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lncRNA PCAT1 might coordinate ZNF217 to promote CRC adhesion and invasion through regulating MTA2/MTA3/Snai1/E-cadherin signaling

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ABSTRACT

LncRNA prostate cancer-associated transcript 1 (PCAT1) is a well-known oncogene, but the mechanisms of exosomes PCAT1 in colorectal cancer (CRC) remain largely unknown. Thus, the mechanisms of exosomes lncRNA PCAT1 were investigated. The expressions of exosomes lncRNA PCAT1 in tissues from stage 0-I and stage II-III CRC patients, and intestinal epithelial cell line FHC and two CRC cell lines, HT29 and HCT8 were measured by real-time quantitative PCR. The effects of lncRNA PCAT1 on adhesion and invasion of two CRC cell lines were investigated by cell-matrix adhesion and transwell assays. In addition, the target of PCAT1 (ZNF217) was validated using an RNA immune precipitation assay. Finally, the protein levels of MTA2, MTA3, SNAI1, and E-cadherin in normal participants, stage 0-I and stage II-III CRC patients, as well as two cell lines with stable ZNF217 knockdown were investigated by western blotting. The plasma exosomal lncRNA PCAT1 was found to be significantly increased in the CRC tissues and cell lines. In addition, lncRNA PCAT1 knockdown significantly inhibited the adhesion and invasion of HT29 and HCT8 cells. RIP assay results showed lncRNA PCAT1 could target ZNF217, and downregulation of lncRNA PCAT1 could decrease the protein expressions of ZNF217 in two CRC cells lines. Moreover, ZNF217 knockdown significantly decreased MTA2, MTA3, and SNAI1 expressions, but increased E-cadherin expressions in both CRC cells lines. Exosomal lncRNA PCAT1 can promote the adhesion and invasion of CRC cells, and PCAT1 overexpression may lead to ZNF217 upregulation that regulates EMT-related MTA2/MTA3/Snai1/E-cadherin signaling.

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Introduction

Colorectal cancer (CRC) is the third frequent malignancy and the second cause of cancer death, only next to lung cancer. GLOBOCAN estimates there are over 1.8 million new CRC cases and 881,000 deaths worldwide in 2018, accounting for 10% of cancer cases and deaths (1). CRC is considered to be a complex disease, which is largely attributed to both environmental and genetic risk factors (2). Commonly, hematochezia is a main clinical symptom for early-stage CRC patients, but it is often ignored until diagnosed at the middle and advanced stage. The five-year survival rate is only 68.4% based on retrospective data of 1,054 CRC patients who underwent radical resection (3). Therefore, early screening and diagnosis are effective to guide prognostication and treatment of CRC.

A large number of transcriptome data are utilized to identify candidate biomarkers of CRC, which offers the possibility for molecularly stratified treatment options for CRC patients (4, 5). Long non-coding RNA (lncRNA) is a kind of RNAs that is longer than 200nt and lacks protein-coding ability, and accumulating evidences have suggested that the alterations of lncRNAs expressions are responsible for the carcinogenesis of CRC, which may be available for CRC diagnosis and treatment (6, 7). In recent years, exosomes biomarkers in particular lncRNAs provide new insights into CRC development (8). Reportedly, lncRNA CCAL expressed by fibroblasts of tumor stroma may transfer to CRC cancer cells via exosomes, thereby suppressing cell apoptosis and promoting chemoresistance of CRC cells (9). CRC cell-derived exosomes can transport RPPH1 into macrophage to promote CRC cells metastasis and proliferation via mediating macrophage M2 polarization. Additionally, exosomal RPPH1 levels in blood plasma are higher in treatment-naive CRC

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patients and display a high diagnostic value (AUC = 0.86) (10). Moreover, the overexpression of lncRNA UCA1 is detected in the serum exosomes of CRC patients, which can transfer UCA1 to CRC cells to enhance the cell proliferation and migration of CRC through regulating the miR-143/MY06 axis (11). However, the research of exosomes in CRC progression via transferring lncRNAs is still limited, which may promote understanding of the pathogenesis of CRC and improve diagnosis and treatment.

LncRNA prostate cancer-associated transcript 1 (PCAT1) is a well-known oncogenic lncRNA located in chromosome 8 and is related to the development of various cancers, including CRC (12, 13). However, the presence of PCAT1 in exosomes and its mechanisms in CRC remain largely unknown. In this study, the expressions of lncRNA PCAT1 in plasma exosome and CRC tissues of CRC patients, as well as in supernatant exosomes of FHC, HT29 and HCT8 cells lines were evaluated using quantitative real-time PCR. In addition, the effects of lncRNA PCAT1 knockdown on adhesion and invasion of two CRC HT29 and HCT8 cell lines were evaluated. Then, the target of PCAT1 was predicted and validated using AGO2 RNA immunoprecipitation. Finally, the protein levels of epithelium-mesenchymal transition (EMT) related markers MTA2, MTA3, SNAI1, and E-cadherin in adjacent tissues, and cancer tissue of stage 0-I and stage II-III CRC patients, as well as in two CRC HT29 and HCT8 cell lines with stable ZNF217 knockdown were investigated. Our findings provide new insights into the molecular mechanism of exosomes PCAT1 on CRC research.

Materials and methods

Chemicals

A total of 50 normal participants who took the routine physical examination, 50 CRC patients at stage 0-I, 50 CRC patients at stage II-III who experienced tumor metastasis or relapse to receive treatment at Shanghai Xuhui District Central Hospital between September 2017 and January 2020 were enrolled in this study. The mean age of participants was 55 aged from 41-70. The study was approved by the ethical committee of Shanghai Xuhui District Central Hospital, and written informed consent was obtained from all participants.

Cell and cell culture

The intestinal epithelial cell line FHC and two CRC cell lines, HT29 and HCT8 were purchased from the Chinese Academy of Sciences (Shanghai, China). All cell lines were grown in DMEM medium (Sigma-Aldrich) added with 10% fetal bovine serum (FBS) and 1% penicillin/streptomycin (100U/ml, J&S information technology, Suzhou, China) at 37˚C in a humidified incubator with 5% CO2.

Exosomes isolation

The serum exosomes from whole blood and supernatant exosomes from cells were isolated, respectively. Prior to isolation of serum exosomes, 10 ml of venous whole blood of each participant was collected and allowed to clot by standing at room temperature for 30 min. Then, the samples were centrifuged at 2000× g at 4 °C for 10 min, followed by second centrifugation at 3000× g at 4 °C for 10 min. Serum exosomes were extracted by the Exosome precipitation kit (System Biosciences, CA, USA). Briefly, a total of 126 μl ExoQuick Exosome precipitation solution was added in 500 μl supernatant, and the mixture was centrifuged at 1500× g at 4 °C for 10 min after leaving it for 30 min at 4 °C. The supernatant exosomes from cells were also isolated by the same kit. Differently, the supernatant of cells was centrifuged at 3000× g at 4 °C for 10 min to remove cells and cell debris, followed by second centrifugation at 3000× g at 4 °C for 20 min. The remained supernatant (200 μl) was mixed with 1 ml ExoQuick Exosome precipitation solution, followed by incubation overnight at 4°C. The mixture was then centrifuged at 1500× g for 30 min at 4 °C. Finally, the exosome precipitation was obtained after centrifugation at 1500× g at 4 °C for 30 min.

Transmission electron microscopy (TEM)

In order to determine the morphology of exosomes, the exosomes were observed by TEM. In brief, exosomes were suspended in 1mL PBS and then 100 KD ultrafiltration centrifugation was conducted at 5000g for 30 min. Following washing three times by PBS, 20 μL suspension was added in a carbon-coated copper grid. After dried at room temperature, the preparations were dyed with phosphotungstic acid and observed with a transmission electron microscope (Tecnai G2 Spirit Bio TWIN, FEI, USA).

Construction of PCAT1 and ZNF217 shRNA vectors and cell transfection

The PCAT1 shRNA targeting 5 –GAAC CTAACTGGACTTTAA-3, ZNF217 shRNA targeting 5'-GCTCACGCCTGTAATCTCA-3' and negative control (NC) shRNA plasmid targeting 5'- TT CTCCGAACGTGTCACGT-3' were constructed (Sangon Biotech) and ligated to the pcDNA3.1 plasmid to construct pcDNA3.1-shPCAT1 and pcDNA3.1-shZNF217 vectors, respectively. For transfection, 1 ×105 cells were plated in six-well plates and transfected with different vectors using Lipofectamine 2000. The cells with stable PCAT1 and ZNF217 knockdown were selected for a future experiment.

Cell adhesion assay

Cell adhesion was measured using a cell-matrix adhesion assay. In brief, about 100 μl cells suspension at a concentration of 5×10-5 were plated on fibronectin pretreated 96-well plate (15μl/well) in duplicate and incubated for 1 h at 37°C. After incubation, cells were washed twice by PBS to remove the non-adhesion cells, and 100 μl new DMEM culture solution containing 0.1% BSA and 20 μl MTT (5mg/ml) was added to each well for continue culture 4h. Then, the crystal was dissolved in DMSO after vibrating for 10 mins. Finally, the count of adherent cells was measured at 490 nm using a microplate reader and was photographed using a microscope at 200 × magnification.

Cell transwell assays

A total of 1 × 105 cells were suspended in 400 μl DMEM medium without FBS and seeded on the top chamber of 24-well plate Transwell inserts, while the lower chambers were added with 600 μl DMEM contained with 20% FBS. Following incubation for 24 h, cells in the upper chamber were removed with cotton swabs. Then, the invaded cells were fixed with methanol and stained with crystal violet. The counts of cells were analyzed by Image J from three random fields captured under a light microscope (200 ×).

RNA immune precipitation (RIP) analysis

Based on our bioinformatics prediction, about 1 × 107 cells (400 μl) were harvested after centrifuging at 1500 rpm for 5 min at 4 °C and then lysed in 400 μl lysis buffer. The supernatant was removed prior to adding a 50 μl magnetic bead. The gathered bead was suspended with 100 μl RIP wash buffer followed by incubating with 1 μl ago2 and control IgG antibodies for 30 min at room temperature, respectively. Then, samples were centrifuged at 14000 rpm for 10 min at 4 °C, and the precleared lysates were incubated with RIP bead antibody complex for 3 h at 4 °C. Next, the mixture that containing 117 μl RIP wash buffer, 15 μl 10% SDS and 10 μl proteinase K was added and incubated for 30 min at 55 °C. Following the RNA-protein complex eluted with RIP wash buffer, RNA was extracted with TRIzol reagent (Invitrogen). For quantitative real-time PCR (qRT-PCR), the steps were the same as the following experiments. GAPDH was used as an internal control. The primers for ZNF217 and GAPDH were designed as: ZNF217: primer F, 5′- ATTCTCCAAATGTGCTGACTGTTC -3' and primer R, 5′- TGCCATGCTGTTAGATAAGTGTTG -3′. GAPDH: primer F, 5' CACCCACTCCTC CACCTTTG 3' and primer R, 5' CCACCACCCTG TTGCTGTAG 3'.

qRT-PCR analysis

Total RNA was isolated from serum exosomes of 50 normal participants, 50 stage 0-I CRC patients, 50 stage II-III CRC patients, and supernatant exosomes of FHC, HT29 and HCT8 cell lines using the SeraMir Exosome RNA Purification Column kit according to manufacturer's instructions, respectively. In addition, the tissue homogenate of 10 adjacent tissue samples, 20 stages 0-I and 20 stage II-III CRC tissue samples were prepared, and the extraction of total RNA from each tissue homogenate was conducted using TRIzol reagent (Invitrogen). Then, the total RNA was reversed using PrimeScript RT reagent Kit (Takara, Japan). SYBR Premix Ex Taq Kit (Takara, Japan) was used to conduct the real-time PCR. GAPDH was used as an internal control. The primers for PCAT1 were as follows. PCAT1: primer F, 5′-ACAGGGAGAAA GGAGATGAC-3' and primer R, 5′- TGGTGAT GTTGCGGTTTG -3′.

Western Blotting

Serum exosomes of each cell group and tissue homogenate of adjacent tissue and stage 0-I and stage II-III CRC samples were lysed in RIPA buffer, and the protein concentration was determined using BCA

Protein Assay kit. Briefly, about 30 μg proteins lysates of exosomes were separated by 12% SDS-PAGE gels and transferred to PVDF membranes. Subsequently, the membranes were incubated overnight at 4 °C with Rabbit polyclonal MTA2 (1:3000, 17554-1-AP, Proteintech), MTA3 (1:1000, 14682-1-AP, Proteintech), SNAI1 (1:5000, 20874-1-AP, Proteintech), and mouse anti-human E-cadherin (1:2000, 60335-1-Ig, Proteintech) and beta-actin as internal reference (1:8000, 60008-1-Ig, Proteintech). The HRP-conjugated secondary antibodies were used for immunostaining and blots were revealed by enhanced chemiluminescence (ECL).

Statistical analysis

All continuous data were expressed as mean ± SD. Comparisons between two groups were conducted using Student's t-test. One-way ANOVA followed by a post hoc Bonferroni test was applied for multiple comparisons. P-value < 0.05 was considered statistically significant. All statistical analyses were performed using SPSS 21.0 software.

Results and discussion

lncRNA-PCAT1 is upregulated in CRC tissues and exosomes derived from CRC patients and cell lines

Exosomes derived from the plasma of early-stage CRC patients were isolated and identified. The results of TEM revealed that the particles had round-shaped vesicles (Fig. 1a) and diameters of about 50 nm sizes of exosomes from early-stage CRC patients (Fig. 1b). Then, to detect the expression of exosomal lncRNA PCAT1, exosomal RNA derived from plasma of stage 0-I, stage II-III CRC patients and normal controls, as well as from normal colonic enterocyte FHC, and two CRC cell lines HT29 and HCT8 was extracted. As a result, the expression of plasma exosomal lncRNA PCAT1 was significantly upregulated in stage 0-I and stage II-III CRC patients than in normal controls and increased in stage II-III CRC patients compared with stage 0-I patients (P < 0.01, Fig. 2a), which was consistent with results in CRC tissues, showing that lncRNA PCAT1 expression was remarkably enhanced in CRC cancer tissues than that in adjacent tissues of stage 0-I CRC and stage II-III CRC patients (Fig. 2b). Similarly, there was a significant increase in CRC HT29 and HCT8 cell lines than in normal enterocyte FHC cell lines (P < 0.01, Fig. 2c).

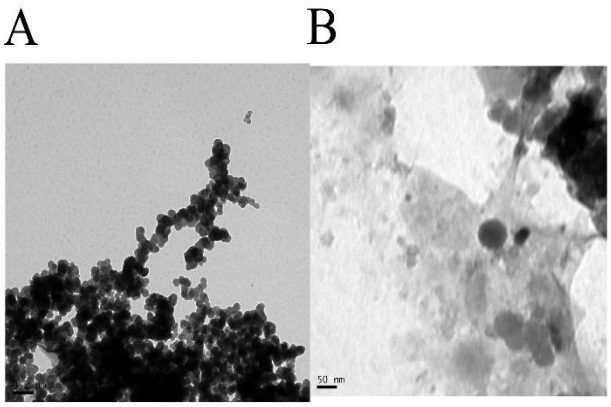

Figure 1. Transmission electron microscopy images of plasma exosomes of CRC patients at scales of 0.2 μm (A) and 50 nm (B).

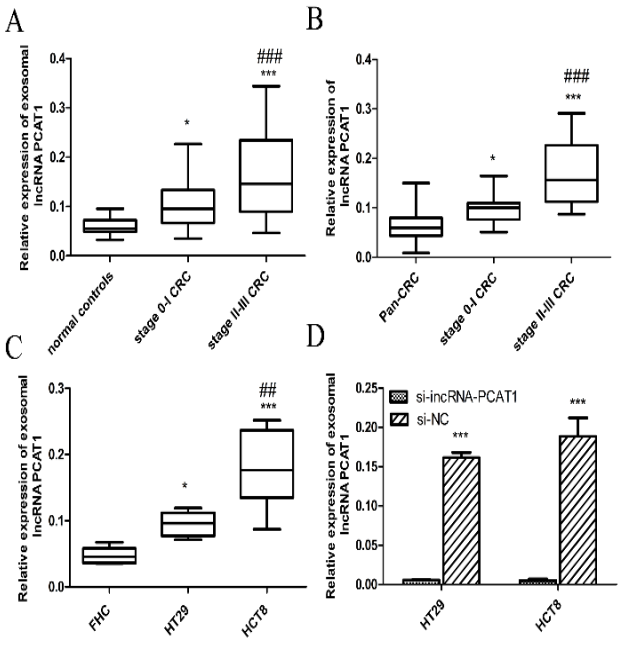

Figure 2. The expression of lncRNA PCAT1 in tissues and cell lines. (a) The overexpression of plasma exosomal lncRNA PCAT1 in stage 0-I and stage II-III CRC patients than in normal controls. (b) lncRNA PCAT1 is upregulated in cancer tissues from stage 0-I and stage II-III CRC. (c) The overexpression of supernatant exosomal lncRNA PCAT1 in CRC HT29 and HCT8 cell lines than in FHC cell line. (d) si-PCAT1 downregulated the expression of lncRNA PCAT1 in exosomes derived from HT29 and HCT8. * p < 0 05 and ***p < 0 001 vs normal controls, FHC cell line, or si-NC. ## p < 0 01, and ### p < 0 001 vs stage 0-I CRC or HT29 cell line.

lncRNA PCAT1 promotes CRC adhesion and invasion

The effects of lncRNA PCAT1 on adhesion and invasion of CRC cell lines were evaluated. At first, the adhesion and invasion abilities of two CRC cell lines HT29 and HCT8 and FHC control cell lines were detected. As expected, the adhesion and invasion abilities of two CRC cell lines HT29 and HCT8 were stronger than the FHC cell line (Fig. 3a). Given that the expression of lncRNA PCAT1 was higher in HT29 and HCT8 cell lines than in FHC cells, we wondered whether lncRNA PCAT1 could influence CRC adhesion and invasion. Then, PCAT1 knockdown in two CRC cell lines was performed. RT-PCT analysis showed that the expression of PCAT1 was significantly reduced and the knockdown efficiency was well (Fig. 2d). In addition, PCAT1 knockdown significantly decreased the adhesion and invasion of HT29 and HCT8 cells Fig. 3b).

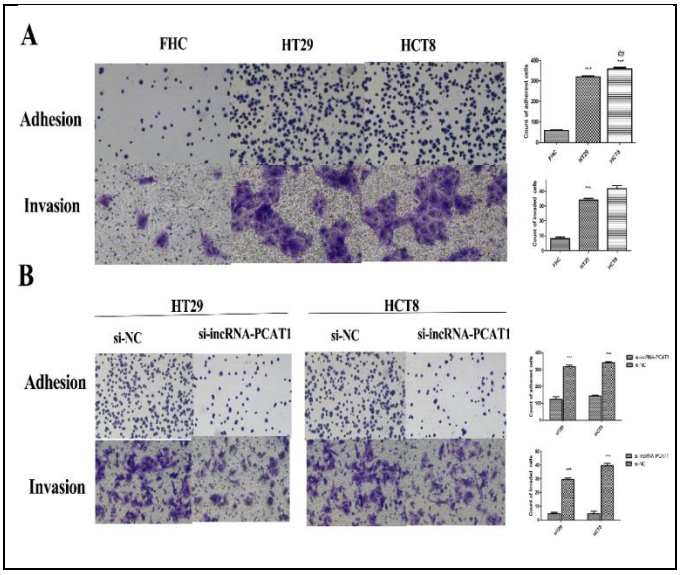

Figure 3. The adhesion and invasion abilities of three cell lines. (a) The adhesion and invasion abilities of two CRC cell lines HT29 and HCT8 are stronger than FHC cell line. (b) PCAT1 knockdown represses the adhesion and invasion of HT29 and HCT8 cells. ***p < 0 001 vs FHC cell line. # p < 0 05 and ## p < 0 05 vs HT29 cell line.

ZNF217 is a potential target of lncRNA PCAT1

Based on the bioinformatic prediction, ZNF217 TF was a potential target of lncRNA PCAT1. To further verify the lncRNA PCAT1-ZNF217 interaction, a RIP assay was conducted. The results showed that the level of ZNF217 was significantly increased in lncRNA PCAT1 immunoprecipitates in stage 0-I CRC and stage II-III CRC tissues samples compared to adjacent tissues, while no ZNF217 expression was detected in control IgG immunoprecipitates (Fig. 4a), demonstrating that lncRNA PCAT1 could target ZNF217.

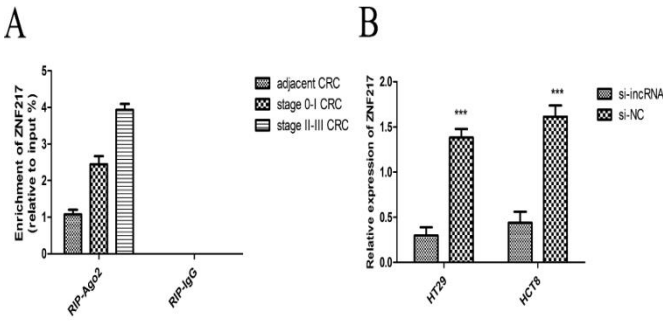

Figure 4. The lncRNA PCAT1-ZNF217 interaction determined by RIP assays and qPCR. (a) ZNF217 RNA levels in immunoprecipitates with ago2 antibodies. The expression level of ZNF217 is presented as fold enrichment relative to IgG. (b) Effects of lncRNA PCAT1 knockdown on ZNF217 expression level. ***p < 0 001.

Effects of lncRNA PCAT1 on the ZNF217 expression level

The influence of lncRNA PCAT1 on ZNF217 expression level was future investigated. The result showed that downregulation of lncRNA PCAT1 could inhibit the expression levels of ZNF217 in both HT29 and HCT8 cells lines (Fig. 4b).

Effects of ZNF217 knockdown on MTA2, MTA3, SNAI1 and E-cadherin proteins expression levels

As lncRNA PCAT1-ZNF217 interaction was validated above, and the downstream molecular mechanisms underlying the tumorigenesis of the PCAT1-ZNF217 complex were further explored. Based on STRING database prediction, ZNF217, MTA2 and MTA3 had interactions with each other. Based on publications, MTA family members may regulate the downstream SNAI1 and E-cadherin expressions in human cancers (14). Thus, the proteins expressions of MTA2, MTA3, SNAI1 and E-cadherin in stage 0-I, stage II-III CRC tissues samples and cell lines were investigated and ZNF217 knockdown in two CRC cells lines was performed to explore the

expressions changes of these molecules. We found that the protein expressions of MTA2, MTA3, and E-cadherin were significantly higher in stage 0-I and stage II-III CRC tissues samples than that in adjacent tissues (P< 0.001, Fig.5a). On the contrary, the expression of SNAI1 was significantly reduced in stage 0-I and stage II-III CRC tissues samples (P< 0.001, Fig.5a). The expression trends of these proteins were consistent in CRC cells than in FHC cells (Fig.5b). Subsequently results revealed that ZNF217 knockdown significantly decreased MTA2, MTA3, and E-cadherin expressions, and increased SNAI1 expressions in both HT29 (P< 0.05, Fig. 6a) and HCT8 (P< 0.05, Fig. 6b) cells, supporting that MTA2/MTA3 was regulated by ZNF217.

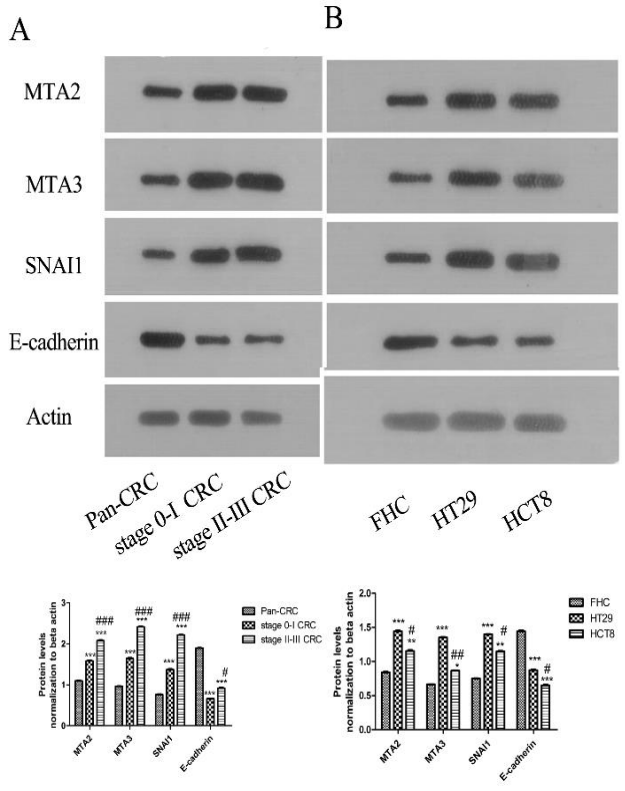

Figure 5. The protein expressions of MTA2, MTA3, SNAI1 and E-cadherin in CRC tissue and cell lines. (a) The upregulation of MTA2, MTA3, and SNAI1, and downregulation of E-cadherin in stage 0-I and stage II-III CRC tissues samples. (b) The upregulation of MTA2, MTA3, and SNAI1, and downregulation of E-cadherin in both HT29 and HCT8 cells. * p < 0 05, * p < 0 01 and ***p < 0 001 vs paracancerous CRC or FHC cell line. # p < 0 01, ## p < 0 01, and ### p < 0 001 vs stage 0-I CRC or HT29 cell line.

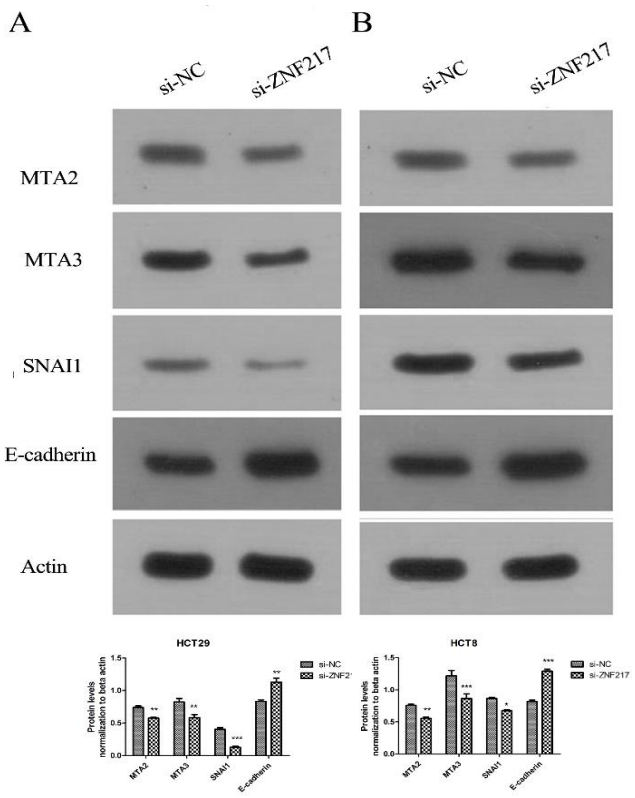

Figure 6. ZNF217 knockdown reduces MTA2, MTA3, and E-cadherin proteins expressions, and increases SNAI1 expression in both HT29 (a) and HCT8 (b) cells. * p < 0 05, * p < 0 01 and ***p < 0 001 vs si-NC.

In recent years, exosomes secreted by various cell types are considered indispensable factors in carcinogenesis and progression of cancers (15). LncRNAs are an emerging group that can be packaged into exosomes to participate in the development of cancer in a variety of ways (16). However, a deeper understanding of the underlying mechanisms of exosomal lncRNAs related to the onset of cancer is still limited. In this study, the plasma exosomal lncRNA PCAT1 was found to be significantly increased in the CRC tissues and cell lines. In addition, lncRNA PCAT1 knockdown significantly inhibited the adhesion and invasion of HT29 and HCT8 cells. Subsequent RIP assay results indicated that lncRNA PCAT1 could target ZNF217, and downregulation of lncRNA PCAT1 could decrease the protein expression level of ZNF217 in both HT29 and HCT8 cells lines. Moreover, ZNF217 knockdown significantly decreased MTA2, MTA3, and SNAI1 expressions, but increased E-cadherin expressions in both HT29 and HCT8 cells lines,

supporting that MTA2/MTA3 was regulated by ZNF217.

An increasing number of research reveals the overexpression of lncRNA PCAT1 can promote cell proliferation, migration and invasion in CRC cells, and suppress cell cycle arrest and apoptosis of CRC cells (17, 18). Especially, PCAT1 knockdown suppressed the drug resistance and reduced the invasiveness of two CRC cell lines Caco-2 and HT-29 via regulating c-Myc expression (19). In this study, we focused on the underlying mechanisms of exosomal lncRNA PCAT1 on CRC tumors and cell lines. We found that lncRNA PCAT1 was upregulated in CRC tissue and cell lines-derived exosomes, which was consistent with the expressions of lncRNA PCAT1 in CRC tumors or cell lines. In addition, Huang et al have reported that PCAT1 is higher in oesophageal squamous cell carcinoma (ESCC) cell-derived exosomes compared to that in normal controls, and may promote cell growth by sponging miR-326 mediated through exosomes (20). Moreover, Huang et al also demonstrate elevated PCAT1 expression in the serum exosomes of advanced ESCC patients than those from stage 1 ESCC patients (20), which was similar to our results in different stages of CRC patients.

Subsequently, our findings revealed that downregulation of lncRNA PCAT1 could decrease the ZNF217 expression in both HT29 and HCT8 cells lines. Zinc Finger Protein 217 (ZNF217) as a DNA-binding TF, is discovered as an oncogenic gene to associate with the development and progression of various cancers such as breast cancer and hepatocellular cancer (21, 22). Reportedly, the expression of ZNF217 is significantly elevated in CRC tissues than in adjacent tissues and ZNF217 knockdown in CRC cells may suppress cell migration and invasion (23). As we know, the lncRNA PCAT1-ZNF217 interaction was first studied in our study. In the consideration of both higher expressions and similar functions in CRC cells, we supposed that lncRNA PCAT1 might coordinate ZNF217 to be responsible for CRC progression. Based on bioinformatic prediction and literature search, several genes (MTA2, MTA3, SNAI1 and E-cadherin) that directly or indirectly related to ZNF217 were future investigated to explore its future mechanisms in CRC cells. Metastasis Associated 1 Family Member 2 and 3 (MTA2 and MTA3) belong to MTA family members, which are components of the nucleosome remodeling and deacetylation (NuRD) complex and directly or indirectly associated with transcriptional regulation (14). Although no study has reported the interactions between ZNF217 and MTA2/MTA3, it has been proved that the NuRD complex can interact with several substoichiometric zinc finger-containing proteins, including ZNF512, ZNF592, and ZNF68 (24). The study suggests an abnormally high expression of MTA2 in CRC tissue and MTA2 is associated with prognosis CRC (25). Additionally, Jiao et al have revealed MTA3 overexpression contributes to cell proliferation, invasion, migration, and cell cycle progression of CRC cells (26). Combined with our findings that ZNF217 knockdown could significantly reduce the expressions of MTA2 and MTA3, we inferred that ZNF217 might contribute to CRC carcinogenesis and development by combining its zinc finger domain to the NuRD complex region of MTA2/MTA3. What's more, it has been reported E-cadherin expression is inhibited by direct binding of ZNF217, which may promote cancer cell migration and invasion (27). Whereas, another mechanism is mentioned, suggesting that MTA2 is involved in pancreatic carcinoma progression through suppressing E-cadherin (28). Interestingly, it has been found that MTA1 can enhance Snai1 and Slug expressions, and silencing of MTA1 may reduce Snai1 and Slug recruitment to the promoter of E-cadherin, thereby leading to a decrease of E-cadherin expression in CRC cells (29). Although it has been reported MTA3 as a repressor of the E-cadherin transcription factor Snail, the major regulator of EMT, and subsequently represses cancer cell invasion and migration. Conversely, MTA can function as oncogene-related properties similarly as MTA1 and MTA2 (30). Notably, ZNF217 is reported to promote EMT in human mammary epithelial cells (31), and PCAT1 is involved in the EMT process in human cancers (32). Taken together, our findings suggested that lncRNA PCAT1 might coordinate ZNF217 to be responsible for CRC progression through regulating MTA2/MTA3/Snai1/E-cadherin, which is implicated in EMT. In this regard, more research is needed to obtain new findings in this area (33-36).

Conclusion

Overall, our results suggest exosomal lncRNA PCAT1 can promote the adhesion and invasion of CRC cells, and the overexpression of PCAT1 leads to upregulation of ZNF217 which regulates EMT-related MTA2/MTA3/Snai1/E-cadherin signaling.

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Conflicts of interest

There are no conflicts of interest to declare.

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