

Effects of propofol on the proliferation and migration of liver cancer cells

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Abstract. Liver cancer is a malignant cancer with worldwide prevalence. It has been reported that cancer cells are usually exposed to a hypoxic microenvironment, which is associated with a poor prognosis in patients with cancer. Propofol is an intravenous anesthetic that is widely used in cancer surgery. The present study aimed to determine the effects of propofol stimulation on the viability, proliferation and migration of liver cancer cells under normoxia and cobalt chloride (CoCl₂)-induced hypoxia. Under normoxia, HepG2 and HCCLM3 cells were randomly divided into six groups as follows: i) Control group; ii) 10 μ M propofol group; iii) 25 μ M propofol group; iv) 50 μ M propofol group; v) 100 μ M propofol group; and vi) DMSO group. Cell viability and proliferation were analyzed using Cell Counting Kit-8 (CCK-8) and 5-ethynyl-2'-deoxyuridine (EdU) assays, respectively, following 24 or 48 h of propofol treatment. In addition, wound healing and Transwell migration assays were used to determine the changes in cell migration. Under CoCl₂-induced hypoxia, the protein levels of hypoxia inducible factor-1 α (HIF-1 α) of HepG2 cells were analyzed using western blotting. Subsequently, CCK-8 and wound healing assays were used to determine the effect of propofol on cell viability and migration. The results of the present study revealed that propofol stimulation had no significant effect on the viability, proliferation and migration of HepG2 and HCCLM3 cells under normoxia. The protein levels of HIF-1 α were significantly upregulated following the treatment with 200 μ M CoCl₂ for 12 h. However, no significant differences were found in the viability and migration of HepG2 cells following the stimulation with propofol in the presence of CoCl₂. In conclusion, the findings of the present study revealed

that propofol exerted no effect on the viability, proliferation and migration of HepG2 and HCCLM3 cells under normoxic and hypoxic conditions.

Introduction

Liver cancer is the sixth most common type of cancer and the fourth leading cause of cancer-associated mortality worldwide, accounting for ~841,000 newly diagnosed cases and 782,000 deaths annually (1). Notably, China accounts for ~1/2 of all cases and deaths (2). In the United States, the morbidity and mortality of liver cancer is growing at a faster rate compared with any other type of cancer (3,4). Surgical resection remains the first-choice treatment option for early-stage patients (5); however, a 5-year recurrence rate as high as 70% has been reported following liver cancer resection (6,7), and there is currently no adjuvant therapy available for patients to prevent or reduce postoperative metastases and the formation of *de novo* tumors (8). Thus, the prognosis and survival outcomes of patients following curative liver cancer resection remain unsatisfactory. It has been reported that cellular and molecular events involved in tumor metastasis may be notably affected during or immediately following surgery (9). However, it has been suggested that during curative resection, the surgery-induced stress response and immunosuppression, in addition to the weakened immune function following surgery, may activate a series of events involved in the metastasis of tumors (10-12). In addition, operative manipulation itself has also been suggested to promote the dissemination of cancer cells (12). Therefore, the perioperative period has been suggested as a window of opportunity to effectively reduce cancer metastasis and recurrence.

Propofol is an anesthetic that has been widely used in the clinic. Recently, the non-anesthetic properties of propofol (13,14), such as its immunomodulatory, neuroprotective, antioxidant and anticancer effects, have been investigated in more detail. Previous studies in multiple cancer types, including liver (15,16), lung (17), breast (18), colorectal (19), gastric (20), prostate (21) and pancreatic cancer (22), have reported that propofol exerts direct anticancer effects by interfering with the biological functions of cancer cells through a variety of mechanisms to inhibit tumor proliferation, invasion and metastasis and promote apoptosis. In addition, propofol

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may also induce indirect effects by affecting the functions of immune cells (23). However, the studies investigating the role of propofol in cancer have reported conflicting results. For example, Zhang *et al.* (24) reported that propofol exerts oncogenic activity by activating NF-E2-related factor 2 (Nrf2), which leads to an increase in the proliferation and invasion of gallbladder cancer cells. In another study by Ecimovic *et al.* (25), the results revealed that propofol had no effect on the proliferation of breast cancer cells. Thus, the association between propofol and cancer progression requires further investigation.

Solid tumor cells usually exist within a hypoxic microenvironment, which is caused by the imbalance of oxygen supply and demand (26). Tumor cells in the hypoxic environment have been observed to undergo a series of adaptive changes, in which the activation of the hypoxia inducible factor-1 α (HIF-1 α) pathway plays an important role. HIF-1 α is rapidly degraded under normoxia, but it can accumulate and be activated under hypoxic conditions (27). The cobalt ion in cobalt chloride (CoCl₂) can replace Fe²⁺ on the active site of prolyl hydroxylase (PHD), which is a key enzyme required for the degradation of HIF-1 α under normoxic conditions. This blocks the activity of PHD and stabilizes HIF-1 α , which subsequently upregulates the protein levels of HIF-1 α and thereby induces its transcriptional activity (28). Therefore, CoCl₂ was used as a chemical hypoxia simulant in the present study.

The present study aimed to determine the effects of propofol on the viability, proliferation and migration of HepG2 and HCCLM3 cells under normoxia and CoCl₂-induced hypoxia *in vitro*.

Materials and methods

Cell culture and treatment. HCCLM3 and HepG2 cells (Kunming Cell Bank of Type Culture Collection) were cultured in DMEM (cat. no. C11995500BT; Gibco; Thermo Fisher Scientific, Inc.) supplemented with 10% FBS (cat. no. 10099141; Gibco; Thermo Fisher Scientific, Inc.) and 1% penicillin-streptomycin (cat. no. P1400; Beijing Solarbio Science & Technology, Co., Ltd.). Both cells were maintained in an incubator with 5% CO₂ at 37°C, and routinely digested with 0.25% Trypsin-EDTA (cat. no. 25200072; Gibco; Thermo Fisher Scientific, Inc.) for subculture upon reaching 80-90% confluence.

The plasma concentration of propofol required for sedation in the intensive care unit is ~2 μ g/ml (11.2 μ M) (29) and 3-8 μ g/ml (16.8-44.8 μ M) for general anesthesia (30). Therefore, the present study used a range of concentrations of propofol (10, 25, 50 and 100 μ M). Under normoxia, HepG2 and HCCLM3 cells were randomly divided into six groups: i) Control group (untreated cells); ii) 10 μ M propofol (cat. no. Y0000016; Sigma-Aldrich; Merck KGaA) group; iii) 25 μ M propofol group; iv) 50 μ M propofol group; v) 100 μ M propofol group; and vi) DMSO (cat. no. D2650; Sigma-Aldrich; Merck KGaA) group. To simulate hypoxia, HepG2 cells were treated with 200 μ M CoCl₂ (cat. no. C8661; Sigma-Aldrich; Merck KGaA) or different concentrations of propofol (10, 25, 50 or 100 μ M) in the presence of 200 μ M CoCl₂ for 12 h. Propofol was dissolved in DMSO, and the

final concentration of DMSO in both the DMSO and propofol groups was 0.05%.

Cell Counting Kit-8 (CCK-8) assay. The viability of HepG2 and HCCLM3 cells was analyzed using a CCK-8 assay (cat. no. CK04; Dojindo Molecular Laboratories, Inc.). Briefly, cells were seeded into 96-well plates at a density of 6x10³ cells/well (for 24 h propofol stimulation) or 4x10³ cells/well (for 48 h propofol stimulation). Under normoxia, the cells were treated with different concentrations of propofol or 0.05% DMSO for 24 or 48 h. For hypoxia simulation, cells were exposed to propofol in the presence of 200 μ M CoCl₂ for 12 h. Following the incubation, 10 μ l CCK-8 solution was added into each well and incubated for a further 1 h. The absorbance of each well was measured at a wavelength of 450 nm using a microplate reader (Epoch2NS; BioTek Instruments, Inc.).

5-ethynyl-2'-deoxyuridine (EdU) assay. Cell proliferation was analyzed using a Click-iT™ EdU Imaging kit (cat. no. C10337; Invitrogen; Thermo Fisher Scientific, Inc.). Briefly, cells were cultured at a density of 5x10⁴ cells/well (for 24 h propofol stimulation) or 3x10⁴ cells/well (for 48 h propofol stimulation) in 24-well plates for 24 h and then treated with different concentrations of propofol or 0.05% DMSO for 24 or 48 h. Following the incubation, the cells were labeled for 2 h with 10 μ M EdU solution and then fixed with 4% paraformaldehyde (Shanghai Lingfeng Chemical Reagent Co., Ltd.) for 15 min at room temperature. Cells were permeabilized with 0.5% Triton X-100 (cat. no. 0694; Beijing Solarbio Science & Technology Co., Ltd.) for 20 min. EdU-incorporated cells and all cells were stained with Alexa Fluor fluorescent dye and Hoechst 33342 solution, respectively, for 30 min at room temperature, away from light. Subsequently, stained cells were visualized in five randomly selected fields of view using a fluorescence microscope (cat. no. DM5500B; Leica Microsystems GmbH), and the percentage of EdU-positive cells was calculated. Analysis was performed using ImageJ version 1.8.0 software (National Institutes of Health).

Wound healing assay. A total of 1x10⁶ cells were seeded into each well of six-well plates and cultured for 24 h. Upon cells reaching 100% confluence, an artificial scratch was made in the middle of the cell monolayer using a 200- μ l pipette tip. The cell medium was subsequently removed and replaced with fresh DMEM containing 10, 25, 50 or 100 μ M propofol or 0.05% DMSO under normoxia. Cells under hypoxia were treated with propofol combined with 200 μ M CoCl₂. The wound closure was observed and images were captured using an inverted microscope (TS100-F; Nikon Corporation) at 0 and 24 h. Analysis was performed using ImageJ version 1.8.0 software.

Transwell migration assay. Analysis of cell migration was conducted using a 24-well Transwell chamber (pore size, 8- μ m; cat. no. 3422; Corning, Inc.). Serum-starved HCCLM3 cells (5x10⁴ cells) suspended in 200 μ l serum-free DMEM containing 10, 25, 50 or 100 μ M propofol or 0.05% DMSO were plated into the upper chamber. The lower chamber was filled with 600 μ l complete medium supplemented with 20% FBS (cat. no. 10099141; Gibco; Thermo Fisher Scientific,

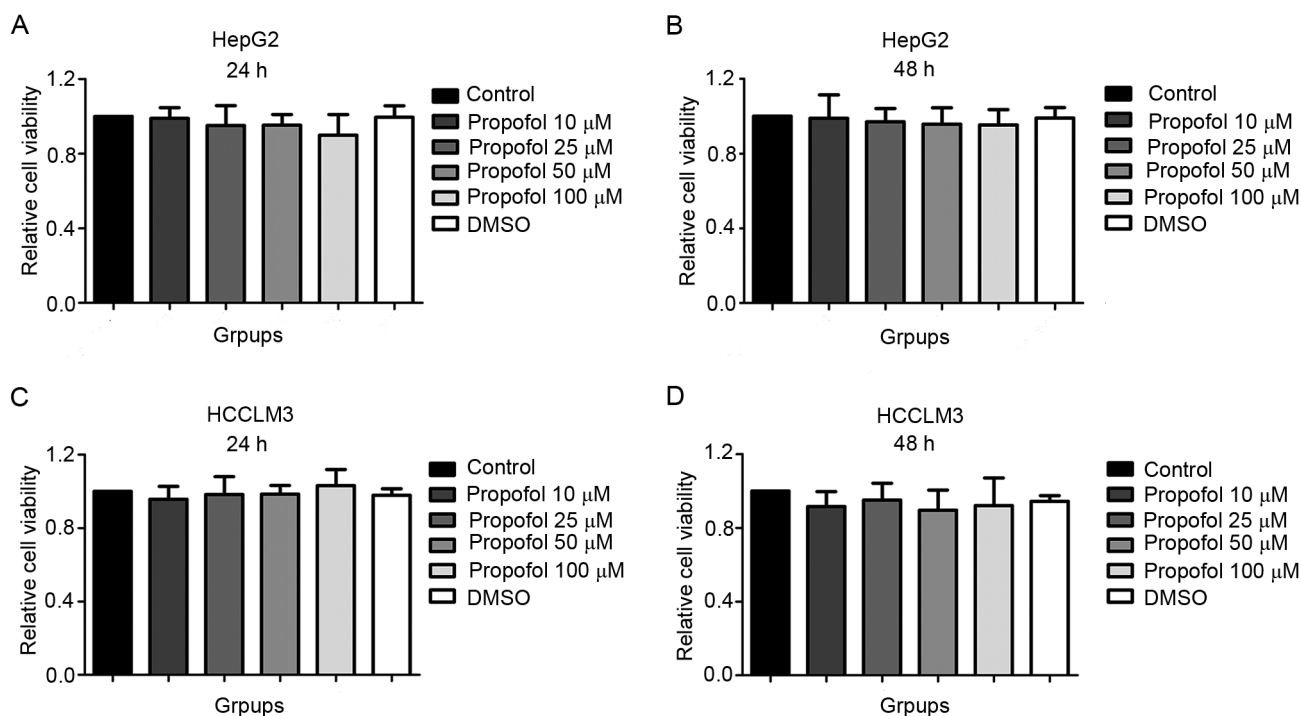


Figure 1. Effect of propofol on the viability of (A and B) HepG2 and (C and D) HCCLM3 cells under normoxia.

Inc.). Following 48 h of incubation at 37°C, the migratory cells in the lower chamber were fixed with 4% paraformaldehyde for 30 min at room temperature and stained with crystal violet solution (cat. no. C0121; Beyotime Institute of Biotechnology) for 30 min at room temperature. The non-migratory cells remaining in the upper chamber were gently removed with a cotton swab. The number of migratory cells was counted in five randomly selected fields of view using an inverted microscope (TS100-F; Nikon Corporation) at a high magnification (x400). Analysis was performed using ImageJ version 1.8.0 software.

Western blotting. HepG2 cells treated with 100, 200 or 300 μM CoCl₂ for 4, 8, 12 or 24 h were lysed on ice with RIPA lysis buffer (cat. no. P0013B; Beyotime Institute of Biotechnology) supplemented with 100 mM phenylmethanesulfonyl fluoride (1%; cat. no. ST506; Beyotime Institute of Biotechnology) and 1% protein phosphatase inhibitor (cat. no. P1260; Applygen Technologies, Inc.). The samples were centrifuged at 12,000 x g for 20 min in a centrifuge (cat. no. 5417R; Eppendorf) at 4°C and the supernatants were collected. The protein concentration was quantified using an enhanced BCA protein assay kit (cat. no. P0010; Beyotime Institute of Biotechnology), according to the manufacturer's protocol, and 30 μg protein/lane was separated via 10% SDS-PAGE. The separated proteins were subsequently transferred onto polyvinylidene fluoride membranes (cat. no. IPVH00010; EMD Millipore) and blocked with 10% non-fat milk for 2 h at room temperature. The membranes were then incubated overnight with the following primary antibodies on a shaker at 4°C: Anti-HIF-1α (rabbit; 1:1,000; cat. no. ab51608; Abcam) and anti-β-tubulin (rabbit; 1:1,000; cat. no. 10068-1-AP; ProteinTech Group, Inc.). Following the primary antibody incubation, the membranes were incubated with a horseradish

peroxidase-conjugated goat anti-rabbit IgG secondary antibody (1:3,000; cat. no. SA00001-2; ProteinTech Group, Inc.) for 1 h at room temperature. Protein bands were visualized using enhanced chemiluminescence western blotting substrates (cat. no. K-12045-D20; Advanta, Inc.). Densitometric analysis was performed using ImageJ version 1.8.0 software.

Statistical analysis. Statistical analysis was performed using GraphPad Prism version 5.0 software (GraphPad Software, Inc.). Data of three or more experimental repeats are presented as the mean ± SD. Data that were normally distributed and had an equal variance were analyzed using a one-way ANOVA, after which a Tukey's test was performed for multiple comparisons. Otherwise, Kruskal-Wallis test was used, followed by Dunn's multiple comparison test. P<0.05 was considered to indicate a statistically significant difference.

Results

Propofol exerts no effect on the viability of HepG2 and HCCLM3 cells under normoxia. The results of the CCK-8 assay revealed that compared with the control (1.00±0.00) and DMSO (0.99±0.06) groups, the viability of HepG2 cells stimulated with 10 (0.99±0.06), 25 (0.95±0.11), 50 (0.95±0.16) or 100 μM (0.90±0.11) propofol for 24 h was not significantly different (Fig. 1A). Similar findings were observed in HepG2 cells following 48 h of propofol stimulation (control, 1.00±0.00; DMSO, 0.99±0.06; 10 μM propofol, 0.99±0.12; 25 μM propofol, 0.97±0.07; 50 μM propofol, 0.96±0.09; 100 μM propofol, 0.95±0.08; Fig. 1B), in HCCLM3 cells following 24 h of propofol stimulation (control, 1.00±0.00; DMSO, 0.98±0.04; 10 μM propofol, 0.96±0.07; 25 μM propofol, 0.98±0.10; 50 μM propofol, 0.98±0.05; 100 μM propofol, 1.03±0.09; Fig. 1C) and in HCCLM3 cells following 48 h of

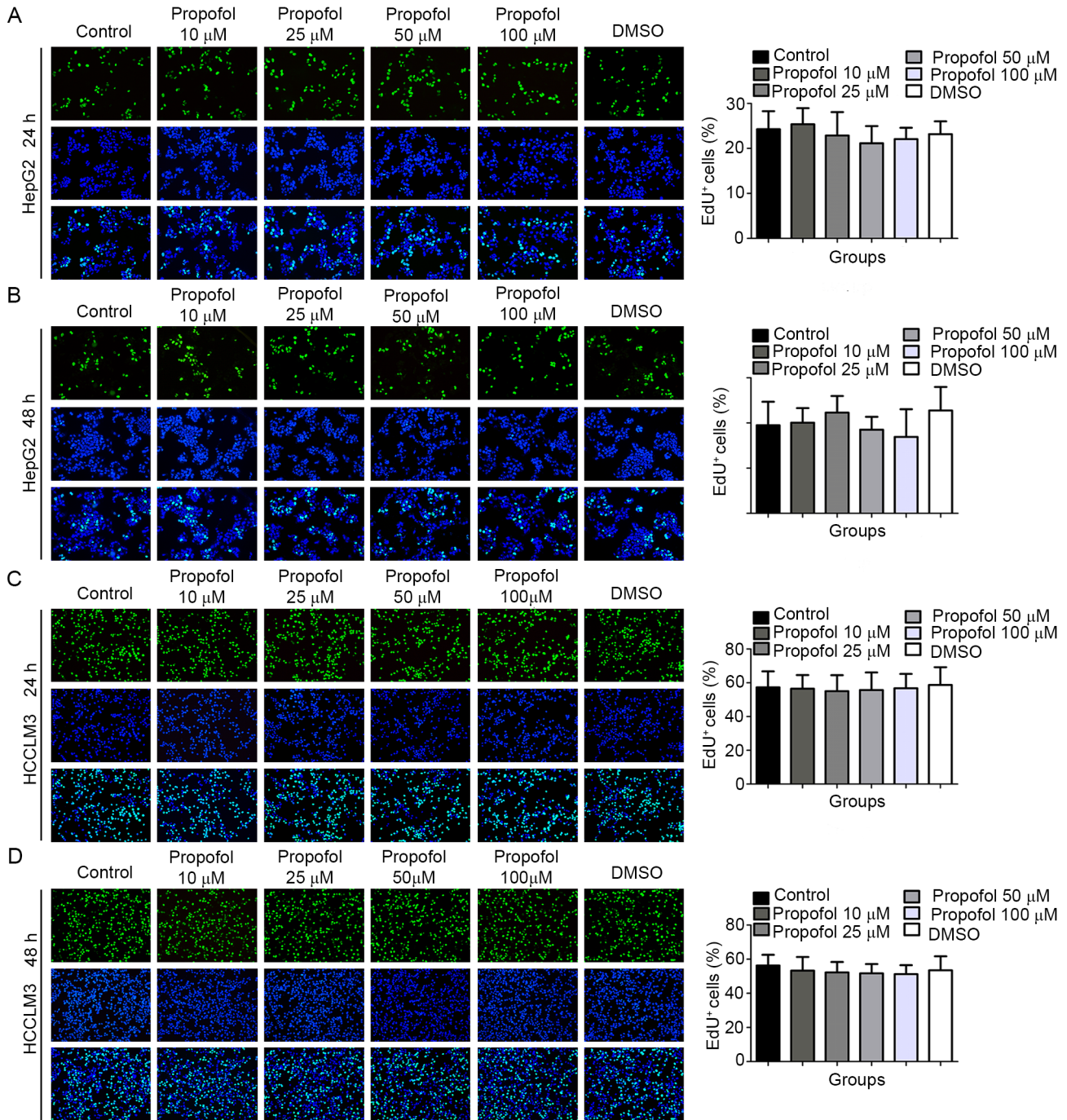


Figure 2. Effect of propofol on the proliferation of (A and B) HepG2 and (C and D) HCCLM3 cells under normoxia. Left panel, representative images (magnification, x200); right panel, statistical analysis of each group. EdU, 5-ethynyl-2'-deoxyuridine.

propofol stimulation (control, 1.00 ± 0.00 ; DMSO, 0.94 ± 0.03 ; 10 μM propofol, 0.92 ± 0.08 ; 25 μM propofol, 0.95 ± 0.09 ; 50 μM propofol, 0.90 ± 0.11 ; 100 μM propofol, 0.92 ± 0.15 ; Fig. 1D). In addition, no statistically significant differences in cell viability were observed between the DMSO and control groups. The above results suggested that propofol exerted no effect on the viability of HepG2 and HCCLM3 cells under normoxic conditions.

Propofol exerts no effect on the proliferation of HepG2 and HCCLM3 cells under normoxia. The results of the EdU assay demonstrated that, compared with the control (24.26 ± 4.01)

and DMSO (23.17 ± 2.84) groups, the percentage of EdU⁺ HepG2 cells following 10 (25.37 ± 3.56), 25 (22.87 ± 5.19), 50 (21.15 ± 3.82) or 100 μM propofol stimulation for 24 h was not statistically significantly different (Fig. 2A). Similar trends were observed in HepG2 cells following 48 h of propofol stimulation (control, 19.48 ± 5.21 ; DMSO, 22.75 ± 5.22 ; 10 μM propofol, 20.07 ± 3.23 ; 25 μM propofol, 22.29 ± 3.67 ; 50 μM propofol 18.52 ± 2.85 ; 100 μM propofol, 16.91 ± 6.13 ; Fig. 2B), in HCCLM3 cells following 24 h of propofol stimulation (control, 57.31 ± 9.45 ; DMSO, 58.69 ± 10.54 ; 10 μM propofol, 56.47 ± 8.12 ; 25 μM propofol, 55.05 ± 9.42 ; 50 μM propofol, 55.72 ± 10.38 ; 100 μM propofol, 56.77 ± 8.49 ; Fig. 2C)

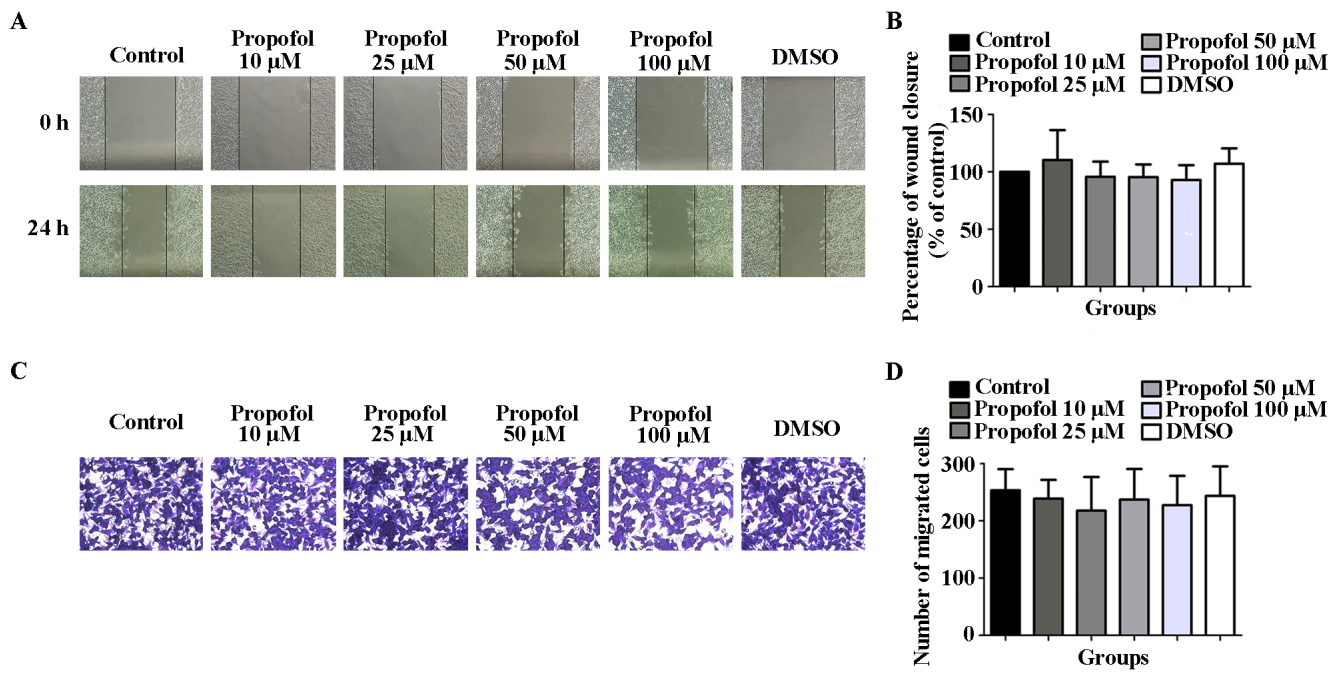


Figure 3. Effect of propofol on the migration of HCCLM3 cells under normoxia. (A) Representative images of the wound healing assay (magnification, x100). (B) Statistical analysis of part (A). (C) Representative images of the Transwell migration assay (magnification, x400). (D) Statistical analysis of part (C).

and in HCCLM3 cells following 48 h of propofol stimulation (control, 56.30 ± 6.29 ; DMSO, 53.51 ± 8.21 ; 10 μM propofol, 53.31 ± 7.98 ; 25 μM propofol, 52.26 ± 6.11 ; 50 μM propofol, 51.69 ± 5.45 ; 100 μM propofol, 51.25 ± 5.25 ; Fig. 2D). The above results indicated that propofol exerted no effect on the proliferation of HepG2 and HCCLM3 cells under normoxic conditions.

Propofol exerts no effect on the migration of HCCLM3 cells under normoxia. The results of the wound healing and Transwell migration assays revealed that, compared with the control (100.0 ± 0.0) and DMSO (107.1 ± 13.37) groups, the wound closure percentage of HCCLM3 cells stimulated with 10 (110.4 ± 25.97), 25 (95.68 ± 13.30), 50 (95.49 ± 10.91) or 100 (92.92 ± 12.83) μM propofol for 24 h was not statistically significantly different (Fig. 3A and B). Similarly, the number of migratory HCCLM3 cells in the propofol groups [10 (239.0 ± 32.72), 25 (217.8 ± 58.77), 50 (237.2 ± 53.40) or 100 (227.5 ± 51.11) μM] following 48 h of stimulation was also not statistically significantly different compared with the control (253.3 ± 36.97) and DMSO (243.6 ± 51.54) groups (Fig. 3C and D). The results suggested that propofol exerted no effect on the migration of HCCLM3 cells under normoxic conditions.

Propofol exerts no effect on the viability and migration of HepG2 cells under CoCl_2 -induced hypoxia. The results of the western blotting analysis indicated that, compared with the control group, the protein levels of HIF-1 α in HepG2 cells treated with 200 and 300 μM CoCl_2 for 24 h were significantly upregulated (1.00 ± 0.00 vs. 1.80 ± 0.60 and 2.12 ± 0.69 , respectively; $P < 0.001$; Fig. 4A and B). In addition, following the treatment of HepG2 cells with 200 μM CoCl_2 , the protein levels of HIF-1 α were upregulated at 4 h and

reached a peak at 12 h ($P < 0.05$ and $P < 0.01$, respectively; Fig. 4C and D), and were subsequently downregulated by 24 h. Therefore, the 12-h time point was selected for subsequent experiments. The effects of propofol on the viability and migration of HepG2 cells under CoCl_2 -induced hypoxia were further investigated. The results demonstrated that compared with the 200 μM CoCl_2 group (1.00 ± 0.00), the viability of HepG2 cells following 12 h of stimulation with 10 (1.07 ± 0.21), 25 (0.99 ± 0.21), 50 (1.01 ± 0.16) or 100 μM (0.99 ± 0.24) propofol in the presence of CoCl_2 was not significantly different (Fig. 4E). Similarly, the wound closure percentage in HepG2 cells following stimulation with 10 (8.35 ± 1.52), 25 (8.55 ± 1.94), 50 (7.24 ± 2.70) or 100 (8.00 ± 2.89) μM propofol for 24 h in the presence of CoCl_2 was also not statistically significantly different compared with the 200 μM CoCl_2 group (7.79 ± 2.01) (Fig. 4F and G). In HCCLM3 cells, pre-experimental results revealed that low concentrations of CoCl_2 (≤ 150 $\mu\text{mol/l}$) failed to induce an increase in HIF-1 α protein levels. An increase in CoCl_2 concentration (>150 $\mu\text{mol/l}$) resulted in morphological changes and the death of HCCLM3 cells (data not shown). The results suggested that the hypoxia model of HCCLM3 cells was not successfully induced by CoCl_2 in this experiment. These above results indicated that propofol exerted no effect on the viability and migration of HepG2 cells under hypoxia induced by CoCl_2 .

Discussion

The main finding of the present study was that different concentrations of propofol (10, 25, 50 and 100 μM) had no effect on the viability, proliferation and migration of HepG2 and HCCLM3 cells *in vitro* under normoxic or CoCl_2 -induced hypoxic conditions. It has been demonstrated that anesthetics

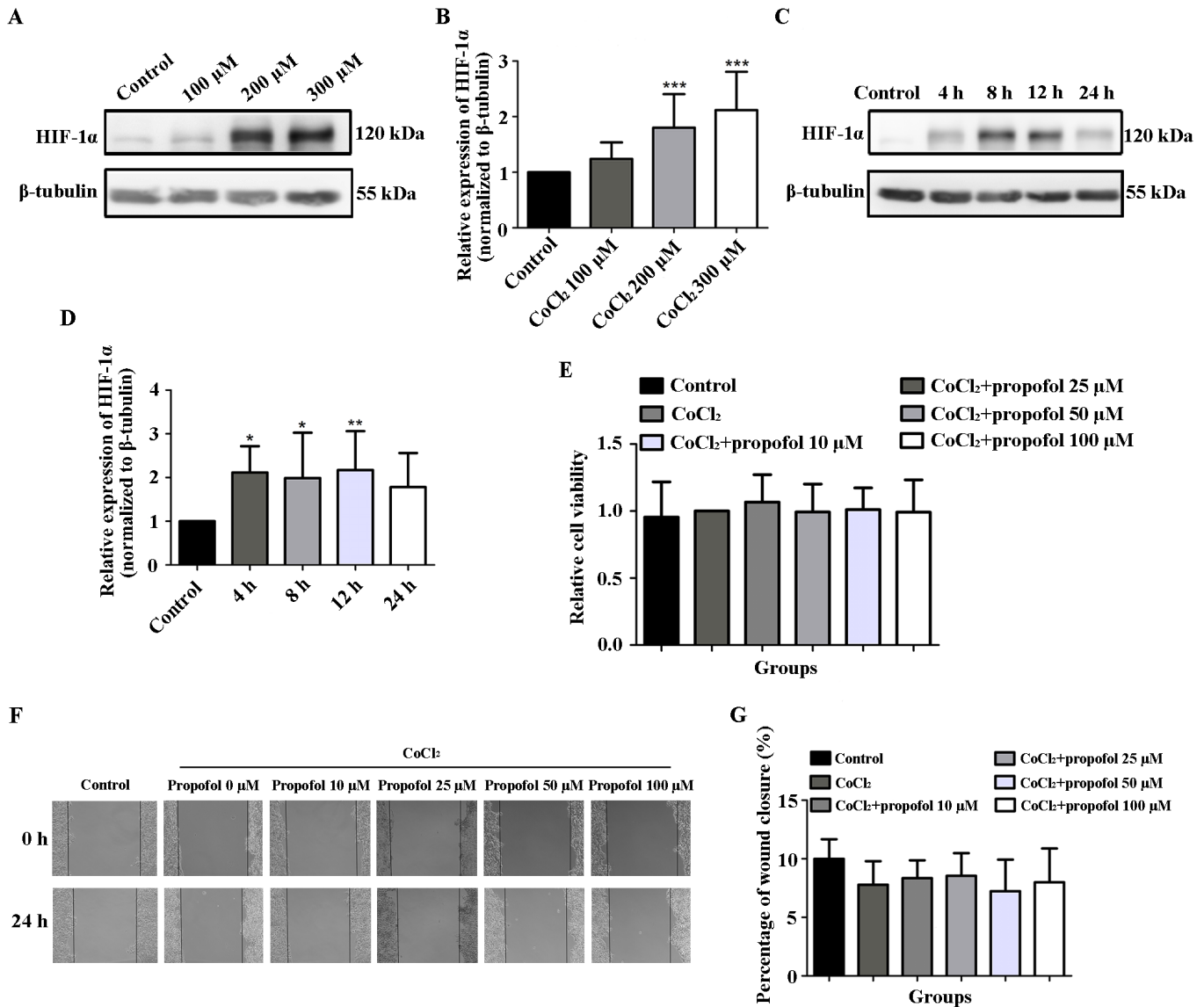


Figure 4. Effect of propofol on the viability and migration of HepG2 cells under CoCl₂-induced hypoxia. HepG2 cells were treated with different concentrations of CoCl₂ (100, 200 or 300 μM) for 24 h. (A) Protein levels of HIF-1α were analyzed. (B) Statistical analysis of the semi-quantification of HIF-1α protein levels in part (A). HepG2 cells were treated with 200 μM CoCl₂ for different durations (4, 8, 12 and 24 h). (C) Protein levels of HIF-1α were analyzed. (D) Statistical analysis of the semi-quantification of HIF-1α protein levels in part (C). HepG2 cells were co-treated with 200 μM CoCl₂ and propofol and (E) cell viability and (F) cell migration (magnification, x100) were determined. (G) Statistical analysis of part (F). *P<0.05, **P<0.01, ***P<0.001 vs. control group. CoCl₂, cobalt chloride; HIF-1α, hypoxia inducible factor-1α.

may affect the long-term prognosis of patients with cancer who have a compromised immune response during major surgery with increased risk of regrowth and metastasis of their malignant tumors (31). Therefore, evaluating the effects of anesthetics on cancer cells could provide a basis for the appropriate use of anesthetics in patients with cancer in order to improve their prognosis. Propofol is one of the most common anesthetics used for both the induction and maintenance of anesthesia in cancer surgery (32), and, undoubtedly, numerous such cases are liver cancer surgery. To date, the effects of propofol on different types of cancer cell cultured *in vitro* remain contradictory (16-18,24,25), and the mechanisms of action of propofol have not been clarified. Therefore, the present study aimed to investigate the effects of propofol on the proliferation and migration of liver cancer cells.

A previous study demonstrated that cancer cells are usually exposed to a hypoxic microenvironment in the body as a result of their rapid proliferation (26). Hypoxia can prompt tumors to metastasize to distant tissues rich in oxygen, thereby facilitating the development of tumor cells with highly aggressive phenotypes (33). One of the most important adaptive responses to hypoxia is the activation of HIF-1α, which can upregulate the expression levels of genes involved in cell proliferation, migration and angiogenesis, and promote the glucose metabolism of tumor cells to shift from oxidative phosphorylation to aerobic glycolysis to provide sufficient energy to meet the needs of rapid tumor proliferation (34). Hypoxia was previously reported to be associated with a poor prognosis in liver cancer (35). Therefore, the present study aimed to investigate the effects of propofol on liver cancer cells under CoCl₂-induced hypoxic conditions. The results

of the present study on the CoCl_2 -induced hypoxia model of HepG2 cells revealed that propofol stimulation had no effect on cell viability and migration.

A previous study found that in prostate cancer PC3 cells, propofol stimulation alone does not alter cell proliferation and migration (21), which is consistent with the findings of the current study. However, contrary to the present findings, numerous studies have demonstrated that propofol exerts significant advantages against liver cancer by decreasing cell proliferation and migration (36-38), while other studies showed that propofol stimulation promotes cancer cell proliferation and migration (24,39). Several other studies have reported that propofol suppresses cell migration without affecting the levels of proliferation (25,40). In contrast to these, Deegan *et al* (41) demonstrated that anesthesia with propofol reduces cell proliferation, but not migration. These aforementioned findings indicated that propofol may not serve an anticancer role in all types of neoplasms. The cell type, propofol concentration and duration of stimulation in the present study were consistent with those implemented in previous studies (37,38). There are several possible explanations for the observed differences between the current results and previous findings. First, owing to the inter- and intratumor heterogeneity and the diversity of liver cancer cells (42), the sensitivity to propofol may vary in different liver cancer cells. Second, it is now widely accepted that tumor proliferation and metastasis are not independent events in tumor cells, as tumors are required to interact with their microenvironment (43). Therefore, changes in the microenvironment of liver cancer cells cultured *in vitro* may affect cell proliferation and migration. Third, propofol may act through a variety of factors such as microRNA (18), transforming growth factor- β (44) and Nrf2 (39). Thus, changes in the expression levels of these factors may also account for the diverse responses of cancer cells to propofol.

There were also several limitations to the present study. For example, the effect of propofol on HIF-1 α protein levels in the CoCl_2 -induced hypoxia model were not investigated. In addition, the effects of propofol on the expression levels of proliferation- and migration-related genes were not determined. The study was also limited because it only included *in vitro* cell experiments, while investigating the effects of propofol in liver cancer model mice *in vivo* or using primary cell cultures from tissue specimens obtained from liver cancer model mice would produce more reliable results and help validate the present findings.

In conclusion, the results of the present study indicated that the anticancer effects of propofol may be questionable and controversial, and further investigations are required to determine the potential of propofol as a targeted anticancer drug. Although several studies have suggested that propofol-based anesthesia may have potential benefits for the survival of cancer patients (45,46), to the best of our knowledge, there is currently no clear evidence to support the selection of propofol as the optimum choice of anesthesia for liver cancer surgery.

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Availability of data and materials

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

Authors' contributions

WS and CL contributed to the design of the study and project administration. CL, QF and JC performed the experiments and analyzed the data. HM performed the statistical analysis. CL interpreted the data. WS, CL and QF confirmed the authenticity of all raw data. CL and WS drafted, reviewed and edited the manuscript. All authors read and approved the final 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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