RETRACTED ARTICLE: Long non-coding RNA CCAT2 acts as an oncogene in osteosarcoma through regulation of miR-200b/VEGF

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ABSTRACT
Background/aim: Colon cancer-associated transcript 2 (CCAT2) is a new IncRNA, which is closely associated with risk of several cancers. The aim of this study was to explore the regulatory mechanism of CCAT2 in osteosarcoma (OSA).

Methods: Cells were transfected with si-CCAT2, microRNA (miR)-200b inhibitor and the corresponding controls. Quantitative reverse transcription polymerase chain reaction (qRT-PCR) was used to measure the expression of CCAT2 and miR-200b in OSA tissues and cell lines. CCK8 and bromodeoxyuridine (BrdU) were conducted to examine cell proliferation. Apoptosis was detected by PI/FITC-Annexin V combining with flow cytometric analysis. Migration and invasion were respectively measured through transwell chambers assays. Western blot was used to examine expressions of relative proteins.

Results: CCAT2 was highly expressed and miR-200b was lowly expressed in OSA tissues and cell lines. Knockdown of CCAT2 suppressed cell proliferation, migration and invasion but induced apoptosis and up-regulation of miR-200b. miR-200b inhibitor weakened the effect of si-CCAT2 on cell progression and cell mobility. Besides, knockdown of CCAT2 blocked the PI3K/Akt and AMPK pathways through up-regulating miR-200b.

Conclusions: The CCAT2/miR-200b/vascular endothelial growth factor (VEGF) axis plays important regulating effect in OSA through the PI3K/Akt and AMPK pathways.

Abbreviations: OSA: Osteosarcoma; nc RNA: Non-coding RNA; Inc RNA: Long chain non-coding RNA; CCAT2: Colon cancer-associated transcript 2; CCK-8: Cell Counting Kit-8; BrdU: Bromodeoxyuridine; PI: Propidium iodide; RT: Reverse transcription

Introduction
Osteosarcoma (OSA) is characterized by high degree of malignancy and poor prognosis, which brings a serious threat to physical and mental health for patients. OSA can occur in any age group, but the incidence of OSA is mainly concentrated in 10–25 years old and the incidence of OSA in males is significantly higher than that in females [1]. The causes of OSA are closely related to previous traumatic history, genetic factors, viral infection and radiation stimulation [2]. The treatment of OSA has been greatly improved in the past few decades and the 5-year survival rate has reached 60–70%. However, many patients appear lung metastasis with poor prognosis [3,4]. Thus, it is urgent to explore more accurate and effective treatment methods.

Non-coding RNA (ncRNA) accounts for about 98% of the total human genome and more and more scholars have carried out in-depth studies on the specific functions of ncRNA. Among them, long chain non-coding RNA (IncRNA) is a kind of RNA whose length is larger than 200 NT [5]. LncRNA can interfere with gene expression at various levels such as epigenetic modification, transcription and post-transcription, thus affecting important biological processes such as chromatin modification, gene transcription and translation, and RNA variable splicing regulation. It also participates in almost all physiological and pathological processes of human [6].

Colon cancer-associated transcript 2 (CCAT2) is a new IncRNA which was discovered by Ling et al. in 2013. CCAT2 consists of 1752 nucleotides and is located at chromosomal 8q24, which is closely associated with risk of several cancers [7]. The expression of CCAT2 in hepatocellular carcinoma [8], breast cancer [9], gastric cancer [10] and some other cancers is significantly increased, and overexpression of CCAT2 can promote the proliferation, invasion, migration and inhibit apoptosis of cancer cells. However, the effect of CCAT2 in OSA has not been clearly elucidated and related regulatory mechanism has not been explored.
Previous study of cancer mechanism found that miRNAs are involved in different stages of tumourigenesis and metastasis [11]. The pathogenesis of OSA is related to gene mutation, abnormal activation of signal pathway, abnormal regulation between miRNAs and IncRNAs and so on [12].

miR-200b is one of the miRNAs which is closely with cancer progression. For example, miR-200b was significantly down-regulated in both breast cancer tissues and cell lines and it has been regarded as prognostic biomarker and tumour suppressor for breast cancer patients [13]. However, Pan et al. reported that miR-200b was overexpressed in colorectal cancer and it can promote the progression of colorectal cancer [14]. Besides that, Liu et al. reported that IncRNA ZFAS1 positively regulated malignant phenotypes by competitively binding the miR-200b and miR-200c and up-regulating BMI1 in OSA [15]. However, the specific effect of miR-200b in OSA has also not been clearly clarified.

In this study, we found that CCAT2 was highly expressed and miR-200b was lowly expressed in OSA cells and the knockdown of CCAT2 effectively suppressed cell proliferation and cell mobility in OSA cells. Further study revealed that the anti-tumour effect of CCAT2 was exerted through the CCAT2/miR-200b/vascular endothelial growth factor (VEGF) axis via the phosphoinositide 3 kinases (PI3K)/protein kinase B (Akt) and AMP activated protein kinase (AMPK) signal pathways, enriching our knowledge about the progression of OSA and providing novel diagnostic markers for OSAs.

Materials and methods

Clinical specimens
Twenty pairs of human OSA tissues and adjacent normal tissues were collected from OS patients who were diagnosed at Qilu Hospital of Shandong University. This study was approved by the Research Medical Ethics Committee of the Qilu Hospital of Shandong University and written informed consent was signed by every OSA patient.

Cell culture and treatment
The human OSA cell lines MG63, U2OS, OS732 and Saos-2 as well as human osteoblast cell lines hFOB1.19 were purchased from the American Type Culture Collection (Manassas, VA) and were cultured in high glucose DMEM (Gibco Co., New York, NY) supplemented with 10% foetal bovine serum (FBS) (Invitrogen, Carlsbad, CA) in a moist atmosphere at 37 °C with 5% CO2.

CCK-8 assay
In brief, cells were seeded in 96-well plate with 5000 cells/well and were maintained at 37 °C incubator overnight. Cell proliferation was assessed by a CCK-8 (Dojindo Molecular Technologies, Gaithersburg, MD). Briefly, after the cells were treated as indicated, the CCK-8 solution was added into each well, and the cultures were incubated for 4 h. Then, the absorbance was measured at 450 nm by microplate reader (BioTek Instruments, Winooski, VT).

Proliferation assay
Cell proliferation was valued by using the bromodeoxyuridine (BrdU, 1 mg/mL, Sigma-Aldrich, St. Louis, MO) adding to the cultured cells 3 h before analysis. At least 1000 cells were counted in each condition and at least five replicates are contained in each condition.

Cell invasion assay
Cell invasion was valued by using a modified two-chamber (Becton-Dickinson, Franklin Lakes, NJ). About 1 × 105 cells/well cells suspended in 200 μL of serum-free medium were seeded on the upper compartment of the chamber and 600 mL of complete medium containing 20% FBS was added to the lower compartment. After 24 h, non-invading cells on the upper membranes were removed with a cotton swab and the invaded cells were fixed with methanol and stained with hematoxylin. The cell numbers were counted by Image J software and photographed randomly from five visual fields per well by an inverted microscope. Each experiment was independently repeated in triplicate.

Cell migration assay
Cells 1.5 × 105 cells/well were seeded in 6-well plates with a dilution of 1.5 × 105 cells/well, and cultured overnight until the cells reached 90% confluence. Wounds on the monolayer cells were made by a 200 μL sterile pipette tip and cell debris was washed by phosphate buffer saline (PBS) gently for at least 3 times. Then, cell migration range was observed and imaged at 0 and 24 h with a digital camera (Leica DFC300FX; Leica Microsystems GmbH, Wetzlar, Germany).

Apoptosis assay
Cell apoptosis analysis was performed using FITC Annexin V/PI Apoptosis Detection Kit I (RiboBio, Guangzhou, China). Briefly, cells were harvested and washed in PBS and fixed in 70% ethanol. Fixed cells were then washed twice in PBS and stained with 5 μL of Annexin V-FITC in the presence of 50 μg/mL RNase A (Sigma-Aldrich, St. Louis, MO) for 15 min, and then incubated with 10 μL of propidium iodide (PI; 10 mg/mL) for 5 min at room temperature in the dark. Flow cytometry analysis was done by using a FACScan flow cytometer (Becton Dickinson, Franklin Lakes, NJ). The data were analysed by using FlowJo software.

qRT-PCR
Total RNA was extracted from OSA tissues and cells using Trizol reagent (Invitrogen, Carlsbad, CA). The concentrations of RNA were valued using a NanoDrop ND-1000 instrument (ThermoFisher, Waltham, MA) at 260 and 280 nm. For the reverse transcription (RT) reaction of the miRNA, the
PrimeScript RT Master Mix (Perfect Real Time; Takara Bio Inc., Tokyo, Japan) was used. PCR reactions were conducted in an ABI PRISM 7900 Real-Time system (Applied Biosystems, Foster City, CA) with the SYBR Premix ExTaq kit (Takara Bio, Inc., Tokyo, Japan). All PCR experiments were performed in triplicate. The results were measured by using the $2^{\Delta\Delta CT}$ method. The level of miR-200b and CCAT2 was normalized to the GAPDH and U6, respectively.

**Western blot**

The protein in OSA tissues and cells was extracted using RIPA lysis buffer (Beyotime Biotechnology, Shanghai, China) and protein concentration was quantified using the BCA™ Protein Assay Kit (Pierce, Appleton, WI). Proteins were separated by 10% sodium dodecyl sulphate polyacrylamide gel electrophoresis (SDS-PAGE) and then transferred to polyvinylidene difluoride (PVDF) membranes (Millipore, Billerica, MA). After blocked with 5% skim milk at room temperature for 2 h, the membranes were incubated with the primary antibodies at 4 °C overnight, followed by wash and incubation with secondary antibody marked by horseradish peroxidase at room temperature for 1 h. After rinsing, the membrane were transferred into the Bio-Rad ChemiDoc™ XRS system following the standard method. β-actin was used as an internal reference.

**Cell transfection**

For the knockdown of CCAT2, specific si-CCAT2 and corresponding control si-NC were synthesized by Gene Pharma (Shanghai, China). At the same time, specific miR-200b inhibitor and its negative control (anti-miR) and miR-200b mimic which was used to knockdown and overexpress miR-200b, respectively were also synthesized by Gene Pharma (Shanghai, China). For cell transfection, cells were grown to 50% confluence, and then transfected using Lipofectamine 2000 (Invitrogen Life Technologies, Carlsbad, CA) according to the manufacturer’s instructions. Transfected cells were harvested 48 h later for subsequent experiments.

**DNA constructs and luciferase reporter assay**

The 3'-UTR WT sequence of VEGF predicted to interact with miR-200b or a MUT sequence within the predicted target sites were inserted into the pGL3 control vector respectively (Promega, Madison, WI). For the luciferase activity analysis, cells were co-transfected with the miR-200b mimic and VEGF WT or MUT. The DharmaFECT Duo transfection reagent (Thermo Fisher Scientific, Waltham, MA) was used for 48 h, and the luciferase assays were performed with the Dual-Glo Luciferase assay (Promega, Beijing, China) according to the manufacturer’s instructions.

**Figure 1.** Expression level of CCAT2 was up-regulated and miR-200b was down-regulated in osteosarcoma (OSA) tissues and cell lines. (A) qRT-PCR analysis was performed to detect the expression of CCAT2 in OSA tissues and the control non-tumour tissues. (B) qRT-PCR analysis was performed to detect the expression of CCAT2 in human OSA cell lines MG63, U2OS, OS732 and Saos-2 as well as human osteoblast cell line hFOB1.19. (C) qRT-PCR analysis was performed to detect the expression of miR-200b in OSA tissues and the control non-tumour tissues. (D) qRT-PCR analysis was performed to detect the expression of miR-200b in human OSA cell lines MG63, U2OS, OS732 and Saos-2 as well as human osteoblast cell line hFOB1.19. All data were presented as mean ± SD ($N = 3$, $* p < .05$, $** p < .01$, $*** p < .001$ vs. control group).
Statistical analysis

All experiments were repeated three times. All statistical assays were measured by SPSS version 17.0 software (IBM, Chicago, IL). The results of multiple experiments were formulated as mean ± SD. Statistical analyses were performed using Graphpad version 6.0 statistical software (GraphPad, San Diego, CA). The difference between two groups was analysed by the Student’s t-test. The p values were calculated using a one-way analysis of variance (ANOVA) and p < .05 was considered as statistically significant.

Results

Expression level of CCAT2 is up-regulated and miR-200b is down-regulated in OSA tissues and cell lines

At first, the expression level of CCAT2 and miR-200b was detected in OSA tissues and cell lines. As was shown, the level of CCAT2 was obviously elevated in OSA tissues compared with the control non-tumour tissues (p < .001, Figure 1(A)). Similarly, the level of CCAT2 was also much elevated in human OSA cell lines MG63, U2OS, OS732 and Saos-2 compared with that in human osteoblast cell lines hFOB1.19 (p < .05 or p < .01, Figure 1(B)). On the contrary, the level of miR-200b was down-regulated in OSA tissues compared with the control non-tumour tissues (p < .01, Figure 1(C)). Similar results were found in MG63, U2OS, OS732 and Saos-2 cell lines compared with the hFOB1.19 cells (p < .05 or p < .01, Figure 1(D)). Thus, we concluded that the expression level of CCAT2 was up-regulated and miR-200b was down-regulated in OSA tissues and cell lines.

Knockdown of CCAT2 suppresses cell proliferation and induces cell apoptosis

To further explore the effect of CCAT2 in OSA, the high expression of CCAT2 in OSA was knocked down by specifically transfected the MG63 and U2OS cells with si-CCAT2 (both
p < .01, Figure 2(A)). At the same time, si-NC was used as a control. Then we found that the expression of cyclic marker protein CyclinD1 and CDK6 was suppressed by si-CCAT2 in MG63 and U2OS cells (p < .05 or p < .01, Figure 2(B–E)). The result detected through BrdU assay also demonstrated that cell proliferation was greatly suppressed in the si-CCAT2 group compared with the control (p < .05 or p < .01, Figure 2(F)). Besides that, cell viability detected through CCK-8 assay was suppressed (both p < .05, Figure 2(G)) and cell apoptosis rate detected through flow cytometry was elevated (p < .01 or p < .001, Figure 2(H)) in the si-CCAT2 group compared with the control. In addition, the expression of cell apoptosis related proteins was valued through western blot. The results showed that the level of cleaved caspase-3 and Bax was up-regulated (p < .01 or p < .001) and the level of Bcl-2 was down-regulated (p < .05 or p < .01) in the si-CCAT2 group compared with the control, identifying that obvious cell apoptosis was induced by si-CCAT2 (Figure 2(I–L)). Thus, we concluded that knockdown of CCAT2 suppressed cell proliferation and induced cell apoptosis in OSA cell lines.

**Knockdown of CCAT2 inhibits cell migration and invasion**

Having known the effect of CCAT2 on cell proliferation and cell apoptosis, the effect of CCAT2 on cell mobility was also further explored. The results of wound healing assay and transwell invasion assay showed that cell migration and cell invasion rate in MG63 and U2OS cells were both greatly suppressed by si-CCAT2 compared with the control group (all p < .01, Figure 3(A,B)). Thus, we concluded that knockdown of CCAT2 suppressed cell migration and invasion.

**Knockdown of CCAT2 elevates the expression level of miR-200b**

Further study found that the expression level of miR-200b was up-regulated by si-CCAT2 compared with the control group, identifying that miR-200b might be a target of CCAT2 in OSA cells (p < .05 or p < .01, Figure 4(A,B)).

**Knockdown of CCAT2 suppresses cell proliferation and cell mobility but induces cell apoptosis through up-regulating miR-200b**

Specific miR-200b inhibitor was used in our following experiments. Then we found that the result of BrdU assay showed that suppressed cell proliferation by si-CCAT2 was then partly rescued by the adding of miR-200b inhibitor in MG63 and U2OS cells (both p < .05, Figure 5(A)). At the same time, the results of western blot also showed that suppressed expression of CyclinD1 and CDK6 in si-CCAT2 group was up-regulated by miR-200b inhibitor (all p < .05, Figure 5(B–E)). In addition, elevated cell apoptosis rate was then suppressed by the combination of miR-200b inhibitor (both p < .05, Figure 5(F)). Corresponding results were shown by western blot assay. The results showed that the promoting effect of si-CCAT2 on the level of cleaved caspase-3 and Bax and the inhibiting effect of si-CCAT2 on the level of Bcl-2 were both offset by the combination of miR-200b inhibitor (all p < .05, Figure 5(G–J)). What is more, the results of the wound healing assay and transwell invasion assay also showed that the inhibiting effect of si-CCAT2 on cell migration and cell invasion was also counteracted by the adding of miR-200b inhibitor (all p < .05, Figure 5(K–L)). Thus, we concluded that knockdown of CCAT2 suppressed cell proliferation and cell mobility but induced cell apoptosis through up-regulating miR-200b.
VEGF is a target of miR-200b in OSA cell lines

Downstream target was further investigated. As was shown, the expression of VEGF was suppressed by si-CCAT2 (both \( p < .01 \)) and was then rescued by miR-200b inhibitor in MG63 and U2OS cells (\( p < .05 \) or \( p < .01 \)) (Figure 6(A–C)). Besides that, the result of luciferase reporter assay showed that the combination of VEGF WT and miR-200b mimic greatly suppressed luciferase intensity and the combination of VEGF MUT and miR-200b mimic had no obvious effect on luciferase intensity, identifying that VEGF is a target of miR-200b in OSA cell lines (\( p < .01 \), Figure 6(D)).

Knockdown of CCAT2 inactivates the PI3K/AKT and AMPK signal pathways through up-regulating miR-200b

Associated signal pathways were also explored. As was shown, the rate of p/t-PI3K and p/t-AKT was suppressed by...
si-CCAT2 ($p < 0.05$ or $p < 0.01$) and was then elevated by the combination of miR-200b inhibitor ($p < 0.05$ or $p < 0.01$), identifying that si-CCAT2 inactivates the PI3K/AKT signal pathway through up-regulating miR-200b (Figure 7(A,B)). Similarly, the rate of p/t-AMPK was suppressed by si-CCAT2 (both $p < 0.01$) and was then elevated by the adding of miR-200b inhibitor (both $p < 0.05$), identifying that si-CCAT2 inactivates the AMPK signal pathway through up-regulating miR-200b (Figure 7(E–H)). Thus, we concluded that the knockdown of CCAT2 inactivated the PI3K/AKT and AMPK signal pathways through up-regulating miR-200b.

**Discussion**

OSA, a highly malignant tumour, mainly occurs in children and adolescents. It may affect the head and neck, reproductive organs, urinary system and limbs of patients, and seriously endanger the limb function and life safety of patients. At present, the treatment of OSA is mostly limited to chemotherapy and surgery. Chemotherapy and surgery can bring serious psychological and physiological injuries to patients, and they are impossible to predict the effect of surgery [16]. Therefore, there is an urgent need to find targeted molecules of OSA cells to inhibit the growth of OSA cells at the cellular and molecular levels, so as to achieve the goal of treating OSA.

The abnormal expression of IncRNA plays an important role in cancer progression. Recently, large amount of researches have demonstrated that many IncRNAs such as HOTAIR, and metastasis associated lung adenocarcinoma transcript 1 (MALAT1) are highly expressed in colorectal cancer, breast cancer, hepatocellular carcinoma and some other cancer tissues and are closely related to a variety of cancer metastasis, thus can be used as targets for cancer early diagnosis, treatment and prognosis prediction [17,18]. CCAT2, as a novel oncogene, is located in the hotspot of tumour-related rs6983267 SNP, which promotes the occurrence and development of kinds of cancers by affecting Wnt and TGF-β signalling pathways [9,19]. Such as Xie et al. reported that CCAT2 was overexpressed in endometrial cancer and knockdown of CCAT2 inhibited endometrial cancer cells growth and metastasis via sponging miR-216b [20]. Ruan et al. also demonstrated that CCAT2 was able to promote the progression of OSA by enhancing cell proliferation via GSK3β/β-catenin signalling pathway [21]. However, what downstream genes are regulated by CCAT2 and which molecular mechanisms are associated with the carcinogenesis need further investigation. However, the specific downstream targeting molecules of CCAT2 in OSA have not been explored before. Similarly, in our study, we found that the expression of CCAT2 was highly up-regulated in OSA tissues and cell lines compared with the control. Our further study also revealed that knockdown of CCAT2 had effectively inhibiting effect on cell proliferation and cell mobility in OSA. Then we set to explore possible target molecules and associated signal pathways in our following experiments.

Studies have shown that miRNAs are closely involved in tumourigenesis, proliferation and differentiation, invasion and migration of cancer cells. Besides that, studies have found that about 50% of the miRNAs are closely related to the origin of cancer genes, suggesting that miRNAs may play an indispensable role in tumourigenesis and development [22].
Figure 7. Knockdown of CCAT2 blocked the PI3K/AKT and AMPK signal pathways through up-regulating miR-200b. (A,B) The expression of PI3K/AKT signal pathway associated proteins p-PI3K, t-PI3K, p-AKT and t-AKT was detected through western blot in MG63 cells. (C,D) The expression of PI3K/AKT signal pathway associated proteins p-PI3K, t-PI3K, p-AKT and t-AKT was detected through western blot in U2OS cells. (E,F) The expression of AMPK signal pathway associated proteins p-AMPK, t-AMPK was detected through western blot in MG63 cells. (G,H) The expression of AMPK signal pathway associated proteins p-AMPK, t-AMPK was detected through western blot in U2OS cells. β-actin was used as internal control. All data were presented as mean ± SD (N = 3, *p < .05, **p < .01 vs. corresponding group).
Many tissue-specific miRNAs can provide clues for the diagnosis of tumours and improve the possibility of cancer prevention and the dysregulation of miRNAs can be rescued by using miRNA mimics and inhibitors, showing promise in pre-clinical development [23]. For example, Zhang et al. reported that miR-124 was down-regulated in prostate cancer specimens and cell lines and overexpression of miR-124 by transient transfection of mimics effectively suppressed cell motility and adhesion in prostate cancer cells [24]. Besides that, there exists a complex regulatory relationship between miRNAs and IncRNAs. Because the transcription process of IncRNA is similar to that of miRNA, including RNA splicing, RNA editing and so on, so miRNA can also match the sequence of IncRNA to a certain degree, thus to regulate the expression level of IncRNA and affect the occurrence and development of disease. IncRNA can also affect the downstream target genes of miRNA by regulating miRNA, thus affecting various biological processes [25]. In addition, Liu et al. found that IncRNA 00312 inhibited the proliferation, invasion and metastasis of cancer cells and overexpression of miRNA-197-3p was able to inhibit the expression of IncRNA 00312. Therefore, the growth of thyroid cancer cells can be controlled by inhibiting the expression of miRNA-197-3p [26]. In this study, we found that the knockdown of CCAT2 elevated the expression of miR-200b and the combination of miR-200b abolished the inhibiting effect of CCAT2 on cell proliferation and cell mobility. Thus, miR-200b may act as a target of CCAT2 in OSA cells and the anti-tumour effect of CCAT2 was performed through up-regulating the expression of miR-200b.

VEGF is an endothelial cell-specific cytokine, which has a great influence on angiogenesis of tumours. It can not only increase the permeability of micro-vessels, but also bind to receptors on vascular endothelial cells to directly stimulate the proliferation and division of endothelial cells, thus promoting the formation of blood vessels and the progression of cancers [27]. In our study, we found that the expression of VEGF in OSA cells was suppressed by the knockdown of CCAT2 and was then elevated by the addition of miR-200b inhibitor. At the same time, the result of luciferase reporter assay further demonstrated the targeting relationship between VEGF and miR-200b. Similarly, Li et al. reported that miR-377 overexpression inhibited the initiation, growth and angiogenesis of ESCC tumours by targeting CD133 and VEGF [28]. Thus, we concluded that VEGF was a target of miR-200b in OSA cells.

Studies have shown that the signal transduction pathway mediated by PI3K is closely related to tumourigenesis. The abnormal activation of PI3K signal transduction pathway is closely related to the occurrence and development of malignant tumours, and plays an important role in the proliferation, migration and metabolism of cancer cells [29]. AMPK widely exists in animals and plants and is a highly conserved serine/threonine binding protein that acts as an energy balancer, playing a key role in regulating energy metabolism, and is of great importance in the growth, proliferation and angiogenesis of tumour cells [30]. Such as Lin et al. reported, interleukin-12 (IL-12) served as an important tumour suppressor in human breast cancer cells through the inhibition of the PI3K/Akt signalling pathway and the activation of the AMPK signalling pathway [31]. In this study, we found that the knockdown of CCAT2 inactivated the PI3K/Akt and AMPK signal pathways and the addition of miR-200b inhibitor partly offset the inhibiting effect of si-CCAT2 on the PI3K/Akt and AMPK signal pathways. Thus, we concluded that the knockdown of CCAT2 inactivated the PI3K/Akt and AMPK signal pathways through up-regulating the expression of miR-200b.

Conclusions

Taken together, we demonstrate that the knockdown of CCAT2 has inhibiting effect on the proliferation, invasion and migration of OSA cells, which through directly targeting and up-regulating miR-200b to inactivate the PI3K/Akt and AMPK signal pathways. Further study also found that VEGF was a target of miR-200b. Our study enriches our knowledge of how IncRNA CCAT2 contributes to the progression of OSA cells, providing novel diagnostic markers for OSA.

Disclosure statement

Authors declare that there is no conflict of interests.

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