Glucose-regulated protein 78, an important biomarker in cancer etiopathogenesis

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Abstract

Glucose-regulated protein 78 (GRP78) is a major ubiquitous endoplasmic reticulum (ER) protein expressed in normal cells. It belongs to the heat shock protein (HSP) family and is induced by a variety of stress conditions. In cancer cells, in addition to its location in the ER, GRP78 is also present in the plasma membrane, serving as a receptor for various ligands. Moreover, the amount of expressed GRP78 is directly correlated with tumor invasiveness and can be used as a biomarker for cancer. The interest in GRP78 is based on its different functions, both in normal and pathological conditions. GRP78 is a marker for ER stress. GRP78 is a central regulator of ER stress due to its major antiapoptotic role, as well as to its capacity of controlling the activation of transmembrane ER stress sensor. Recent studies have demonstrated that GRP78 plays an important role in the development, progress and chemoresistance of tumors.
Keywords: *GRP78, endoplasmic reticulum, stress, cancer*

Introduction

Protein synthesis is regulated intracellularly by multiple mechanisms aimed at ensuring the acquisition of the unique three-dimensional conformation characteristic of the biologically active protein[1,2].

Protein biosynthesis and maturation depend on the efficiency of the polypeptide chain folding process, which in turn depends on the interaction of the polypeptide chain with molecular chaperones residing in the endoplasmic reticulum (ER). Chaperones located outside the ER are involved in the response of eukaryotic organisms to stress conditions[3].

The ER is an essential cell organelle at the level of which many proteins that play an important role in various biochemical processes are synthesized[4]. ER stress is an adaptive reaction, which normally occurs in any cell that is required to process more molecules than usual per time unit. In this situation, part of the incompletely processed molecules (i.e. folding, coiling, post-translational molecular packing) might generate an adaptive reaction in ER, consisting of: detection of molecular crowding, of conformational defects or excessive volume of molecules, which might prevent molecular traffic through this structure[5].

Glucose-regulated protein 78 (GRP78) is a resident of the ER and an important regulator of the unfolded protein response (UPR). It is expressed in normal adult organs such as the brain, lungs and liver, but it is strongly expressed in malignant tumors[6,7].

The interest in GRP78 is based on its different functions, both under normal and pathological conditions. GRP78 is responsible for intracellular calcium regulation, protein shaping in ER stress and cell survival by an immediate response to insults, having antiapoptotic properties[7].

The accumulation of non-functional, incorrectly folded proteins in the lumen of the ER disturbs organelle homeostasis[8]. In the presence of such a stress situation, the ER reacts by a cascade of events known as the unfolding protein response - UPR, in the attempt to adapt to the new conditions by the restoration of homeostasis[9]. The first reaction consists of translation repression, followed by the activation of signaling pathways that lead to the increased production of chaperones[10]. Signaling mechanisms are initiated in the lumen of the ER by the activation of transmembrane proteins with a role in the control of folding. Three transmembrane proteins of the smooth ER: protein kinase ribonucleic acid (RNA)-like ER kinase (PERK), activating transcription factor-6 (ATF6), and inositol-requiring enzyme 1α (IRE1α) monitor its "health"[11,12,13]. In physiological conditions, GRP78 is bound to the three proteins, maintaining it in their inactive form. During protein accumulation in the ER lumen, GRP78 is dissociated from PERK, ATF6 and IRE1α, allowing their activation and triggering of UPR signals[14].

ER stress has been extensively studied because it can contribute to the development of severe diseases such as neurodegenerative diseases, type 2 diabetes mellitus or various forms of cancer[15]. Recent studies have demonstrated that ER stress leads to the alteration of lipid metabolism and the induction of hepatic steatosis, as some components of the signaling system through the UPR also play a role in the regulation of lipid metabolism by increasing the synthesis of certain enzymes involved in lipogenesis[16].

Proapototic Signals

Neoplastic progression is a process in several stages resulting from genetic changes that lead to the pro-

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gressive transformation of normal cells into malignant cells, inhibiting proapoptotic signals in time. The rapid proliferation of cancer cells requires an increase of ER activity to facilitate the folding, assembling and transmembrane transport of proteins, thus subjecting the ER to stress. The stress to which ER is subjected is the insufficient vascularization and the rapid growth of tumor cells which result in hypoxia, on the one hand, and nutrient deprivation, which affects protein glycosylation and adenosine triphosphate production, on the other hand[17,18].

Cancer cells are characterized by an altered glucose metabolism and the tumor microenvironment is marked by insufficient blood flow and hypoxia, all these triggering ER stress.

Under these circumstances, tumor cells express excess GRP78 and the pro-survival characteristics of GRP78 play a positive role in tumor progression and chemoresistance[19,20]. GRP78 is involved in the protection of tumor cells from cytotoxic damage and apoptosis, helping them to survive under stress conditions such as oxygen and nutrient deprivation[20].

Over-expression of GRP78

Recent studies indicate that GRP78 is over-expressed in several cancer cell lines, contributing to tumor invasion and metastasis. GRP78 was detected in response to various cancer treatments, including photodynamic therapy[21].

Pootrakul and contributors showed that in patients with prostate cancer, GRP78 expression was strongly associated with the presence of castration-resistant tumor cells[22]. This and other studies demonstrate that an increase in GRP78 was correlated with a higher risk of cancer recurrence and a general reduction of patient survival[22,23], therefore GRP78 might be an important prognostic indicator for the development of castration resistance and recurrences in patients with prostate cancer. Similarly, GRP78 excess was correlated with the progression of melanoma[24].

In the case of breast cancer, the detection of predictive factors for tumor chemoresistance is necessary for the improvement of adjuvant treatment. A retrospective analysis showed that about 67% of the subjects diagnosed with breast cancer had high GRP78 levels in their tumors before chemotherapy, and in patients treated with adriamycin alone, GRP78 was correlated with a shorter time period until the onset of recurrence[25]. GRP78 can predict an adequate response to adjuvant taxane treatment in breast cancer[25,26]. However, the interaction between GRP78 and UPR pathways in adriamycin/taxane therapy remains to be established[26].

GRP78 over-expression has been reported to be a biomarker of digestive tumors. The increase of GRP78 detection is directly proportional to the stage and prognosis of esophageal adenocarcinoma[27], gastric and colorectal cancer, GRP78 being one of the stress response proteins, which plays an important role in tumor biology, such as the regulation of apoptosis and the maintenance of intracellular calcium balance[28,29]. A recent report describing a group of 262 patients treated surgically for colorectal adenocarcinoma finds a positive association between GRP78 expression and invasion depth[28].

Su and colleagues performed a study in a group of 44 patients with hepatocellular carcinoma and showed that GRP78 excess in the cells of hepatocellular carcinoma favors its invasion both *in vitro* and *in vivo*[30].

Regarding renal carcinoma, GRP78 plays a role in the protection of renal carcinoma cells from hypoxia and hypoglycemic stress induced by antiangiogenic therapy. The reduction of GRP78 expression levels is a therapeutic target in the management of renal carcinoma[31].

Insulin and insulin-like growth factor-1 stimulate protein synthesis, antiapoptotic proliferation and signaling by the induction of mitogen activated protein kinase and phosphatidylinositide 3-kinases/protein kinase B/mammalian target of rapamycin pathway. GRP78 expression is a key target downstream of insulin, recent evidence suggesting that GRP78 and the global protein balance of the ER can regulate the insulin sensitivity of the organism and protect the cells during acute stress. Obesity and type 2 diabetes mellitus are metabolic disorders characterized by insulin resistance and hepatic steatosis. The presence of ER stress in the context of metabolic syndrome has been documented[32,33], and the presence of chaperones could be a key in the regulation of insulin sensitivity and glucose homeostasis. The administration of chemical chaperones in obese mice decreased ER stress markers, restored glucose levels and insulin homeostasis, while weight gain and the increase of hepatic insulin sensitivity were reduced[34]. GRP78 mRNA was diminished in adipose tissue in patients with gastric bypass after weight loss, which means that the relationship between obesity induced by ER stress and metabolic dysfunction is present also in humans[35].

Obesity is a risk factor for endometrial cancer[36]. However, the prognostic utility of obesity for the development of endometrial cancer is in progress. The association of obesity with endometrial cancer represents a double molecular injury on the ER, therefore the triggered unfolded protein response is presents both at tumor and adipocyte level. This double injury is also determined by the association of endometrial cancer with inflammation, evidenced by the presence of inflammatory factors in blood circulation (i.e. adiponectin, leptin, resistin, omentin, tumor-necrosis factor etc.)[37].

Future Directions

Despite the strong association between obesity and endometrial cancer, it is not clear whether ER stress occurs in adipocytes in these patients. The results of the recent studies have demonstrated that GRP78 levels in visceral adipocytes are correlated with the stage of the disease and patient survival, and might have clinical utility as a predictor for endometrial cancer. ■

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