Surgical Neurology International

OPEN ACCESS

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Editor: **James I. Ausman,** MD, PhD University of California, Los Angeles, CA, USA

Original Article

The role of metabolic therapy in treating glioblastoma multiforme

Joseph C. Maroon, Thomas N. Seyfried[1], Joseph P. Donohue, Jeffrey Bost

Departments of Neurosurgery, University of Pittsburgh Medical Center, Suite 5C, 200 Lothrop St., Pittsburgh, PA, [1]Biology, Boston College, 140 Commonwealth Ave., Chestnut Hill, MA, USA

E-mail: Joseph C. Maroon - maroonjc@upmc.edu; Thomas N. Seyfried - thomasseyfried@bc.edu; Joseph P. Donohue - jdonohue@conncoll.edu; *Jeffrey Bost - bostj@upmc.edu
*Corresponding author

Received: 24 July 14 Accepted: 31 December 14 Published: 16 April 15

This article may be cited as:
Maroon JC, Seyfried TN, Donohue JP, Bost J. The role of metabolic therapy in treating glioblastoma multiforme. Surg Neurol Int 2015;6:61.
Available FREE in open access from: http://www.surgicalneurologyint.com/text.asp?2015/6/1/61/155259

Abstract

Glioblastoma multiforme (GBM) is an aggressive and nearly uniformly fatal malignancy of the central nervous system. Despite extensive research and clinical trials over the past 50 years, very little progress has been made to significantly alter its lethal prognosis. The current standard of care (SOC) includes maximal surgical resection, radiation therapy and chemotherapy and temozolomide (TMZ), including the selective use of glucocorticoids for symptom control. These same treatments, however, have the potential to create an environment that may actually facilitate tumor growth and survival. Research investigating the unique metabolic needs of tumor cells has led to the proposal of a new metabolic treatment for various cancers including GBMs that may enhance the effectiveness of the SOC. The goal of metabolic cancer therapy is to restrict GBM cells of glucose, their main energy substrate. By recognizing the underlying energy production requirements of cancer cells, newly proposed metabolic therapy is being used as an adjunct to standard GBM therapies. This review will discuss the calorie restricted ketogenic diet (CR-KD) as a promising potential adjunctive metabolic therapy for patients with GBMs. The effectiveness of the CR-KD is based on the "Warburg Effect" of cancer metabolism and the microenvironment of GBM tumors. We will review recent case reports, clinical studies, review articles, and animal model research using the CR-KD and explain the principles of the Warburg Effect as it relates to CR-KD and GBMs.

Key Words: Adjunctive cancer therapy, calorie restriction, glioblastoma multiforme, ketogenic diet, metabolic cancer therapy

Access this article online

Website: www.surgicalneurologyint.com

DOI: 10.4103/2152-7806.155259

Quick Response Code:

INTRODUCTION

Glioblastoma multiforme (GBM) is a highly aggressive malignant tumor of the central nervous system that arises from astrocytes. Primary or *de novo* glioblastomas are the most common and aggressive form, while secondary forms are somewhat less aggressive. Despite billions of research dollars, innumerable clinical trials and untold associated morbidity, the prognosis of afflicted patients from the time of diagnosis to death remains dismal and virtually unchanged over the past 50 years. The standard of care (SOC) includes maximal safe resection followed by radiation therapy, which extends the median survival from 6 to 12.1 months. The addition of temozolomide (TMZ) adds another 2.5 months (median) to the survival time.[1] The increased survival benefit from

TMZ is remarkable in light of recent findings showing that TMZ actually increases the number of driver mutations in GBM tumors.[23] The effectiveness of TMZ has been variable and Hegi *et al.* suggest that genetic factors, specifically the epigenetic silencing (meythlation) of the MGMT promoter explain these observations. Their research showed that GBM patients containing a silenced (methylated) MGMT promoter benefited from TMZ, whereas those who did not have a methylated MGMT promoter did not show as much of a benefit.[21] The addition of TMZ to radiotherapy in a randomized did, however, extend median survival time compared with standalone radiotherapy.[46] These findings may become useful as genomic profiling and personalized medicine become integrated into the SOC.

GBM is a heterogeneous condition consisting of various subtypes with different genetic alterations and gene expression patterns. Thus, no single therapy as presently used will be efficacious across all subtypes. Wilson *et al.* recently concluded in their review article on the state of the art therapeutics for the treatment of GBM that "Failure of conventional treatments combined with its poor prognosis highlights the need for novel approaches for GBM."[52] Indeed, there is clearly a need for novel safe, effective, and perhaps radical strategies to supplement our current SOC, or to be used *de novo*.

For over 75 years it has been known that there is a fundamental metabolic and molecular difference between cancerous cells and normal somatic cells. One major metabolic difference is how cancer and normal cells undergo cellular respiration, glucose metabolism, and energy production. This insight to cancer energy metabolism has recently been exploited through the use of a novel adjunctive cancer therapy known as the calorie restricted ketogenic diet (CR-KD). In this article, we will review studies investigating the KD, CR-KD and cancer metabolism to provide a better understanding of cancer energy metabolism and the potential use of metabolic and dietary therapies for the future treatment of GBM.

A novel approach: The "Warburg Effect" and cancer glycolysis

In 1931, German scientist Otto Warburg won the Nobel Prize for his significant work in cellular respiration, specifically for "his discovery of the nature and mode of action of the respiratory enzyme."[35] He further demonstrated that malignant cells could survive and proliferate even in hypoxic environments, and was the first to propose that all cancers arise from irreversible damage to mitochondria and cellular respiration. Cancer is, Warburg proposed, a metabolic disease.[48,49] Although modern research unanimously proposes that it is mutations in tumor suppressor and oncogenes that cause this metabolic shift, he was correct in hypothesizing that cancer is distinctively characterized by abnormal metabolism of glucose.

Glycolysis, the breakdown of glucose into 2 pyruvate, 2 H^+, 2 net Adenosine Triphosphate (ATP), two NADH and lactic acid, occurs in nearly all living organisms.[34] This metabolic breakdown of glucose occurs in the cytosol of eukaryotes, where under aerobic conditions, pyruvate is oxidized to produce 36 ATP through the citric acid cycle (CAC) and oxidative phosphorylation (OxPhos). Under anaerobic conditions, pyruvate is reduced to lactate.[10] This process is known as Fermentation [Figure 1]. Oxygen inhibits fermentation in healthy cells and in doing so regulates glycolysis. This is known as the 'Pasteur effect'. In many invasive cancer cells, however, aerobic fermentation is observed. This metabolic shift is known as the "Warburg Effect". Under these conditions, increased rates of fermentation necessitate increased glycolytic rates and increased consumption of glucose in many cancer cells.[19]

It is important to recognize that all respiring cells use aerobic glycolysis to produce pyruvate, which is then completely oxidized in the mitochondria. Much of the pyruvate that is produced in tumor cells through aerobic glycolysis is fermented to lactate rather than oxidized in the mitochondria. It is aerobic fermentation that distinguishes the tumor cell from the normal cell.[42]

Tumor cells undergoing aerobic fermentation produce ATP in the cytosol and consume significantly more glucose than healthy cells, but much less efficiently. Aerobic fermentation produces a net 2 ATP compared with the

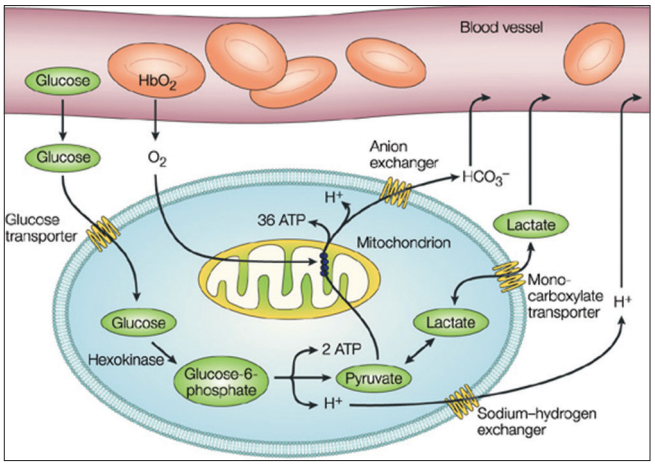

Figure 1:[18] Glucose is transported into the cell where it undergoes glycolysis, the metabolism of glucose to 2 pyruvate, 2 H^+, 2 NADH, 2 H_2O and 2 net ATP. In healthy cells, the pyruvate subsequently enters the mitochondria where it is converted to Acetyl-CoA, which enters the citric acid cycle (CAC) producing the proton donors for the electron transport chain (ETC) that produces approximately 36 ATP via ATP Synthase. Under anaerobic conditions, pyruvate is fermented to lactate. In cancer cells, however, this conversion is observed to occur under aerobic conditions as well, this is known as the "Warburg effect"

approximately net 36 ATP produced from the CAC and OxPhos. Glycolytic rates 200 times higher than normal cells have been observed. This aberrant bioenergetics and dependency on glucose has become a hallmark of cancer.

Over the half-century following Warburg's hypotheses, significant research has been conducted into cancer mitochondrial activity, metabolism, and bioenergetics. Increased rates of glycolysis and aerobic fermentation have been observed in many cancer cell lines evidenced by increased expression of glycolytic enzymes, glucose transporters, lactate production, and glucose consumption.[20,50,51,56,57]

Prolonged dependence on glycolysis and fermentation (nonoxidative energy metabolism) has also been shown to induce genomic instability, which could further increase genomic mutations.[41,44] This dependency of cancer cells, especially those of GBM tumors, on glucose for energy may provide a window for therapeutic management of cancer.

Molecularly targeted therapies: Aiming at a moving target?

In their review, Wilson *et al*. focus on types of genetic alterations from GBM cells that lead to the overexpression of receptor protein kinases (RPKs) such as EGFR and PDGFR.[52] This overexpression leads to abnormal, unregulated tumor proliferation, and growth. Wilson *et al*. propose genetic therapy with small inhibitory molecules to inhibit the known growth factors and angiogenic pathways. This inhibitory treatment could potentially downregulate the overexpression of these protein kinases. They also propose targeting the inhibition of angiogenic pathways induced by the overexpression of vascular endothelial growth factor (VEGF) gene. These genetic alterations along with numerous other malignant genetic mutations are continually evolving under the influence of the individual's genotype and environmental and epigenetic factors. Wilson *et al*. recognize that GBM is genetically "highly heterogeneous" with "recurrent and nonrecurrent" associated genetic conditions.[52] GBMs are incredibly difficult to treat efficiently with molecular targeted therapy in the brief therapeutic window that exists before this aggressive cancer mutates and spreads beyond reconciliation.[12] The authors did not acknowledge, however, that these mutations and genetic alterations might be ever changing, and profoundly influenced by metabolic factors.[15,45]

The function of DNA repair enzymes, impaired in cancer cells, and the integrity of the nuclear genome are dependent on normal mitochondrial function and ATP production. Seyfried has proposed that genomic instability and hence the thousands of genetic abnormalities may be downstream epiphenomena of damaged or insufficient respiration and mitochondrial dysfunction.[41,44] He hypothesizes that external factors damage the mitochondria directly, resulting in mitochondrial dysfunction initiating subsequent downstream signaling sequence in the nucleus activating tumor suppressor and oncogenes that begin the cancer cycle. This hypothesis emphasizes the importance of mitochondrial function to the integrity of the cell. It is clear that mitochondrial health defines metabolic activity and in many ways influences and regulates gene expression.[41,56]

Research has elucidated relationships between specific tumor suppressor genes, oncogenes, and glucose metabolism.[56] The MYC oncogene, for example, directly targets numerous glycolytic enzymes, while SRC and RAS oncogenes regulate glycolysis through the activity of Hypoxic Inducible Factor 1 (HIF1). The tumor suppressor gene p53 regulates hexokinase II, the first enzyme in glycolysis as well as TIGAR, a glycolysis regulator.[9,31,56] These genes upregulate glycolysis in GBM cells, thus increasing their dependency on glucose. Nutrient deprivation, namely in the form of glucose, may provide a mechanism to exploit these mutations by denying cancer cells the very "fuel" upon which they depend.

Are we pouring gasoline on a fire? The role of glucose and glutamine in brain tumor progression

Glucose serves as the primary metabolic fuel of GBM cells and is required in high amounts for tumor cell glycolysis. The metabolic shift to glycolysis renders these cancer cells even more dependent upon glucose. Indeed, human glioblastoma cells expressing constitutively active AKT undergo apoptotic death when deprived of glucose.[17] Glutamine has also been shown to be an important nutrient. It serves as an alternative substrate for the Krebs cycle during aerobic glycolysis. Glutamine, which has elevated levels in cancer cell lines, has also been characterized as an important regulatory factor for the production of macromolecules that enable tumorigenesis, proliferation, and progression.[56]

With this is mind now consider how current GBM SOC may induce an environment that may actually facilitate tumor cell growth, tumor cell survival, and a greater likelihood of tumor recurrence [Figure 2].[44] Novel adjunctive metabolic treatments are now being explored to exploit glucose dependent cancer cell metabolism, mitochondrial dysfunction, and hypoxic environment characteristic of neoplastic cells.

Paradox of current standard of care

Traumatic surgical intervention, radiation therapy, and chemotherapy have all been extensively documented to increase tissue inflammation and blood glucose and glutamate levels.[44] The amino acid glutamate is a major excitatory neurotransmitter, which is rapidly cleared in the astrocytes where it is converted to glutamine following synaptic release to prevent excitotoxic damage.

It is metabolized to glutamine for delivery back to neurons. The glutamate–glutamine cycle maintains low extracellular levels in normal neural parenchyma. Neoplastic Glioma cells, however, especially after surgical manipulation and resulting inflammation, secrete glutamate in high concentrations, contributing to neuronal excitotoxicity and tumor invasion.[44] New studies make it increasingly clear that glutamate and its receptors play a major role in the progression and development of cancer including gliomas.[11] Indeed, GBM cells lines and fresh surgical specimens of GBM have shown a 100-fold lower uptake of glutamate and a 3-fold increase in glutamate release by active transport.[55] Abnormal glutamate levels have been associated with GBM cells that lack the principle glutamate transport proteins, EAAT1 (GLAST), and EAAT2 (GLT-1).[54] This inability of malignant GBM cells to maintain appropriate glutamate levels induces excitotoxicity, which has been proposed to enable the rapid proliferation of GBMs.

Elevated extracellular levels of glutamine contribute to tumor cell growth, proliferation, and cell transformation.[27,47] Radiation and chemotherapies according to the SOC not only cause tumor cell death but also induce necrosis and inflammation, both of which increase glutamate and glutamine levels. Surgery, radiation, and chemotherapy markedly increase tumor-associated macrophages/monocytes, (TAM) which release profuse pro-inflammatory and pro-angiogenic factors favorable to tumor growth.[44]

In addition to glutamate levels, the current SOC may contribute to hyperglycemia, providing GBM cells with elevated glucose, the very fuel upon which they depend. Several laboratory and clinical studies have documented that persistent hyperglycemia in patients with GBM is directly correlated with decreased survival independent of the degree of disability, tumor grade, diabetes or prolonged dexamethasone use. In other words, the higher the blood sugar, the quicker the demise.[16] Therapeutic steroids commonly prescribed to GBM patients are also known to increase blood glucose furthering hyperglycemia and fueling cancer glycolysis.[44] Additionally, radiation therapy is known to upregulate P13K/Akt signaling pathways, which drive glioma glycolysis and therapeutic drug resistance, and further contribute to elevated blood sugar levels. Together, these treatments may exacerbate the GBM microenvironment by inducing a microenvironment that may be facilitative for tumor recurrence.

The current SOC including steroids, surgical resection, radiation, and chemotherapy may provide some initial therapeutic, however, the incredibly poor prognosis indicates indisputably that new approaches must be evaluated to render a new efficacious SOC. A careful examination of the current SOC reveals a certain paradox; the treatments disrupt the blood–brain barrier, elevate glucose, glutamate and glutamine levels, and contribute to the inflammatory process—thus adding more "fuel" to the neoplastic fire. This paradox leaves treating physicians in a plight where the immediate treatment needs of the patient must be balanced with the pursuit of finding an efficacious long-term therapeutic management strategy for the treatment of GBM [Figure 2].

The metabolic management of glioblastomas: The ketogenic diet, ketones, and calorie restriction

The KD has been successfully used for over 90 years in the treatment of drug-resistant refractory seizures in children with epilepsy.[8] Recently, there has been success in the treatment of refractory seizures in adults as well.[26] The diet requires a marked reduction in carbohydrate intake and the use of fat as the main source of energy to induce ketosis. Based on the Warburg effect as a means to restrict glucose substrate for cancer cells, Seyfried and others have reported in animal studies using this diet to induce ketosis to inhibit cancer cell growth [Figure 3]. Along with these successful animal cancer models and recent human case reports using the KD as an adjunct

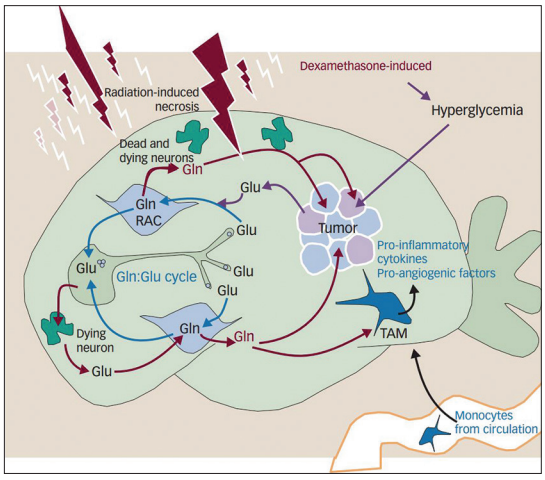

Figure 2[27]: Used with permission of Thomas N. Seyfried. Cellular environment induced by the current standard of care for Glioblastoma multiforme

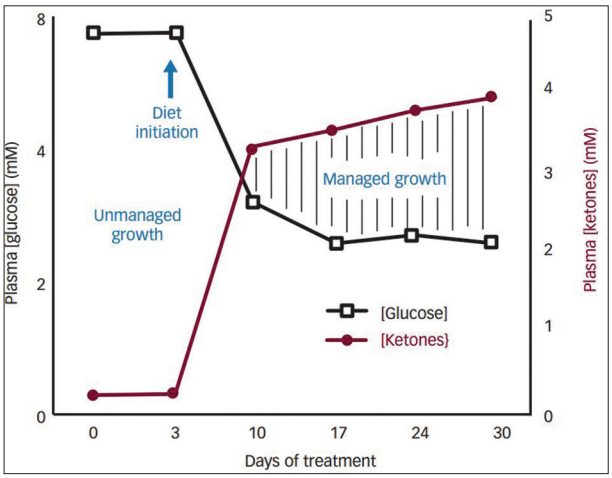

Figure 3[29]: Biomarkers for CR-KD patients

cancer treatment, several phase I studies in the US and Europe have been initiated to study the KD to help slow GBM tumor growth and prolong cancer survival.[42]

In addition to a low carbohydrate KD, Seyfried *et al.* have proposed adding calorie restriction (CR) to further reduce cancer cellular metabolism. This CR-KD shifts energy production in the liver to ketones, an alternative energy source to glucose. Using this diet, Seyfried has reported inducing tolerable ketosis in mice, lowering blood glucose levels and profoundly reducing brain tumor size.[43]

Mammals have evolved to utilize ketones produced in the liver as an alternative energy source. Normal metabolic pathways can reconvert circulating ketone bodies (excluding acetone), derived from fatty acids in the liver, to acetyl CoA, which subsequently initiates the CAC in the mitochondria. GBM cancer cells cannot utilize ketones in this way, and remain largely dependent on glucose as their metabolic substrate.[18,32,36,53] Glucose depletion through the CR-KD would, therefore, deprive GBM cells of their critical energy supply.

A CR-KD takes advantage of evolutionary conserved traits enabling survival during times of food scarcity. The human body has evolved enduring mechanisms to convert fat stores in the liver to beta hydroxybutyrate and acetoacetate—the primary ketone bodies. Ketones can serve as an alternative source to glucose for human energy metabolism. Cancer cells, however, are glucose dependent, and lack this evolutionary versatility to survive on ketone bodies when glucose substrate is deficient.[44]

Cancer cell culture studies done thus far have confirmed that reduced levels of glucose as an energy substrate can "starve" human astrocytomas, reduce angiogenesis, and diminish production of inflammatory cytokines. Several clinical case studies investigating the use of KD as a metabolic approach to GBM have reported improved survival rates. At this time there are no randomized studies that have shown whether CR-KD can statistically enhance progression-free survival or preserve normal function in thus diagnosed with GBM. Klement and Kammerer in a recent review article have proposed a role for carbohydrate restriction in the treatment and prevention of cancer.[25] Furthermore, Champ *et al.* described how the KD could reduce the somatic adverse effects of radiation therapy and proposed that CR may enhance overall therapeutic efficacy.[13]

CR, in addition to KD, can provide an additional advantage by activating sirtuin genes that have been shown to inhibit tumor proliferation. For example, SIRT 1 and Nrf-2 genes are both activated by CR. The SIRT 1 gene has been shown to inhibit neurodegeneration and neoplastic activity.[29,37,38] Nrf-2 activates more than 200 additional genes that are antitumorigenic. Experimental CR regimens have reduced circulating levels of IGF-1, VEGF, and cytokines, leading to decreased growth factor signaling, fewer vascular perturbations, and decreased inflammation.[22]

In addition to CR, other dietary approaches have been shown to induce anticancer metabolic changes. Sulforphane is a sulfur-containing molecule found naturally in cruciferous vegetables, such as broccoli, brussel sprouts, andcabbage, which has been shown to have anticancer and antimicrobial properties. Dietary antioxidants, such as curcumin (from turmeric) and Resveratrol (from grapes and red wine) have also shown powerful anticancer properties.[29]

Implementation and challenges with the restricted calorie ketogenic diet

Despite seeming potential of the metabolic approach using glucose and CR, the practical application of this therapy has been difficult in the human case studies and clinical trials done thus far [Table 1]. The ideal CR-KD, as proposed by Seyfried *et al.* aims to restrict calories (kcal) to approximately 1000–1500 a day based on weight loss not to exceed 20% and limit carbohydrates in order to maintain the blood glucose range between 50 and 65 mg/dL. Carbohydrates would be severely limited to induce ketosis. Protein would also be limited as higher levels of protein can block the production of ketones. The diet would therefore be high in medium-chain triglyceride (MCT) fats and mirror a typical KD used for medication resistant seizures. These fats are usually obtained from avocados, nuts, butter and coconut oil and other good fats and can be used to supplement MCT prepackaged supplements and ketogenic meals designed specifically for these requirements. Based on animal model data, the suggested human ideal ketotic state would allow plasma ketone bodies in the range of 2–4 mM.[44]

The CR-KD in practice would require frequent monitoring of blood glucose and ketone levels to maintain the targeted therapeutic levels. Additionally, enough calories and glucose are required to limit symptoms of hypoglycemia and ketoacidosis seen with blood ketone levels greater than 15 mM. In a recent clinical trial, subjects did experience fatigue and significant weight loss that required diet alterations.[14] Intensive dietary instruction and support is required throughout the course of the therapy, and blood ketone and glucose levels as well as other biomarkers are monitored to track the effectiveness of the diet. Tumor size and other GMB symptoms are monitored to track the progress of the cancer and the effectiveness of the treatment.[14]

Clinical case reports using KD in human cancer

In 1995, Nebeling *et al.* reported on two female children with nonresectable advanced stage brain tumors. Both patients were previously treated with chemotherapy and radiation. They were placed on the KD only, using a MCT oil-based diet, which was reported to effectively manage tumor growth and enhance progression-free survival.[33] The results of this study are somewhat confounded by the

Table 1: Current and past case studies and clinical trials evaluating metabolic therapy and the ketogenic diet for adjunctive treatment of GBM and other malignant cancers

Year	Cancer studied	Treatments rendered	Primary end-point	Secondary end-point	Outcome	Reference
2014-2016 (projected)	Glioblastoma multiforme (newly diagnosed)	Ketogenic diet with Temozolomide during course of radiation therapy	KD compliance indicated by patient tolerance, blood ketones and glucose. Determine if decreasing blood glucose and increasing ketones will enhance effects of standard radiation therapy	Overall survival, time to recurrence and quality of life	N/A	Scheck AC, Barrow Neurological research, Phoenix Arizona[7]
2013-2015 (projected)	Glioblastoma multiforme (refractory/ end-stage)	KD consisting of 4:1 (fat): (Protein + carbohydrate) weight ratio with 1600 kcal restriction. Dietary supplementation with vitamins, calcium, phosphorous, zinc, and selenium to DRI standard	Evaluate the safety of KD as adjunctive treatment (over 1 year) of GBM. Evaluate compliance, fasting lipid, serum glucose and insulin levels	Pilot data on efficacy of KD as adjunctive therapy of GBM. Evaluate tolerability of KD	N/A	Klein P, Mid-Atlantic Sleep Center in collaboration with the Mid-Atlantic Epilepsy and Sleep Center, Bethesda, MD[6]
2013-2015 (projected)	Glioblastoma multiforme (recurrent)	Calorie-restricted KD and transient fasting during and after reirradiation. Restriction of carbohydrates supported by "Tavarlin" drink	Progression-free survival rate 6 months after reirradiation	Feasibility of calorie-restricted ketogenic diet and transient fasting. Overall survival, ketosis, frequency of seizures, quality of life, depression and response	N/A	Rieger J, Johann Wolfgang Goethe University Hospitals[5]
2011	Advanced metastatic malignant tumors (different origins)	KD (less than 70 g CHO per day) with normal groceries and were provided with a supply of food additives to mix a protein/fat shake to simplify the 3-month intervention period	Quality of life (assessed by EORTC QLQ-C30 (version 2)), serum and general health parameters. Ability to tolerate the KD	Metabolic changes monitored by urinary ketone bodies	For various reasons, only 5 of 16 patients completed the 3-month period. Those who finished experienced ketosis and all had stable disease (progression free) after course of diet	Melanie Schmidt *Nutr Metab*, University of Wuerzburg, Germany[40]
2010	Glioblastoma multiforme	Prior to and during standard therapy, a restricted 4:1 (fat: Carbohydrate + protein) ketogenic diet that delivered about 600 kcal/ day, supplemented with vitamins and minerals. The patient was followed using MRI and positron emission tomography with fluoro-deoxy-glucose (FDG-PET)	Efficacy of restricted KD in reducing tumor progression and recurrence	Overall survival of older patient with GBM	After 2 months treatment no discernable brain tumor tissue was detected using either FDG-PET or MRI imaging. Reduced levels of blood glucose and elevated levels of urinary ketones. Tumor recurrence was found 10 weeks after suspension of diet therapy	Zuccoli G, *Nutr Metab* (Lond)[58]
1995	Advanced stage malignant Astrocytoma tumors (pediatric)	Ketosis maintained by consuming a 60% medium chain triglyceride oil-based diet	Tumor glucose metabolism assessed by Positron Emission Tomography, comparing (Fluorine-18) 2-deoxy-2-fluoro-D-glucose uptake at the tumor site before and following the trial period	N/A	Blood glucose levels declined to low-normal levels and blood ketones were elevated twenty to thirty fold. PET scans indicated a 21.8% average decrease in glucose uptake at the tumor site in both subjects	Nebeling LC, University Hospitals of Cleveland, *J Am Coll Nutr*[33]

KD: Ketogenic diet, FDG: Fluoro-deoxy-glucose, PET: Positron emission tomography, MRI: Magnetic resonance imaging, CHO: Carbohydrates, GBM: Glioblastoma multiforme

fact that neither patient described by Nebeling *et al.* had showed tumor progression at the time of enrollment in the dietary regimen. More recently, the ERGO trial published in 2014 examined the feasibility of the KD alone in 20 patients with recurrent GBM. In this study, one patient achieved a minor response and two patients had stable disease after 6 weeks.[39] In this study, no CR was applied and patients were instructed to always eat to satiety. The authors did suggest CR and lower glucose levels might have enhanced the therapeutic effects of the KD.[44]

In 2010, researchers published a case report on a single 65-year-old female patient with GBM that was placed on the CR-KD. The patient had already received standard radiation treatment and chemotherapy. She was restricted to 600 kcal/day and her glucocorticoids were stopped.[58] After 2 months on the diet, the patient showed a significant reduction in blood glucose with an elevation of urine ketones, and experienced a 20% weight reduction. Most importantly, the tumor could no longer be imaged on magnetic resonance imaging (MRI) or positron emission tomography (PET) scanning and clinically she improved. No new tumor progression was seen during the duration of the CR-KD. Ten weeks after the strict diet was terminated, the tumor returned and standard chemotherapy was given. Despite the initial success of the metabolic therapy, the patient died after the diet was stopped and tumor returned. This was the first fully documented treatment of GBM with a CR-KD.

In 2011, German researchers evaluated the CR-KD in 16 subjects with end-stage malignant tumors of various types who had exhausted all standard cancer therapies. Of the 16 subjects, 5 were able to complete the 3-month CR-KD treatment and all 5 experienced no tumor progression while on the diet.[40] Both the ERGO study and the study authored by Champ *et al.* demonstrated improved tolerability and compliance of the KD alone without CR.[13,39] Presently there are at least three clinical trials under way evaluating this metabolic approach to brain tumors using varying applications of both KD and CR diet.[2-4] [Table 1].

To make the CR-KD more tolerable for patients and improve compliance, drugs targeting energy metabolism are being proposed that could potentially allow higher carbohydrate consumption. 2-Deoxyglucose is a glucose mimetic that targets glucose metabolism and glycolysis by impeding the cellular uptake of glucose.[24] Seyfried *et al.* found that CR-KD with 2-DG was more effective than a restricted calorie diet alone to inhibit the growth of astrocytomas in mice models.[30] Additionally, the pyruvate analog 3-bromopyruvate (3BP) has also been shown to be a potent inhibitor of glycolysis, and may be considered for clinical use. 3BP selectively enters tumor cells because of their elevated levels of monocarboxylic acid transporters not present in normal cells, and then interrupts ATP production by blocking glycolysis and OxPhos, but only in malignant cells. Together, drugs of this nature should be evaluated for their potential to alleviate the metabolic difficulty of the diet, ensuring the maximum potential effectiveness of the treatment.[28]

CONCLUSION

Despite extensive research and clinical trials over the past 50 years, very little progress has been made to significantly alter the lethal prognosis of GBM brain tumors. This lack of an effective SOC obliges a reexamination of the disease and our current approach and has inspired the pursuit of novel therapeutics. In fact, current treatments may in part lead to enhanced tumor growth by increasing the metabolic fuel for cancer cells. Due to the current lack of effective long-term data we are not advocating the intervention as a standalone therapy, but we believe there is now a sound scientific basis for evaluating metabolic therapy as an adjunct for the treatment of malignant brain tumors. By recognizing the underlying unique energy production requirements of cancer cells, nutritional strategies are proposed to induce ketosis and reduce glucose levels to restrict cancer cell growth.

In this setting, clinicians specialized in metabolism and nutrition should be included with the neuro-oncology teams treating patients with GBM. This approach may be particularly worthy for patients who are nonsurgical by choice or because of technical reasons, and as an adjunct to radiation and chemotherapy treatments. Complimentary treatments using antiglycolytic drugs, selected tumor suppressing nutrients and the CR-KD should be considered in clinical trials as alternative or as adjunctive treatment to standard cancer therapies. Metabolic therapy, particularly the CR-KD, may enhance cancer treatment protocols by reducing glucose and glutamate levels, thus possibly extinguishing the neoplastic "fire" of GBM.

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