Synergistic effects of olaparib combined with *ERCC1* on the sensitivity of cisplatin in non-small cell lung cancer

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Abstract. Non-small cell lung cancer (NSCLC) is a common malignant tumor. ERCC excision repair 1 endonuclease non-catalytic subunit (ERCC1) is a key mediator of nucleotide excision repair. The present study aimed to explore the synergistic effects of the poly(ADP-ribose) polymerase (PARP) inhibitor olaparib combined with *ERCC1* on the sensitivity of NSCLC cells to cisplatin. Preliminary experiments were performed to identify the optimal concentrations of cisplatin and olaparib for cellular treatment and subsequently NCI-H1299 and SK-MES-1 cells were treated with 20 μg/ml cisplatin combined with 50 μg/ml olaparib and 50 μg/ml cisplatin combined with 70 μg/ml olaparib, respectively. Subsequently, transfections were carried out to overexpress or knockdown the expression of *ERCC1* in NSCLC cell lines, including NCI-H1299 and SK-MES-1. The transfection efficiency was evaluated using reverse transcription-quantitative PCR and western blotting. The results demonstrated that cells with *ERCC1* overexpression and *ERCC1* knockdown were successfully constructed. Finally, the cell viability and apoptosis were determined using the Cell Counting Kit-8 and Annexin V-FITC cell apoptosis assays, respectively. In NCI-H1299 or SK-MES-1 cells treated with cisplatin combined with olaparib for 24 h, the cell viability significantly increased following *ERCC1* overexpression compared with the GV230 group ($P<0.05$), but significantly inhibited following *ERCC1* knockdown compared with the siRNA-NC group ($P<0.05$). However, ERCC1 overexpression or knockdown had the opposite effect on apoptosis. In conclusion, olaparib combined with *ERCC1* expression may enhance the sensitivity of cisplatin in NSCLC. These findings may provide novel insight for the improvement of platinum drug sensitivity and treatment of NSCLC.

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Abbreviations: NSCLC, non-small cell lung cancer; NER, nucleotide excision repair; ERCC1, ERCC excision repair 1 endonuclease non-catalytic subunit; PARP1, poly(ADP-ribose) polymerase 1; FBS, fetal bovine serum; NC, negative control; CCK-8, Cell Counting Kit-8.

Key words: non-small cell lung cancer, cisplatin, PARP inhibitor, olaparib, ERCC1, sensitivity

Introduction

Lung cancer is a leading cause of tumor-related mortality worldwide (1). Every year, 1.8 million people are diagnosed with lung cancer, and 1.6 million people die as a result of the disease, as well as 5-year survival rates vary from 4-17% depending on stage and regional differences (2). Non-small cell lung cancer (NSCLC) accounts for ~85% of lung cancer cases, and most deaths from lung cancer can be attributed to NSCLC (3). After radical surgery, >60% of patients with early-stage NSCLC can experience *in situ* recurrence of the tumor or distal metastasis (4). Recently, adjuvant chemotherapy based on platinum drugs has been widely applied to the clinical treatment of NSCLC after surgery (5). Platinum drugs can destroy the structure of DNA by forming platinum-DNA complexes, thereby contributing to the apoptosis of tumor cells (6). However, the long-term use of platinum drugs leads to drug resistance, which is one of the major obstacles of cancer treatment (7). Thus, an improved understanding of the mechanisms of platinum drug resistance, as well as the development of methods that can overcome resistance, is necessary to improve the prognosis of patients with cancer.

As the major target of platinum drugs is DNA, the sensitivity/resistance to these drugs may be affected by the ability of cells to recognize and repair DNA damage (8). Nucleotide excision repair (NER) is the major pathway for the removal of platinum-DNA adducts (9). ERCC excision repair 1, endonuclease non-catalytic subunit (ERCC1), a key component of NER, is involved in interstrand cross-linking repair, double-strand break repair, homologous recombination and telomere maintenance (10). Increasing evidence has indicated that the differential expression of *ERCC1* may be a cause of cell resistance to platinum drugs. Selvakumaran *et al* (11) found that downregulation of *ERCC1* altered the DNA repairing ability of cisplatin-resistant ovarian cancer cells and increased their sensitivity to cisplatin. In a previous meta-analysis, patients with lung cancer that had low/negative *ERCC1* expression had a higher response to platinum drugs and longer

median survival time compared with those with high/positive *ERCC1* expression (12).

In addition, poly(ADP-ribose) polymerase 1 (PARP1), is a sensor for DNA strand break that responds to platinum-induced DNA damage and participates in DNA repair (13). PARP inhibitors can improve the sensitivity of tumor cells to chemotherapy drugs or directly kill tumor cells through a homozygous lethal mechanism (14,15). A randomized clinical study demonstrated that the PARP inhibitor olaparib significantly increased the sensitivity to platinum drugs and prolonged the median progression-free survival time of breast cancer (16). However, the effects of PARP inhibitors combined with *ERCC1* expression on the sensitivity of platinum drugs remain unclear in NSCLC.

Hence, the present study aimed to investigate whether the expression of *ERCC1* enhanced the sensitivity of platinum drugs in combination with PARP inhibitors, thereby improving the prognosis of NSCLC. These results may provide novel insight for the improvement of platinum drug sensitivity and treatment of NSCLC.

Materials and methods

Cell culture. NSCLC cell lines, including the NCI-H1299 (adenocarcinoma) and SK-MES-1 (squamous carcinoma) cell lines were purchased from the Cell Resource Center, Shanghai Institute of Biotechnology, Chinese Academy of Sciences. NCI-H1299 and SK-MES-1 cells were cultured in RPMI-1640 medium (Thermo Fisher Scientific, Inc.) with 10% fetal bovine serum (FBS; Thermo Fisher Scientific, Inc.) and minimum essential medium (Thermo Fisher Scientific, Inc.) with 10% FBS, respectively. Both cell lines were incubated in an incubator with 5% CO_2 at 37˚C.

Cell transfection. Small interfering (si)RNA-negative control (NC, non-targeting; forward, 5'-UUCUCCGAACGUGUC ACGUTT-3' and reverse, 5'-ACGUGACACGUUCGGAGA ATT-3'), siRNA-ERCC1-1 (GCCCTTATTCCGATCTACA), siRNA-ERCC1-2 (CGACGTAATTCCCGACTAT), and siRNA-ERCC1-3 (CCGTGAAGTCAGTCAACAA) were designed and synthesized by Guangzhou RiboBio Co., Ltd. GV230 and GV230-ERCC1+ were purchased from Shanghai Genechem Co., Ltd. Cell transfection was performed as previously described (17). NCI-H1299 or SK-MES-1 cells were cultured in serum-free medium, then seeded into 6-well plates ($5x10^5$ cells/well). Next, 3.5 μg GV230, 3.5 μg GV230-ERCC1+, 50 nM siRNA-NC, 50 nM siRNA-ERCC1-1, 50 nM siRNA-ERCC1-2 or 50 nM siRNA-ERCC1-3 were transfected into the cells at 24±2˚C for 20 min using Lipofectamine 2000® (Invitrogen; Thermo Fisher Scientific, Inc.) according to the manufacturer's instructions. After transfection at 37˚C for 6 h, the medium was replaced with 10% serum-containing medium and cultured at 37˚C for another 48 h. Total RNA and total protein of the cells from different cells were extracted. The transfection efficiency was evaluated by determining the expression of ERCC1 using reverse transcription-quantitative (RT-q) PCR and western blotting.

RT-qPCR. Total RNA was extracted from the transfected cells ($5x10^5$ cells/well) using TRIzol® reagent (Thermo Fisher Scientific, Inc.) according to the manufacturer's protocol. Total RNA was reverse transcribed into cDNA using the PrimeScript™ II 1st Strand cDNA Synthesis kit (Takara Bio, Inc.). The temperature protocol used for reverse transcription was 37˚C for 60 min and 85˚C for 5 sec. Subsequently, qPCR was performed using SYBR Premix EX Taq (2X; Thermo Fisher Scientific Inc.), and the primer sequence of *ERCC1* was as follows: Forward, 5'-TTGTCCAGGTGGATGTGAAA-3' and reverse, 5'-GCTGGTTTCTGCTCATAGGC-3'. The qPCR thermocycling conditions were as follows: 95˚C for 3 min; 95˚C for 10 sec; followed by 40 cycles at 60˚C for 30 sec and 60˚C for 30 sec. The mRNA expression of ERCC1 was quantified using the $2^{-\Delta\Delta Cq}$ method (18) and normalized to the reference gene GAPDH forward, 5'-AGACAGCCGCATCTTCTTGT-3' and reverse, 5'-CTTGCCGTGGGTAGAGTCAT-3'.

Western blotting. Total protein was isolated from transfected cells ($5x10^5$ cells/well using radioimmunoprecipitation assay protein lysis buffer (Beyotime Institute of Biotechnology). Protein concentrations were measured using a Bicinchoninic Acid Protein Assay kit (Wuhan Boster Biological Technology, Ltd.) following the manufacturer's protocol. Protein samples (20 μg) were separated by 10% SDS-PAGE and transferred to PVDF membranes. After blocking with 5% skimmed milk for 2 h at 37˚C, the membranes were incubated with anti-ERCC1 antibody (1:2,000; cat. no. 14586-1-AP; ProteinTech Group, Inc.) and anti-β-actin antibody (1:10,000; cat. no. 66009-1-Ig; ProteinTech Group, Inc.) overnight at 4˚C. After washing 3 times with PBST (0.05% Tween-20 in PBS), the membranes were incubated with goat anti-rabbit mouse IgG (1:10,000; cat. no. 115-035-003; Jackson ImmunoResearch Laboratories, Inc.) at 37˚C for 2 h. After 3 washes, protein bands were visualized using the ECL assay kit (Beyotime Institute of Biotechnology) and analyzed using Image-Pro Plus software v.6.0, (Media Cybernetics Inc.).

Cell viability assay. Cell viability of NCI-H1299 and SK-MES-1 cells was determined using the Cell Counting Kit-8 (CCK-8; Beyotime Institute of Biotechnology). The schematic workflow of the cellular experimentation is presented in Fig. 1. Briefly, different concentrations of cisplatin (Sigma-Aldrich; Merck KGaA) and PARP inhibitor olaparib (Selleck Chemicals) were prepared in dimethyl sulfoxide (Beijing Solarbio Science & Technology Co., Ltd.). The control, *ERCC1*-overexpressing and *ERCC1*-knockdown cells were seeded into 6-well plates ($5x10^5$ cells/well), then treated with cisplatin alone or in combination with olaparib at 37KC, as shown in Fig. 1. Following treatment for 24 h, 10 μl of CCK-8 reagent (Beyotime Institute of Biotechnology) was added to the cells and incubated at 37˚C for 2 h. Absorbance was detected at 450 nm using a microplate reader.

Cell apoptosis assay. Effects of *ERCC1* expression on apoptosis in NCI-H1299 and SK-MES-1 cells were determined using an Annexin V-FITC cell apoptosis assay kit (Beyotime Institute of Biotechnology) according to the manufacturer's instructions. The NCI-H1299 or SK-MES-1 cells with *ERCC1* overexpression or interference ($1x10^4$ cells/well) were treated with cisplatin and olaparib at 37˚C for 24 h as detailed in Fig. 1. Subsequently, the cells were harvested and resuspended in

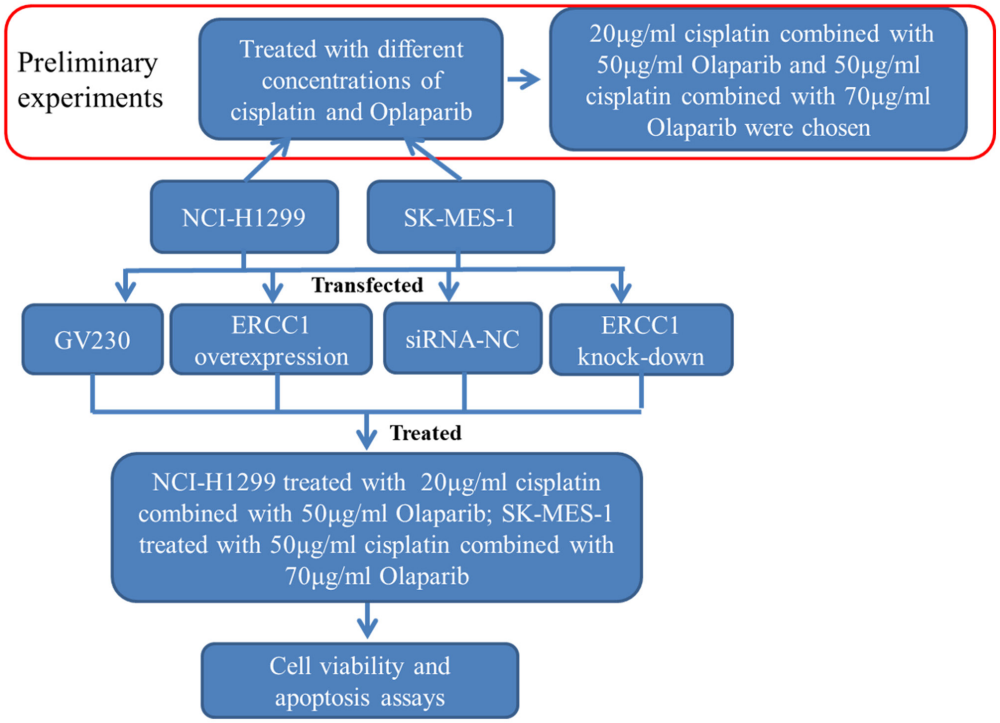

Figure 1. Schematic workflow diagram of cellular experiments. NCI-H1299 and SK-MES-1 cells were treated with different concentrations of cisplatin and oplaparib to screen the optimal concentrations of cisplatin and olaparib. Subsequenttly, the cells were transfected with si-ERCC1 and *ERCC1* overexpression plasmid and the cell viability and apoptosis were determined. ERCC1, ERCC excision repair 1 endonuclease non-catalytic subunit; si, small interfering; NC, negative control.

pre-cooled phosphate-buffered saline. After centrifugation at 1,000 x g for 5 min at room temperature, 195 μl binding buffer and 5 μl Annexin V-FITC (20 μg/ml) were added to the cells. After incubation at room temperature for 30 min in the dark, the cells were stained with 5 μl of propidium iodide (PI; 50 μg/ml) and incubated at room temperature in the dark for 10 min. Finally, flow cytometry (FACSCalibur; Becton-Dickinson and Company) was used to observe cell apoptosis, and the apoptosis rate (early plus late apoptosis) was calculated using the CellQuest software v.4.0 (Becton-Dickinson and Company).

Statistical analysis. Each experiment was performed in triplicate. All data are expressed as the mean ± standard deviation. GraphPad Prism 5.0 (GraphPad Software, Inc.) was used for statistical analysis. For multiple comparisons, one-way analysis of variance followed by Bonferroni correction was performed. Student's t-test with unpaired test was applied for comparisons between two groups. P<0.05 was considered to indicate a statistically significant difference.

Results

Selecting optimal concentrations of cisplatin and olaparib for cellular treatment. To select the optimum concentration of cisplatin, different concentrations were used to treat NSCLC cell lines for 24 h. In the NCI-H1299 cell line, when the cisplatin concentration was 10 μg/ml, the cell viability was significantly inhibited compared with the control group (P<0.05) and gradually decreased with an increase in cisplatin concentration (Fig. 2A). In the SK-MES-1 cell line, when the cisplatin concentration was 30 μg/ml, the cell viability began to be significantly inhibited compared with the control group (P<0.05), and when the concentration of cisplatin was 50 μg/ml, cell viability was further suppressed compared with the control group (P<0.05; Fig. 2B). Thus, in subsequent experiments, 20 and 50 μg/ml cisplatin were applied to the NCI-H1299 and SK-MES-1 cell lines, respectively.

To confirm the concentrations required for the PARP inhibitor olaparib, the NSCLC cell lines were treated with cisplatin combined with different concentrations of olaparib. When the NCI-H1299 and SK-MES-1 cell lines were treated with 20 and 50 μg/ml cisplatin, respectively, cell viability was significantly decreased compared with that of the control group (P<0.05; Figs. 2C and D). However, in the NCI-H1299 cell line, the cell viability was significantly lower after 50 μg/ml olaparib combined with cisplatin administration compared with after cisplatin treatment alone (P<0.05; Fig. 2C). Similarly, in the SK-MES-1 cell line, when the concentration of olaparib was 70 μg/ml, the cell viability was further significantly reduced compared with that of the cells treated without olaparib (P<0.05; Fig. 2D). These results indicated that in subsequent experiments, the NCI-H1299 and SK-MES-1 cells should be treated with 20 μg/ml cisplatin combined with 50 μg/ml olaparib and 50 μg/ml cisplatin combined with 70 μg/ml olaparib, respectively.

Cell transfection efficiency analyses by RT-qPCR and western blotting. To evaluate the transfection efficiency, the expression level of ERCC1 in NSCLC cell lines was determined by RT-qPCR and western blotting. In the NCI-H1299 cell line, western blot analysis demonstrated that the expression of ERCC1 significantly decreased following transfection with siRNA-ERCC1-1/2/3 compared with the siRNA-NC group (P<0.05) and that the siRNA-ERCC1-1 group had the

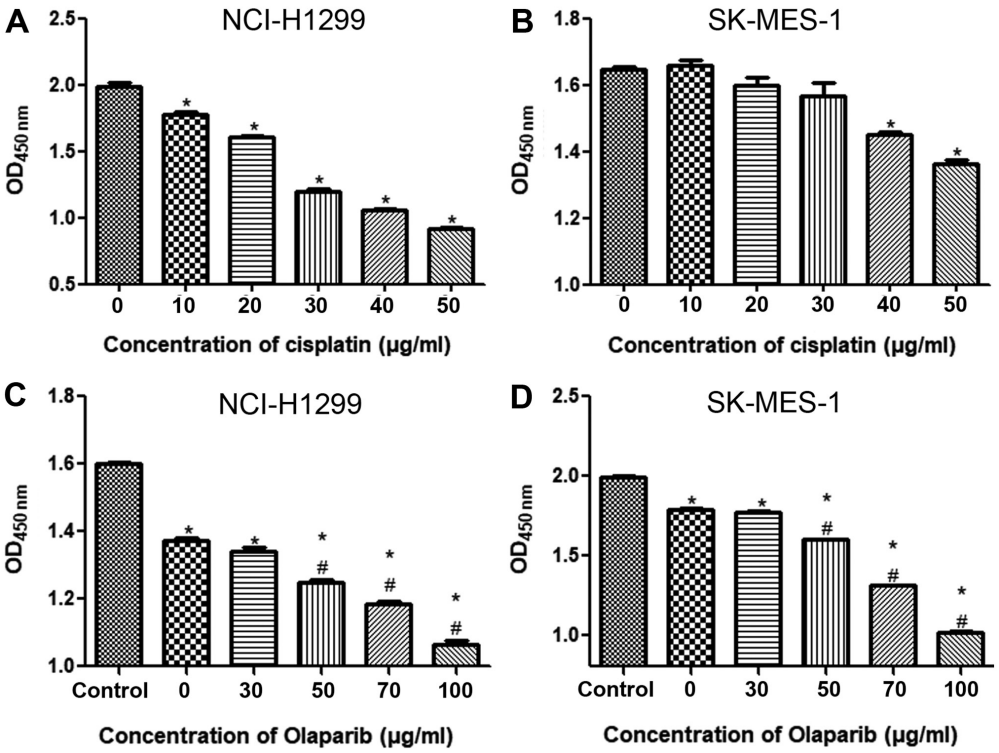

Figure 2. Selecting optimal concentrations of cisplatin and olaparib for cellular treatment. Cell viability of (A) NCI-H1299 cells treated with different concentrations of cisplatin, (B) SK-MES-1 cells treated with different concentrations of cisplatin, (C) NCI-H1299 cells treated with 20 μg/ml cisplatin combined with different concentrations of PARP inhibitor olaparib. The control group was comprised of untreated cells; 0 μg/ml group consisted of cells treated with 20 μg/ml cisplatin combined with 0 μg/ml olaparib. (D) SK-MES-1 cells treated with 50 μg/ml cisplatin combined with different concentrations of PARP inhibitor Olaparib. The control group was comprised of untreated cells; 0 μg/ml group consisted of cells treated with 20 μg/ml cisplatin combined with 0 μg/ml olaparib. *P<0.05 vs. control group; #P<0.05 vs. 0 μg/ml olaparib group. OD, optical density; PARP, poly(ADP-ribose) polymerase.

best transfection efficiency as the *ERCC1* mRNA expression was the lowest in the siRNA-ERCC1-1 group compared with the siRNA-NC, si-RNA-ERCC1-2 and si-RNA-ERCC1-3 groups (Fig. 3A and B). In addition, RT-qPCR was performed to determine the mRNA expression levels of *ERCC1* in the GV230, GV230-ERCC1+, siRNA-NC, and siRNA-ERCC1-1 groups. The expression level of *ERCC1* in the GV230-ERCC1+ group was significantly higher compared with that in the GV230 group, while the expression level was significantly downregulated in the siRNA-ERCC1-1 group compared with that in the siRNA-NC group (both P<0.05; Fig. 3C). The trend of ERCC1 mRNA expression in the SK-MES-1 cell line detected by RT-qPCR was similar to that in the NCI-H1299 cell line (Fig. 3D). These results suggested that the NCI-H1299 or SK-MES-1 cells with *ERCC1* overexpression and *ERCC1* knockdown were successfully generated.

Effects of ERCC1 combined with olaparib on the cell viability of cisplatin-treated cells. In the NCI-H1299 cell line, after the cells were treated with 20 μg/ml cisplatin combined with 50 μg/ml olaparib for 24 h, cell viability was significantly increased in the GV230-ERCC1+ group compared with the GV230 group, but significantly decreased in the siRNA-ERCC1 group compared with the siRNA-NC group (both P<0.05; Fig. 4A). Additionally, the change in cell viability in the SK-MES-1 cell line was in accordance with that in the NCI-H1299 cell line (Fig. 4B). Indeed, after the SK-MES-1 cells were treated with 50 μg/ml cisplatin in combination with 70 μg/ml olaparib, the cell viability was enhanced in *ERCC1*-overexpressing cells, whereas it was inhibited in *ERCC1*-knockdown cells (P<0.05; Fig. 4B). These results suggested that *ERCC1* combined with olaparib may enhance the sensitivity of NCI-H1299 and SK-MES-1 cells to cisplatin by affecting cell viability.

Effects of ERCC1 combined with olaparib on the cell apoptosis of cisplatin-treated cells. Effects of *ERCC1* on the apoptosis of cells treated with cisplatin and olaparib were determined using flow cytometry. The flow cytometry dotplots evaluating the apoptosis of the NCI-H1299 and SK-MES-1 cell lines are shown in Figs. 5A and 6A, respectively. In the NCI-H1299 cell line, the cell apoptosis rates in the GV230, GV230-ERCC1+, siRNA-NC, and siRNA-ERCC1 groups were 67.88±2.1%, 50.14±1.53%, 6.3±0.29% and 11.7±0.98%, respectively (Fig. 5B). These results demonstrated that after NCI-H1299 cells were treated with 20 μg/ml cisplatin combined with 50 μg/ml olaparib for 24 h, the cell apoptosis rate significantly decreased in *ERCC1*-overexpressing cells compared with the GV230 group, and markedly increased in *ERCC1*-knockdown cells compared with si-NC group (P<0.05; Fig. 5B). In addition, when the SK-MES-1 cells were treated with 50 μg/ml cisplatin in combination with 70 μg/ml olaparib, the trend of cell apoptosis rate in the SK-MES-1 cell line was similar to that in the NCI-H1299 cell line (Fig. 6B). In summary, after the treatment of NSCLC cells with cisplatin and olaparib, cell apoptosis was inhibited in *ERCC1*-overexpressing cells, but enhanced *ERCC1*-knockdown cells.

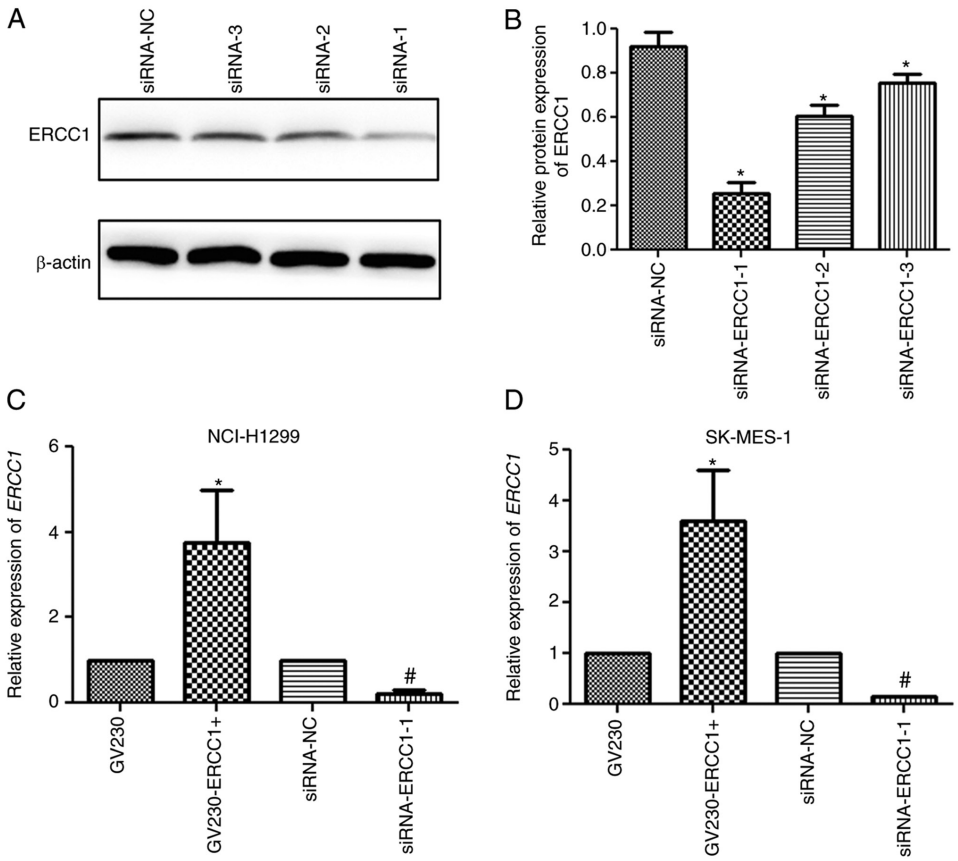

Figure 3. Cell transfection efficiency analyses by RT-qPCR and western blotting (A) Protein expression level of ERCC1 in the NCI-H1299 cell line, measured by western blotting analysis. (B) Gray analysis of the western blotting for the NCI-H1299 cell line. *P<0.05vs. siRNA-NC group. (C) mRNA expression level of *ERCC1* in the NCI-H1299 cell line. (D) mRNA expression level of *ERCC1* in the SK-MES-1 cell line. *P<0.05 vs. GV230 group; #P<0.05 vs. siRNA-NC group. ERCC1, ERCC excision repair 1 endonuclease non-catalytic subunit; si, small interfering; NC, negative control; RT-q, reverse transcription-quantitative.

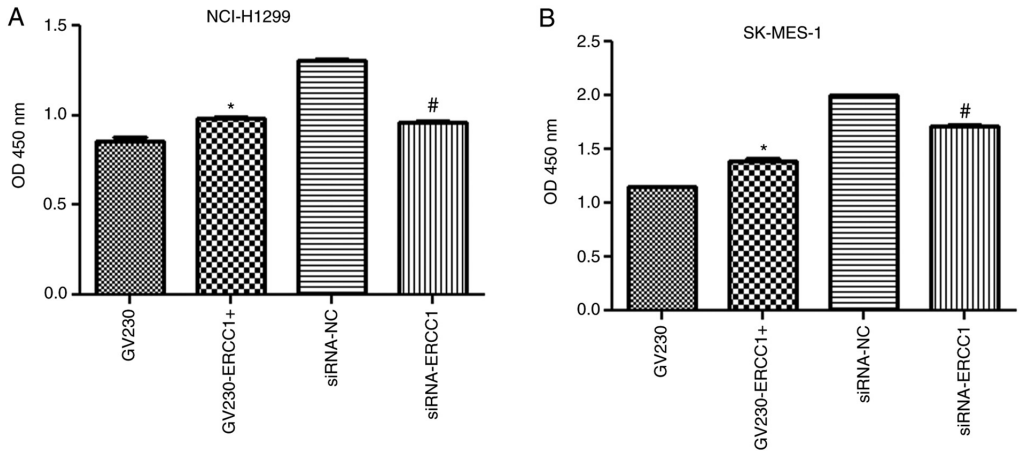

Figure 4. Effects of *ERCC1* on the viability of cells treated with cisplatin and olaparib in the (A) NCI-H1299 and (B) SK-MES-1 cell lines. *P<0.05 vs. GV230 group; #P<0.05 vs. siRNA-NC group. ERCC1, ERCC excision repair 1 endonuclease non-catalytic subunit; si, small interfering; NC, negative control; OD, optical density.

Discussion

NSCLC is one of the most common malignant tumors and is harmful to human health and life (19). Platinum-based chemotherapy is usually used to assist in the treatment of NSCLC (20). However, clinical treatment is not always satisfactory due to the progression of drug resistance (21). Hence, there is an urgent need to develop new ways to combat drug resistance and improve the sensitivity of these drugs. The expression of *ERCC1* regulates DNA damage induced by platinum drugs and is a candidate biomarker for predicting the sensitivity of platinum drugs (22,23). Additionally, olaparib can enhance the sensitivity of platinum drugs by inhibiting PARP-related pathways (24).

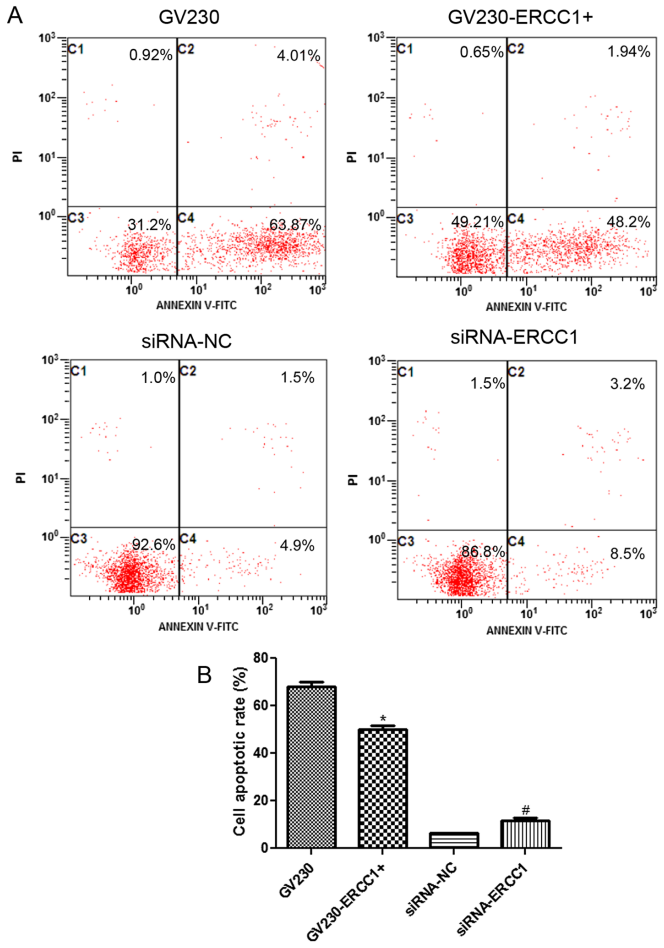

Figure 5. Effects of *ERCC1* on the apoptosis of NCI-H1299 cells treated with cisplatin and olaparib. (A) Representative flow cytometry images. (B) Cell apoptosis rate in different groups of treated cells. *P<0.05 vs. GV230 group; #P<0.05 vs. siRNA-ERCC1 group. ERCC1, ERCC excision repair 1 endonuclease non-catalytic subunit; si, small interfering; NC, negative control; PI, propidium iodide.

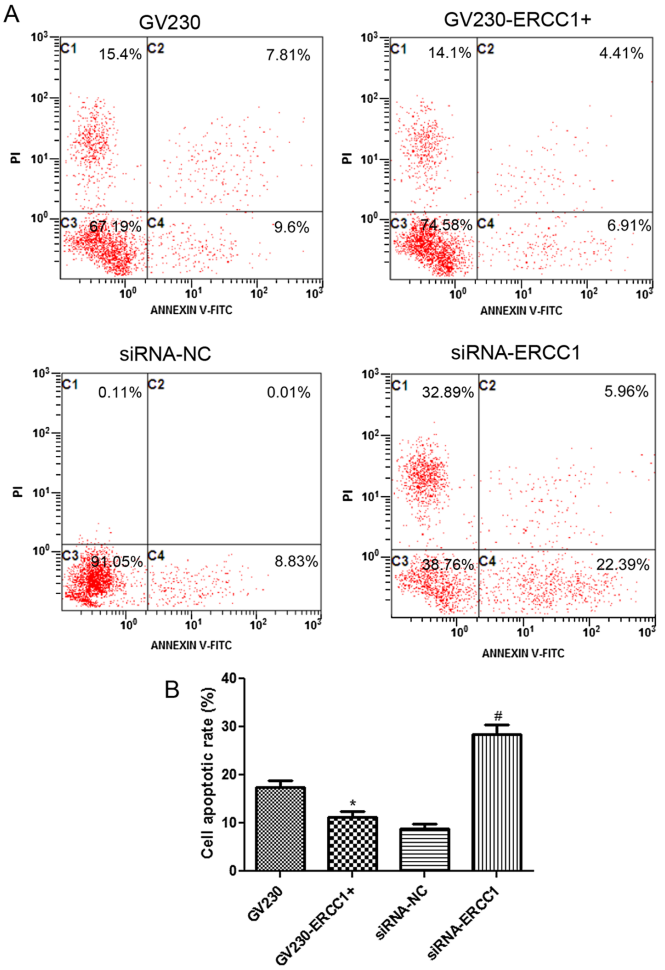

Figure 6. Effects of *ERCC1* on the apoptosis of SK-MES-1 cells treated with cisplatin and Olaparib. (A) Representative flow cytometry images. (B) Cell apoptosis rate in different groups of treated cells. *P<0.05 vs. GV230 group. #P<0.05 vs. siRNA-ERCC1 group. ERCC1, ERCC excision repair 1 endonuclease non-catalytic subunit; si, small interfering; NC, negative control; PI, propidium iodide

In the present study, 50 μg/ml olaparib combined with 20 μg/ml cisplatin significantly inhibited the viability of NCI-H1299 cells, while 70 μg/ml olaparib combined with 50 μg/ml cisplatin further inhibited the viability of SK-MES-1 cells. The findings of the present study indicated that olaparib can enhance the sensitivity of cisplatin in NSCLC. Ledermann and Pujade-Lauraine (25) demonstrated that olaparib increased the sensitivity of platinum-based drugs and prolonged the progression-free survival time of relapsed ovarian cancer, which is consistent the results of the present study. The present study further demonstrated that olaparib combined with cisplatin can inhibit the viability of NSCLC cell lines.

In the present study, to further explore the synergistic effects of *ERCC1* expression combined with olaparib on the sensitivity of cisplatin, NSCLC cell lines with *ERCC1* overexpression and knockdown were successfully constructed. In the NCI-H1299 or SK-MES-1 cells with *ERCC1* knockdown, olaparib combined with cisplatin inhibited cell viability and promoted cell apoptosis. In the present study, the trends of cell viability and apoptosis in the cells with ERCC1 overexpression were opposite to those in cells with ERCC1 knockdown. The low expression of some genes related to DNA damage repair, including *ERCC1*, cyclin dependent kinase 1 (*CDK1*), serine/threonine protein kinase CHK1 (*CHK1*), CHK2 DNA damage checkpoint kinase (*CHK2*), BRCA1 DNA repair associated (*BRCA1*), sperm hammerhead 2 (*SH2*), ATM serine/threonine kinase (*ATM*), RAD51 recombinase (*RAD51*), MRE11 homolog, double strand break repair nuclease (*MRE11*), phosphatase and tensin homolog (*PTEN*), X-ray repair cross complementing 1 (*XRCC1*), damage specific DNA binding protein 1 (*DDB1*), and XPA binding protein 2 (*XAB2*), can improve the sensitivity of PARP inhibitor therapy (26-28). A study by Xie *et al* (29) demonstrated that patients with NSCLC with low expression of both *ERCC1* and *PARP1* had the best prognosis in platinum-based chemotherapy compared to patients with NSCLC. Based on the findings of the aforementioned and present studies, it can be hypothesized that *ERCC1* expression may have an effect on the sensitivity of olaparib to cisplatin and there may be a synergistic effect between PARP inhibitors and *ERCC1* knockdown in NSCLC. Additionally, cisplatin-induced DNA damage triggers G2/M cell cycle arrest by activating checkpoint signaling (30). Hence, it can be hypothesized that olaparib combined with *ERCC1* knockdown may mediate the proliferation and apoptosis of tumor cells by regulating DNA damage repair. However, this needs to be investigated in future studies.

In addition, another study indicated that the predictive effect of *ERCC1* expression on cisplatin treatment may be associated with the histological types of lung cancer (31). In the present study, olaparib combined with *ERCC1* overexpression or *ERCC1* knockdown enhanced the sensitivity of cisplatin in adenocarcinoma cells (NCI-H1299) and squamous carcinoma cells (SK-MES-1), thereby regulating cell viability and apoptosis. Pierceall *et al* (31) indicated that *ERCC1* expression in patients with squamous cell carcinoma displayed significant predictive value, while in patients with adenocarcinoma it was not significant. This was somewhat different from the findings of the present study, possibly due to the joint action of ERCC1 and other genes related to DNA damage repair. *BRCA1* expression is also considered a predictive biomarker for the sensitivity of platinum drugs and is associated with the expression of *PARP* in NSCLC (32,33). Further studies need to be performed to explore the combined effects of different genes related to DNA damage repair on the sensitivity/resistance of platinum drugs.

However, there are some limitations to our study. The relationship between Olaparib combined with ERCC1 and DNA damage repair in NSCLC requires further investigation, and all underlying mechanisms need to be further explored. Additionally, the combination effects of Olaparib and *ERCC1* knockdown should be further verified *in vivo*; moreover, further studies associated with preclinical or clinical models are also required.

In conclusion, in the present study, olaparib combined with *ERCC1* knockdown enhanced the sensitivity of cisplatin, which inhibited cell viability and promoted cell apoptosis in NSCLC cell lines. The findings of the present study provide evidence for drugs that block *ERCC1* function during the treatment of NSCLC and may assist in the future development of new therapeutic strategies with olaparib combined with *ERCC1* for the treatment of NSCLC.

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Availability of data and material

The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

Authors' contributions

HH conceived the study. KX performed the experiments. KX, XN and SL collected and analyzed the data. KX and HH drafted the manuscript and HH and GZ revised it for important intellectual content. HH supervised the study. HH and KX confirm the authenticity of all the raw data. All authors have read and approved the final version of the manuscript.

Ethics approval and consent to participate

Not applicable.

Patient consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

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