Article

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PCGF2 Acts as an Oncogenic Driver in Colon Cancer through the Upregulation of CENPE

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Background: Colon cancer (CC) is a highly prevalent malignancy that contributes significantly to global morbidity and mortality. The polycomb group ring finger 2 (PCGF2) has been identified as a relevant factor influencing the outcomes of CC. At the same time, the centromere-associated protein E (CENPE) is implicated in promoting carcinogenesis and adversely affecting the survival of tumor patients. The primary objective of this study was to elucidate the precise impact of PCGF2 on CC and unravel the underlying mechanisms associated with CENPE.

Methods: Human normal colon epithelial cells and CC cells were utilized to investigate the differential expression of PCGF2 and CENPE. CC cell line LOVO was exploited and transfected for PCGF2 regulation. Subsequently, cell viability and proliferation were assessed using the cell counting kit 8 (CCK-8) and colony forming assay. Cell viability and proliferation were assessed using the terminal deoxynucleotidyl transferase (TdT) dUTP nick-end labeling (TUNEL) assay, while cell migration and invasion capabilities were determined using the transwell assay, and mRNA levels of cell cycle-related genes were measured for evaluating cell cycle activation. In addition, mice were used for *in vivo* experiments to investigate the progression of CC cells with different levels of PCGF2. Moreover, GSK-923295 was used to inhibit CENPE, followed by the evaluation of cell progression.

Results: PCGF2 and CENPE were upregulated in CC cell lines ($p < 0.001$), and upregulation/downregulation of PCGF2 led to the upregulation and downregulation of CENPE ($p < 0.001$). The upregulation/downregulation of PCGF2 led to an increase/decrease in viability, proliferation, migration, and invasion while suppressing/enhancing apoptosis in LOVO cells ($p < 0.001$), promoting cell progression. The tumor progression of LOVO cells with PCGF2 knockdown was slower ($p < 0.001$). The PCGF2-promoting LOVO cell progression was disrupted when CENPE was inhibited, presented by the reversely decreased viability, proliferation, migration, invasion, and cell cycle activation, and increased apoptosis ($p < 0.001$).

Conclusion: PCGF2 promotes CC cell progression by upregulating CENPE, providing PCGF2 inhibition and CENPE inhibition as potential therapeutic targets for treating CC.

Keywords: colon cancer; PCGF2; CENPE

Introduction

Among all diseases in the world, cancer is the second most fatal one [1]. In 2021, it has been reported that there were 1,898,160 cases of cancers and 608,570 deaths related to cancers [2]. Among all cancer types, colon cancer (CC) is one of the most prevalent cancers and significantly contributes to morbidity and mortality across the globe [3]. Despite the stipulated options of diagnosis and treatments, the prognosis of patients with CC is not always satisfactory, and the death number remains high [1]. Therefore, novel and viable therapy alternatives are urgently needed to treat CC and improve the quality of life of patients.

It has been observed that genes differently expressed are associated with tumor initiation and progression [4]. Moreover, CC is a multifaceted disease caused by the numerous genetic and epigenetic changes aggregated in the colon at the cellular level [5]. Therefore, molecular alterations and underlying pathways could be potential therapeutic targets for treating CC. The polycomb group ring finger 2 (PCGF2) has been proven to act as a driver. It represents a potential therapeutic target in various types of cancers and tumors, including melanoma [6], breast cancer [7,8], prostate cancer [9] and head and neck squamous cell carcinoma [10]. Moreover, PCGF2 has also been identified as closely related to the prognosis of CC [11], indicating that PCGF2 could potentially serve for CC therapies. As the precise impact of PCGF2 on CC and its underlying mechanisms remain elusive, the present study conducted experiments to understand PCGF2 functions in CC better.

Additionally, it has been previously reported that the cell-cycle-related centromere-associated protein E (CENPE), which showed carcinogenic effects in tumors, was upregulated in almost all tumors, was related to worse patient survival, and was able to act as an oncogene in various cancers and as a promising biomarker for both can-

cer diagnosis and prognosis [12]. Especially, CENPE was found to be significantly upregulated in human colon cancer and showed a remarkable prognostic value in CC [12,13], demonstrating its critical role in CC.

In the current investigation, we exploited CC cells to scrutinize the impact of PDGF2 on the progression of colon cancer. Moreover, we delved into the role of cell cycle- and cancer-related CENPE in the context of PDGF2 functions in CC. The results of this study provided potential therapeutic strategies based on PDGF2 and CENPE for treating CC.

Materials and Methods

Cell Culture

All cell lines with short tandem repeat (STR) authentications were purchased from iCell Bioscience Inc. (Shanghai, China), including human normal colon epithelial cells NCM460 (iCell-h373) and CC cell lines of LOVO (iCell-h126), HCT-116 (iCell-h071), HT-29 (iCell-h078) and Caco-2 (iCell-h032). Cells were cultured in Roswell Park Memorial Institute (RPMI)-1640 (iCell-0002, iCell Bioscience Inc., Shanghai, China), containing 10% fetal bovine serum (FBS; iCell-0002, iCell Bioscience Inc., Shanghai, China) and 1% antibiotics (iCell-15140-122, iCell Bioscience Inc., Shanghai, China) under the condition of 37 °C, 5% CO_2 and 70–80% humidity. The mycoplasma test was performed to avoid cell contamination before cells were used. When inhibiting CENPE, GSK-923295 (15 nM; HY-10299, MedCHemExpress, Monmouth Junction, NJ, USA) was added to the cell culturing medium.

Cell Transfection

All lentiviral plasmids were constructed by GeneChem (Shanghai, China), including the PCGF2 overexpression plasmid pCMV-tag5a-PCGF2 (OE-PCGF2; inserted by the full-length of human *PCGF2* DNA, ENSG00000277258) and blank plasmid (OE-NC) for upregulating PCGF2, and plasmids inserted by the short hairpin (sh)RNA against PCGF2 (sh-PCGF2; 5′-CACCGCCTCTCCATCGAATTCTACGCGAACGTAGAATTCGATGGAGAGGC-3′) as well as plasmid inserted by the negative control shRNA (sh-NC; 5′-CACCGCACTGATTTCAAATGGTGCTATTCGAAAATAGCACCATTTGAAATCAGTG-3′) for downregulating PCGF2. When cells reached a confluence of 70%, they were mixed with plasmids (2 µg) and Lipofectamine™ 3000 reagent (L3000001, Invitrogen, Carlsbad, CA, USA) and incubated for 24 hours. The transfection efficiencies were evaluated by determining PCGF2 mRNA and protein expression levels.

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

Total RNA was isolated using a TRIzol Kit (DP424, Tiangen, Beijing, China) and reversely transcribed into cDNA using the cDNA Synthesis Kit (KR116, Tiangen, Beijing, China). The qRT-PCR reaction mixture was prepared using the SYBR Green qRT-PCR Kit (D7268S, Beyotime, Shanghai, China), and the reaction was performed in a LightCycler96 thermocycler (Roche, Shanghai, China). The mRNA expression levels were processed by the $2^{-\Delta\Delta CT}$ method. Primer sequences are as follows: *PCGF2* (forward 5′-GTGTGAGTCAGTCAGCGACA-3′, reverse 5′-CAGTGGCCCATCCAGTTCAT-3′), *CENPE* (forward 5′-ATGCAGGAGCAGAGAGTGTG-3′, reverse 5′-TTGCACTCAGGCACATCCTT-3′), cyclin-dependent kinase 2 (*CDK2*; forward 5′-GGCCTTGGGCTATTTGGACT-3′, reverse 5′-TAGGAGGTGGACGTCAGAGG-3′), cyclin-dependent kinase 4 (*CDK4*; forward 5′-GCCTCGAGATGTATCCCTGC-3′, reverse 5′-TCCCGACTCCTCCATCTCAG-3′), Cyclin D3 (forward 5′-CATGTACCCGCCATCCATGA-3′, reverse 5′-CAGCTCTGTGAGCTCATCCC-3′), glyceraldehyde-3-phosphate dehydrogenase (*GAPDH*; forward 5′-ATGACCACAGTCCATGCCATCAC-3′, reverse 5′-AGGTCCACCACCCTGTTGCTGTA-3′).

Western Blot

Total proteins in cells were extracted by the Radio Immunoprecipitation Assay (RIPA) (P0013, Beyotime, Shanghai, China) and were separated on 10% SDS-PAGE gel (P0012, Beyotime, Shanghai, China). After proteins were transferred to membranes (IPVH15150, Millipore Sigma, Billerica, MA, USA), they were incubated at 4 °C with primary antibodies for PCGF2 (1:1000 dilution, Cat. No. ab5267, Abcam, Cambridge, MA, USA), CENPE (1:1000 dilution, Cat. No. ab264251, Abcam, Cambridge, MA, USA), and GAPDH (1:1000 dilution, Cat. No. ab8245, Abcam, Cambridge, MA, USA). After overnight incubation, membranes were incubated with the horseradish-peroxidase (HRP) goat anti-mouse IgG secondary antibody (1:1000 dilution, Cat. No. A0216, Beyotime, Shanghai, China). Finally, proteins were visualized using an enhanced chemiluminescence kit (P0018S, Beyotime, Shanghai, China) and photographed by the imaging system (ChemiDoc XRS+, Bio-Rad, Shanghai, China). The gray values were analyzed using ImageJ software (1.48, National Institutes of Health, Rockville, MD, USA).

Cell Counting Kit 8 (CCK-8) Assay

Cells (2.5×10^3 cells/mL) were seeded into 96-well plates and incubated for 24 h. Afterward, a CCK-8 solution (10 µL; G4103-1ML, Servicebio, Wuhan, China) was added to the cells, followed by a 2-h incubation. Finally, the absorbance (450 nm) was measured by a microplate reader (DR-200Bs, Diatek, Jiangsu, China).

Colony Forming Assay

Cells were digested by trypsin (T1300, Solarbio, Beijing, China) and were added into 24-well plates (1500

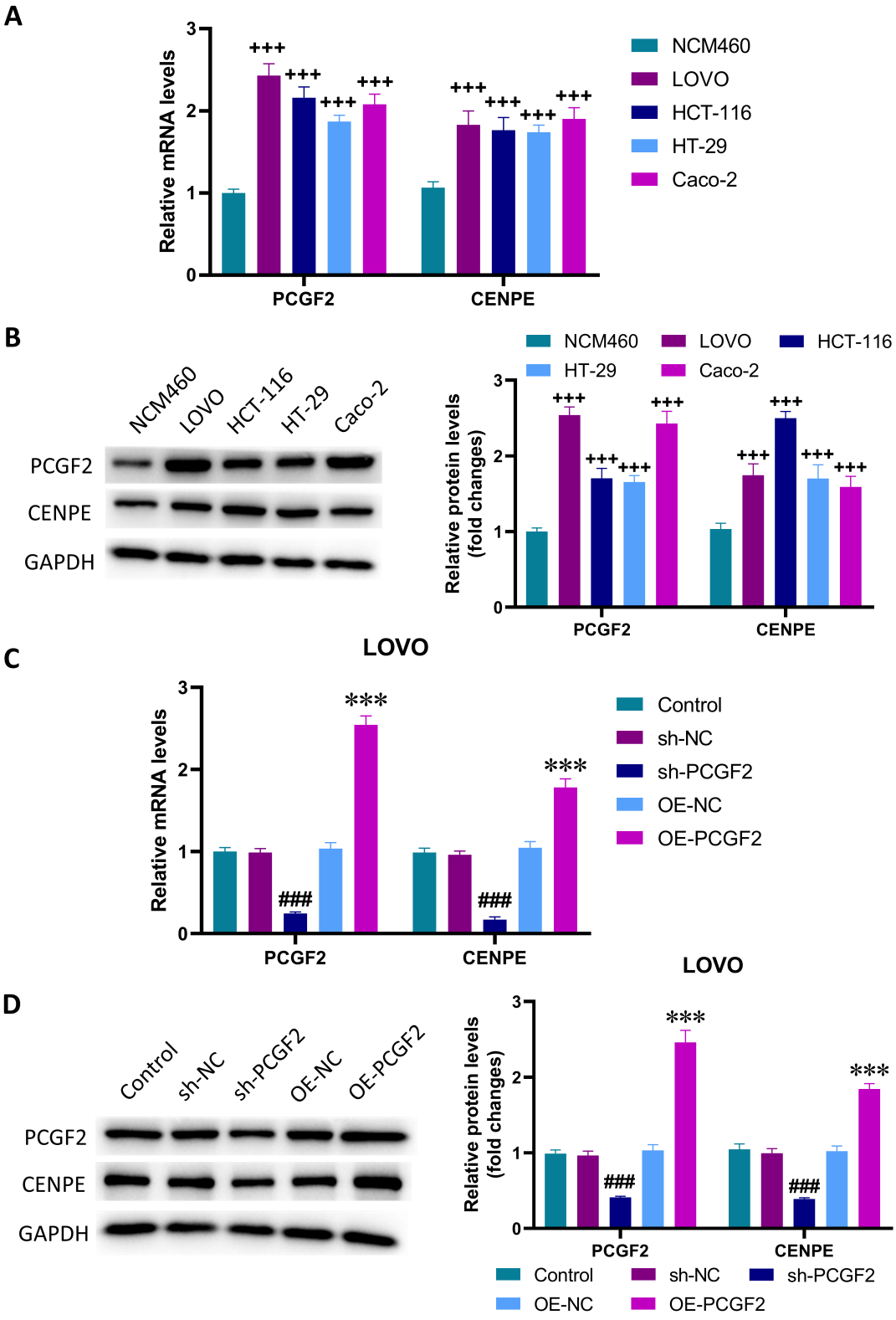

Fig. 1. Polycomb group ring finger 2 (PCGF2) was overexpressed in colon cancer (CC) cells and positively regulated cell-cycle-related centromere-associated protein E (CENPE) expression. (A) mRNA and (B) protein expressions of PCGF2 and CENPE in normal colon epithelial cells (NCM460) and in CC cell lines (LOVO, HCT-116, HT-29, Caco-2). (C) mRNA and (D) protein levels of PCGF2 and CENPE in LOVO cells after transfection. N = 5. +++$p < 0.001$ vs. NCM460; ###$p < 0.001$ vs. sh-NC; ***$p < 0.001$ vs. OE-NC.

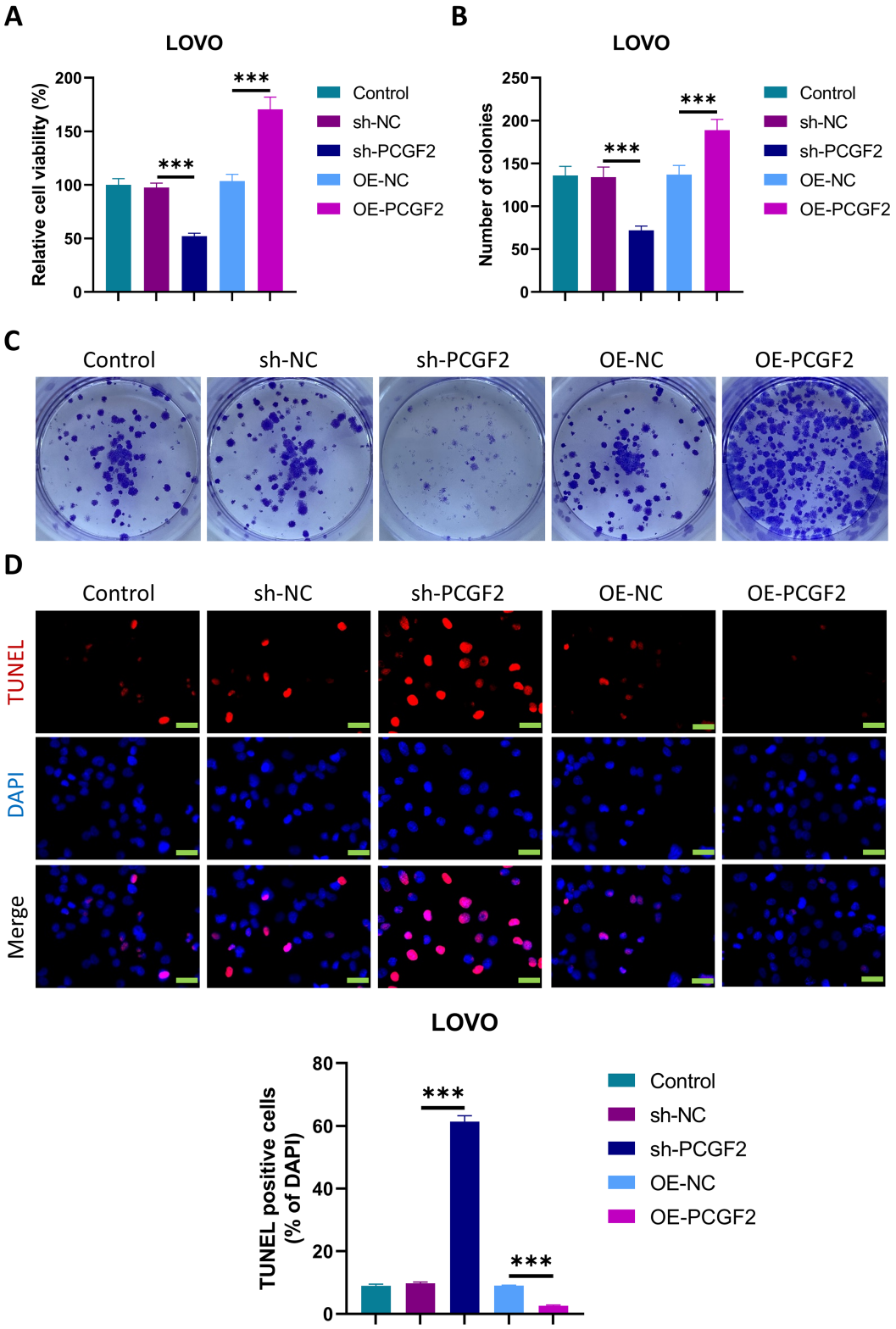

Fig. 2. PCGF2 promoted the proliferation while suppressing apoptosis of CC cells. (A) Cell counting kit 8 (CCK-8) assay results. (B) Quantification and (C) representative images of colony forming assay. (D) Terminal deoxynucleotidyl transferase (TdT) dUTP nick-end labeling (TUNEL) assay results (scale: 50 µm). N = 5. ***$p < 0.001$.

cells/well). After cells were incubated at 37 °C with 5% CO_2 for 14 days, they were fixed using 4% paraformaldehyde (P0099, Beyotime, Shanghai, China) for 30 min and stained by 0.1% crystal violet dye (G1064, Solarbio, Bei-

jing, China) for 30 min. Images of cell colonies were observed by microscopy (CKX53, OLYMPUS, Tokyo, Japan) and captured by the camera (SLR, Nikon, Tokyo, Japan).

Transwell Assay

The upper layer of the Transwell chamber (3422, Corning, NY, USA) was loaded with 300 µL serum-free medium containing 1×10^5 cells. In contrast, the lower layer was loaded with 500 µL medium containing 10% FBS (FCS500, EXcellBio Co., Ltd., Shanghai, China) to induce cell migration to the other side. After incubation for 24 h, cells were fixed using 4% paraformaldehyde (P0099, Beyotime, Shanghai, China) for 30 min and stained by 0.1% crystal violet dye (G1064, Solarbio, Beijing, China) for 30 min. Finally, the cells were examined using microscopy (CKX53, OLYMPUS, Tokyo, Japan). For the invasion assay, Matrigel (356234, BD Biosciences, Franklin Lakes, NJ, USA) was pre-coated on the upper layer, followed by a 2-h incubation. The rest of the procedures were the same as the migration assay.

Terminal Deoxynucleotidyl Transferase (TdT) dUTP Nick-End Labeling (TUNEL)

A TUNEL assay was used to evaluate cell apoptosis. After cells were digested with protease K (39450-01-6, Roche, Basel, Switzerland), they were treated with the TUNEL reagent (11684817910, Roche, Basel, Switzerland) at 4 °C overnight and subsequently stained by DAPI (C1002, Beyotime, Shanghai, China). Cell slides were sealed and observed using microscopy (CKX53, OLYMPUS, Tokyo, Japan).

Animal Experiments

A total of 20 BALB/c nude mice were purchased from Beijing Vital River Laboratory Animal Technology Co., Ltd. (Beijing, China) and maintained for 1 week at 37 °C, under standard 12-h light/dark cycles, with adequate food and water. Subsequently, mice were randomly divided into 2 groups, with 10 mice in each group. Mice in the sh-NC/sh-PCGF2 group were intraperitoneally injected at the left proximal tibia with a 10 µL mixture of sh-NC/sh-PCGF2 transfected LOVO cells and Matrigel (356234, BD Biosciences, Franklin Lakes, NJ, USA) (1.5×10^7 cells/mL). After mice were normally cultured for 28 days, they were euthanized with 1.5% pentobarbital sodium (160 mg/kg), and the tumors were collected. The mass of each tumor was measured, and the volume of each tumor was calculated by the formula of $V = 1/2 \times a \times b^2$, in which the a and b are the long and short diameters of the tumor, respectively. The study was approved by the Experimental Animal Ethics Committee of Ezhou Central Hospital (approval number: [2023]-02).

Statistical Analysis

Data were presented as mean ± standard deviation, and statistics were analyzed by GraphPad Prism 7.0 software (GraphPad Software Inc., San Diego, CA, USA). The group comparison analyses were performed using analysis of variance (ANOVA) with the Newman-Keuls post hoc tests. Statistical significance was considered when $p < 0.05$.

Results

PCGF2 Overexpression in CC Cells Positively Influenced the Expression of CENPE

As shown, PCGF2 exhibited higher expression in CC cell lines compared to normal colon epithelial cells (NCM460) at both mRNA (Fig. 1A, $p < 0.001$) and protein levels (Fig. 1B, $p < 0.001$). At the same time, CENPE was also upregulated in CC cells compared to NCM460 (mRNA: Fig. 1A, $p < 0.001$; protein: Fig. 1B, $p < 0.001$). These observations suggested an association of both PCGF2 and CENPE with CC. In addition, after the transfection for regulating PCGF2 expression in CC LOVO cells, sh-PCGF2-transfected LOVO cells showed downregulated PCGF2 compared to sh-NC-transfected LOVO cells (mRNA: Fig. 1C, $p < 0.001$; protein: Fig. 1D, $p < 0.001$). Conversely, OE-PCGF2-transfected LOVO cells exhibited upregulated PCGF2 compared to OE-NC-transfected LOVO cells (mRNA: Fig. 1C, $p < 0.001$; protein: Fig. 1D, $p < 0.001$), confirming the successful transfection. We also found that when PCGF2 was downregulated in sh-PCGF2 transfected LOVO cells, the CENPE was also downregulated (mRNA: Fig. 1C, $p < 0.001$; protein: Fig. 1D, $p < 0.001$), while upregulation of CENPE was observed in OE-PCGF2 transfected LOVO cells (mRNA: Fig. 1C, $p < 0.001$; protein: Fig. 1D, $p < 0.001$). These results suggested a positive regulatory role of PCGF2 in CENPE expression.

PCGF2 Enhanced the Proliferation and Inhibited Apoptosis of CC Cells

Results depicted in Fig. 2A revealed a decrease in cell viability of sh-PCGF2 transfected LOVO cells compared with sh-NC transfected LOVO cells ($p < 0.001$). In contrast, cell viability increased for OE-PCGF2 transfected LOVO cells compared to OE-NC transfected LOVO cells ($p < 0.001$), indicating that PCGF2 elevated the viability of CC cells. We also found that the sh-PCGF2 transfected LOVO cells formed fewer cell colonies compared to sh-NC transfected LOVO cells (Fig. 2B,C, $p < 0.001$), while OE-PCGF2 transfected LOVO cells formed more cell colonies compared to OE-NC transfected LOVO cells (Fig. 2B,C, $p < 0.001$), indicating that PCGF2 increased cell proliferation of CC cells. Moreover, the results from the TUNEL assay (Fig. 2D) demonstrated an increase in cell apoptosis of sh-PCGF2 transfected LOVO cells compared to sh-NC trans-

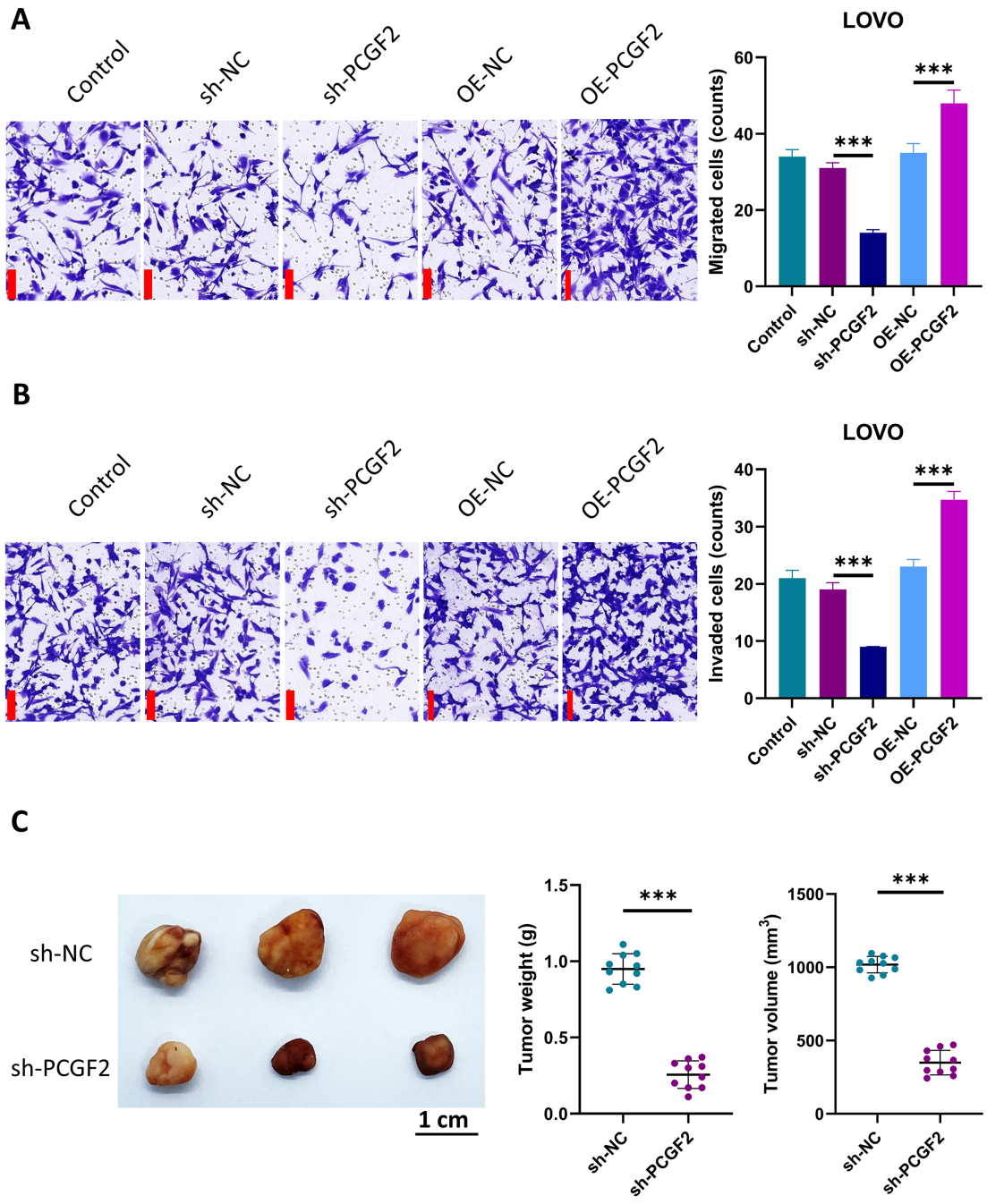

Fig. 3. PCGF2 promoted migration and invasion while preventing cell cycle arrest of CC cells. Results of transwell assays for (A) migration (N = 5) and (B) invasion (N = 5; scale: 50 µm). (C) Tumors progressed from differently transfected LOVO cells (N = 10). ***$p < 0.001$.

fected LOVO cells ($p < 0.001$). In contrast, cell apoptosis decreased for OE-PCGF2 transfected LOVO cells compared to OE-NC transfected LOVO cells ($p < 0.001$), indicating that PCGF2 suppressed apoptosis of CC cells. All these results demonstrated that PCGF2 could act as a CC driver and enhance the progression of CC cells by promoting cell growth and inhibiting cell death.

PCGF2 Promoted the Migration and Invasion, and PCGF2 Knockdown Suppressed CC Progression

The transwell assay for migration (Fig. 3A) and invasion (Fig. 3B) were performed. The results showed that the sh-PCGF2 transfected LOVO cells migrated less ($p < 0.001$) and invaded less ($p < 0.001$) compared to sh-NC transfected LOVO cells, while OE-PCGF2 transfected LOVO cells migrated more ($p < 0.001$) and invaded more ($p < 0.001$) compared to OE-NC transfected LOVO cells,

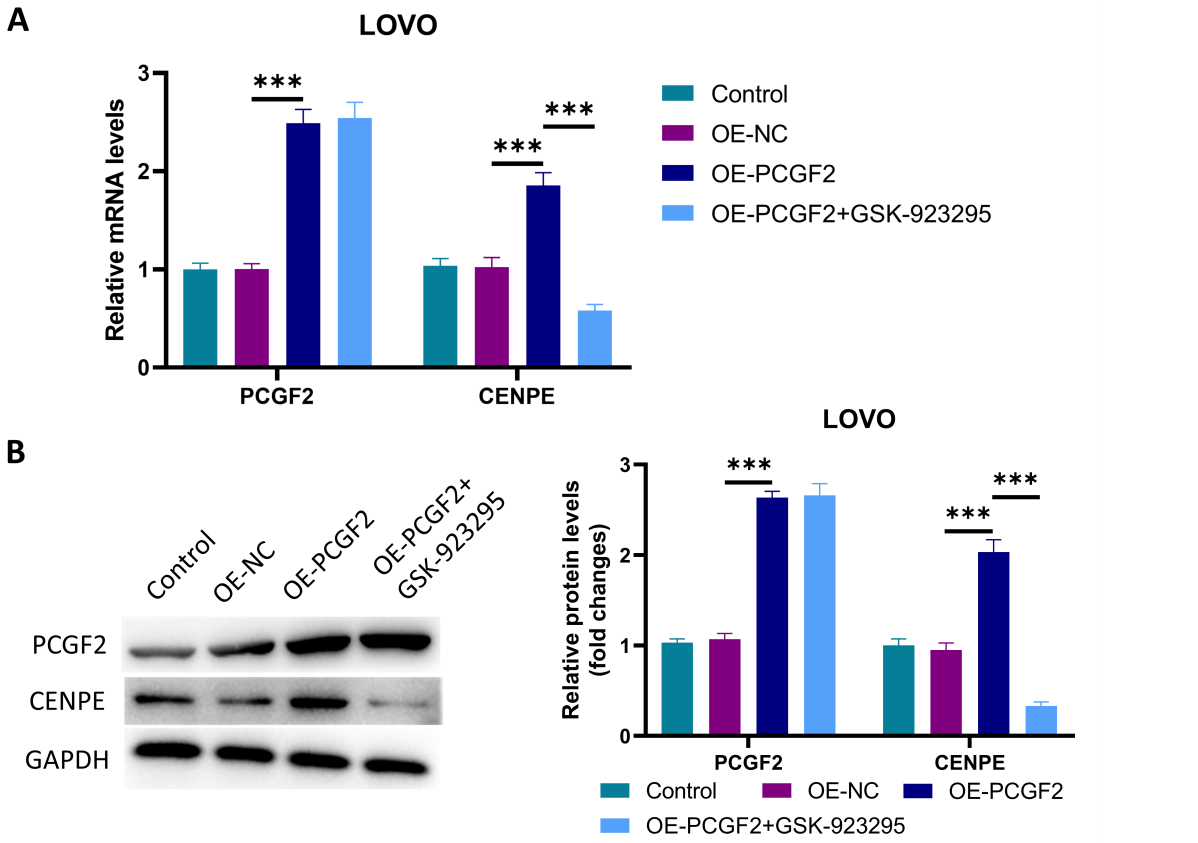

Fig. 4. The CENPE was inhibited by the inhibitor GSK-923295. (A) mRNA levels and (B) protein levels of PCGF2 and CENPE differently treated LOVO cells. N = 5. ***$p < 0.001$.

indicating that PCGF2 promoted the metastasis of LOVO cells, thus facilitating the progression of CC cells. The *in vivo* tumor progression analysis (Fig. 3C) indicated significantly lower tumor progression levels of sh-PCGF2 transfected LOVO cells compared to sh-NC transfected LOVO cells ($p < 0.001$), indicating that the PCGF2 knockdown prevented the CC progression and could potentially be used as the strategy during treating CC.

The CENPE was Inhibited by the Inhibitor GSK-923295

The *CENPE* (Fig. 4A) was upregulated in OE-PCGF2 transfected LOVO cells compared with those in OE-NC transfected cells ($p < 0.001$). This upregulation was subsequently reversed by the CENPE-specific inhibitor GSK-923295 in the OE-PCGF2+GSK-923295 group ($p < 0.001$). Conversely, when *PCGF2* mRNA was upregulated in the OE-PCGF2 group (Fig. 4A, $p < 0.001$), it remained at a high expression level without being affected by the CENPE inhibitor. Similarly, western blot analysis in Fig. 4B further confirmed that the protein level of CENPE was increased in OE-PCGF2 transfected LOVO cells compared with in OE-NC transfected cells ($p < 0.001$) while was decreased by GSK-923295 in OE-PCGF2+GSK-923295 group ($p < 0.001$). Meanwhile, PCGF2 protein was increased in the OE-PCGF2 group ($p < 0.001$) and was not affected by the CENPE inhibitor, indicating that the expression of CENPE did not regulate the expression of PCGF2. These findings proved that we established a LOVO cell model with both increased PCGF2 and decreased CENPE in the OE-PCGF2+GSK-923295 group, and by combining with results in Fig. 1, we proved that the CENPE was regulated and was downstream of PCGF2.

PCGF2 Stimulated the Proliferation and Suppressed Apoptosis of CC Cells by Upregulating CENPE

The CCK-8 assay results (Fig. 5A) demonstrated that the cell viability of OE-PCGF2 transfected LOVO cells increased compared with OE-NC transfected LOVO cells ($p < 0.001$), and this effect was reversed by CENPE inhibition. These findings indicated that PCGF2 enhanced the viability of CC cells only when CENPE was upregulated. Additionally, OE-PCGF2 transfected LOVO cells formed more cell colonies than OE-NC transfected LOVO cells (Fig. 5B,C, $p < 0.001$), and this effect was reversed by inhibiting CENPE. These findings demonstrated that PCGF2 could not increase the proliferation of CC cells if CENPE was inhibited. Furthermore, the TUNEL assay results (Fig. 5D) showed that the cell apoptosis of OE-PCGF2 transfected LOVO cells decreased compared with OE-NC

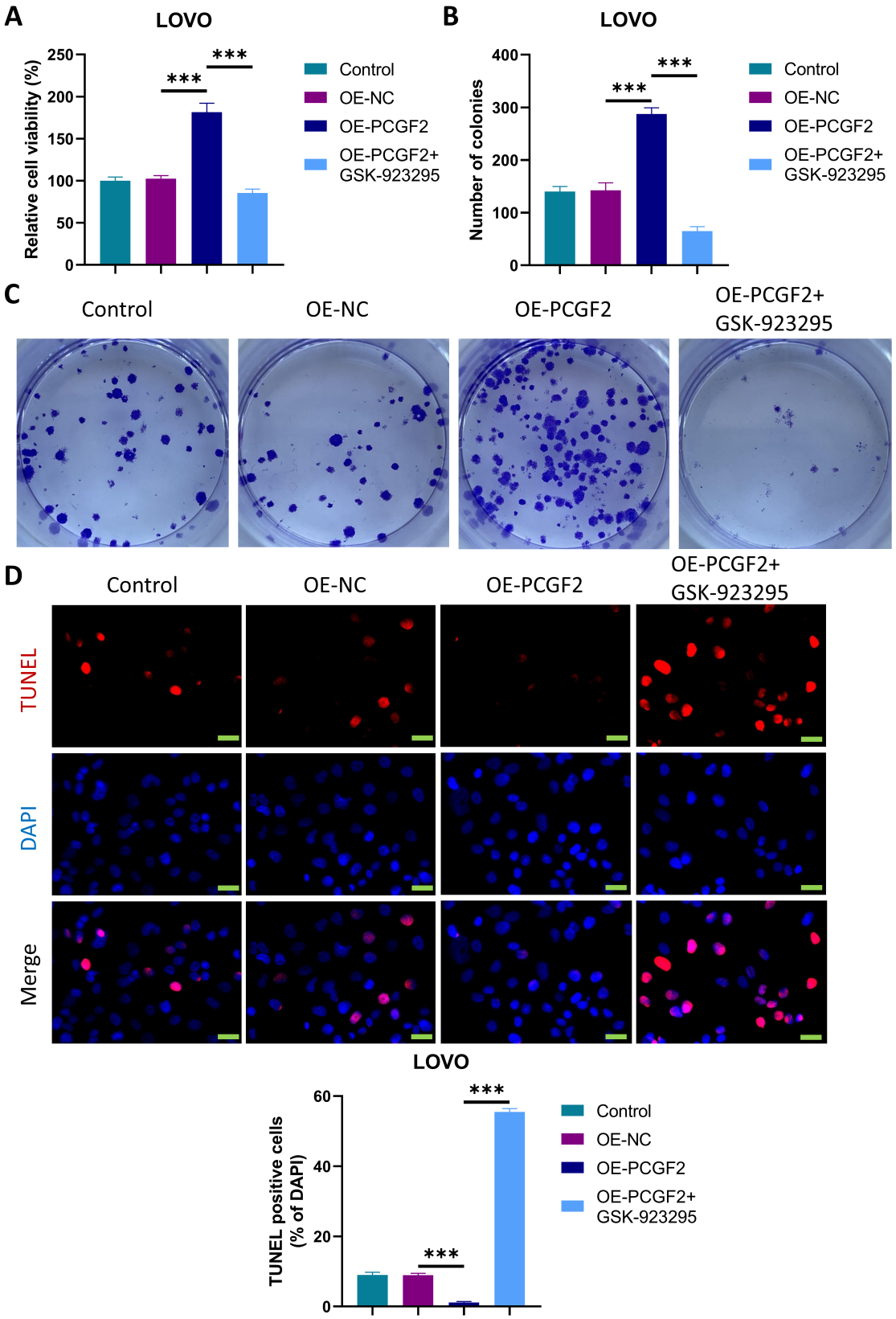

Fig. 5. PCGF2 enhanced CC cell proliferation and inhibited apoptosis by upregulating CENPE. (A) Results of CCK-8 assay. (B) Quantification and (C) representative images of colony forming assay. (D) Results of TUNEL assay (scale: 50 μm). N = 5. ***$p <$ 0.001.

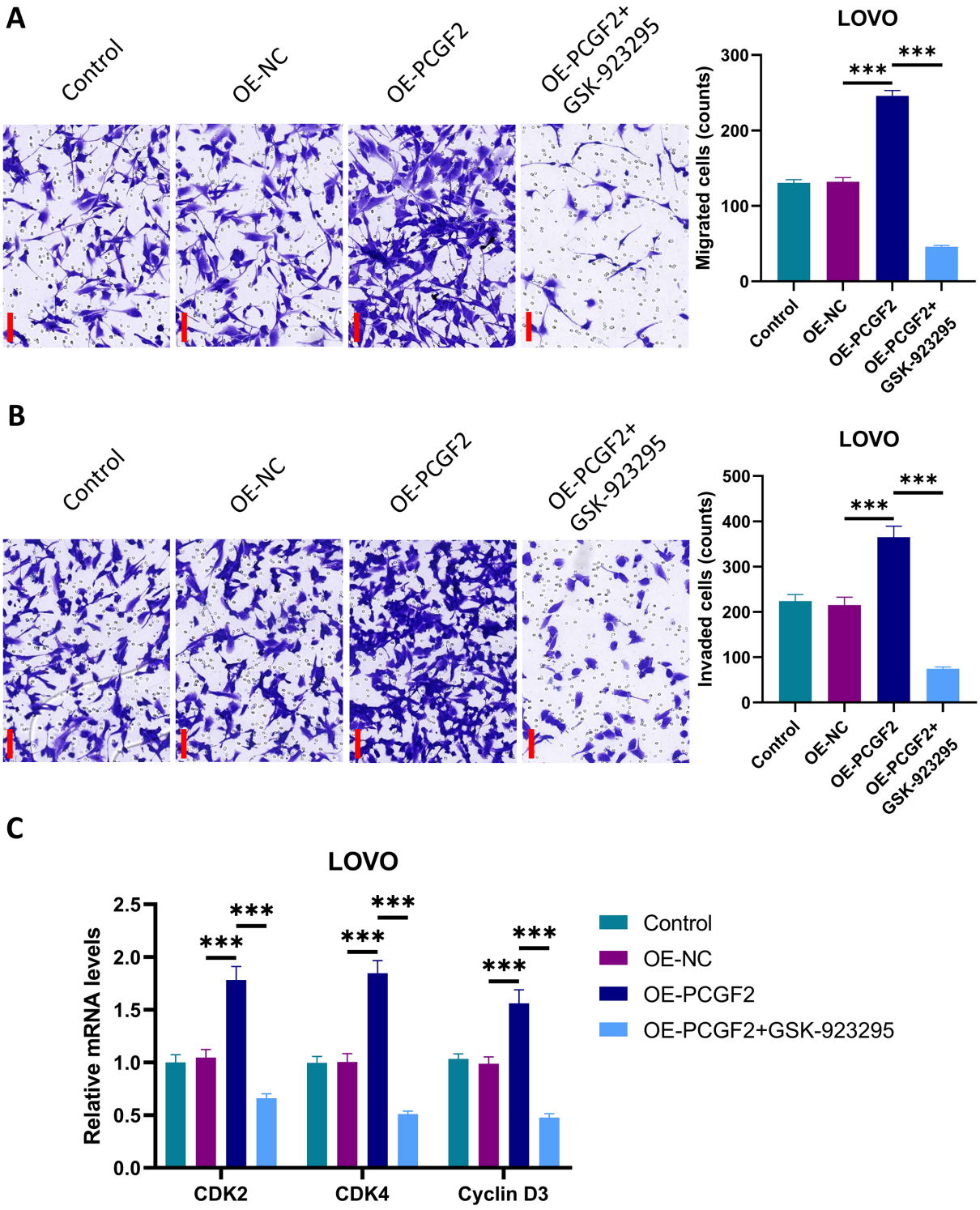

Fig. 6. PCGF2 promoted the migration and invasion of CC cells while preventing cell cycle arrest, achieved through the upregulation of CENPE. Results of transwell assays for (A) migration and (B) invasion (scale: 50 μm). (C) mRNA levels of cell cycle-related genes. N = 5. ***$p < 0.001$.

transfected LOVO cells ($p < 0.001$), and this effect was also reversed by CENPE inhibitor, indicating that PCGF2 suppressed apoptosis of CC cells when CENPE was upregulated. Overall, these results showed that PCGF2 promoted the progression of CC cells by upregulating CENPE. If the CENPE was inhibited, the effect of PCGF2 on promoting CC would be disrupted.

PCGF2 Promoted the Migration and Invasion of CC Cells while Preventing Cell Cycle Arrest through the Upregulation of CENPE

The transwell assay for migration (Fig. 6A) and invasion (Fig. 6B) revealed that the OE-PCGF2 transfected LOVO cells exhibited increased migration ($p < 0.001$) and invasion ($p < 0.001$) compared to OE-NC transfected LOVO cells, which were all reversed when CENPE was inhibited (migration: $p < 0.001$; invasion: $p < 0.001$). These results illustrated that PCGF2 promoted the metastasis of LOVO cells when CENPE was not inhibited. The results depicted in Fig. 6C showed that the cell cycle-related *CDK2*, *CDK4*, and Cyclin D3 were increased in OE-PCGF2 transfected LOVO cells compared to those in OE-NC transfected LOVO cells ($p < 0.001$), which were all reversed by CENPE inhibitor ($p < 0.001$). These findings suggested that the PCGF2 enhanced cell cycle activation in CC cells by successfully upregulating CENPE.

Discussion

In the present study, we demonstrated that PCGF2 functions as a driver of CC and promotes the progression of CC cells by upregulating CENPE. This study acquired a major breakthrough in our understanding of the effect of PCGF2 on CC. However, further in-depth explorations, including investigations and verifications by exploiting multiple cell lines and animals, would be needed for better understanding and clinical applications.

We observed that the PCGF2 enhanced the viability, proliferation, and metastasis of CC LOVO cells and, at the same time, inhibited cell apoptosis. These findings proved that PCGF2 facilitated the progression of CC cells and potentially promoted the CC progression, consistent with previous studies that considered PCGF2 to be related to poor prognosis in CC [11,14]. Moreover, it has been previously claimed that PCGF2 decreased immune cell migration by regulating cytokines, chemokines, and chemokine receptors [15]. Therefore, the promotion of CC by PCGF2 might be linked to immune system suppression, an aspect warranting further exploration. Moreover, the abnormal cell cycle has been proven to be one of the critical mechanisms underlying tumorigenesis, rendering cell cycle regulators targets during anticancer therapies [16]. In this study, we found that the tumor progression of CC cells with PCGF2 knockdown was suppressed, highlighting the potential of PCGF2 as a therapeutic target for treating CC.

Furthermore, our study demonstrated that the impact of PCGF2 on CC promotion was related to the upregulated CENPE. The CENPE, functioning as a microtubule plus-end-directed kinetochore motor protein, plays crucial roles in chromosome congression and alignment, segregation, spindle assembly checkpoints, and spindle microtubule capture [17]. It has been proved that the CENPE participated in multiple cancer types [18,19], and the CENPE inhibitor GSK923295 has shown remarkable anticancer activity *in vivo* models [20]. Moreover, CENPE inhibition suppresses chromosome alignment and prevents microtubules from attaching to kinetochores, leading to cell cycle arrest [20]. This aligns with our findings, where CENPE inhibition lowered the cell cycle activation, which had previously been enhanced by PCGF2 upregulation. In this study, we demonstrated that PCGF2 promoted CC cell progression through upregulating CENPE, suggesting that the inhibitors of PCGF2 and CENPE could be used in combination as an effective treatment for treating CC. However, the limitations of this study should be addressed in future studies. Firstly, the results of this study should be further verified by more cell lines and additional *in vivo* studies. Moreover, more mechanisms by which PCGF2 affects CC and is related to CENPE or cell cycle should be investigated in future studies.

Conclusion

To sum up, PCGF2 was proven to promote the progression of CC cells by upregulating CENPE, making PCGF2 inhibition combined with CENPE inhibition a potentially effective therapy for CC.

Availability of Data and Materials

Data to support the findings of this study are available on reasonable request from the corresponding author.

Author Contributions

QL, XC and ZL performed the research. JT and WY provided help and advice on the experiments. ZL, WY and JT contributed to the analysis and interpretation of the data. All authors contributed to the drafting and critical revision of the manuscript. All authors read and approved the final manuscript. All authors have participated sufficiently in the work to take public responsibility for appropriate portions of the content and agreed to be accountable for all aspects of the work in ensuring that questions related to its accuracy or integrity.

Ethics Approval and Consent to Participate

The study was approved by the Experimental Animal Ethics Committee of Ezhou Central Hospital (approval number: [2023]-02).

Acknowledgment

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This research received no external funding.

Conflict of Interest

The authors declare no conflict of interest.

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