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Differential Toxic Mechanisms of 2-Deoxy-D-Glucose *versus* 2-Fluorodeoxy-D-Glucose in Hypoxic and Normoxic Tumor Cells

METIN KURTOGLU, JOHNATHAN C. MAHER, and THEODORE J. LAMPIDIS

ABSTRACT

The dependence of hypoxic tumor cells on glycolysis as their main means of producing ATP provides a selective target for agents that block this pathway, such as 2-deoxy-D-glucose (2-DG) and 2-fluoro-deoxy-D-glucose (2-FDG). Moreover, it was demonstrated that 2-FDG is a more potent glycolytic inhibitor with greater cytotoxic activity than 2-DG. This activity correlates with the closer structural similarity of 2-FDG to glucose than 2-DG, which makes it a better inhibitor of hexokinase, the first enzyme in the glycolytic pathway. In contrast, because of its structural similarity to mannose, 2-DG is known to be more effective than 2-FDG in interfering with N-linked glycosylation. Recently, it was reported that 2-DG, at a relatively low dose, is toxic to certain tumor cells, even under aerobic conditions, whereas 2-FDG is not. These results indicate that the toxic effects of 2-DG in selected tumor cells under aerobic conditions is through inhibition of glycosylation rather than glycolysis. The intention of this minireview is to discuss the effects and potential clinical impact of 2-DG and 2-FDG as antitumor agents and to clarify the differential mechanisms by which these two glucose analogues produce toxicity in tumor cells growing under anaerobic or aerobic conditions. *Antioxid. Redox Signal.* 9, 1383–1390.

INTRODUCTION

ALTHOUGH GLUCOSE IS COMMONLY THOUGHT OF as a central molecule for supplying energy to the cell, its six- carbon skeleton is also used in forming the sugar backbones of DNA and RNA precursors, as well as other metabolic intermediates necessary for cellular growth, replication, and survival. The metabolic fate of glucose is dictated by slight chemical alterations in each of its carbons (*i.e.*, epimerization at the second carbon produces mannose, which is involved in glycosylation of proteins, whereas isomerization from aldose to ketose generates fructose, used by the glycolytic pathway). Since the early 1990s when it was clearly demonstrated that oncogenic transformation coincides with an increase of glucose metabolism (19), studies with sugar analogues that inhibit various metabolic pathways have accelerated (24, 35, 46). These studies have begun to clarify the roles of each individual pathway in supplying the high demand of tumor cells for glucose-derived anabolic intermediates and energy.

2-Deoxyglucose: metabolism and metabolic blocks

2-DG has been recognized as an antagonist of glucose metabolism since the early 1950s (4, 10). Its structure is identical to that of D-glucose, except that the C-2 hydroxyl group is replaced with hydrogen. Early studies demonstrated that 2-DG was an effective inhibitor of both aerobic and anaerobic glucose fermentation in yeast (53), underscoring its efficacy as a glycolytic inhibitor. Metabolic analyses showed that, like glucose, 2-DG is taken up through the glucose transporters (GLUTs) and phosphorylated by hexokinase (HK) to form 2-DG-6-phosphate (2-DG-6-P). However, although glucose-6-phosphate (G-6-P) progresses through the glycolytic pathway, 2-DG-6-P accumulates within the cell and is not metabolized

[1]University of Miami, Miller School of Medicine and Sylvester Cancer Center, Miami, Florida.

further (4, 67, 77). 2-DG-6-P was found to compete with glucose for phosphoglucose isomerase (PGI) in the subsequent reversible glycolytic step through which G-6-P is converted to fructose-6-phosphate (53, 77). Thus, the primary means by which 2-DG was thought to block glycolysis was by competitively inhibiting PGI (4, 77).

However, more recent studies have demonstrated that a secondary metabolic effect of 2-DG is noncompetitive inhibition of HK by 2-DG-6-P (9, 33). Although this effect is similar to the feedback inhibition of HK by G-6-P, the K_i for 2-DG-6-P is well in excess of that for G-6-P, which could explain why earlier reports concluded that 2-DG-6-P did not inhibit HK (67). It is likely that treatment of cells with 2-DG leads to a buildup of 2-DG-6-P to concentrations capable of HK inhibition. This activity could explain the outcome of studies in which cells treated with 2-DG showed reduced intracellular levels of G-6-P (69).

The Warburg effect and 2-DG

In the 1920s, Otto Warburg observed a hallmark of cancer cells: they produce high levels of lactate and use glycolysis to generate energy, even in the presence of oxygen (76). This effect, known as aerobic glycolysis or the "Warburg effect," suggested that tumor cells may be inherently sensitive to glycolytic inhibition, and spawned studies to evaluate the antitumor activity of 2-DG as an inhibitor of this pathway. It was found that treatment with 2-DG inhibited the growth of a variety of tumor types in rodents (2, 6, 17, 40, 65, 66), which led to early trials of 2-DG administration as a single agent in human cancer patients (39). However, the clinical trials were unsuccessful and generated adverse side effects associated with the high drug concentrations required to mimic the antitumor activity observed in the animal studies (1, 4, 31, 32, 68). Although the Warburg hypothesis has been elegantly confirmed by positron emission tomography (PET) (34, 72), the ineffectiveness of 2-DG monotherapy in killing the tumor is more likely due to glycolysis not being essential for cell survival. Warburg proposed that tumor cells were glycolytic because of defective mitochondria (23, 78, 79); however, subsequent studies have not established defective mitochondria as a hallmark of tumorigenesis (24). Accordingly, the Warburg effect does not appear to predispose tumor cells to the toxic effects of 2-DG, because most of these cells have normal capacities for using alternative carbon sources through oxidative phosphorylation in the presence of O_2. Rather, accelerated glycolysis appears to provide a selective advantage to oncogenically driven proliferating cells by supplying the metabolic intermediates that are essential precursors for growth and replication. Moreover, it has been proposed that because glycolysis (despite its reduced efficiency) also produces ATP at a significantly faster rate than oxidative phosphorylation, it allows cancer cells more effectively to compete for limited fuel sources (56). Thus, in the presence of oxygen and sufficient amounts of nutrients, inhibition of glycolysis should only growth inhibit, but not kill tumor cells by decreasing the synthesis of glycolytic intermediates. In contrast, when oxygen is unavailable for oxidative phosphorylation, the utilization of fats and proteins is hampered, and tumor cells become dependent on glycolysis for survival.

Low intratumoral oxygen resulting from attenuated blood flow due to poor or aberrant vasculature or both (5, 15) within certain areas of tumors is known to occur in more than a third of all cancers (29). Additionally, tumor hypoxia may be elevated because of anemia, a condition typically associated with chemotherapy or radiation treatment (26). Clinically, hypoxia is known to reduce the efficacy of both radiotherapy and chemotherapy. The mechanisms underlying such resistance include limitation of molecular O_2 necessary for radiation- and chemotherapy-induced DNA damage (26, 27, 36), increased production of nucleophilic substances that competitively inhibit alkylating agents, increased activity of DNA repair enzymes (26), and the expression of both hypoxic stress and antiapoptotic proteins (26, 60, 70). Perhaps most important, hypoxia reduces cell-cycle progression and decreases the cytotoxic effects of chemo- and radiotherapies that preferentially target rapidly proliferating cells (4, 25, 26, 42).

Although a low-oxygen environment creates multifactorial resistance to therapy, it also leads to a primordial metabolic switch, which offers a therapeutic window for selectively targeting slow-growing hypoxic tumor cells found in most solid tumors. In contrast, slow-growing normal cells that compose most tissues and organs receive normal oxygen levels and do not require glycolysis to survive. As mentioned earlier, unlike aerobic normal cells, tumor cells under hypoxia cannot use alternate carbon sources (*i.e.*, amino acids and fatty acids) for the production of ATP and rely solely on glycolysis for energy. This metabolic alteration renders hypoxic cells sensitive to the toxic effects of glycolytic inhibition, whereas those under normal O_2 are relatively unaffected by such treatment (Fig. 1). These differential responses to glycolytic inhibitors were demonstrated in three distinct models of anaerobic metabolism including a chemical model (A), that uses mitochondrial inhibitors to block oxidative phosphorylation; a genetic model (B), in which osteosarcoma cells (ρ^0) are devoid of mitochondrial DNA and therefore can not undergo oxidative phosphorylation; and an environmental model (C), in which tumor cells are grown under decreased levels of oxygen (41, 45, 47). Our results showed that 2-DG treatment causes profound cell death and inhibition of the S to G2 phase transition in all three models. In contrast, aerobic cells with functional mitochondria were able to survive glycolytic inhibition.

The overall goal of the *in vitro* studies using the different models of anaerobic metabolism was to stimulate interest in developing and applying the concept of using glycolytic inhibitors to enhance the activity of current chemotherapeutic protocols. A report from our laboratory showed that 2-DG treatment increased the efficacy of chemotherapy in mouse xenograft models (47). Thus, data from our *in vitro* and *in vivo* studies support the hypothesis that 2-DG should be an effective adjunct therapy for human tumors (41, 45, 47). In February of 2004, we initiated the first phase I clinical trial of 2-DG in combination with docetaxel to target both the hypoxic, chemotherapy-resistant cells and the aerobic cells within solid tumors, [Protocol 2003121: A Phase I dose escalation trial of 2-deoxy-D-glucose (2-DG) alone and in combination with docetaxel in subjects with advanced solid malignancies]. Similarly, 2-DG in combination with fractionated radiation has proven to be efficacious in mouse xenograft models and clinical studies (51).

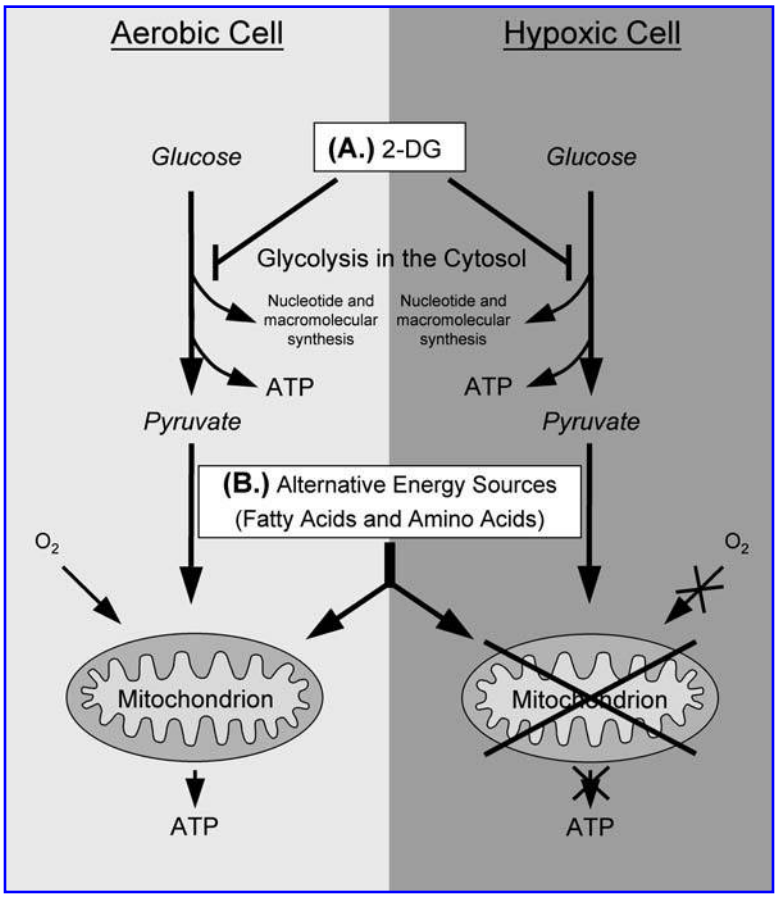

FIG. 1. Consequences of blocking glycolysis by 2-DG in aerobic versus hypoxic cells. In an aerobic cell, if glycolysis is inhibited by 2-DG (**A**), ATP cannot be generated by this pathway. However, because oxygen is available, amino or fatty acids or both (**B**) can act as alternative carbon sources for ATP production through oxidative phosphorylation in the mitochondria. In contrast, when glycolysis is blocked in a hypoxic cell, these other carbon sources cannot be used because oxygen is unavailable for oxidative phosphorylation. Thus, when a hypoxic cell is treated with 2-DG, it has no alternative means for generating ATP and will thereby succumb to this treatment.

Hypoxic-inducible factor confers a level of resistance to 2-DG by upregulating hexokinase

Interestingly, in the environmental model of anaerobiosis, wherein cells are grown under reduced oxygen tension, the cytotoxic and cell-cycle effects of 2-DG were less than those found in models A and B. These diminished effects may result from a low level of oxidative phosphorylation that is possible at 0.1–0.5% O_2. Another possibility is that hypoxia-inducible factor-1 (HIF-1) (71, 74), activated by hypoxia, is protective (50). HIF-1 is a central modulator of adaptive cellular responses and promotes survival under hypoxia. This transcription factor induces expression of >60 genes that play a role in adaptation to decreased oxygen tension. Because such genes include those that are involved in glucose transport and glycolysis (49, 64, 71, 75), it is likely to affect 2-DG toxicity in tumor cells growing under hypoxia.

A plausible explanation of increased resistance to 2-DG in the presence of HIF-1 is that greater amounts of glycolytic enzymes induced by this transcription factor (63, 64) require higher concentrations of 2-DG to block glycolysis effectively. Additionally, because the cytotoxic activity of 2-DG in hypoxic cells is believed to be through the inhibition of glycolytically derived energy, the sensitivity of an hypoxic cell to 2-DG should parallel the amount of ATP depletion by this agent. Indeed, it was recently shown that HIF-1 activation diminishes the effects of 2-DG on ATP depletion (44).

Because 2-DG inhibits the glycolytic pathway at HK and PGI (67, 77), HIF-induced increases in either of these enzymes could, in principle, contribute to 2-DG resistance. Results from our laboratory as well as others demonstrate that intracellular amounts of PGI are not markedly altered in the presence of HIF-1 (22, 44), whereas HK expression increases in an HIF-1–dependent manner. These results are consistent with a role for HK in conferring resistance to 2-DG in hypoxic cells.

2-DG is currently in clinical trials, and it is important to understand precisely the mechanisms of resistance that may develop in tumor cells. We found that the knockdown of HIF-1 with a selective siRNA significantly increased the sensitivity of cells under hypoxia to 2-DG (52), suggesting that inhibition of HIF-1 may improve the clinical efficacy of glycolytic inhibitors. HIF-1 is widely believed to promote tumorigenesis (8, 48, 57, 58, 63), and specific inhibitors are under development for use in cancer treatment (18, 28, 43, 59, 63). Combining such inhibitors with 2-DG may provide a synergistic strategy for targeting the chemotherapy and radiation-resistant, hypoxic cell populations found in most solid tumors.

2-FDG is more potent than 2-DG in inhibiting glycolysis and killing hypoxic cells

The chemical properties of fluorine in 2-FDG are known to resemble more closely the hydroxyl group of the second carbon in glucose than the hydrogen at the same position in 2-DG.

It is therefore expected that 2-FDG should be a better substrate for hexokinase, suggesting two possible scenarios: (a) FDG could bind to the catalytic site of hexokinase better than 2-DG, resulting in increased levels of 2-FDG-6-P and enhanced inhibition of the next enzyme in the glycolytic pathway, PGI; or (b) FDG-6-P interacts with the allosteric site of hexokinase with higher affinity than 2DG-6-P and is a more effective inhibitor of this enzyme. Although hydrogen bonding between the sugar analogues and the catalytic site of hexokinase is in favor of 2-FDG over 2-DG, no significant difference in ΔG for this site was found for these analogues (38). However, the binding energy of 2-FDG-6-P for the allosteric site was significantly lower than that of 2-DG-6-P and more closely resembled the ΔG of glucose-6-P. These molecular modeling analyses suggested that 2-FDG, in comparison with 2-DG, might prove to be a better inhibitor of hexokinase and thereby glycolysis, which should make it more toxic to hypoxic tumor cells (Fig. 2). The latter possibility is substantiated by the findings that 2-FDG is two- to threefold more potent than 2-DG in reducing lactate levels, and this correlates with increased killing of tumor cells growing under chemical or environmental models of hypoxia (38). Whereas it is possible that the transport rate of these analogues also contributes to their differential potencies, molecular modeling and biochemical and toxicity assays support 2-FDG as a more potent inhibitor of glycolysis and thereby a more effective antitumor drug than 2-DG.

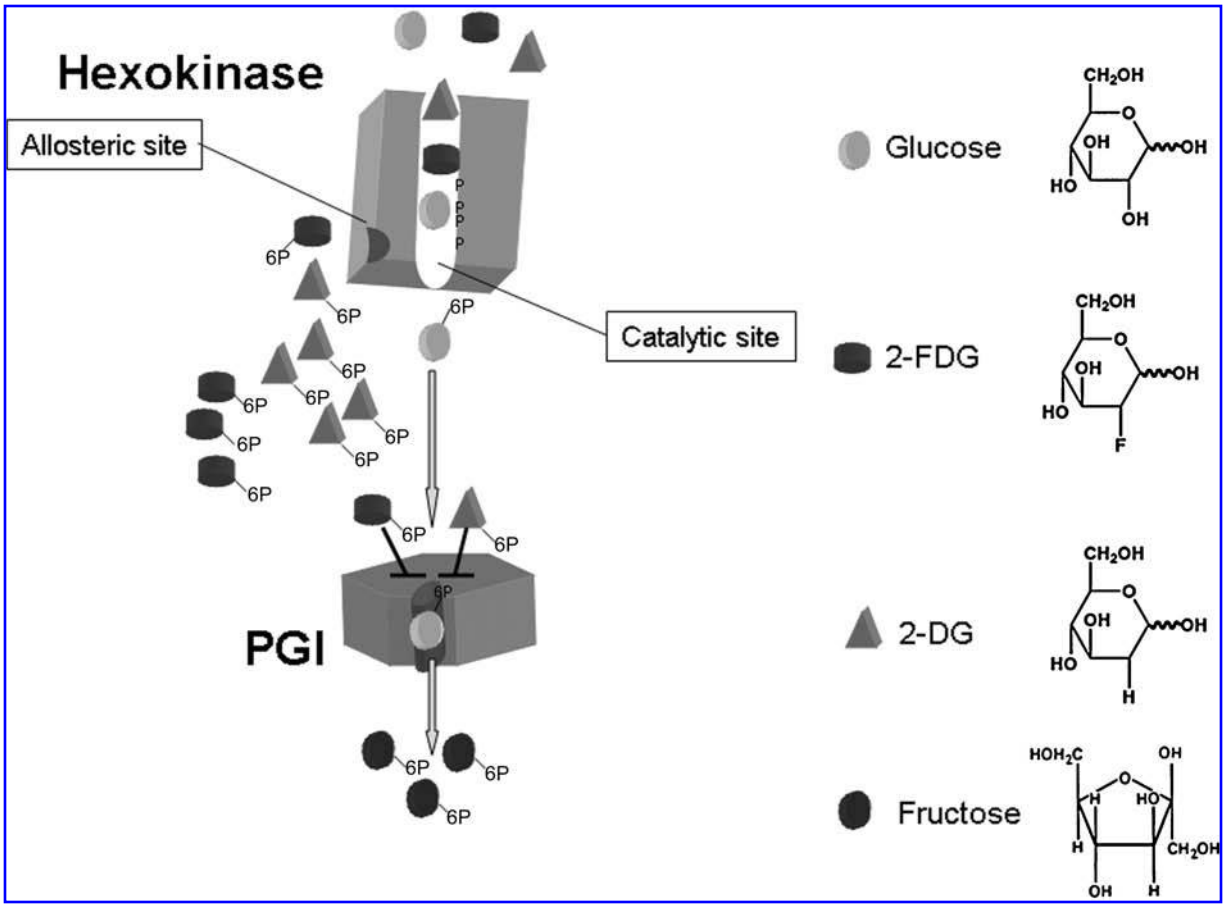

FIG. 2. 2-FDG is a better inhibitor of glycolysis than 2-DG. When glucose enters the cytosol, its sixth carbon is phosphorylated by hexokinase, yielding glucose-6-phosphate, which cannot diffuse out of the cell because of its negative charge. Subsequently, this intermediate is converted to fructose-6-phosphate by the second enzyme of the glycolytic pathway, phosphoglucoisomerase (PGI). Similar to glucose, both 2-DG and 2-FDG are trapped inside the cell by phosphorylation of their sixth carbon; however, both 2-DG-6-phosphate and 2-FDG-6-phosphate cannot be used by PGI, leading to competitive inhibition of this enzyme. Therefore, the *primary block* in glycolysis induced by either 2-DG or 2-FDG depends on the concentration of their 6-phosphate metabolites, which is a measure of the first reaction catalyzed by hexokinase. In this regard, molecular modeling studies showed that the difference in the affinity of 2-DG *versus* 2-FDG to the catalytic site of hexokinase was comparable, indicating that their phosphorylation rate and thereby ability to block PGI is similar. Conversely, a *secondary metabolic block* of these analogues comes from noncompetitive inhibition of hexokinase via binding of their 6-phosphate derivatives to the allosteric site of the enzyme. This effect is similar to the feedback inhibition of hexokinase by glucose-6-phosphate. When the affinity of 2-DG-6-phosphate for binding to this regulatory site was compared with that of 2-FDG-6-phosphate, the latter analogue was found to have a significantly higher binding energy, suggesting that 2-FDG is better than 2-DG in inhibiting hexokinase and thereby glycolysis.

2-DG toxicity in selected tumor types growing under normoxia correlates with interference with N-linked glycosylation

Recently, we found that 2-DG is toxic to a select number of tumor cell lines, even in the presence of O_2. This result was surprising since we had previously found that both tumor and nontumor cells were resistant to 2-DG–mediated death in the presence of oxygen (41, 45). One possibility to explain this result is that some tumor cells are sensitive to 2-DG in the presence of O_2 because of defective mitochondrial oxidative phosphorylation, as suggested by Warburg (76). This possibility seems unlikely, because oxygen consumption is similar in 2-DG–sensitive and 2-DG–resistant cells (37). Therefore, 2-DG toxicity must arise by another mechanism in the sensitive cells.

A clue to the mechanism came from a series of articles published in the late 1970s, wherein it was demonstrated that in certain viruses, N-linked glycoprotein synthesis is inhibited by sugar analogues, including 2-DG (11, 12, 61, 62). It was found that these analogues inhibit the assembly of lipid-linked oligosaccharides, resulting in the disruption of mannose-type protein glycosylation. Structurally, 2-DG resembles mannose as well as glucose, and in the process of N-linked glycosylation, this analogue was shown to mimic mannose in its stepwise addition to the lipid-linked oligosaccharide chain. In the first step of N-linked glycosylation, mannose is activated by covalent reaction with guanosine diphosphate (GDP) or dolichol phosphate (Dol-P). It was demonstrated in these early studies that during the assembly of lipid-linked oligosaccharides, 2-DG undergoes conversion to 2-DG-GDP and competes fraudulently with mannose-GDP for addition onto *N*-acetyl-glucosamine residues, catalyzed by GDP-mannosyltansferase. Furthermore, it was reported that the intracellular conjugation of 2-DG to GDP and Dol-P results in depletion of these precursors, thereby further disrupting normal oligosaccharide formation (13). Thus, the aberrant oligosaccharides produced result in decreased synthesis of viral glycoproteins. In contrast, the fluorine group at the gluco-configuration in 2-FDG restricts this sugar analogue to resembling glucose. Thus, it is not surprising that 2-FDG does not inhibit mannose incorporation into oligosaccharides (11). Overall, these studies concluded that 2-DG is more potent than 2-FDG in disrupting N-linked glycosylation.

Further support that 2-DG interferes with N-linked glycosylation predominantly by competition with mannose metabolites came from results that showed that low amounts of exogenous mannose, but not glucose, blocked the disruption of lipid-linked oligosaccharides (LLO) assembly by 2-DG (11). Furthermore, the mannose analogue, 2-fluoro-deoxy-D-mannose (2-FDM), was similar to 2-DG in that it inhibited mannosyltransferases and suppressed mannose incorporation into LLO. Interestingly, however, 2-FDM was found to be less effective than 2-DG in interfering with LLO assembly because 2-DG, but not 2-FDM, incorporated into the oligosaccharide chain. It was therefore concluded that the order of potency in disrupting N-linked glycosylation in viral glycoprotein synthesis was 2-DG > 2-FDM > 2-FDG (11, 12, 61).

We found that these sugar analogues display a similar order of potency (*i.e.,* 2-DG > 2-FDM > 2-FDG) in killing selected tumor cell lines growing under normoxic conditions. Moreover, exogenous mannose was shown to reverse 2-DG toxicity completely. These findings, and the fact that 2-FDG is a better inhibitor of glycolysis than 2-DG, strongly suggest that interference with N-linked glycosylation, and not inhibition of glycolysis, is the mechanism by which these sugar analogues are toxic to normoxic tumor cells (Fig. 3) (37).

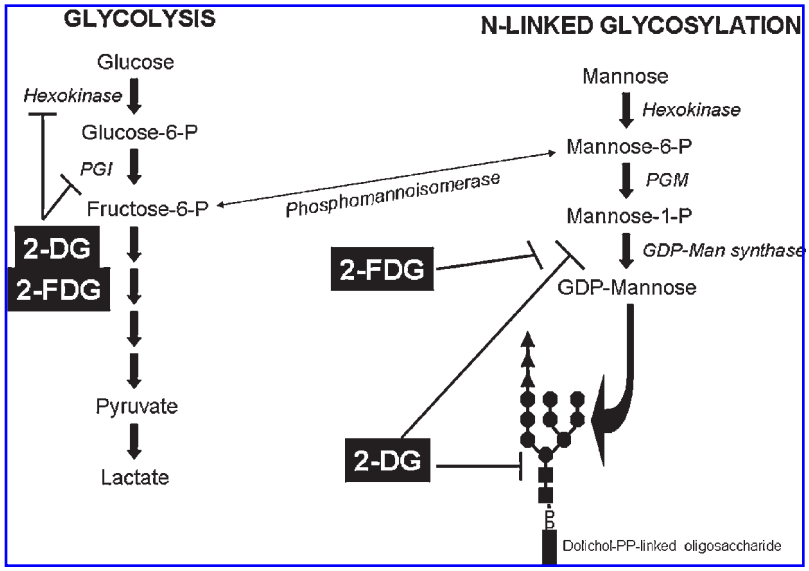

FIG. 3. Summary illustration of the differences between 2-DG and 2-FDG in their inhibitory effects on glycolysis and N-linked glycosylation. 2-FDG blocks the first enzyme of glycolysis, hexokinase, better than 2-DG. However, because 2-DG resembles mannose as well as glucose, it has profound inhibitory effects on mannose metabolism, including incorporation of this sugar into dolichol-pyrophosphate (lipid)-linked oligosaccharide, which is the precursor for N-linked glycosylation. In contrast, the fluorine group in 2-FDG restricts it from resembling mannose and, therefore, does not have a direct inhibitory effect on mannose incorporation into lipid-linked oligosaccharide. However, it can decrease the metabolites necessary for transport of mannose from the cytosol into the endoplasmic reticulum, which results in less N-linked glycosylation disruption than that by 2-DG. The greater activity of 2-DG *versus* 2-FDG on N-linked glycosylation correlates with its toxic activity in selected tumor types growing under normal oxygen tension, which can be reversed by addition of exogenous mannose. Additionally, it is important to note that intracellular glucose can be converted to mannose by isomerization to fructose through phosphoglucoisomerase (PGI) and then epimerization to mannose by phosphomannoisomerase (PMI), and therefore theoretically should reverse 2-DG–induced cell death. However, because 2-DG also blocks PGI, glucose cannot be converted to mannose, which can explain its inability to reverse 2-DG toxicity.

Interference with N-linked glycosylation by 2-DG induces an unfolded protein response leading to apoptosis via GADD153/CHOP

It is well established that inhibition of N-linked glycosylation prevents the normal folding of proteins and promotes retention of these proteins in the ER with activation of the unfolded protein response (UPR) (16, 55). The UPR is similar to the p53-mediated repair process that is activated by DNA damage: both mediate death or survival pathways, depending on the severity of the damage and the efficacy of repair. Thus, if UPR fails to establish homeostasis, ER stress-specific apoptotic pathways such as the one mediated by GADD153/CHOP are activated (3, 73). Indeed, it was found that GADD153/CHOP levels are increased in tumor cells undergoing aerobic death by 2-DG (37), further implicating interference with glycosylation as the mechanism of 2-DG toxicity.

Because UPR was originally shown to be induced by glucose starvation (16, 55), it was assumed that 2-DG also induced the UPR by inhibiting glucose utilization. However, our results, as well as those of Schwarz *et al*. (11, 12, 37, 61), indicate that 2-DG also depletes growing oligosaccharides of mannose and incorporates into the chain, making 2-DG potentially more potent than glucose deprivation. This idea is supported by the findings that neither low glucose nor 2-FDG is toxic to these selected tumor cells under normoxia. Moreover, Kang *et al*. (30) showed that high doses of 2-DG, but not low glucose, can disturb O-glycosylation of cytosolic proteins, including Sp1, which led to growth inhibition or cell death in some tumor types (30). Overall, these results suggest that application of 2-DG as a single antitumor agent should be revisited, because the ability of this sugar analogue to mimic mannose appears to confer it with a unique toxic activity in certain tumor cell types.

Do differences in PMI activity correlate with cellular sensitivity to 2-DG under normoxia?

The molecular pathways underlying heightened sensitivity to 2-DG in some tumors remain unknown. However, one possibility is that these tumors have defective pathways for generating mannose from glucose. For example, patients with a deletion in the phosphomannose isomerase (PMI) gene are deficient in intracellular mannose and require dietary mannose supplements to survive (20, 21, 52, 54). PMI converts glucose-6-phosphate to mannose-6-phosphate, which is subsequently converted to GDP-mannose and incorporated onto growing lipid-linked oligosaccharide chains. A deletion in PMI was shown to cause glycosylation syndrome 1b, which resulted in hypoglycosylation of serum glycoproteins, leading to thrombosis and gastrointestinal disorders in patients identified with this defect (20, 21, 52, 54). Addition of mannose to the diet was shown to alleviate the patient's symptoms as well as normalize his glycoproteins. Thus, a deficiency in or downregulation of this enzyme could explain both the toxicity of 2-DG and 2-FDM, as well as the ability of exogenous mannose to reverse these toxic effects in sensitive tumor cell lines. In the absence of PMI, cells would be dependent on exogenous mannose (present in serum) for the synthesis of N-linked oligosaccharide precursors. Mannose concentrations in the serum of mammals (50–60 μg/ml), or in the medium used for *in vitro* studies, are known to be significantly less than the concentration of glucose (14). Thus, in cells with deleted or downregulated PMI, low doses of 2-DG and 2-FDM could favorably compete with the small amounts of mannose present in serum, and thereby interfere with the addition of this sugar onto the oligosaccharide chains. In contrast, cells with normal PMI can produce GDP-mannose from glucose, and therefore, much higher doses of 2-DG or 2-FDM would be necessary to cause complete disruption of oligosaccharide assembly. This could explain why most of the cells that have been tested are resistant to 2DG under normoxic conditions. Further studies are necessary to assess this possibility.

CONCLUDING REMARKS

Differences in sugar metabolism between normal and tumor cells have been recognized for >70 years. However, only recently are these differences being effectively exploited for therapeutic purposes. Development of the PET scan reinvigorated investigations on the relation between oncogenic transformation and glucose metabolism, which set the stage for further drug development. As a consequence, clinical trials targeting slow-growing tumor cells in hypoxic areas of solid tumors with glycolytic inhibitors are now under way. Although a variety of antiglycolytic agents are being introduced, 2-DG and 2-FDG hold promise as antitumor agents through the targeting of two divergent metabolic pathways; glycolysis and glycosylation. The slight dissimilarity in the chemical structures of these analogues appears to produce a significant difference in their effects on cellular metabolism. Further investigations on how sugar analogues affect glucose metabolism in normal and tumor cells should lead to new ways of treating cancer patients with relatively nontoxic agents.

ABBREVIATIONS

2-DG, 2-deoxy-D-glucose; 2-FDG, 2-fluoro-deoxy-D-glucose; 2-FDM, 2-fluoro-deoxy-D-mannose; GLUT, glucose transporter; HIF-1, hypoxia-inducible factor-1; HK, hexokinase; PGI, phosphoglucoisomerase; PMI, phosphomannoisomerase.

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Address reprint requests to:
Theodore J. Lampidis
1550 NW 10th Ave
PAP Bldg, Room 131
Miami, FL 33136

E-mail: tlampidi@med.miami.edu

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