European Review for Medical and Pharmacological Sciences 2018; 22: 5364-5370

Fulvestrant inhibits the glycolysis of prolactinoma GH3 cells by downregulating IRE1/XBP1 signaling pathway

C. WANG[1], J.-L. XU[2], Y. WEN[1], D.-Z. ZHANG[1], X. WANG[1], L. CHANG[1], G.-F. LI[1], L.-Y. XIE[1], J. SU[1], X.-X. ZHANG[1], C.-L. TAN[1]

[1]Department of Neurosurgery, Harbin Medical University Cancer Hospital, Harbin, Heilongjiang Province, China
[2]Department of Internal Medicine-Oncology, Harbin Medical University Cancer Hospital, Harbin, Heilongjiang Province, China

Abstract. – OBJECTIVE: We aimed to evaluate the effects of fulvestrant on the glycolysis of prolactinoma GH3 cells, and reveal the potential regulatory mechanisms.

MATERIALS AND METHODS: Prolactinoma cell line GH3 was treated with different concentrations of fulvestrant (0, 0.12, 0.25, 0.5 and 1 ng/ml) for 4 h. siRNAs XBP1s and XBP1u were constructed to treat GH3 cells. The expression levels of XBP1s, XBP1u, IRE1, PKM2 and GRP78 of GH3 cells were detected by Western blot. Meanwhile, the glycolytic activity of GH3 cells, including the glucose uptake, ATP/ADP, and lactate production were detected.

RESULTS: The expression levels of XBP1s and XBP1u were significantly inhibited by fulvestrant in a dose-dependent manner. The glucose uptake, ATP/ADP and lactate production of GH3 cells were significantly inhibited by fulvestrant as well as siRNA XBP1s and XBP1u (p < 0.05). Western blot analysis suggested that the expression levels of IRE1, PKM2 and GRP78 were significantly decreased in GH3 cells treated by fulvestrant as well as siRNA XBP1s and XBP1u, compared with those in normal control ($p < 0.05$).

CONCLUSIONS: Fulvestrant could inhibit the glycolysis of GH3 cells by downregulating IRE1/XBP1 signaling pathway, and this process was closely related with the downregulation of PKM2.

Key Words:
Fulvestrant, Prolactinoma, Glycolysis, IR.

Introduction

Prolactinoma is a noncancerous tumor frequently occurring in pituitary gland[1]. It is usually accompanied with amenorrhea, galactorrhea and infertility in females, and sexual dysfunction and sellar mass in males[2]. Bromocriptine is a dopamine agonist, which is commonly used in treating prolactinoma. It can effectively suppress the prolactin secretion and reduce the tumor size[3]. However, nearly 10% prolactinoma patients are resistant to bromocriptine[4]. Therefore, searching of novel molecular therapies for prolactinoma is urgently needed.

As an important source of ATP, active glycolytic metabolism is a significant biochemical feature of malignant tumors[5]. Tumor cells prefer glycolysis especially under aerobic conditions[6]. Therefore, inhibition of glycolysis has become an effective strategy in the treatment of tumors. It has been reported that methyl jasmonate can induce necrosis and apoptosis of hepatocellular carcinoma cells via the inhibition of glycolysis[7]. The cell proliferation in colorectal cancer is inhibited by miR-1 through the inhibition of SMAD3-mediated glycolysis[8]. Extracellular matrix protein Reelin promotes myeloma progression by facilitating tumor glycolysis[9]. These results suggested that the inhibition of glycolysis may exhibit great potential in the treatment of prolactinoma. The antiestrogen agents exhibit suppressive effects on prolactinoma[10]. Fulvestrant, an estrogen receptor (ER) antagonist, is commonly used in the treatment of hormone receptor (HR)-positive metastatic breast cancer[11]. Meanwhile, the inhibitory role of fulvestrant on prolactinoma has also been revealed by various researches. For example, it has been reported that fulvestrant can inhibit the proliferation and prolactin secretion of prolactinoma MMQ and GH3 cells *in vitro*[12]. The tumor proliferation and prolactin secretion are inhibited by fulvestrant

Corresponding Author: Chunlei Tan, MD; e-mail: wggdgdg233@sina.com

on F344 rat models of prolactinoma[13]. However, the inhibitory effects and mechanisms of fulvestrant on glycolysis of prolactinoma cells have not been revealed.

In this study, the effects of fulvestrant on the glycolysis of prolactinoma GH3 cells were evaluated. Meanwhile, siRNA XBP1s and XBP1u were constructed to analyze the potential action mechanisms of fulvestrant with XBP1. Our findings might be useful to reveal the molecular mechanisms involved in the inhibitory effects of fulvestrant on the glycolysis of prolactinoma cells.

Materials and Methods

Cell Culture and Treatments

Prolactinoma cell line GH3 (purchased from Sun Yat-Sen University, Guangzhou, Guangdong, China) was maintained in Dulbecco's Modified Eagle Medium (DMEM, BD Pharmingen, San Diego, CA, USA) containing 10% fetal bovine serum (FBS) at 37°C with 5% CO_2. GH3 cells (1×10^5/ml) were inoculated in 96-well plate, and fulvestrant (No. 129453-61-8, Hangzhou Jihan Chemical Co., Ltd., Zhejiang, China) at the concentrations of 0.12, 0.25, 0.5 and 1 ng/ml was added into each well with three replicates for each concentration. After 4 h of culturing at 37°C, cells were collected for Western blot and glycolysis analysis. Cells without the treatment of fulvestrant were considered as the control (0 ng/ml).

Vectors Construction and Transfection

Two siRNA vectors of XBP1s and XBP1u were constructed. Simply, special complementation fragments of XBP1s and XBP1u were amplified via PCR with specific primers (XBP1s-F: 5'-GTTAGGATCCCGCCATGACCCCGCAGCTTCTCCTGGCCC-3', XBP1s-R: 5'-ACCTCCACCTGAACCTCCACCACCCGAACCACCTCCACCCTTG CCCAGGCTCATTGTGGC-3'; XBP1u-F: 5'-GATAGGAACCCGCCA TGACGGCGCAGCGTCTCCTGGCCG-3', XBP1u-R: 5'-GCCTCCACCTACCTGCACCACCCGTACCACCTCCACCCTTGCAAAGCTCATCCTGGC-3') by using the cDNA library of human myeloid leukemia cell line KG-I as template. PCR products were purified by 1% agarose gel electrophoresis, and inserted into the vector of pcDNA3.1(+) (conserved in our laboratory) using T4 ligase (TaKaRa, Dalian, China) (Restriction site: BamH I and XbaI). After transformed into DH5a (conserved in our laboratory), positive clones were identified by sequencing (Invitrogen, Carlsbad, CA, USA). Before transfection, a total of 2×10^5 GH3 cells were seeded in 6-well plates and cultured for 24 h. Next, these cells were transfected with siRNA XBP1s and XBP1u by using Lipofectamine 2000 (Invitrogen, Carlsbad, CA, USA) according to the manufacturer's protocol. GH3 cells transfected with empty pcDNA3.1(+) were considered as control.

Western Blot

The total proteins of GH3 cells in different groups were extracted by lysis buffer. After 10 min of centrifugation (12000 rpm) at 4°C, the proteins were separated by SDS-polyacrylamide gel electrophoresis (PAGE) on 12% polyacrylamide gels, and transferred to a polyvinylidene-fluoride membrane (PVDF, Millipore, Billerica, MA, USA). The membrane was blocked with 5% skim milk for 1 h, and special diluted primary antibodies (anti-XBP1, 1:500, LSBio, Shanghai, China; anti-IRE1, 1:1000, Novus, CT, USA; anti-PKM2, 1:500, Abcam, Cambridge, MA, USA; anti-GRP78, 1:500, Abcam, Cambridge, MA, USA) were added and incubated overnight at 4°C. After washed with Tris-buffered saline with 1% and Tween-20 (TBST-20) for three times, horseradish peroxidase (HRP)-conjugated goat anti-rabbit antibody (1:5000, Hangzhou Huaan Biotechnology Co., Ltd., Zhejiang, China) was added and incubated for 1 h at 25°C. The samples were finally washed with TBST for 3 times, and the protein brands were colored by enhanced-chemiluminescence (ECL) reagent (Hangzhou Huaan Biotechnology Co., Ltd., Zhejiang, China). The expression levels of proteins were quantified by using a UV gel imager (FluorChem M, Globalebio Co., Ltd., China).

Glucose Uptake Detection

A total of 2×10^5 GH3 cells were seeded in 12-well plates, and cultured for 24 h in serum-free DMEM at 37°C with 5% CO_2. After washed with phosphate-buffered saline (PBS), cells were cultured for 2 h in DMEM containing 1% FBS. Then, these cells were incubated in 1 ml PBS containing 200 nmol/l insulin for 30 min, followed by 1 μCi/ml 2-deoxy-[3H]-D-glucose for 5 min. Subsequently, 20 μmol/l cytochalasin B were immediately added into these cells to avoid intracellular diffusion of glucose. After washed with precooled PBS for 3 times, cells

were lysed by 10 min of incubation with 400 µl 1% SDS. The cell lysate was gathered in a glass fiber filter paper by cell harvester, and the dried filter paper was put in a measuring bottle. Next, after adding 4 ml scintillation solutions, the glucose uptake of GH3 cells was determined by the counts per minute (CPM) on a liquid scintillation counter (LSC-7200, Hitachi, Tokyo, Japan).

ATP/ADP Detection

A total of 1×10^6 GH3 cells in different groups were cultured for 24 h in Dulbecco's Modified Eagle Medium (DMEM) at 37°C with 5% CO_2. After being centrifuged at 800 rpm for 5 min and washed twice with PBS, cells were collected and counted. The cell membrane was broken by 10 min of incubation with 500 µl perchloric acid (0.1 mmol/l). Then, 250 µl supernatant were collected by 20 min of centrifugation at 1500 rpm, followed by mixing with 50 µl Na_2CO_3 (0.5 mmol/L) After 10 min of centrifugation at 1500 rpm, 50 µl supernatant were used for detection. The contents of ATP/ADP were detected by HPLC with hypersil C18 (250 mm×4.6 mm, 5 µm). HPLC conditions included 20 µl sample size, 25°C temperature, 0.1 mmol/l KH2PO4 (pH 6.0) mobile phase, 0.9 ml/min liquid flow and 254 nm detection wavelength.

Lactate Production Detection

A total of 2×10^6 GH3 cells in different groups were cultured for 48 h in DMEM at 37°C with 5% CO_2. After 10 min of centrifugation at 800 rpm, 50 µl supernatant were added into 96-well plates, and then 50 µl reaction mixture (46 µl buffer, 2 µl substrate and 2 µl lactolase) were added into each well. Followed by 30 min of incubation, the optical density at 450 nm (OD450) of each well was detected by using a Microplate Reader (HBS-1096A, DeTie, Nanjing, China). The concentrations of lactate (C) produced by GH3 cells were calculated as followed: C = La/Sv (nmol/µl) where La represents the content of lactate calculated by a standard curve, and Sv represents the sample volume.

Statistical Analysis

All data were expressed as mean ± Standard Deviation (SD). Statistical analysis was performed by SPSS version 17.0 (SPSS Inc., Chicago, IL, USA). Comparison between different groups was determined by *t*-test. *p*-value < 0.05 was considered to be significantly different.

Results

The Expression Levels of XBP1s and XBP1u were Inhibited by Fulvestrant in GH3 Cells

The effects of fulvestrant on the expression of XBP1s and XBP1u were evaluated on GH3 cells. The expression levels of XBP1s and XBP1u were significantly inhibited by fulvestrant in a dose-dependent manner ($p < 0.05$) (Figure 1). When the concentration of fulvestrant reached to 1 ng/ml, approximately 5 times of reduction was observed on the expression levels of XBP1s and XBP1u (Figure 1).

The Glycolytic Activity of GH3 Cells was Inhibited by Fulvestrant through Downregulating XBP1s and XBP1u

In order to reveal the relations between fulvestrant and the expression of XBP1, siRNA XBP1s and XBP1u were transformed into GH3 cells. Western blot showed that the expression levels of XBP1s and XBP1u in GH3 cells were significantly reduced by siRNA XBP1s and XBP1u (28% and 37%, $p < 0.05$).

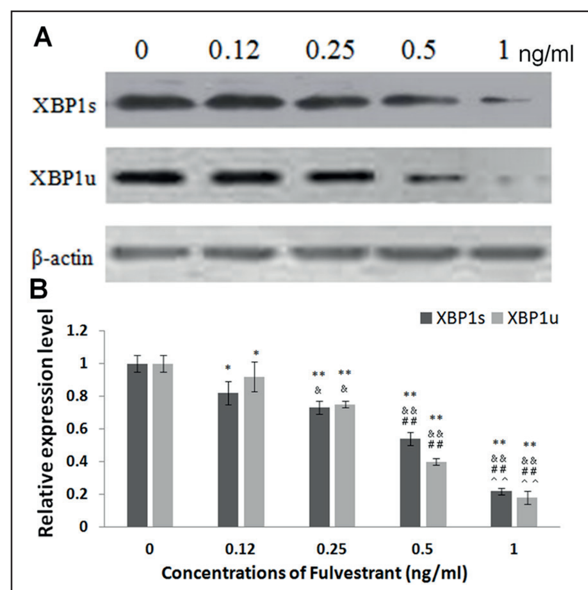

Figure 1. The expression of XBP1s and XBP1u on GH3 cells treated by different concentrations of fulvestrant (0, 0.12, 0.25, 0.5 and 1ng/ml) detected by Western blot (***A*** and ***B***). * and ** represented significantly different at $p < 0.05$ and 0.01 when compared with normal control (0 ng/ml). & and && represented significantly different at $p < 0.05$ and 0.01 when compared with GH3 cells treated with 0.12 ng/ml Fulvestrant. ## and ^^ represented significantly different at $p < 0.05$ when compared with GH3 cells treated with 0.25 and 0.5 ng/ml fulvestrant, respectively.

After the treatment of fulvestrant, the glucose uptake, ATP/ADP and lactate production of GH3 cells were significantly inhibited in a dose-dependent manner ($p < 0.05$). At the concentration of 1 ng/ml, the glucose uptake, ATP/ADP and lactate production were reduced by fulvestrant for about 40%, 45% and 40%, respectively, compared with those in normal control (0 ng/ml) (Figure 2A). In consistent with fulvestrant, significantly reduced glucose uptake, ATP/ADP and lactate production were also observed on GH3 cells treated by siRNA XBP1s and XBP1u ($p < 0.05$). Meanwhile, there was no significant difference on glucose uptake, ATP/ADP and lactate production between GH3 cells treated by siRNA XBP1s/XBP1u and those treated with 1 ng/ml fulvestrant (Figure 2B). Since fulvestrant could inhibit the expression levels of XBP1s and XBP1u, these results indicated that the inhibitory effect of fulvestrant on the glycolytic activity of GH3 cells might be related with the downregulation of XBP1s and XBP1u.

The Expression Levels of IRE1, PKM2 and GRP78 in GH3 were Inhibited by Fulvestrant through Downregulating XBP1s and XBP1u

In order to further investigate the regulatory mechanisms of fulvestrant on GH3 cells, the protein expression of IRE1, PKM2 and GRP78 was detected by Western blot. The expression levels of IRE1, PKM2 and GRP78 were significantly decreased by fulvestrant in GH3 cells in a dose-dependent manner ($p < 0.05$) (Figure 3).

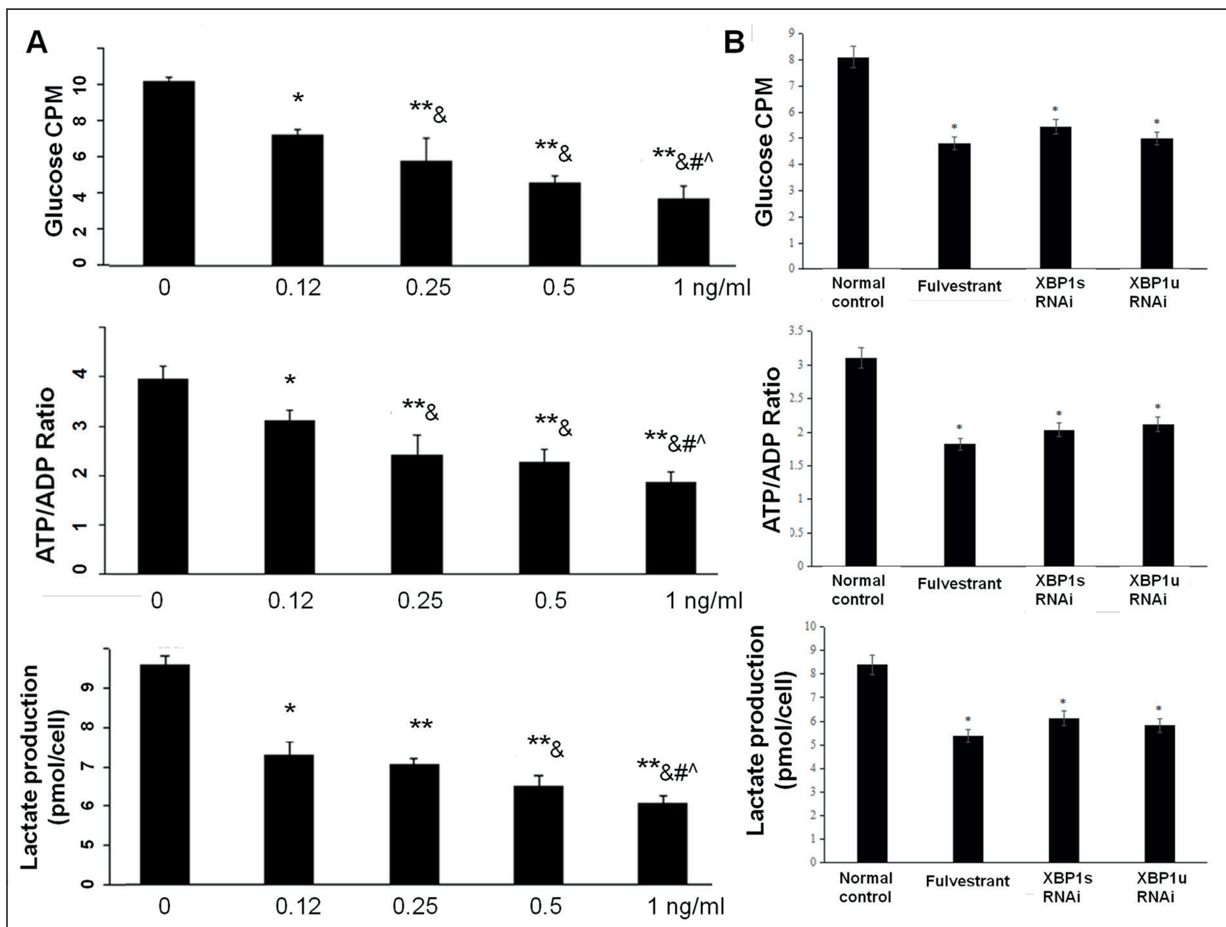

Figure 2. The glucose uptake, ATP/ADP and lactate production of GH3 cells. ***A)*** GH3 cells treated by different concentrations of fulvestrant (0, 0.12, 0.25, 0.5 and 1ng/ml). * and ** represented significantly different at $p < 0.05$ and 0.01 when compared with normal control (0 ng/ml). &, # and ^ represented significantly different at $p < 0.05$ when compared with GH3 cells treated with 0.12, 0.25 and 0.5 ng/ml fulvestrant, respectively. ***B)*** GH3 cells treated by fulvestrant (1 ng/ml), siRNA XBP1s and XBP1u. * represented significantly different at $p < 0.05$ when compared with GH3 cells treated with 1 ng/ml respectively.

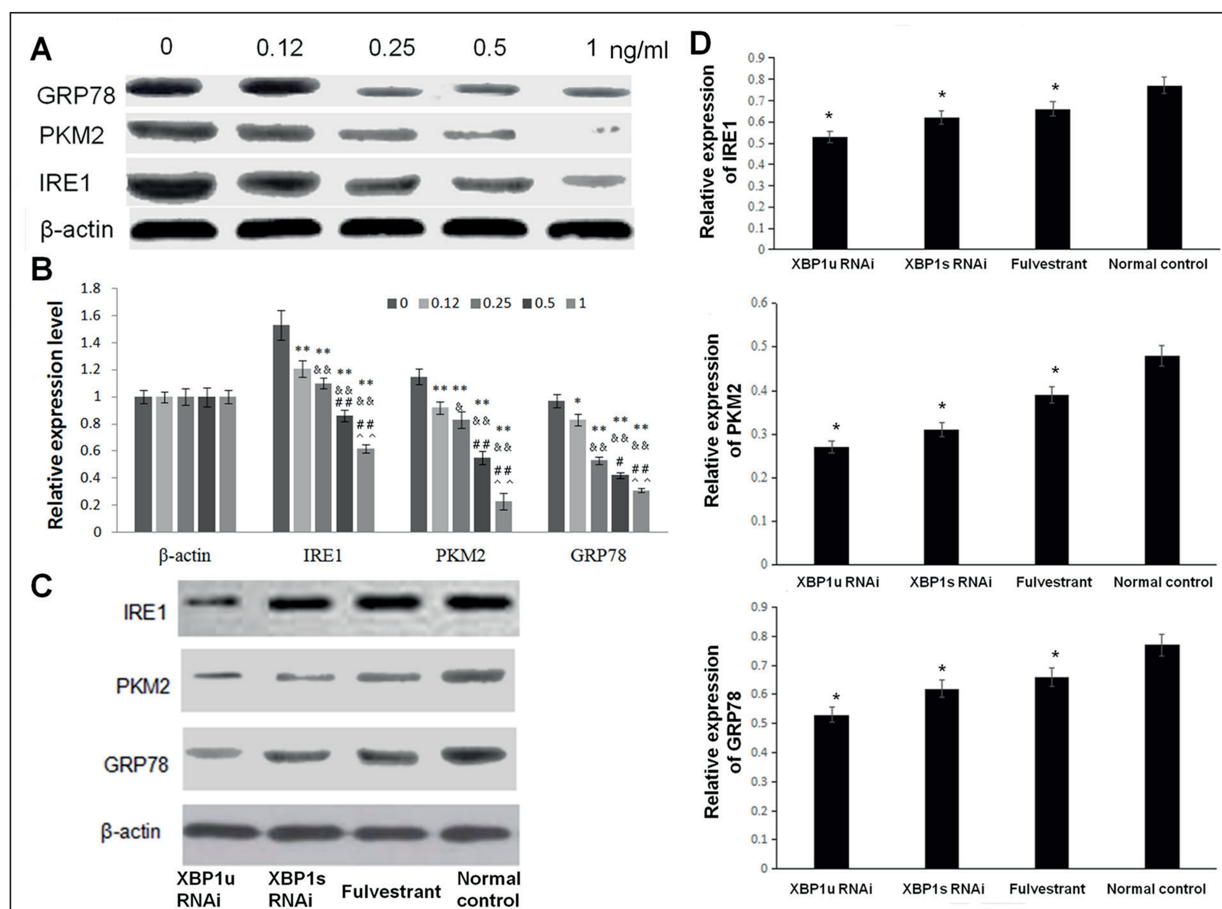

Figure 3. The expression of IRE1, PKM2 and GRP78 on GH3 cells treated by different concentrations of fulvestrant (0, 0.12, 0.25, 0.5 and 1ng/ml) through Western blot (***A*** and ***B***). * and ** represented significantly different at $p < 0.05$ and 0.01 when compared with normal control (0 ng/ml). & and && represented significantly different at $p < 0.05$ and 0.01 when compared with GH3 cells treated with 0.12 ng/ml fulvestrant. # and ## represented significantly different at $p < 0.05$ and 0.01 when compared with GH3 cells treated with 0.25 ng/ml fulvestrant. ^^Represented significantly different at $p < 0.05$ when compared with GH3 cells treated with 0.5 ng/ml fulvestrant. The expression of IRE1, PKM2 and GRP78 on GH3 cells treated by Fulvestrant, siRNA XBP1s and XBP1u through Western blot (***C*** and ***D***). *Represented significantly different at $p < 0.05$ when compared with normal control (0 ng/ml).

When the concentration of fulvestrant reached to 1 ng/ml, approximately 40%, 55% and 30% reduction was observed on the expression of IRE1, PKM2 and GRP78, respectively. Meanwhile, the expression levels of IRE1, PKM2 and GRP78 were also reduced in GH3 cells treated by siRNA XBP1s and XBP1u compared with those in normal control ($p < 0.05$). The expression of IRE1, PKM2 and GRP78 of GH3 cells treated by 1 ng/ml fulvestrant was similar with those treated by siRNA XBP1s and XBP1u. As fulvestrant could inhibit the expression of XBP1s and XBP1u, these phenomena indicated that the inhibitory effects of fulvestrant on glycolysis of GH3 cells may closely related with the downregulation of IRE1, PKM2 and GRP78.

Discussion

Prolactinoma constitutes the most prevalent hormone-secreting pituitary adenomas[14]. As a typical ERα antagonist, fulvestrant exhibits great therapeutic potential on prolactinoma[15]. It has been reported that fulvestrant can inhibit the cell proliferation and prolactin secretion, and promote the apoptosis of prolactinoma cells[16]. Fulvestrant inhibited the tumor proliferation and prolactin secretion on F344 rat models of prolactinoma[13]. In this study, the glucose uptake, ATP/ADP and lactate production of GH3 cells were significantly inhibited by the treatment of fulvestrant in a dose-dependent manner. Our findings indicated that fulvestrant could significantly inhibit the

glycolysis of prolactinoma cells. Since most tumors exhibited high glycolysis metabolism, the inhibited glycolysis may directly contribute to the apoptosis of prolactinoma cells.

The action mechanisms of fulvestrant on prolactinoma are complex, which have been reported to be associated with various factors, such as WIF-1[13], matrix metalloproteinase-9 (MMP-9), B cell leukemia/lymphoma-2 (BCL-2)[17], pituitary tumor transforming gene (PTTG) and transforming growth factor β1 (TGFβ1)[18]. In this study, we found the expression levels of XBP1s and XBP1u were significantly inhibited by fulvestrant in a dose-dependent manner. Meanwhile, the glucose uptake, ATP/ADP, and lactate production were also found significantly reduced in GH3 cells treated by siRNA XBP1s and XBP1u. These results indicated that fulvestrant might inhibit glycolysis of prolactinoma cells by downregulating XBP1s and XBP1u. XBP1, a transcription factor of the unfolded protein, is important in the proper functioning of the immune system and cellular stress response[19]. Recently, scholars have shown that XBP1 played an important role in tumor progression[20-23]. It has been reported that loss of XBP1 severely inhibited the growth of human fibrosarcoma cells in a hypoxic microenvironment [21]. XBP1 overexpression was significantly correlated with the tumor stage, lymph node metastasis and poor outcome of esophageal squamous cell carcinoma[22]. Low expression of XBP1 induced by ERβ was associated with decreased survival of breast cancer cells[23]. Our findings suggested that the oncogene role of XBP1 on prolactinoma was closely related with the inhibition of glycolysis. Meanwhile, fulvestrant might inhibit the glycolysis of prolactinoma cells via downregulating XBP1.

IRE1/XBP1 pathway is a conserved unfolded protein response pathway involved in endoplasmic reticulum stress[20]. Under non-stressed conditions, IRE1 is associated with glucose GRP78 in an inactive state. However, the activation of IRE1 can remove an intron from the XBP1 and then recover the endoplasmic reticulum stress[24]. Until now, various researches have shown that IRE1/XBP1 signaling pathway was important in tumor progression[20,25,26]. In this study, the regulatory mechanisms of fulvestrant on glycolysis of GH3 cells were further analyzed, and Western blot showed that the expression of IRE1 and GRP78 was significantly reduced by siRNA XBP1s and XBP1u. It has been reported that the prolonged unfolded protein response activation reduced the glycolysis and mitochondrial metabolism via IRE1 signaling[27]. GRP78 was implicated in the modulation of tumor aerobic glycolysis by promoting autophagic degradation of IKKβ[28]. Our findings were consistent with these researches, and further illustrated that the reduced glycolysis induced by siRNA XBP1s and XBP1u was related with the IRE1/XBP1 pathway. Besides, PKM2 is a limiting glycolytic enzyme that catalyzes the final step in glycolysis. PKM2 plays an important role in tumor metabolism and growth. Besides, it has been reported to be able to promote the glycolysis and inhibit the oxidative phosphorylation in glioblastoma cells[29]. Meanwhile, PKM2 nuclear translocation induced by iNOS-derived nitric oxide promoted the glycolysis in ovarian cancer[30]. The antioxidant uncoupling protein 2 sensitized pancreas cancer cells to glycolysis inhibition via stimulating PKM2 expression[31]. In consistent with these findings, significantly reduced expression of PKM2 was observed on GH3 cells with low glycolysis (treated by siRNA XBP1s and XBP1u). The low expression of XBP1s and XBP1u might inhibit the glycolysis of GH3 cells through downregulating PKM2.

Conclusions

We found that fulvestrant was able to inhibit the glycolysis of GH3 cells by downregulating IRE1/XBP1 signaling pathway. Meanwhile, this process was closely related with the downregulation of PKM2. However, this study was still limited in cell experiment, and further researches on the anti-glycolysis effects of fulvestrant and related regulatory mechanisms on animal model were still needed.

Conflict of Interest

The Authors declare that they have no conflict of interests.

References

1) Auriemma RS, Grasso LF, Pivonello R, Colao A. The safety of treatments for prolactinomas. Expert Opin Drug Saf 2016; 15: 503-512.
2) Fukuoka H, Cooper O, Mizutani J, Tong Y, Ren SG, Bannykh S, Melmed S. HER2/erbB2 receptor signaling in rat and human prolactinoma cells: strategy for targeted prolactinoma therapy. Mol Endocrinol 2011; 25: 92-103.
3) Triantafilo N, Castro-Gutierrez V, Rada G. Cabergoline or bromocriptine for prolactinoma? Medwave 2016; 16: e6545.

4) Bai J, Gui S, Zhang Y. Suppression of MMQ cells by fulvestrant: possible mechanism of action and potential application for bromocriptine-resistant prolactinomas. J Clin Neurosci 2013; 20: 721-725.
5) Porporato PE, Dhup S, Dadhich RK, Copetti T, Sonveaux P. Anticancer targets in the glycolytic metabolism of tumors: a comprehensive review. Front Pharmacol 2011; 2: 49.
6) Pak K, Cheon GJ, Nam HY, Kim SJ, Kang KW, Chung JK, Kim EE, Lee DS. Prognostic value of metabolic tumor volume and total lesion glycolysis in head and neck cancer: a systematic review and meta-analysis. J Nucl Med 2014; 55: 884-890.
7) Wang CF, Wang YQ, Huang FZ, Nie WP, Liu XY, Jiang XZ. Association between reversal of multidrug resistance by methyl jasmonate and P-glycoprotein atpase activity in hepatocellular carcinoma. J Int Med Res 2013; 41: 964.
8) Xu W, Zhang Z, Zou K, Cheng Y, Yang M, Chen H, Wang H, Zhao J, Chen P, He L, Chen X, Geng L, Gong S. MiR-1 suppresses tumor cell proliferation in colorectal cancer by inhibition of Smad3-mediated tumor glycolysis. Cell Death Dis 2017; 8: e2761.
9) Qin X, Lin L, Cao L, Zhang X, Song X, Hao J, Zhang Y, Wei R, Huang X, Lu J, Ge Q. Extracellular matrix protein Reelin promotes myeloma progression by facilitating tumor cell proliferation and glycolysis. Sci Rep 2017; 7: 45305.
10) Zhong HD, Tong XG. Experimental studies in vitro on antiestrogen therapy of pituitary prolactinoma. Trace Elements Science 2005-04.
11) Croxtall JD, McKeage K. Fulvestrant: a review of its use in the management of hormone receptor-positive metastatic breast cancer in postmenopausal women. Drugs 2011; 71: 363-380.
12) Guo Y, Wang W, Knosp E, Wu Y, Zhao Y. Inhibitory effects of fulvestrant on the growth of estrogen-induced prolactinoma in Fischer 344 rats and its possible mechanism. J Neurol Sci 2013; 333: e727.
13) Cao L, Gao H, Gui S, Bai G, Lu R, Wang F, Zhang Y. Effects of the estrogen receptor antagonist fulvestrant on F344 rat prolactinoma models. J Neurooncol 2014; 116: 523-531.
14) Molitch ME. Diagnosis and treatment of pituitary adenomas: a review. JAMA 2017; 317: 516-524.
15) Nagykalnai T, Landherr L, Laczo I, Piko B. [fulvestrant (faslodex(r)) for hormone sensitive breast cancer. A review]. Magy Onkol 2015; 59: 251-257.
16) Li D, Gao H, Gong L, Zhang Y. Effects of fulvestrant on proliferation and secretion of prolactinoma GH3 cells. Chinese Journal of Minimally Invasive Neurosurgery 201317.
17) Leng L, Zhang Y. Effects of an estrogen receptor antagonist on proliferation, prolactin secretion and growth factor expression in the MMQ pituitary prolactinoma cell line. J Clin Neurosci 2011; 18: 1694-1698.
18) Lv H, Li C, Gui S, Sun M, Li D, Zhang Y. Effects of estrogen receptor antagonist on biological behavior and expression of growth factors in the prolactinoma MMQ cell line. J Neurooncol 2011; 102: 237-245.
19) Guo FJ, Lin EA, Liu P, Lin J, Liu C. XBP1U inhibits the xbp1s-mediated upregulation of the iNOS gene expression in mammalian ER stress response. Cell Signal 2010; 22: 1818.
20) Hay MP, Jiang D, Kozak M, Niwa M, Koong AC. Inhibition of the IRE1α-XBP1 pathway: a new approach to targeting the tumour microenvironment. International Tumor Microenvironment Workshop 2015.
21) Romero-Ramirez L, Cao H, Nelson D, Hammond E, Lee AH, Yoshida H, Mori K, Glimcher LH, Denko NC, Giaccia AJ, Le QT, Koong AC. XBP1 is essential for survival under hypoxic conditions and is required for tumor growth. Cancer Res 2004; 64: 5943-5947.
22) Xia T, Tong S, Fan K, Zhai W, Fang B, Wang SH, Wang JJ. XBP1 induces MMP-9 expression to promote proliferation and invasion in human esophageal squamous cell carcinoma. Am J Cancer Res 2016; 6: 2031-2040.
23) Rajapaksa G, Nikolos F, Bado I, Clarke R, Gustafsson JA, Thomas C. ERB decreases breast cancer cell survival by regulating the IRE1/XBP-1 pathway. Oncogene 2015; 34: 4130-4141.
24) Chen Y, Brandizzi F. Ire1: ER stress sensor and cell fate executor. Trends Cell Biol 2013; 23: 547-555.
25) Koong AC, Chauhan V, Romero-Ramirez L. Targeting XBP-1 as a novel anti-cancer strategy. Cancer Biol Ther 2006; 5: 756-759.
26) Henkel A, Green RM. The Unfolded Protein Response in fatty liver disease. Semin Liver Dis 2013; 33: 321-329.
27) van der Harg JM, van Heest JC, Bangel FN, Patiwael S, van Weering JR, Scheper W. The UPR reduces glucose metabolism via IRE1 signaling. Biochim Biophys Acta 2017; 1864: 655-665.
28) Li Z, Wang Y, Newton IP, Zhang L, Ji P. GRP78 is implicated in the modulation of tumor aerobic glycolysis by promoting autophagic degradation of IKKβ. Cell Signal 2015; 27: 1237-1245.
29) Han D, Wei W, Chen X, Zhang Y, Wang Y, Zhang J, Wang X, Yu T, Hu Q, Liu N, You Y. NF-kB/RELA-PKM2 mediates inhibition of glycolysis by fenofibrate in glioblastoma cells. Oncotarget 2015; 6: 26119-26128.
30) Li L, Zhu L, Hao B, Gao W, Wang Q, Li K, Wang M, Huang M, Liu Z, Yang Q. iNOs-derived nitric oxide promotes glycolysis by inducing pyruvate kinase M2 nuclear translocation in ovarian cancer. Oncotarget 2017; 8: 33047-33063.
31) Brandi J, Cecconi D, Cordani M, Torrens-Mas M, Pacchiana R, Dalla Pozza E, Butera G, Manfredi M, Marengo E, Oliver J, Roca P, Dando I, Donadelli M. The antioxidant uncoupling protein 2 stimulates hnRNPA2/B1, GLUT1 and PKM2 expression and sensitizes pancreas cancer cells to glycolysis inhibition. Free Radic Biol Med 2016; 101: 305-316.