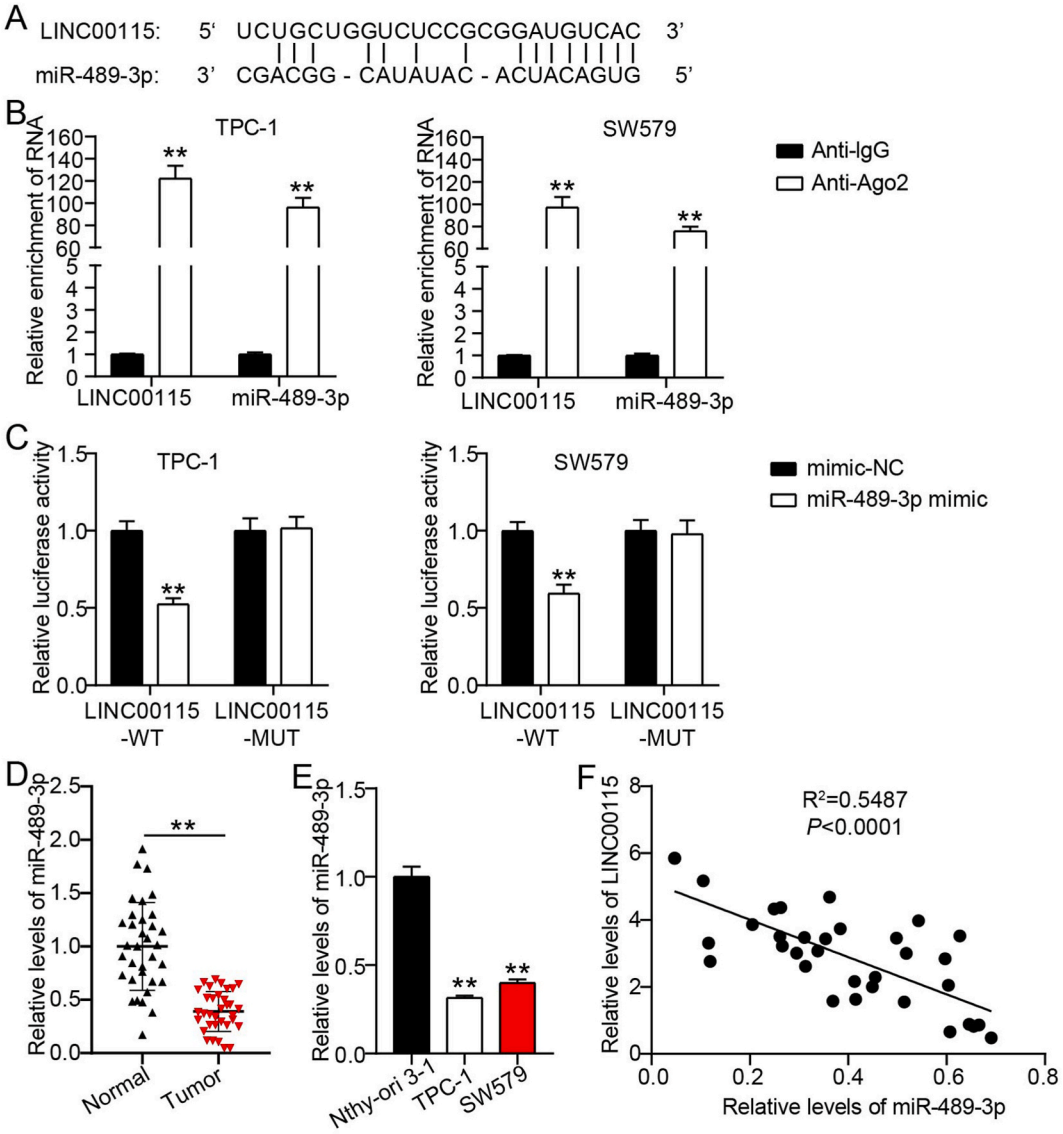

Fig. 3. The relationship between LINC00115 and miR-489-3p. A. The binding site of LINC00115 on miR-489-3p by ENCORI database. B. RIP result shows the binding between LINC00115 and miR-489-3p. **P < 0.001 vs Anti-lgG. C. Dual-luciferase reporter results show the binding between LINC00115 and miR-489-3p. **P < 0.001 vs mimic-NC. D. The relative level of miR-489-3p in THCA tumor and corresponding paracarcinoma normal tissues (n = 34). **P < 0.001. E. miR-489-3p relative level in normal human thyroid cell line Nthy-ori-3-1 and THCA cell lines. **P < 0.001 vs Nthy-ori 3-1. F. The confirmation of correlation between LINC00115 and miR-489-3p in THCA tissues.

3.3. miR-489-3p targeted EVA1A and downregulated EVA1A expression in THCA

Subsequently, we attempted to establish the link between *EVA1A* and *miR-489-3p* in patients with THCA. After searching the ENCORI database, binding sites were predicted between *miR-489-3p* and *EVA1A* (Fig. 5A). The activity of luciferase in the WT-*EVA1A* group was reduced by the *miR-489-3p* mimic (Fig. 5B). In addition, the relative expression levels of *EVA1A* were markedly elevated in THCA tissues than those in controls (Fig. 5C). *EVA1A* expression was consistently and significantly augmented in THCA cells than in Nthy-ori-3-1 cells (Fig. 5D). Moreover, *miR-489-3p* and *EVA1A* expression levels were negatively correlated in THCA tissue samples (Fig. 5E). These observations elucidate that *miR-489-3p* may function in THCA by targeting *EVA1A*.

EVA1A knockdown suppresses THCA progression and cancels the impact of *miR-489-3p* inhibition on THCA via Hippo signaling pathway.

Finally, to assess the effect of *EVA1A* on THCA progression, *EVA1A* was knocked down using si-*EVA1A*. The relative protein levels of *EVA1A* in THCA cells were reduced after si-*EVA1A* transfection, but were elevated after *miR-489-3p* inhibitor transfection (Fig. 6A). We then checked cell proliferation, apoptosis, and migration. As shown in Fig. 6B–D, THCA cells transfected with si-*EVA1A* exhibited

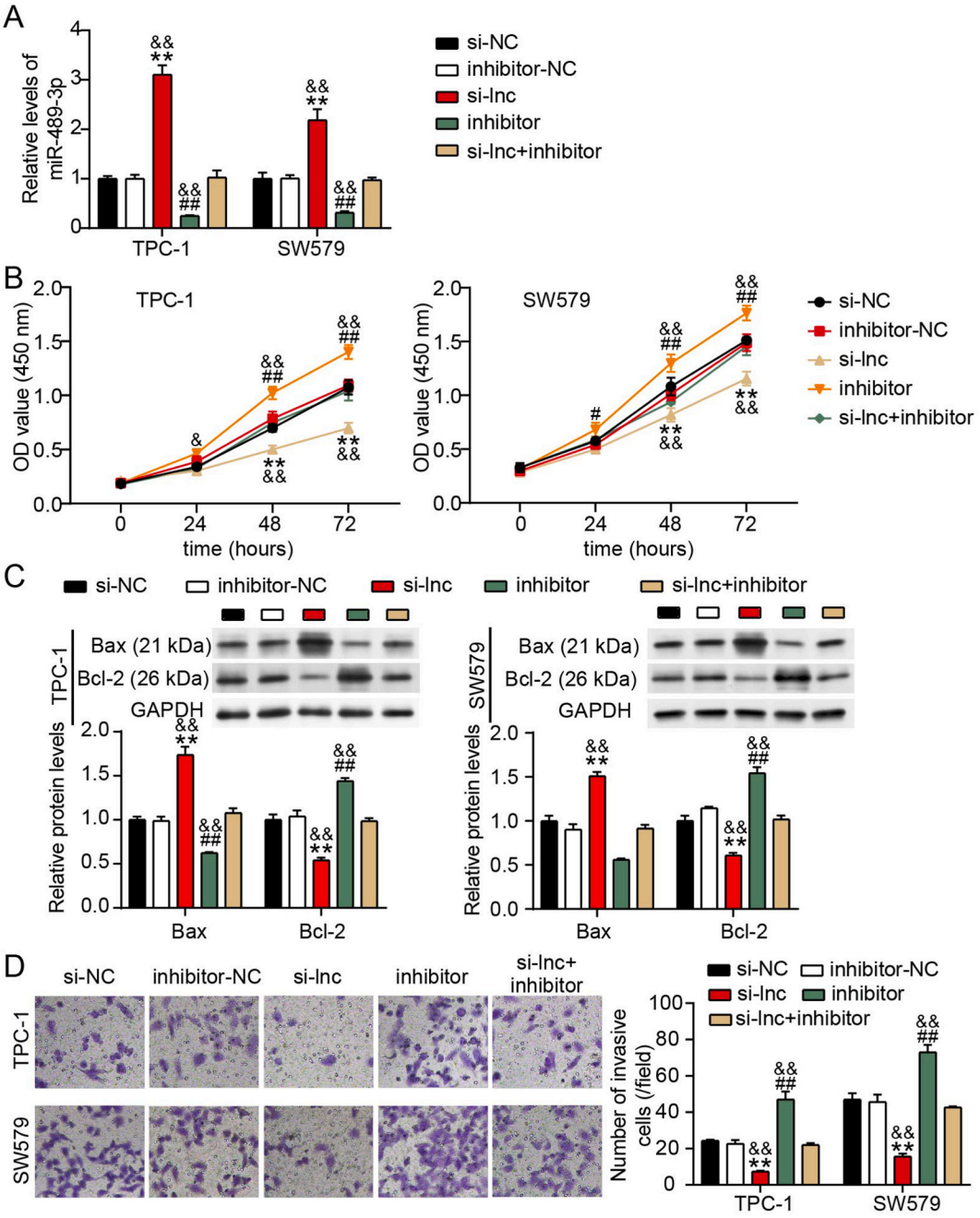

Fig. 4. miR-489-3p inhibitor revised the effect of si-LINC00115. A. The miR-489-3p level in TPC-1 and SW579 transfected with si-LINC00115 (si-lnc), miR-489-3p inhibitor, si-lnc + miR-489-3p inhibitor or corresponding NC. B. The proliferation of the transfected cells mentioned above. C. The protein expression of Bax and Bcl-2 in the transfected cells mentioned above. D. The images and quantitative analysis for Transwell assay of the transfected cells mentioned above. *P < 0.05, **P < 0.001 vs si-NC; and $^{\&}$P < 0.05, $^{\&\&}$P < 0.001 vs inhibitor-NC; and $^{\#}$P < 0.05, $^{\#\#}$P < 0.001 vs si-lnc + inhibitor.

decreased proliferation, enhanced apoptosis, and reduced migration compared to those transfected with siRNA-NC. In addition, si-*EVA1A* significantly reduced the impact of the *miR-489-3p* inhibitor on cell viability, migration, and apoptosis. Detection of the key proteins of the Hippo signaling pathway via western blotting disclosed that si-*EVA1A* lowered the YAP and TAZ relative protein levels and also reduced the *miR-489-3p* inhibitor's positive effects on their relative protein levels (Fig. 6E). Overall, these data revealed that *EVA1A* knockdown attenuates THCA malignancy and reverses the positive effects of *miR-489-3p* inhibition on THCA cells via the Hippo signaling pathway.

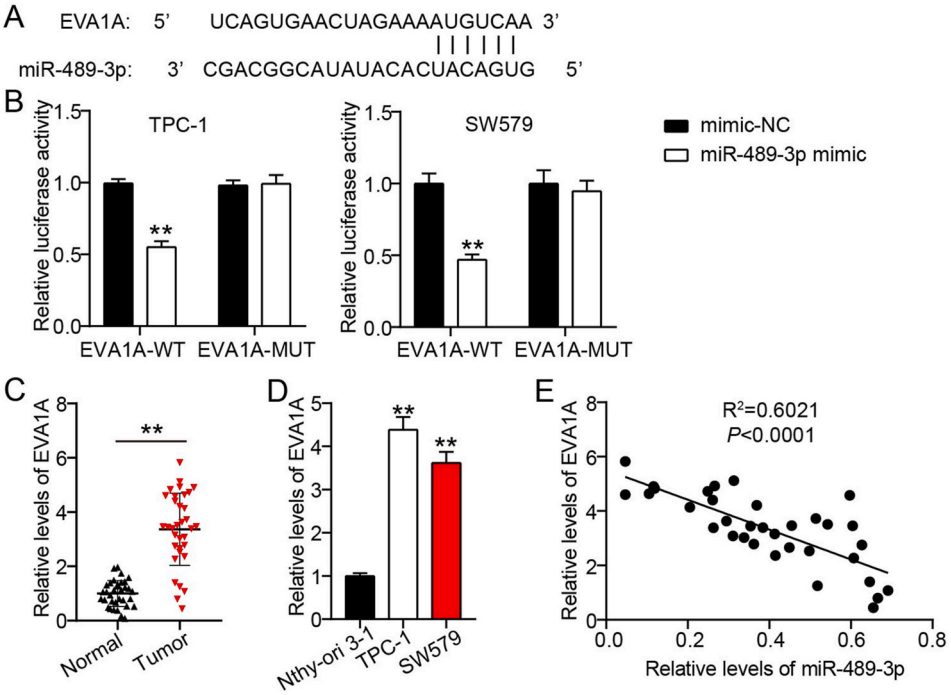

Fig. 5. The relationship between miR-489-3p and EVA1A. A. The prediction of miR-489-3p binding site on EVA1A by ENCORI database. B. The binding among LINC00115 and miR-489-3p via DLR. **P < 0.001 vs mimic-NC. C. The relative level of EVA1A in THCA tumor and corresponding paracarcinoma normal tissues (n = 34). **P < 0.001. D. EVA1A level in normal human thyroid cell line (Nthy-ori-3-1) as well as THCA cell lines. **P < 0.001 vs Nthy-ori 3-1. E. The association between miR-489-3p and EVA1A in THCA.

4. Discussion

It is widely accepted that lncRNAs are crucial regulators of various cancers [18,19]. In this study, we identified *LINC00115* as an oncogene involved in THCA progression. Further investigation showed that *miR-489-3p* was an *LINC00115* target. *LINC00115* also targeted *EVA1A*, which eventually regulate the Hippo signaling pathway. Finally, we revealed that the positive role of *LINC00115* in THCA was dependent on the modulation of the Hippo signaling pathway by the *miR-489-3p/EVA1A* axis.

LINC00115 had been observed as an oncogene in some cancers, such as glioma [13], breast cancer [20], CRC [14], and cervical cancer [21]. It was also documented to be overexpressed in lung cancer and shown to promote lung cancer cell invasion and proliferation [22]. However, the effects of *LINC00115* on THCA remain unclear. We are the first to uncover upregulated expression of *LINC0015* in THCA tissues as well as cells, and that knocking it down significantly inhibited THCA cell migration and proliferation but promoted apoptosis. These results clearly implied the involvement of *LINC00115* in THCA progression; however, the exact mechanism requires further clarification.

lncRNAs exert biological effects by interacting with a variety of molecules, including mRNAs, miRNAs, and proteins [23]. A search of the ENCORI database showed that *miR-489-3p* is a predicted target of *LINC00115*. By performing cellular experiments, we illustrated that *LINC00115* targets *miR-489-3p*, which is in line with a previous investigation done in CRC cells [14]. Furthermore, *miR-489-3p* was identified as an important tumor suppressor in human cancers [24–26]. In non-small cell lung cancer (NSCLC), sponging of *miR-489-3p* by the lncRNA *MIR503HG* suppressed cell proliferation and enhanced apoptosis [27]. While the functional role of *miR-489-3p* was not explored in THCA, we predicted that it may play an inhibitory role based on previous research findings. Our results verified the downregulation of *miR-489-3p* in THCA cells and its inhibition strongly promoted the migration and proliferation of THCA cells and reduced their apoptosis. Interestingly, *LINC00115* knockdown effects on THCA cell growth and apoptosis were effectively overturned by inhibition of *miR-489-3p*. Consistent with our hypothesis and the findings of a previous study on CRC [14], our data clearly indicated that *LINC00115* exerts its effect on THCA by targeting *miR-489-3p*.

Further examination of the downstream regulatory mechanism by cellular assays revealed that *EVA1A* was a target gene of *miR-489-3p*. *EVA1A* is a newly discovered protein-coding RNA associated with the plasmid reticle and lysosomes [28]. It has been demonstrated to serve significant roles in various malignancies, including breast cancer [29], glioblastoma [30], hepatocellular carcinoma [31] and NSCLC [32] by inducing autophagy and apoptosis. In this study, we established upregulated expression of *EVA1A* in THCA. We subsequently knocked down *EVA1A* in THCA cells and observed that it markedly prevented cell proliferation and promoted apoptosis. Our data are congruent with those of a recent investigation, which elucidated that downregulation of *EVA1A* was shown to inhibit papillary thyroid cancer malignant properties [33]. Additionally, we verified that the effects of *miR-489-3p* inhibition on THCA cells were partially reversed by *EVA1A* knockdown via the Hippo signaling pathway. Consequently, we confirmed that

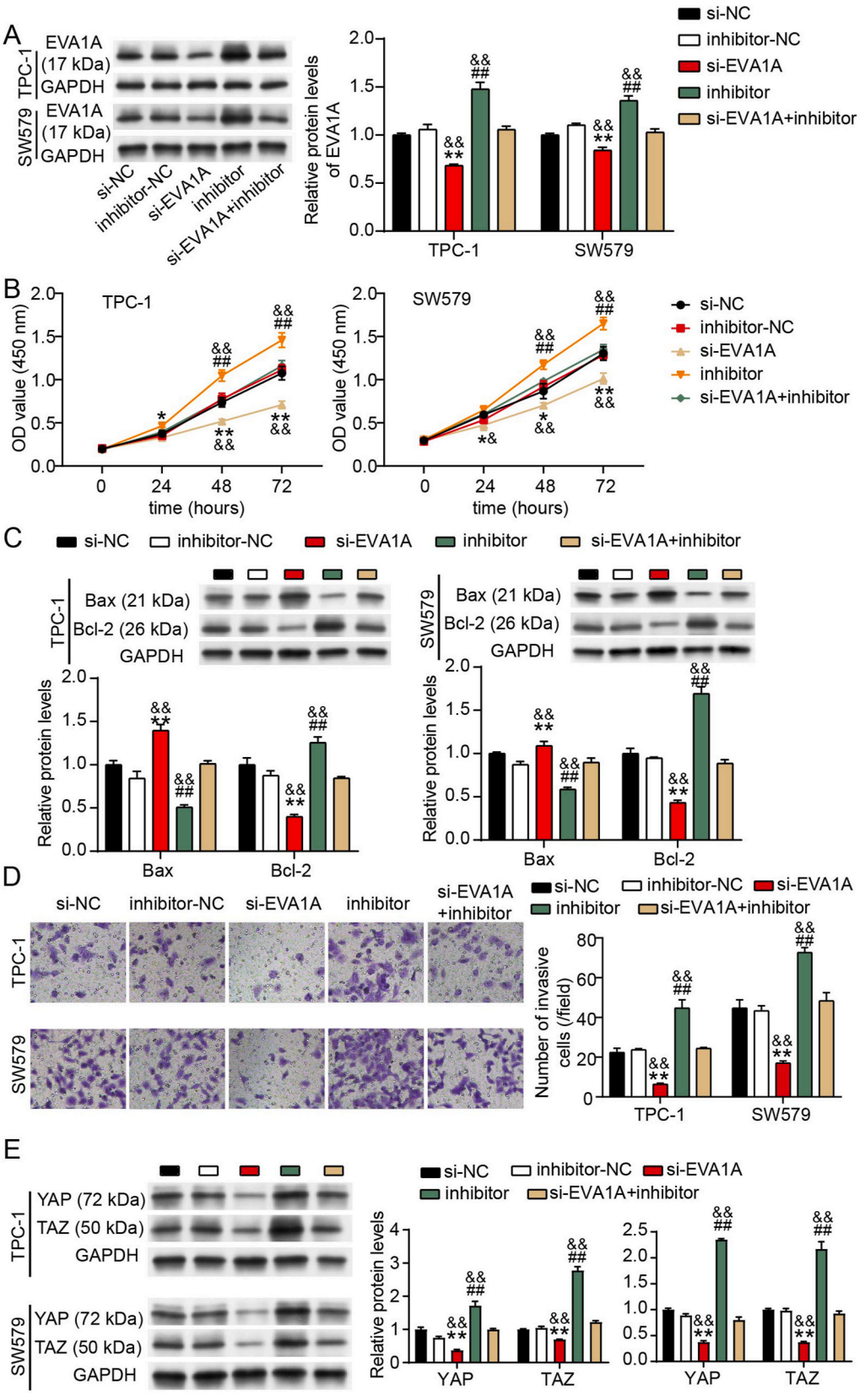

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Fig. 6. Effects of EVA1A knockdown on THCA cells, miR-489-3p inhibition, and Hippo signaling pathway. A. The protein expression of EVA1A in TPC-1 and SW579 transfected with si-EVA1A, miR-489-3p inhibitor, si-EVA1A + miR-489-3p inhibitor or corresponding NC. B. Proliferation of the transfected cells mentioned above. C. Bcl-2 and Bax protein expression in the transfected cells mentioned above. D. The images and quantitative analysis for Transwell assay of the transfected cells mentioned above. E. Western blotting detected the key proteins of Hippo signaling pathway in the transfected cells mentioned above. *P < 0.05, **P < 0.001 vs si-NC; and $^{\&}$P < 0.05, $^{\&\&}$P < 0.001 vs inhibitor-NC; and $^{\#\#}$P < 0.001 vs si-EVA1A + inhibitor.

miR-489-3p inhibits THCA development by targeting *EVA1A* as a downstream target via regulation of the Hippo signaling pathway.

5. Conclusions

In conclusion, we are first to report that *LINC00115* is a potent oncogene in THCA. *LINC00115* aggravates THCA development by targeting *miR-489-3p* and *EVA1A* regulators to regulate the Hippo signaling pathway. Thus, the *LINC00*115/miR-*489-3p/EVA1A* axis might be an effective target for THCA treatment. In the future, the clinical value and therapeutic potential of the *LINC00*115/miR-*489-3p/EVA1A* axis in THCA should be further confirmed.

Funding

Not applicable.

Ethics approval

The Ethics Committee of Wuhan Red Cross Hospital (Wuhan, China) approved this study. The ethics approval number is 2019003. Clinical tissue samples are processed in full agreement with the Helsinki's ethical principles declaration. All patients have completed the consent form in writing.

Consent to participate

All patients signed written informed consent.

Consent for publication

Participants approved publishing this work.

Material and data availability

All data produced or analyzed throughout the research is contained in this paper.

CRediT authorship contribution statement

Jie Cao: Funding acquisition, Formal analysis, Data curation, Conceptualization. **Wei Kong:** Supervision, Software, Resources, Project administration. **Xiangli Xiao:** Resources, Project administration, Investigation, Conceptualization.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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None.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.heliyon.2024.e30331.

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