GRP78 Regulates Apoptosis, Cell Survival and Proliferation in 5-Fluorouracil-resistant SNUC5 Colon Cancer Cells

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Abstract. 5-Fluorouracil (5-FU) is an effective anticancer drug. However, the development of drug resistance has limited its chemotherapeutic efficacy. To address this problem, we investigated the expression of glucose-regulated protein (GRP78, 78 kDa) in 5-FU-resistant colorectal cancer (CRC) cells (SNUC5/5FUR). GRP78 was highly expressed in the SNUC5/5FUR cells compared to wild-type SNUC5 cells. In the presence of 5-FU, GRP78 knockdown induced apoptosis via activation of caspase-3 and poly(ADP-ribose)-polymerase 1. GRP78 also inhibited the production of intracellular reactive oxygen species by regulating stress-associated signaling pathways. Furthermore, GRP78 enhanced cell survival and proliferation via activation of the phosphatidylinositol-3-kinase–AKT–mammalian target of rapamycin axis and cell cycle-associated proteins. These effects were blocked upon GRP78 knockdown, which indicates that GRP78 is involved in the development of 5-FU resistance in these CRC cells. Therefore, a combination of chemotherapy and GRP78-specific targeting may counteract 5-FU resistance in CRC cells.

Colorectal cancer (CRC) is the second leading cause of cancer-related deaths in men and the third leading cause in women (1). Despite improvements in surgical techniques, development of chemotherapy, and molecular targeting therapy, cancer treatment remains a therapeutic challenge. Additionally, drug resistance is a major factor that limits the effectiveness of chemotherapy, which is the main therapeutic strategy for cancer treatment (2). Potential mechanisms of drug resistance include increased drug efflux, mutation of drug targets, defective DNA damage repair, activation of alternative signaling pathways, and evasion from cell death cascades; however, a complete understanding of the mechanisms underlying drug resistance remains unclear (2). Therefore, the discovery and targeting of novel predictive biomarkers has a key role to play in effective CRC therapy.

The 78-kDa glucose-regulated protein (GRP78), also referred to as binding immunoglobulin protein and heat-shock protein family A member 5, belongs to the heat-shock protein 70 family and acts as a stress-inducible molecular chaperone (3). GRP78 also regulates various cellular processes, such as translocation of newly synthesized polypeptides across the endoplasmic reticulum (ER) membrane, regulating calcium homeostasis, acting as an ER stress sensor, and targeting misfolded proteins for ER-associated degradation (4). In cancer cells, GRP78 functions as a multifunctional cell-surface receptor that is involved in promoting cell proliferation and viability (5). Moreover, due to its anti-apoptotic effect, GRP78 is induced in a wide variety of cancer cell lines, including drug-resistant ones (6-8). Notably, GRP78 expression is significantly higher in colon cancer than in colon adenoma and normal tissues (9). GRP78 down-regulation results in colon cancer sensitization to drug-induced apoptosis (10). These observations highlight a potentially important role of GRP78 in cancer therapy.

5-Fluorouracil (5-FU) is widely used as a chemotherapeutic agent in the treatment of a wide variety of cancer types. 5-FU-based therapy significantly increases both the response and survival rates in patients with colon cancer (11). 5-FU causes cytotoxicity in CRC cells via inhibition of thymidylate synthase, incorporation of 5-FU derivatives into DNA, inhibition of RNA synthesis, and genotoxic stress-induced apoptosis (12). Although treatment with 5-FU is beneficial in treating cancer, 5-FU resistance has restricted its therapeutic efficacy in patients with CRC. In order to improve the therapeutic efficacy of 5-FU and elucidate the mechanism(s) underlying the development of 5-FU resistance in CRC cells, we examined the expression of GRP78 in the 5-FU-resistant CRC cell line SNUC5/FUR. In
addition, we explored whether GRP78 regulates CRC cell survival, proliferation, and apoptosis upon 5-FU treatment and the underlying mechanism of 5-FU resistance through regulation of GRP78-mediated apoptosis and proliferation signal pathway.

Materials and Methods

SNUC5 culture. Wild-type (SNUC5) and 5-FU-resistant (SNUC5/5FUR) CRC cells were procured from Chosun University (Gwangju, Republic of Korea). The SNUC5 and SNUC5/5FUR cells were cultured in RPMI-1640 medium (Gibco BRL, Gaithersburg, MD, USA) containing 10% (v/v) fetal bovine serum (FBS, Gibco BRL). Both cell lines were incubated in 5% CO2 at 37°C in a humidified incubator.

Treatment of SNUC5 and SNUC5/5FUR cells. The SNUC5 and SNUC5/5FUR cells were washed twice with phosphate-buffered saline (PBS), and fresh RPMI-1640 medium supplemented with 1% FBS was added. Apoptosis, cell proliferation, and signaling pathway components were investigated after pretreating the cells with 5-FU (140 μM).

Immunocytochemistry. Untreated SNUC5 and SNUC5/5FUR cells were cultured on glass slides in 24-well plates. Next, the cells were washed three times for 5 min each with PBS and fixed with 4% paraformaldehyde (Sigma–Aldrich, St. Louis, MO, USA). Immunofluorescence staining was performed using the primary antibody against GRP78 (Santa Cruz Biotechnology, Santa Cruz, CA, USA) and Alexa-488 secondary antibody (Thermo Fisher Scientific, Waltham, MA, USA). Nuclei were stained with 4′,6′-diamidino-2-phenylindole (DAPI; Sigma–Aldrich). The immunostained samples were observed using a confocal microscope (Olympus, Tokyo, Japan).

Western blot assay. Homogenates of treated and untreated SNUC5 and SNUC5/5FUR cells (total protein, 20 μg) were separated using 8-12% sodium dodecyl sulfate-polyacrylamide gel electrophoresis. Proteins were transferred onto nitrocellulose membranes. After washing the membranes with a solution containing Tris-HCl (10 mM, pH 7.6), NaCl (150 mM), and Tween 20 (0.05%), the membranes were pre-incubated with skimmed milk (5%) for 1 h at room temperature and then incubated with primary antibodies against GRP78, cleaved caspase-3 (C-caspase-3), cleaved poly(ADP–ribose)-polymerase 1 (C-PARP1), p38, phospho-p38 (p-p38), c-JUN N-terminal kinase (JNK), p-JNK, p53, p-p53, ataxia-telangiectasia mutated (ATM), p-ATM, phosphatidylinositol 3-kinase (PI3K), p-PI3K, AKT, p-AKT, mammalian target of rapamycin (mTOR), p-mTOR, cyclin-dependent kinase 2 (CDK2), cyclin E, CDK4, cyclin D1, and β-actin (Santa Cruz Biotechnology). The membranes were washed three times and the primary antibodies were detected using goat anti-rabbit/anti-mouse IgG conjugated with horseradish peroxidase (Santa Cruz Biotechnology). The protein bands were visualized by enhanced IgG conjugated with horseradish peroxidase (Santa Cruz Biotechnology). The membranes were washed three times and the primary antibodies were detected using goat anti-rabbit/anti-mouse IgG conjugated with horseradish peroxidase (Santa Cruz Biotechnology). The protein bands were visualized by enhanced chemiluminescence (GE healthcare, Chicago, IL, USA).

Down-regulation of GRP78 expression by RNA interference. Treated and untreated SNUC5 cells (2×10^5 cells) were seeded in 60-mm dishes and transfected with small interfering RNA (siRNA) in serum-free RPMI-1640 medium (Gibco BRL) using Lipofectamine 2000, according to the manufacturer’s instructions (Thermo Fisher Scientific). At 48-h post-transfection, total protein was extracted and expression of GRP78 was assessed using western blot analysis. The siRNA used for down-regulating GRP78 (si-GRP78) and the scrambled oligonucleotide sequence (si-scr) were synthesized by Bioneer (Daejeon, Korea).

Flow cytometry. To confirm the expression of GRP78, SNUC5 and SNUC5/5FUR cells were incubated with primary antibody to GRP78 for 1 h on ice. After thoroughly washing, the cells were incubated with fluorescent dye-conjugated secondary antibody for 30 min on ice. Next, the cells were washed with PBS. To measure apoptosis, SNUC5/5FUR cells were stained with propidium iodide (PI) and annexin V–fluorescein isothiocyanate (FITC). To investigate the intracellular production of reactive oxygen species (ROS), cells of both lines were stained with dihydroethidium for 30 min at 37°C. To calculate the proportion of cells in each phase of the cell cycle, cells were stained with PI. Each sample was quantitatively analyzed using CyFlow Cube 8 (Sysmex Partec, Münster, Germany) and FSC Express (De Novo Software, Los Angeles, CA, USA).

Statistical analysis. Results were expressed as the mean±standard error of the mean (SEM) and analyzed using one-way analysis of variance (ANOVA). For comparing the means of more than three groups, the Bonferroni–Dunn test was used. Data were considered to be significantly different at values of p<0.05.

Results

Expression of GRP78 in 5-FU-resistant CRC cells. To understand the role of GRP78 in the development of drug resistance in cancer cells, we assessed GRP78 expression in wild-type and 5-FU-resistant SNUC5 CRC cells. Western blot analysis revealed that the level of GRP78 was significantly higher in SNUC5/5FUR cells than in SNUC5 cells (Figure 1A). Flow cytometric analysis showed that the percentage of GRP78-positive cells was significantly higher in SNUC5/5FUR cells than in SNUC5 cells (Figure 1B). Immunofluorescence staining also showed that the expression of GRP78 was significantly higher in SNUC5/5FUR cells than in SNUC5 cells (Figure 1C). These results confirmed high GRP78 expression in the 5-FU-resistant CRC cells.

Effect of GRP78 on apoptosis in CRC cells. It has been reported that the level of GRP78 is high in several cancer cell lines and that it regulates the balance between survival and apoptosis in these cells (3). To understand whether GRP78 regulates apoptosis in CRC cells treated with an anticancer drug, we measured apoptosis and GRP78 expression in SNUC5/5FUR cells. The level of GRP78 significantly decreased in the SNUC5/5FUR cells upon transfection with si-GRP78 (Figure 2A).

After treatment of SNUC5/5FUR cells with 5-FU, the levels of C-caspase-3 and C-PARP1 were analyzed using western blot analysis (Figure 2B). The levels of C-caspase-3 and C-PARP1 significantly increased in the SNUC5/5FUR cells.
cells treated with 5-FU compared to those treated with PBS (Figure 2C). In addition, GRP78 knockdown significantly increased the expression of C-caspase-3 and C-PARP1 in SNUC5/5FUR cells treated with and without 5-FU treatment (Figure 2C). Furthermore, flow cytometric analysis using PI and annexin V–FITC staining revealed that knockdown of GRP78 induced significant apoptosis of SNUC5/5FUR cells upon treatment with 5-FU (Figure 2D and E). These data suggest that the level of GRP78 regulates 5-FU-induced apoptosis of drug-resistant CRC cells.

**GRP78 regulates 5-FU-induced oxidative stress in drug-resistant CRC cells.** Excessive ROS production induces apoptosis (13). We evaluated the effect of GRP78 on ROS production in the drug-resistant CRC cells upon 5-FU treatment using flow cytometry. After treatment with 5-FU for 24 h, ROS production significantly increased in SNUC5/5FUR cells transfected with si-GRP78 compared to those transfected with si-scr (Figure 3A). To confirm whether GRP78 mediates the phosphorylation of oxidative stress-associated proteins in SNUC5/5FUR cells upon 5-FU treatment, we assessed the phosphorylation of p38, JNK, p53, and ATM. Pretreatment with si-GRP78 induced a significant increase in the phosphorylation of oxidative stress-associated proteins in SNUC5/5FUR cells after treatment with 5-FU (Figure 3C-E). These findings indicate that GRP78 controls intracellular ROS production upon 5-FU treatment by regulating the phosphorylation of oxidative stress-associated proteins.

**GRP78 activates the AKT pathway following 5-FU treatment.** The AKT pathway plays a pivotal role in regulating cell survival signals, as well as apoptosis-inducing factors (14). We investigated whether GRP78 is involved in 5-FU-mediated apoptosis by regulating the AKT pathway. For this, we analyzed the phosphorylation of PI3K, AKT, and mTOR in 5-FU-treated SNUC5/5FUR cells (Figure 4A). Pretreatment with si-GRP78 significantly reduced the phosphorylation of PI3K, AKT, and mTOR in 5-FU-treated SNUC5/5FUR cells (Figure 4B). The phosphorylation of PI3K and AKT significantly decreased in si-GRP78-transfected SNUC5/5FUR cells compared to control SNUC5/5FUR cells (Figure 4B). These results reveal that GRP78 is involved in the inhibition of 5-FU-induced apoptosis, by regulating the PI3K–AKT–mTOR signaling pathway.

**GRP78 is involved in the proliferation of 5-FU-resistant CRC cells.** To explore the effect of GRP78 on the proliferation of 5-FU-resistant CRC cells, the expression of cell cycle-associated proteins CDK2, cyclin E, CDK4, and cyclin D1 were assessed in SNUC5/5FUR cells (Figure 5A). Treatment with 5-FU significantly reduced the levels of CDK2, cyclin E, CDK4, and cyclin D1 were assessed in SNUC5/5FUR cells (Figure 5A). Treatment with 5-FU significantly reduced the levels of CDK2, cyclin E, CDK4, and cyclin D1 in SNUC5/5FUR cells (Figure 5B). In addition, pretreatment with si-GRP78 induced a significant decrease in the expression of these proteins in 5-FU-treated SNUC5/5FUR cells (Figure 5B). Furthermore, flow cytometric analysis of PI-stained 5-FU-treated SNUC5/5FUR cells showed that GRP78 knockdown led to a significant decrease in the proportion of cells in the S-phase (Figure 5C and D). These results indicate that
Figure 2. Effect of glucose-regulated protein (GRP78) on 5-fluorouracil (5-FU)-induced apoptosis of and 5-fluorouracil-resistant (SNUC5/SUR) colorectal cancer cells. A: Western blot analysis of GRP78 in SNUC5/SUR cells after transfection with scrambled (si-scr) or GRP78 siRNA (si-GRP78). The expression of GRP78 was normalized relative to that of β-actin. Values represent the mean±standard error of the mean (SEM). **Significantly different at p<0.01 compared si-scr. B,C: Western blot and densitometric analysis of cleaved caspase-3 (C-caspase-3) and cleaved poly(ADP ribose)-polymerase 1 (C-PARP1) expression after transfection of SNUC5/SUR cells with si-scr or si-GRP78, followed by treatment with phosphate-buffered saline (PBS, control) or 5-FU (140 μM) for 48 h. Significantly different at *p<0.05 and **p<0.01 compared to PBS-treated SNUC5/SUR cells pretreated with si-scr; #p<0.05 and ## p<0.01, compared with PBS-treated SNUC5/SUR cells pretreated with si-GRP78; $p<0.05 and $$ p<0.01 compared to 5-FU-treated SNUC5/SUR cells pretreated with si-scr. D: Apoptosis was measured by flow cytometry using propidium iodide (PI) and annexin V–fluorescein isothiocyanate staining. E: Percentage of apoptotic cells in different treatment groups. Values represent the mean±SEM. Significantly different at **p<0.01 compared to PBS-treated SNUC5/SUR cells pretreated with si-scr; *p<0.05 and **p<0.01 compared with PBS-treated SNUC5/SUR cells pretreated with si-GRP78; $p<0.01 compared to 5-FU-treated SNUC5/SUR cells pretreated with si-scr. The expression levels were determined by densitometry, and normalized relative to the expression level of β-actin.
Figure 3. Glucose-regulated protein (GRP78) regulates reactive oxygen species (ROS) production and stress-associated signaling pathway components in 5-fluorouracil (5-FU)-treated 5-fluorouracil-resistant (SNUC5/5FUR) colorectal cancer cells. A: Flow cytometric analysis of dihydroethidium-stained cells to detect intracellular ROS production in SNUC5/5FUR cells pretreated with scrambled (si-scr) or GRP78 siRNA (si-GRP78), and subsequently treated with 5-FU for 48 h. Grey lines indicate the negative control. Black lines indicate the cells stained with dihydroethidium. B, C: Western blot and densitometric analysis illustrating the levels of phosphorylated p38 (p-p38) and c-JUN N-terminal kinase (p-JNK) in SNUC5/5FUR cells transfected with si-scr or si-GRP78 and subsequently treated with phosphate-buffered saline (PBS) or 5-FU (140 μM) for 48 h. Significantly different at *p<0.05 and **p<0.01 compared to PBS-treated SNUC5/5FUR cells pretreated with si-scr; ^p<0.05 and ^#p<0.01 compared to PBS-treated SNUC5/5FUR cells pretreated with si-GRP78; §§p<0.01 compared with 5-FU-treated SNUC5/5FUR cells pretreated with si-scr. D, E: Western blots and densitometric analysis showing the expression levels of phosphorylated p53 (p-p53) and ataxia-telangiectasia mutated (p-ATM) in SNUC5/5FUR cells transfected with si-scr or si-GRP78 and subsequently treated with PBS or 5-FU (140 μM) for 48 h. Significantly different at p<0.01 compared to PBS-treated SNUC5/5FUR cells pretreated with **si-scr; or ## si-GRP78; §§p<0.01 compared to 5-FU-treated SNUC5/5FUR cells pretreated with si-scr. The expression levels were determined by densitometry, and normalized relative to the expression level of β-actin. Values represent the mean±SEM.
GRP78 is involved in the proliferation of 5-FU-resistant CRC cells by regulating the expression of cell cycle-associated proteins.

**Discussion**

In this study, we showed that the expression of GRP78 was high in 5-FU-resistant CRC cells, resulting in the augmentation of cell survival and proliferation post-5-FU treatment. In addition, GRP78 controlled the production of ROS in response to 5-FU-mediated oxidative stress. Knockdown of GRP78 induced apoptosis and inhibited proliferation of SNUC5/5FUR cells by regulating the apoptosis-, survival-, and proliferation-associated signaling pathways.

GRP78 is known to be highly expressed in tumor cells. It plays an important role in tumor cell proliferation, angiogenesis, metastasis, and resistance to anticancer drugs (15). In particular, GRP78 is involved in the development of therapeutic resistance in several types of cancer cell. High expression of GRP78 regulates anti-estrogen resistance in breast cancer cells (16). Elevated levels of GRP78 are also associated with trastuzumab and doxorubicin resistance in breast cancer cells (17). In ovarian cancer cells, GRP78 had...
Figure 5. Glucose-regulated protein (GRP78) is involved in proliferation of 5-fluorouracil (5-FU)-treated 5-fluorouracil-resistant (SNUC5/SFU) colorectal cancer cells via regulation of cell cycle-associated proteins. A, B: Western blot and densitometric analysis for analyzing the levels of cyclin-dependent kinase 2 (CDK2), cyclin E, CDK4, and cyclin D1 in SNUC5/SFU cells transfected with scrambled (si-scr) or GRP78 siRNA (si-GRP78), and subsequently treated with phospho buffered saline (PBS) or 5-FU (140 μM) for 48 h. Significantly different at *p<0.05 and **p<0.01 compared with PBS-treated SNUC5/SFU cells pretreated with si-scr; ##p<0.01 compared to PBS-treated SNUC5/SFU cells pretreated with si-GRP78; $$ p<0.01 compared to 5-FU-treated SNUC5/SFU cells pretreated with si-scr. The expression levels were determined by densitometry, and normalized relative to the expression level of β-actin. C, D: Flow cytometric analysis for PI staining to assess G0/G1, S, and G2/M phase populations in SNUC5/SFU cells treated with si-scr or si-GRP78, and subsequently treated with 5-FU. Significantly different at *p<0.05 and **p<0.01 compared with PBS-treated SNUC5/SFU cells pretreated with si-scr; $$$p<0.01 compared with PBS-treated SNUC5/SFU cells pretreated with si-GRP78; $$$p<0.01 compared with 5-FU-treated SNUC5/SFU cells pretreated with si-scr. Values represent the mean±SEM.
an anti-senescence effect against cisplatin treatment via regulation of the ATM pathway and induction of calcium efflux from the ER to the cytoplasm (18). These findings suggest that GRP78 could be targeted for overcoming the problem of drug resistance in cancer cells.

Recent studies have suggested that an elevated GRP78 level can protect cancer cells from the cytotoxic effects of chemotherapeutic agents, such as doxorubicin, taxol, and 5-FU (8, 19, 20). We observed that GRP78 knockdown resulted in the increase of apoptosis of 5-FU-treated SNUC5/5FUR cells via activation of the apoptosis-associated signaling proteins, such as caspase-3 and PARP1. In addition, GRP78 regulated the production of intracellular ROS by modulating the stress-associated signaling pathway proteins, including p38, JNK, p53, and ATM. In Chinese hamster ovary, human leukemia, and bladder carcinoma cell lines, GRP78 overexpression induced resistance to cytotoxic drugs (such as topoisomerase inhibitors) and induced apoptosis via suppression of caspase-7 activation (7). In the DLD-1 colon cancer cells, GRP78 knockdown enhanced apoptosis and intracellular ROS production via down-regulation of the AKT pathway and activation of protein phosphatase 2A, following epirubicin treatment (21). In glial cells, GRP78 enhanced cellular antioxidant levels through suppression of lipid peroxidation, following hydrogen peroxide exposure (22). These findings indicate that 5-FU-mediated apoptosis is regulated by GRP78. This suggests that 5-FU-induced increase in GRP78 expression results in the inhibition of 5-FU resistant CRC cells via regulation of intracellular ROS production.

The PI3K–AKT–mTOR signaling axis plays an important role in cell survival and growth under various physiological and pathological conditions (23). In particular, this pathway plays a key role in regulating cancer biology, since cancer cells usually reside in harsh environments, such as with low oxygen, pH, and nutrient supply (23). Our results demonstrated that the PI3K–AKT–mTOR signaling pathway was activated in SNUC5/5FUR cells and knockdown of GRP78 reduced the phosphorylation of PI3K, AKT, and mTOR. GRP78 acts as a co-receptor or signal-transducing receptor for soluble ligands, including α2-macroglobulin, tumor differentiation factor, vaspin, T-cadherin, and Cripto (24). The binding of GRP78 to these ligands promotes the activation of the PI3K–AKT pathway, resulting in increased cell survival, adhesion, and migration (24). The resistance of breast and prostate cancer cells to chemotherapy is associated with the localization of GRP78 to the cell surface, where it binds to PI3K, which is an activator of AKT (25). Overexpression of GRP78 enhances the formation of phosphatidylinositol (3,4,5)-trisphosphate (PIP3), which is present downstream of PI3K; however, mutation of the N-terminal region of GRP78 reduces the binding of GRP78 to PI3K and PIP3 (25). In prostate cancer cells, blocking of cell surface GRP78 by anti-GRP78 reduced the growth of cells via inhibition of the PI3K–AKT pathway (26). In addition, GRP78 promoted breast cancer cell survival via activation of mTOR-regulated pro-survival autophagy. Our results suggest that the 5-FU-induced increase in GRP78 expression promotes survival of 5-FU resistant CRC cells via regulation of the PI3K–AKT–mTOR signaling axis.

We also revealed that the 5-FU-induced increase in GRP78 expression increased the proliferation of SNUC5/5FUR cells; however, GRP78 knockdown inhibited cell proliferation by regulating expression of cell cycle-associated proteins CDK2, cyclin E, CDK4, and cyclin D1. GRP78 was found to enhance cell proliferation and survival during mouse embryonic development (27). Overexpression of GRP78 increased cell proliferation in chondrocyte development by enhancement of the S-phase population (28). In breast cancer cells, GRP78 facilitated cell proliferation through activation of the Janus kinase 2/signal transducer and activator of transcription 3 pathway (29). In addition, inhibition of GRP78 and GRP94 reduced cellular proliferation in a human gastric cancer cell line (30). These findings suggest that the 5-FU-induced increase in GRP78 expression plays a key role in the development of 5-FU resistance in CRC cells.

Taken together, our findings demonstrate that drug-induced expression of GRP78 prevents apoptosis and production of intracellular ROS. It also enhances cell survival and proliferation of drug-resistant CRC cells via regulation of the apoptosis-, survival-, and cell cycle-associated signaling pathways. These findings indicate that GRP78 could be an important biomarker for drug-resistant CRC. In conclusion, the development of resistance in colon cancer cells against chemotherapeutic agents might be combated by targeting GRP78. This may be used as a novel strategy for treating colon cancer.

Conflicts of Interest

The Authors have no conflicts of interest to declare with regards to this study.

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References


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