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Press-pulse: a novel therapeutic strategy for the metabolic management of cancer

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Abstract

Background: A shift from respiration to fermentation is a common metabolic hallmark of cancer cells. As a result, glucose and glutamine become the prime fuels for driving the dysregulated growth of tumors. The simultaneous occurrence of "Press-Pulse" disturbances was considered the mechanism responsible for reduction of organic populations during prior evolutionary epochs. Press disturbances produce chronic stress, while pulse disturbances produce acute stress on populations. It was only when both disturbances coincide that population reduction occurred.

Methods: This general concept can be applied to the management of cancer by creating chronic metabolic stresses on tumor cell energy metabolism (press disturbance) that are coupled to a series of acute metabolic stressors that restrict glucose and glutamine availability while also stimulating cancer-specific oxidative stress (pulse disturbances). The elevation of non-fermentable ketone bodies protect normal cells from energy stress while further enhancing energy stress in tumor cells that lack the metabolic flexibility to use ketones as an efficient energy source. Mitochondrial abnormalities and genetic mutations make tumor cells vulnerable metabolic stress.

Results: The press-pulse therapeutic strategy for cancer management is illustrated with calorie restricted ketogenic diets (KD-R) used together with drugs and procedures that create both chronic and intermittent acute stress on tumor cell energy metabolism, while protecting and enhancing the energy metabolism of normal cells.

Conclusions: Optimization of dosing, timing, and scheduling of the press-pulse therapeutic strategy will facilitate the eradication of tumor cells with minimal patient toxicity. This therapeutic strategy can be used as a framework for the design of clinical trials for the non-toxic management of most cancers.

Keywords: Glucose, Glutamine, Mitochondria, KETONE bodies, Diet, Warburg effect, Cancer metabolism, Glutaminolysis, Hyperbaric oxygen

Background

According to the paleobiologists, Arens and West, the simultaneous occurrence of "Press-Pulse" disturbances was considered the mechanism responsible for the extinction of organic populations during prior evolutionary epochs [1]. A "press" disturbance was considered a chronic environmental stress on all organisms in an ecological community. The press disturbance promoted extinction through habitat loss, reduced reproduction, and restriction of range and resources. Press disturbances would force a biological community into a new equilibrium where previously important species become non-viable. A press disturbance would shift the adaptive landscape to favor the fittest species while eliminating the weakest species. In contrast to the press disturbances, "pulse" disturbances were considered acute events that disrupted biological communities to produce high mortality [1]. Through extensive mortality in the immediate aftermath of the event, a pulse disturbance could cause extinction. However, survival of some species could occur following a pulse disturbance, as the physical and biotic environments would eventually recover to their pre-disturbance equilibria [1]. It was only when both the press and the pulse disturbances coincided that mass extinction of species, without recovery, was possible. We describe how a modification of the press-pulse concept can be adopted as a therapeutic

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strategy for the possible eradication of tumor cells. The press-pulse concept should be best considered in light of current views on the origin of cancer.

The origin of cancer

Cancer is a systemic disease involving multiple time- and space-dependent changes in the health status of cells and tissues that ultimately lead to malignant tumors [2]. Neoplasia involving dysregulated cell growth is the biological endpoint of the disease [3, 4]. Tumor cell invasion into surrounding tissues and their spread (metastasis) to distant organs is the primary cause of morbidity and mortality of most cancer patients [5–9]. Data from the American Cancer Society show that the rate of increase in cancer deaths/year (3.4%) was two-fold greater than the rate of increase in new cases/year (1.7%) from 2013 to 2017 [10, 11]. Indeed, cancer is predicted to overtake heart disease as the leading cause of death in Western societies. The failure to clearly define the origin of cancer is responsible in large part for the failure to significantly reduce the cancer death rate from treatments and in developing cancer prevention strategies [12].

Cancer is generally considered a genetic disease where random somatic mutations underlie the origin and progression of the disease [4, 13–16]. This general view is now under serious reconsideration in light of major inconsistencies with the gene theory [2, 3, 12, 14, 17–24]. Emerging evidence from the cancer genome projects shows that most malignant tumors are remarkably heterogeneous [2, 15, 16, 25–27]. This degree of heterogeneity will confound attempts to exploit genomic defects for effective therapies. Moreover, the majority of genetic mutations are considered downstream epiphenomena of dysregulated energy metabolism [2, 20, 28]. In contrast to the extensive genetic heterogeneity seen in tumors, most if not all neoplastic cells within tumors share the common metabolic malady of aerobic fermentation that arises ultimately from dysregulated oxidative phosphorylation [2, 17, 29–33]. In light of these findings, cancer can also be recognized as a metabolic disease.

Methods

Aerobic fermentation: a common metabolic malady of tumor cells

Most cells of the body oxidize glucose to CO_2 and water for energy production. Before entering the mitochondria for complete oxidation, glucose is first split into two molecules of pyruvate through the Embden–Meyerhof–Parnas glycolytic pathway in the cytosol. As most cells are bathed in oxygen, the production of pyruvate occurs through aerobic glycolysis [34]. Under hypoxia, however, much of the pyruvate is reduced to lactic acid in order to maintain cell ATP production. Aerobic fermentation, on the other hand, involves the production of lactic acid under normoxic conditions. As the Pasteur effect should reduce lactic acid fermentation under normoxia, persistent lactic acid production in the presence of adequate oxygen is indicative of abnormal respiration [35]. Otto Warburg first proposed that all cancers arise from damage to cellular respiration. As a result, cancer cells increase their capacity to produce lactic acid even in the presence of oxygen in order to compensate for their insufficient respiration [31, 36].

Although Warburg's hypothesis on the origin of cancer has created confusion and controversy [37–40], his hypothesis has never been disproved. The Crabtree effect and the high oxygen consumption rate seen in some tumor cells have confused the picture of defective oxidative phosphorylation in tumor cells. The Crabtree effect is an artifact of the in vitro environment and involves the glucose-induced suppression of respiration with a corresponding elevation of lactic acid production even under hyperoxic (pO^2 = 120–160 mmHg) conditions associated with cell culture, [41, 42]. Also, the oxygen consumption seen in tumor cells is not always linked to ATP production through oxidative phosphorylation and cannot therefore be used alone as evidence of normal respiration [29, 43–48]. It can be difficult to accurately measure mitochondrial respiratory function in cultured cells unless appropriate controls are used, as the in vitro environment can alter mitochondrial function [41, 49]. These issues have confounded the interpretation of Warburg's findings despite his attempts to clarify the issues [32, 48, 50]. Nevertheless, the Warburg theory of insufficient aerobic respiration remains as the most credible explanation for the origin of tumor cells [2, 37, 51–57].

The main points of Warburg's theory are; 1) insufficient respiration is the predisposing initiator of tumorigenesis and ultimately cancer, 2) energy through glycolysis gradually compensates for insufficient energy through respiration, 3) cancer cells continue to produce lactic acid in the presence of oxygen, and 4) respiratory insufficiency eventually becomes irreversible [2, 31, 32, 36, 58, 59]. Warburg referred to the phenomenon of enhanced glycolysis in cancer cells as "aerobic fermentation" to highlight the abnormal production of lactic acid in the presence of oxygen [31, 32, 36, 58, 59]. Efraim Racker coined the term "Warburg effect", which refers to the aerobic glycolysis that occurs in cancer cells [60]. Although Warburg insisted that aerobic glycolysis confuses the issue of insufficient respiration as the origin of cancer [31, 32], some in the cancer metabolism field have persisted in thinking that aerobic glycolysis (Warburg effect) is a central issue in cancer metabolism [39, 61]. Warburg clearly demonstrated that aerobic fermentation (aerobic glycolysis) is an effect, and not the

cause, of insufficient respiration [36]. Hence, the targeting of fermentable fuels becomes of prime importance for cancer management.

Substantial evidence exists showing that many cancers avidly consume glucose and produce lactic acid [62–67]. The diagnostic procedure of ^{18}F-deoxyglucose positron emission tomography (FDG-PET) is considered evidence for the elevated use of glucose by some tumors [66]. Elevated glucose consumption would be expected for any glucose-dependent cell with quantitative or qualitative abnormalities in mitochondria, as enhanced fermentation would be needed to compensate for the insufficient respiration [43, 68]. Indeed, all tumor cells that have been examined to date contain abnormalities in the content or composition of cardiolipin, the signature lipid of the inner mitochondrial membrane that regulates oxidative phosphorylation [69–74]. Mammalian cells containing abnormalities in cardiolipin cannot respire effectively and will therefore need to increase energy production through fermentation reactions [41, 70, 73, 75–78]. This fact cannot be overemphasized considering arguments that tumor cells can have normal respiration [39, 61, 79]. The expression of immature cardiolipin linked to reduced Complex I activity in the inner mitochondrial membrane of tumorigenic and non-tumorigenic cells suggests that many proliferative cells grown in culture obtain energy through fermentation rather than through oxidative phosphorylation despite the appearance of normal oxygen consumption [41, 43]. The cardiolipin abnormalities found in tumor cells provide direct support for Warburg's central theory. In addition to cardiolipin abnormalities, Pedersen also showed that some degree of abnormality could be found in the number, structure, or function of tumor cell mitochondria providing further support for Warburg's theory [68]. The evidence supporting Warburg's original theory comes from a broad range of cancers and is now overwhelming [2, 36, 53, 80–85]. Hence, respiratory insufficiency, arising from any number mitochondrial defects, can contribute to the fermentation metabolism seen in tumor cells.

Although the abnormal energy metabolism and mitochondrial abnormalities seen in most cancers could arise in part through oncogenic modulation of metabolism [4, 39, 86], the data from the nuclear and mitochondrial transfer experiments suggest that oncogene changes are effects, rather than causes, of tumorigenesis [2, 14, 24, 87, 88]. Normal mitochondria can suppress tumorigenesis, whereas abnormal mitochondria can enhance tumorigenesis [14, 87]. The results from these experiments must be viewed together, as results from any given single experiment are not capable of overturning the gene theory [14]. Recent advances in CRISPR/Cas9 technology might help to generate nuclei with changes in specific tumor-associated genes to further evaluate the influence of gene mutations and mitochondrial function on tumorigenesis. The acquisition of dysfunctional mitochondria in macrophages through fusion hybridization with non-metastatic tumor cells provides a compelling argument for the origin of those cancer cells that become metastatic [5, 89–91]. We recently showed how all of the Hanahan & Weinberg hallmarks of cancer, including the genomic mutations, could be linked either directly or indirectly to mitochondrial dysfunction [2, 56, 92].

Amino acid fermentation could also drive cancer metabolism

As the result of insufficient aerobic respiration, cancer cells must rely primarily on fermentation metabolism to maintain energy balance and viability. Besides substrate level phosphorylation in the cytoplasm through lactic acid fermentation, TCA cycle substrate level phosphorylation can also produce significant amounts ATP [93–98]. In addition to glucose, cancer cells also rely heavily on glutamine for growth and survival [99–102]. Glutamine is anapleurotic and can be rapidly metabolized to glutamate and then to α-ketoglutarate for entry into the TCA cycle. In addition to serving as a carbon/nitrogen source for tumor cell growth, glutamine also plays a role in cancer cell survival and growth through enzymatic release of ammonia into the microenvironment [103]. The TCA cycle succinate thiokinase reaction could generate the majority of cellular ATP through substrate level phosphorylation under hypoxia or in tumor cells with defective oxidative phosphorylation [78]. Mitochondrial ATP production through TCA cycle substrate level phosphorylation, using glutamine as a substrate, could give the appearance that mitochondrial energy metabolism is normal in some cancer cells especially in combination with oxygen consumption and CO_2 production. Although Warburg did not address the role of TCA cycle substrate level phosphorylation in his original work [31, 36], an increase in TCA cycle substrate level phosphorylation would be expected in cells with OxPhos deficiencies, just as lactic acid fermentation is expected in cells with this deficiency. Further studies will be needed to substantiate the role of glutamine fermentation in cancer cells.

Glucose and glutamine act synergistically for driving rapid tumor cell growth. Glutamine metabolism can produce ATP from the TCA cycle under aerobic conditions. Glutamine is also a nitrogen donor for nucleotide biosynthesis and can serve as precursor for lipid synthesis under hypoxic conditions [104, 105]. We also found that only minor amounts of glutamine are metabolized to lactic acid under either normoxia or hypoxia in the VM-M3 invasive glioblastoma cells consistent with

findings in other tumor cells [105–107]. We suggest that the metabolism of glucose and glutamine for energy will depend on the physiological state of the tumor microenvironment, and will be of greater significance in tumors with an aggressive Warburg phenotype. We found that glutamine targeting can be effective in managing systemic metastatic cancer in the VM/Dk mice [108].

Amino acid fermentation can generate energy through TCA cycle substrate level phosphorylation under hypoxic conditions [94, 96, 97, 109, 110]. Succinate is a waste product of amino acid fermentation that can enhance inflammation as well as inhibit a family of prolyl hydroxylases, which facilitate Hif-1α degradation through the von Hippel–Lindau (VHL) gene product [111–113]. Through its action on several glycolytic pathways, Hif-1α stabilization enhances aerobic fermentation [114–116]. It can be difficult to determine, however, the degree to which mitochondrial ATP production in tumor cells arises from coupled respiration or from TCA cycle substrate level phosphorylation [94, 98].

Several byproducts of amino acid fermentation can also accumulate in the tumor microenvironment including acetate, glutamate, alanine, succinate, and ammonia. Although acetate has been considered a potential fuel for supporting tumor cell growth [117, 118], acetate levels are generally low in the circulation [119]. Jaworski et al. recently provided a comprehensive discussion on the potential role of acetate in tumor metabolism [120]. It should be recognized that with the exception of glucose and glutamine, none of the other potential fuels needed for tumor cell fermentation would likely be available in sufficient quantities to drive robust tumor cell growth. As many amino acids are synthesized from glucose and glutamine, targeting glucose and glutamine will deprive the microenvironment of fermentable fuels. Hence, the restriction of glucose and glutamine becomes of prime importance for targeting tumor cell growth and survival. The role of glucose and glutamine in driving tumor cells energy metabolism is shown in Fig. 1.

Tumor cell energy metabolites from cannibalism and phagocytosis

Emerging evidence indicates that macrophages, or their fusion hybridization with neoplastic stem cells, are the origin of metastatic cancer cells [5, 89, 121–124]. Radiation therapy can enhance fusion hybridization that could increase risk for invasive and metastatic tumor cells [91, 125]. Cannibalism and phagocytosis of cellular debris are well known characteristics of macrophages and of myeloid cancer cells with macrophage properties [121, 126–131]. Shelton showed that glioblastoma cells with myeloid properties could survive in Matrigel (extracellular matrix material) in the absence of added glucose and glutamine [132]. The gradual accumulation of lactate in the media suggested that the glioblastoma cells survived through lysosomal digestion and aerobic fermentation of glycoconjugates present in the Matrigel. Glioblastoma cell death occurred immediately following the addition of chloroquine, which neutralizes lysosomal acidity and digestion [132]. Shelton's findings are consistent with the more recent findings of Kamphorst et al. in showing that pancreatic ductal adenocarcinoma cells could obtain glutamine under nutrient poor conditions through lysosomal digestion of extracellular proteins [133]. It will therefore become necessary to also target lysosomal digestion, under reduced glucose and glutamine conditions, to effectively manage those invasive and metastatic cancers that express cannibalism and phagocytosis.

Genome integrity and energy metabolism

Emerging evidence indicates that the function of DNA repair enzymes and the integrity of the nuclear genome are dependent to a large extent on the energy derived from normal respiration [134–142]. Previous studies in yeast and mammalian cells show that disruption of aerobic respiration can cause mutations (loss of heterozygosity, chromosome instability, and epigenetic modifications etc.) in the nuclear genome [28, 141, 143, 144]. A protracted reliance on fermentation causes oxidative stress leading to the production of reactive oxygen species (ROS) mostly through the mitochondrial coenzyme Q couple [145]. In addition to their role in oncogenic signaling, excess ROS production damages mitochondrial function, and can be both carcinogenic and mutagenic [146, 147]. The somatic mutations and genomic instability seen in tumor cells thus arise from a protracted reliance on fermentation energy metabolism and a disruption of redox balance through excess oxidative stress.

We recently discussed how a transition from respiration to fermentation could account for Szent-Gyorgi's "Oncogenic Paradox", i.e., the process by which various provocative agents (radiation, inflammation, hypoxia, carcinogenic chemicals, age, germline mutations, etc.) could produce cancer through a common pathological mechanism [2, 148]. Mukherjee and Cairns also struggled to explain the oncogenic paradox [149, 150]. All of these provocative cancer-causing agents damage respiration thus forcing the cells to rely more heavily on energy generated through fermentation for survival. According to the mitochondrial metabolic theory of cancer, the large genomic heterogeneity seen in tumor cells arises as a consequence, rather than as a cause, of mitochondrial dysfunction [2, 14, 28]. A therapeutic strategy targeting the metabolic abnormality common to most tumor cells should therefore be more effective in managing cancer than would a strategy targeting genetic

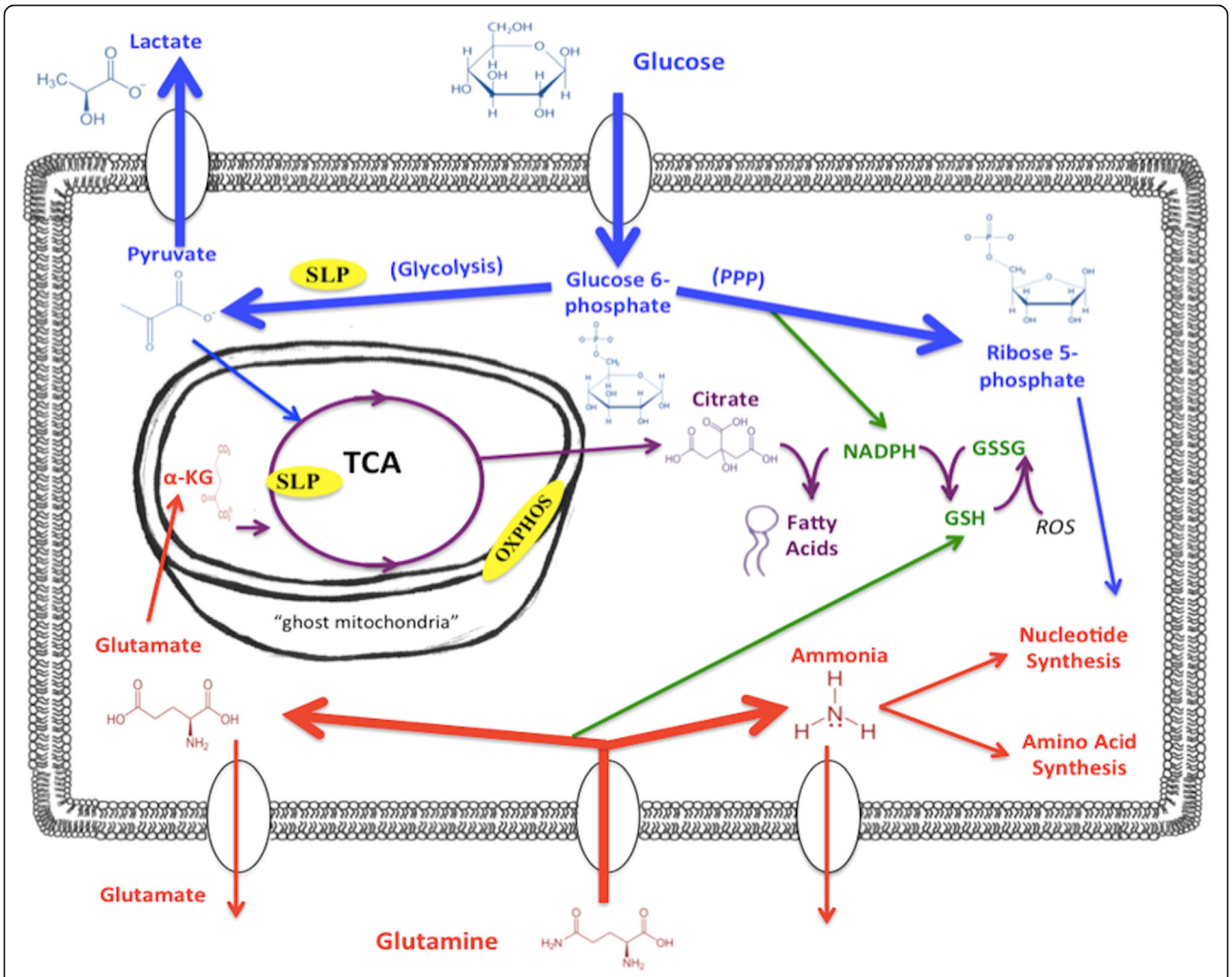

Fig. 1 Targeting Glucose and Glutamine for the Metabolic Management of Cancer. Cancer cells are largely dependent on glucose and glutamine for survival and growth. Energy through fermentation metabolism (substrate level phosphorylations, SLP) in glycolysis and the tricarboxylic acid cycle (TCA) will compensate for reduced energy through oxidative phosphorylation (OxPhos) that occurs in tumor cells. The yellow ovals indicate the three source of cellular ATP production. Glucose carbons can be used for both the glycolytic and pentose phosphate (PPP) pathways to supply ATP and precursors for lipid and nucleotide synthesis, as well as for glutathione production. Glutamine provides its amide nitrogen for ammonia and nucleotide synthesis. The glutamine-derived glutamate provides anapleurotic carbons to the TCA cycle through α-KG for protein synthesis while also providing ATP through TCA cycle SLP. TCA cycle substrate level phosphorylation through the succinate thiokinase reaction can generate significant cellular ATP under hypoxia especially in tumor cells with defective respiration [78]. The glutamine-derived glutamate is also used for glutathione production that protects tumor cells from oxidative stress. Glucose and glutamine targeting will thus make cancer cells vulnerable to oxidative stress therapies. The simultaneous targeting of glucose and glutamine through the press-pulse therapeutic strategy will starve tumor cells of energy production while blocking their ability to synthesize proteins, lipids, and nucleotides. Glucose and glutamine can also be generated internally through the lysosomal digestion of phagocytosed glycoconjugates and proteins (see text). An elevation of non-fermentable ketone bodies through, calorie restriction, ketogenic diets, or supplementation will provide normal cells with an alternative energy source to glucose while also protecting them from oxidative stress. Ghost mitochondria are those containing little or no inner mitochondrial membranes (cristae), which are essential for normal OxPhos function [67, 282, 283]

mutations that vary widely between tumors of the same histological grade and even within the same tumor.

Human evolution and adaptive versatility

Rick Potts, a paleoanthropologist at the Smithsonian Institution, suggested that the evolutionary success of our species has been due largely to the germline inheritance of traits that bestowed adaptive versatility [151–153]. Adaptability was defined in terms of, 1) the ability of an organism to persist through major environmental shifts, 2) to spread to new habitats, and 3) to respond in novel ways to its surroundings [153]. These characteristics were honed over millions of years and enabled humans to adapt rapidly to abrupt changes in the physical environment including changes in moisture, temperature, food resources etc. Adaptability to

abrupt environmental change is a property of the genome, which was selected for in order to ensure survival under environmental extremes [65, 154].

Potts' hypothesis is an extension of Darwin's original theory (Chapter IV, Natural Selection) and can be applied to the individual cells of the organism, which exist as an integrated society of cells [65, 154]. The success in dealing with environmental stress and disease is therefore dependent on the integrated action of all cells in the organism. Further, this integrated action depends on the flexibility of each cell's genome, which responds to both internal and external signals according to the needs of the organism. More specifically, only those cells possessing flexibility in nutrient utilization will be able to survive under nutrient stress. Environmental forcing has therefore selected those genomes most capable of adapting to change in order to maintain metabolic homeostasis [65, 152, 153, 155]. This concept was first discussed in relationship to the management of brain cancer [65].

The widely held notion that tumor cells have a growth advantage and are more fit than normal cells are in contrast to Darwin's theory of evolution and also to Potts' theory of adaptive versatility [65, 153, 154]. It is difficult to conceive how a random accumulation of somatic mutations could enhance the adaptability and fitness of cancer cells. It is important to recognize that mutations in *p53*, *K-Ras*, and *Raf* impact negatively on mitochondrial energy efficiency thus making cells with these mutations less metabolically flexible than normal cells [28, 44, 53, 135, 156–159]. Indeed activating mutations in *K-Ras* target mitochondria, thus enhancing glycolysis [53, 160]. Enhanced glycolysis will make tumor cells appear more metabolically fit than normal cells in hypoxic environments [161, 162]. Most normal cells, however, cannot grow in hypoxia and will often die in hypoxic environments due to respiratory failure. Tumor cells are more fit than normal cells to survive in the hypoxic niche of the tumor microenvironment. Hypoxic adaptation of tumor cells allows for them to avoid apoptosis due to their metabolic reprograming following a gradual loss of respiratory function [31, 32, 162, 163]. The high rates of tumor cell glycolysis and glutaminolysis will also make them resistant to apoptosis, ROS, and chemotherapy drugs [163]. Despite having high levels of ROS, glutamate-derived from glutamine contributes to glutathione production that can protect tumor cells from ROS [164]. As long as the tumor cells have access to the metabolic fuels needed for glycolysis and TCA cycle substrate level phosphorylation (glucose and glutamine, respectively) they will give the appearance of having a growth advantage over most normal cells [2]. According to Darwin and Potts, mutations that bestow a selective advantage are those that will enhance survival under environmental stress. If the multiple pathogenic point mutations, chromosomal rearrangements, and mitochondrial abnormalities confer a fitness or survival advantage to tumor cells, then survival under environmental stress and nutrient deprivation should be better in tumor cells than in normal cells [165]. This is not what actually happens, however, when the hypothesis is tested.

For example, when mice or people with tumors are placed under energy stress using dietary energy reduction (glucose restriction), many tumor cells die while normal cells survive. Indeed, the health and vitality of the normal cells improves with time under dietary energy reduction while hyper-glycolytic tumor cells undergo energetic crisis triggering apoptotic death [166, 167]. Support for this contention comes from studies of treating brain tumors with dietary energy stress [114, 168–174]. It is clear that adaptability to environmental stress is greater in normal cells than in tumor cells, as normal cells can transition from the metabolism of glucose to the metabolism of ketone bodies when glucose becomes limiting. Mitochondrial oxidative phosphorylation is less robust in tumor cells than in normal cells while glucose utilization through lactic acid fermentation is greater in tumor cells than in normal cells. Targeting glucose availability will therefore cause greater death in the tumor cells than in the normal cells. Mitochondrial respiratory chain defects will prevent tumor cells from using ketone bodies for energy [145]. Consequently, glycolysis-dependent tumor cells are less adaptable to metabolic stress than are the normal cells. This vulnerability can be exploited for targeting tumor cell energy metabolism [160, 163].

It is also possible that therapeutic energy stress could restore the microenvironment thus reversing abnormal energy metabolism and growth behavior in tumor cells not containing genetic mutations [19, 175]. In contrast to dietary energy reduction, radiation and toxic drugs can damage the microenvironment and transform normal cells into tumor cells while also creating tumor cells that become highly resistant to drugs and radiation. Drug-resistant tumor cells arise in large part from the damage to respiration in bystander pre-cancerous cells. These cells are often those that eventually become heavily dependent on fermentation for energy.

The greater adaptability of normal cells than tumor cells to energy stress is predicted based on the theories of Darwin and Potts [154]. Metabolic flexibility allows the organism to respond in a coordinated way to environmental stress and limited substrate availability. Energy stress will force all normal cells to work together for the survival of the organism [154]. Pathogenic mutations and genomic instability will reduce adaptability and metabolic flexibility under energy stress. The greater the genomic instability in tumor cells, the less will be their adaptability to stress. This concept is similar to that of Nowell's except in viewing genomic instability as a liability

rather than as an advantage to progression [154, 176]. Because energy generated through substrate level phosphorylation is greater in tumor cells than in normal cells, tumor cells are more dependent than normal cells on the availability of fermentable fuels (glucose and glutamine) [94]. With few exceptions, most normal cells shift energy metabolism from glucose to ketone bodies and fats when placed under energy stress from glucose deprivation, insulin deficiency, and prolonged fasting. This shift is the result of adaptive versatility and genomic stability, which is lacking in the tumor cells but is prominent in cells and tissues with robust mitochondrial function.

Tumor cells will have difficulty surviving and growing, regardless of their complement of genomic changes, if fermentable fuels become restricted in the microenvironment. Ketone bodies and fats are non-fermentable fuels [177]. Tumor cells have difficulty using ketone bodies and fats for fuel when glucose is reduced [57, 178–180]. Although some tumor cells might appear to oxidize ketone bodies by the presence of ketolytic enzymes [181], it is not clear if ketone bodies and fats can provide sufficient energy for cell viability in the absence of glucose and glutamine. The studies in immunocompetent syngeneic mice and xenografts with brain tumors are proof of concept that tumor cells are less adaptable than normal cells when placed under energy stress [114, 170, 171, 182–184]. Apoptosis under energy stress is greater in tumor cells than in normal cells [170]. The multiple genetic defects in tumor cells will reduce genomic flexibility thus increasing the likelihood of cell death under environmental stress that would lower glucose and elevate ketone bodies. Regardless of when or how genomic defects become involved in the initiation or progression of tumors, these defects can be exploited for tumor management or resolution [12].

Results

Press-pulse: a therapeutic strategy for the gradual elimination of cancer cells

Mark Vincent suggested how a Press-Pulse strategy could be used to target tumor cells [185]. We have now expanded this concept to show how a press-pulse therapeutic strategy can be used for the non-toxic management and possible resolution of cancer. A calorie restricted ketogenic diet or dietary energy reduction creates chronic metabolic stress in the body. This energy stress acts as a press disturbance; the effects of which would be greater in the tumor cells than in the normal cells due to their dependency on fermentation energy metabolism, mitogens, anabolic signaling (IGF-1, mTOR, etc.), elevated redox stress, and mutational load. Drugs that target availability of glucose and glutamine would act as pulse disturbances in causing an acute reduction of these tumor-dependent fuels [186]. Hyperbaric oxygen therapy can also be considered another pulse disturbance in elevating ROS to a greater degree in tumor cells than in normal cells, thus promoting cancer cell death through redox stress [40]. Normal cells readily transition to ketone body metabolism for protection against ROS damage and oxidative stress. The goal therefore is to produce a therapeutic strategy that can more effectively manage cancer than can the toxic cancer therapies currently used in most standards of care. The following examples illustrate the potential of press-pulse therapeutic strategies for cancer management.

Calorie restriction and restricted Ketogenic diets: a press disturbance

Calorie restriction, water-only fasting, and restricted ketogenic diets reduce circulating glucose and insulin levels while elevating circulating levels of ketone bodies. Ketogenic diets (KDs) are low carbohydrate-high fat diets that are widely used to reduce refractory epileptic seizures in children [187, 188]. The KD can more effectively reduce glucose and elevate blood ketone bodies than can CR alone making the KD potentially more therapeutic against tumors than CR [174, 189, 190]. The protein and fat composition of the KD differs from that of Atkins-type diets in having comparatively less protein and more fat than the Atkins diets. This is important as several amino acids found in proteins can be deaminated to form pyruvate, which can then be metabolized to form glucose through gluconeogenesis [191]. Campbell showed that tumor growth in rats is greater under high protein (>20%) than under low protein content (<10%) in the diet [192]. Protein amino acids can be metabolized to glucose through the Cori cycle. The fats in KDs used clinically also contain more medium chain triglycerides than do Atkins diets. Consequently, blood glucose levels will be lower and ketone body levels will be higher with KDs than with Atkins-type diets. Calorie restriction, fasting, and restricted KDs are anti-angiogenic, anti-inflammatory, and pro-apoptotic and thus can target and eliminate tumor cells through multiple mechanisms [114, 166, 171, 174, 182, 193, 194]. Ketogenic diets can also spare muscle protein, enhance immunity, and delay cancer cachexia, which is a major problem in managing metastatic cancer [195–198].

The therapeutic effects of KDs used alone or in combination with other therapies have been documented in preclinical studies for several cancer models including neuroblastoma [199, 200], lung cancer [201], prostate cancer [202, 203], breast and ovarian cancers [204, 205], head & neck cancers [204], colon cancer [206], and pancreatic cancer [198]. These preclinical studies are

also motivating case reports and pilot studies in humans with brain cancer and other cancers [169, 181, 207–214]. It is clear from these studies and other studies in children and adults with cancer that KDs are generally safe and well tolerated [168, 212, 213, 215–217], These observations are also consistent with decades of research obtained from evaluation of children treated with KDs for epilepsy management [218]. Information on ketogenic diets can be obtained from the Charlie Foundation web site (https://www.charliefoundation.org).

We recently developed the Glucose/Ketone Index calculator (GKIC) to assess the potential therapeutic effects of various low-carbohydrate and KDs for brain cancer management [189]. The GKIC is a simple tool that measures the ratio of blood glucose to blood ketones and can help monitor the efficacy of metabolic therapy in preclinical animal models and in clinical trials for malignant brain cancer or for any cancer that expresses aerobic fermentation. GKI values of 1.0 or below are considered therapeutic, though therapeutic benefit appears to be associated more with elevated ketone bodies and suppression of insulin than with reduced glucose [190, 215]. However, the elevation of ketone body levels is generally greater when blood glucose levels are lower than when glucose levels are higher [174, 219, 220]. The GKI can therefore serve as a biomarker to assess the therapeutic efficacy of various diets in a broad range of cancers.

Reduced glucose availability and suppression of insulin signaling will produce chronic energy stress on those tumor cells that depend primarily on glucose for growth and survival. It is important to remember that insulin drives glycolysis through stimulation of the pyruvate dehydrogenase complex [221, 222]. Reduced levels of glucose will also reduce substrates for both the glycolytic and the pentose phosphate pathways thus reducing cellular energy, and the synthesis of glutathione and nucleotide precursors (Fig. 1).

The water-soluble ketone bodies (D-β-hydroxybutyrate and acetoacetate) are produced largely in the liver from adipocyte-derived fatty acids and ketogenic dietary fat. Ketone bodies bypass glycolysis and directly enter the mitochondria for metabolism to acetyl-CoA [223]. In contrast to fatty acid metabolism, which generates both NADH and $FADH_2$, ketone body metabolism generates only NADH [145]. Moreover, ketone body metabolism does not induce mitochondrial uncoupling in contrast to metabolism of saturated fatty acids [145]. The metabolism of D-β-hydroxybutyrate in normal cells will therefore increase the redox span between Complexes I and III, thus increasing the delta G' of ATP hydrolysis while, at the same time, reducing ROS formation through the Complex II coenzyme Q couple [224, 225]. Due to mitochondrial defects, tumor cells cannot exploit the therapeutic benefits of burning ketone bodies as normal cells would. Indeed, racemic mixtures of D-/L-ketone bodies can be toxic to tumor cells under both low and high glucose conditions [57, 190]. Fine et al. found that uncoupling protein 2 is overexpressed in tumor cells, but not in normal control cells [226]. This finding provides a plausible molecular mechanism by which ketone bodies spare normal cells but suppresses growth in cancer lines.

In contrast to D-β-hydroxybutyrate, L-β-hydroxybutyrate is beta-oxidized thus producing both NADH and $FADH_2$. $FADH_2$ will deliver electrons to Complex III, which can increase the semiquinone of Q, the half-reduced form. The Q semiquinone will react with molecular oxygen to form the superoxide O_2^- free radical [145]. Therapeutic ketosis with racemic ketone esters can also make it feasible to safely sustain hypoglycemia for inducing metabolic stress on cancer cells [227]. Hence, mixtures of L- and D-ketone esters have the potential to both enhance oxidative stress in tumor cells while reducing oxidative stress in normal cells, respectively [145, 228]. There is also evidence showing that ketone bodies can inhibit histone deacetylases (HDAC) [229]. HDAC inhibitors play a role in targeting the cancer epigenome [230]. Deregulated inflammation is also considered to be one of the hallmarks of cancer. Therapeutic ketosis reduces circulating inflammatory markers, and ketones directly inhibit the NLRP3 inflammasome, an important pro-inflammatory pathway linked to carcinogenesis and an important target for cancer treatment response [231]. There are no adverse side effects of short-term therapeutic ketosis, but relatively mild adverse effects have been noted in some children with epilepsy after long-term use of ketogenic diets including constipation, kidney stones, electrolyte imbalances, and bone fracture [218]. These adverse effects were easily managed with various supplements and pale in comparison to the adverse effects produced from current standards of care [232]. In general, there are no currently known cancer drugs that embody the therapeutic properties of ketone bodies.

Psychological stress reduction: a press disturbance

Chronic psychological stress is known to promote tumorigenesis through elevations of blood glucose, glucocorticoids, catecholamines, and insulin-like growth factor (IGF-1) [233, 234]. In addition to calorie-restricted ketogenic diets, psychological stress management involving exercise, yoga, music etc. also act as press disturbances that can help reduce fatigue, depression, and anxiety in cancer patients and in animal models [235–238]. Ketone supplementation has also been shown to reduce anxiety behavior in animal models [239]. The mechanism of action of psychological stress management for cancer control would largely involve reductions in blood glucose levels that contribute to tumor growth.

Restricted ketogenic diet used with 2-Deoxyglucose

Calorie restriction or therapeutic fasting is anti-angiogenic, anti-inflammatory, and pro-apoptotic, and thus targets multiple cancer hallmarks [114, 166, 167, 170, 171, 182, 240–243]. This physiological state also enhances the efficacy of chemotherapy and radiation therapy, while reducing the side effects [244–246]. Indeed, lower dosages of chemotherapeutic drugs can be used when administered together with calorie restriction or restricted ketogenic diets (KD-R). We showed a synergistic interaction between a KD-R and the glycolysis inhibitor 2-deoxyglucose (2-DG) for the metabolic management of the syngeneic CT-2A malignant mouse glioma [247]. It was interesting to find that 2-DG (25 mg/kg) had no therapeutic effect on CT-2A tumor growth when administered alone to mice on a standard high carbohydrate diet, but had a powerful therapeutic effect when administered with a KD-R. Indeed, this relatively low dose of 2-DG became somewhat toxic when used with the KD suggesting that lower dosing of some tumor-targeting drugs could also be effective when administered with KD-R. Besides 2-DG, a range of other glycolysis inhibitors might also produce similar therapeutic effects when combined with the KD-R including 3-bromopyruvate, oxaloacetate, and lonidamine [58, 186, 248–250]. In the example here the KD-R is the press making cancer cells selectively vulnerable to death and the 2-DG is the pulse, which could be used intermittently or cycled to avoid toxicity.

Ketogenic diet used with radiation therapy

Adrienne Scheck and colleagues showed that the therapeutic efficacy of radiotherapy against the orthotopically grown GL261 mouse glioma could be greatly enhanced when combined with a commercially available ketogenic diet [183]. Mice fed the KetoCal ketogenic diet had elevated levels of β-hydroxybutyrate and an increased median survival of approximately 5 days relative to animals maintained on a high-carbohydrate standard diet alone. A synergistic interaction of the KD diet plus radiation was seen, as no bioluminescent signal was detected in 9 of 11 that received the combined treatment. Furthermore, no signs of tumor recurrence were seen for over 200 days when the treated mice were switched to the SD 101 days after tumor implantation. These findings suggest tumor resolution in some of the mice treated with the combined therapy. In this example, the KD is the press and radiotherapy is the pulse. It is important to recognize, however, that the radiotherapy used in glioma patients can damage the respiration of normal cells and increase availability of glutamine in the microenvironment, which can increase risk of tumor recurrence especially when used together with the steroid drug dexamethasone [31, 251–253].

A Ketogenic diet used with hyperbaric oxygen therapy

Poff and colleagues demonstrated that hyperbaric oxygen therapy (HBOT) enhanced the ability of the KD to reduce tumor growth and metastasis [40]. Evidence in animal models and in humans suggests that HBOT may have a modest anti-cancer effect when used alone [254], but appears most efficacious when it is used in combination with standard care. Indeed, HBOT has proven effective when used prior to radiation therapy for GBM [255]. The mechanism of HBOT in tumor management is not yet clear, but saturating the tumor with oxygen could reverse hypoxia and suppresses growth [254, 256] HBOT also increases oxidative stress and membrane lipid peroxidation of GBM cells in vitro [257]. The effects of the KD and HBOT can be enhanced with administration of exogenous ketones, which further suppressed tumor growth and metastasis [190]. Besides HBOT, intravenous vitamin C and dichloroacetate (DCA) can also be used with the KD to selectively increase oxidative stress in tumor cells [258, 259]. Recent evidence also shows that ketone supplementation may enhance or preserve overall physical and mental health [260, 261], which are often compromised due to disease progression and standard of care therapies. Under these conditions the KD with exogenous ketones serve as the press, while HBOT serves as the pulse. Although HBOT and radiotherapy kill tumor cells through oxidative stress, HBOT is less toxic to normal cells than is radiotherapy.

Calorie restriction used with glutamine targeting for metastatic cancer

Some tumors use glucose as a prime fuel for growth, whereas other tumors use glutamine as a prime fuel [102, 186, 262–264]. Glutamine-dependent tumors are generally less detectable than glucose-dependent under FDG-PET imaging, but could be detected under glutamine-based PET imaging [265]. Glutamine targeting should have therapeutic benefit against those tumors that depend on glutamine for growth and survival. We found that the highly metastatic VM-M3 tumor cells are dependent primarily on availability of glutamine for growth and ability to spread systemically [108]. The glutaminase inhibitor DON (6-diazo-5-oxo-L-norleucine) has shown therapeutic benefit in the clinic, as long as toxicity can be managed [186, 266]. DON could work best when combined with inhibitors of glycolysis such as lonidamine [186]. In addition to DON, other glutamine inhibitors ((bis-2-(5-phenylacetamido-1,2, 4-thiadiazol-2-yl)ethyl sulfide, BPTES, or CB-839) could also be therapeutic in targeting glutamine-dependent tumors [267]. A greater attention to possible adverse effects will be needed for glutamine targeting than for glucose targeting, as glutamine is involved with several essential

physiological functions especially for cells of the immune system [268, 269]. It might therefore be necessary to also periodically schedule glutamine supplementation with glutamine targeting to obtain maximum therapeutic benefit while protecting immune system function.

The VM-M3 tumor is an excellent model system for evaluating the role of glutamine as a metabolic driver of invasive and metastatic cancer. The VM-M3 tumor arose spontaneously in the brain of its syngeneic immunocompetent VM/Dk inbred mouse host [270]. The tumor was classified as a glioblastoma (GBM) based on histological appearance, invasive growth behavior in brain, and systemic metastasis when give access to extraneural sites [271–277]. The neoplastic VM-M3 tumor cells share several characteristics with mesenchymal microglia/macrophages, which are abundant in GBM and use glutamine as a major fuel [278, 279]. Although calorie restriction could partially reduce distal invasion of VM-M3 tumor cell in brain and reduce primary tumor growth in flank, CR did not prevent systemic metastasis despite causing reduction in blood glucose and elevation of ketone bodies [108, 280]. However, DON had a major effect in reducing both primary tumor size and systemic metastasis indicative of the importance of glutamine in driving this tumor [108]. A synergistic interaction was also seen when DON was combined with calorie restriction [281]. Modifications of DON scheduling, timing, and dosing would be needed to improve efficacy and reduce toxicity. In this example, CR is the press and DON is the pulse. As glutamine is a major fuel of immune cells, glutamine targeting should be effective in reducing most metastatic cancers that have characteristics of macrophages and other immune cells [121].

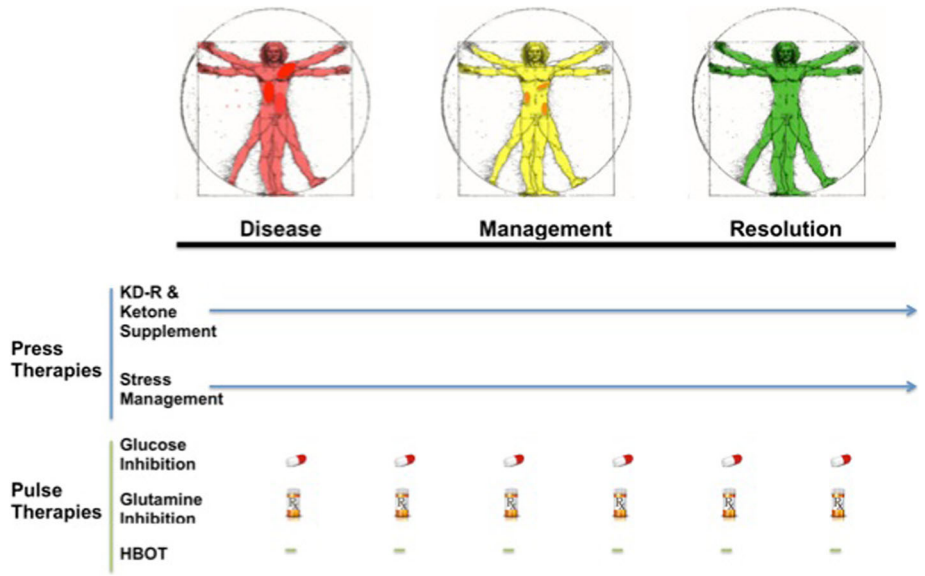

Fig. 2 Illustration of the Press-Pulse Therapeutic Strategy for Cancer Management. The "Press-Pulse" therapeutic strategy considers cancer as a singular systemic disease regardless of the specific tissue or organ system containing invasive or metastatic tumor cells. This strategy is designed to target the glucose and glutamine dependency of tumor cells, while enhancing the metabolic efficiency in normal cells. Press therapies are designed to reduce systemic glucose availability while elevating blood levels of ketone bodies, which tumor cells cannot effectively use for energy generation. This approach pits the metabolic demands of normal cells against those of the mutated tumor cells, which are less capable than normal cells in adapting to metabolic stress from nutrient deprivation. Ketone body supplements could further reduce glucose levels while enhancing the respiratory energy metabolism in normal cells. Stress management techniques together with exercise could further stress tumor cell metabolism while improving general health. The press therapies would be designed to work synergistically with acute pulse therapies to further target glucose and glutamine metabolism. HBOT will work together with the press therapies to selectively increase oxidative stress in tumor cells. The spacing between the various pulse therapies is designed to stress tumor cell metabolism while minimizing toxicity to normal body cells. This therapeutic strategy will target the fermentation metabolism common to most tumor cells, thus gradually degrading tumor burden. The progressive color change in the Vitruvian man drawing from red (diseased with darker red spots indicative of metastatic lesions), to yellow (reduced metastasis), to green (resolution) symbolizes a gradual metabolic management and resolution of cancer. The pill symbol is indicative of glycolysis targeting that could be delivered orally. The Rx symbol is indicative of glutamine targeting that could be delivered intravenously. Pulse therapies would terminate with evidence of management or resolution while press therapies could continue under modification or adjustment (*arrow*). Optimization of dosing, timing, and scheduling of the press-pulse therapeutic strategy will facilitate the eradication of tumor cells with minimal patient toxicity. This therapeutic strategy can be used as a framework for the design of clinical trials for the majority of cancers. HBOT, hyperbaric oxygen therapy; KD-R, calorie restricted ketogenic diet

Optimization of scheduling, timing, and dosing

The success of the press-pulse therapeutic strategy for the metabolic management of cancer will depend on optimization of the scheduling, dosing, and timing of the various diets, drugs, and procedures used in order to achieve maximum synergistic interactions (Fig. 2). Scheduling will involve the order in which the chosen pulses are delivered to the subject while under dietary therapy. Timing will determine when and for how long the presses and pulses are given (number/day,/week,/month etc.). Dosing will identify the optimal drug dosages needed to kill tumor cells while preventing or minimizing systemic toxicity. Scheduling for each of these variables can be adjusted for the age, sex, and general health status of the subject. The strategy should degrade tumor cell populations gradually to prevent tumor lysis syndrome, which could cause excessive toxicity. Tumor imaging procedures involving FDG-PET, magnetic resonance imaging (MRI), and computed tomography perfusion (CTP), as well as analysis of serum cancer biomarkers should be helpful in assessing therapeutic success. The goal of the press-pulse therapeutic strategy is to improve progression-free and overall survival from cancer without producing adverse effects from the treatment.

Discussion & Conclusions

Many of the current treatments used for cancer management are based on the view that cancer is a genetic disease. It is clear from the cancer death statistics that most current therapies are wanting in their ability to reduce the yearly death rate or to manage the disease without toxicity. Emerging evidence indicates that cancer is a mitochondrial metabolic disease that depends on availability of fermentable fuels for tumor cell growth and survival. Glucose and glutamine are the most abundant fermentable fuels present in the circulation and in the tumor microenvironment. The press-pulse therapeutic strategy is designed to target availability of glucose and glutamine thus starving tumor cells of their most important fuels and increasing their vulnerability to oxidative stress and apoptotic death. Low-carbohydrate, high fat-ketogenic diets coupled with glycolysis inhibitors will reduce metabolic flux through the glycolytic and pentose phosphate pathways needed for synthesis of ATP, lipids, glutathione, and nucleotides. DON and other similar glutamine inhibitors will deprive proliferating tumor cells of the glutamine needed for TCA cycle anaplerosis, and synthesis of glutathione, nucleotides, and proteins. Lysosomal targeting with chloroquine or similar drugs will reduce glucose and glutamine production following digestion of phagocytosed glycoconjugates and proteins. Glutamine targeting will require careful adjustments, however, as glutamine is a key metabolite needed for the immune system and for other physiological functions. Hyperbaric oxygen therapy combined with the calorie restricted ketogenic diet will kill tumor cells through apoptotic and anti-angiogenic mechanisms while also reducing inflammation in the tumor microenvironment and systemically. It is our view that the "Press-Pulse" paradigm is a compelling and parsimonious therapeutic strategy for effectively managing the vast majority of malignant cancers with minimal toxicity, as this approach will target the major energy pathways responsible for tumor cell growth and survival while enhancing the energetic efficiency of normal body cells and tissues.

Abbreviations

2-DG: 2-deoxyglucose; CR: Calorie restriction; DON: 6-diazo-5-oxo-L-norleucine; FAD: Flavin adenine dinucleotide; GBM: Glioblastoma multiforme; GKI: Glucose Ketone Index; HBOT: Hyperbaric oxygen therapy; KD-R: Restricted Ketogenic Diet; NAD: Nicotinamide adenine dinucleotide; ROS: Reactive Oxygen Species; SLP: Substrate level phosphorylation; TCA: Tricarboxylic acid

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Authors' contributions

TNS wrote most of the manuscript with the assistance of DPD, GY, and JCM. All authors read and approved the final manuscript.

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References

1. Arens NC, West ID. Press-pulse: a general theory of mass extinction? Paleobiology. 2008;34(4):456–71.
2. Seyfried TN, Flores RE, Poff AM, D'Agostino DP. Cancer as a metabolic disease: implications for novel therapeutics. Carcinogenesis. 2014;35(3):515–27.

3. Sonnenschein C, Soto AM. Somatic mutation theory of carcinogenesis: why it should be dropped and replaced. Mol Carcinog. 2000;29(4):205–11.
4. Hanahan D, Weinberg RA. Hallmarks of cancer: the next generation. Cell. 2011;144(5):646–74.
5. Seyfried TN, Huysentruyt LC. On the origin of cancer metastasis. Crit Rev Oncog. 2013;18(1–2):43–73.
6. Sporn MB. The war on cancer. Lancet. 1996;347(9012):1377–81.
7. Fidler IJ. The pathogenesis of cancer metastasis: the 'seed and soil' hypothesis revisited. Nat Rev Cancer. 2003;3(6):453–8.
8. Lazebnik Y. What are the hallmarks of cancer? Nat Rev Cancer. 2010;10(4):232–3.
9. Tarin D. Cell and tissue interactions in carcinogenesis and metastasis and their clinical significance. Semin Cancer Biol. 2011;21(2):72–82.
10. Siegel RL, Miller KD, Jemal A. Cancer Statistics, 2017. CA Cancer J Clin. 2017; 67:7–30.
11. Siegel R, Naishadham D, Jemal A. Cancer statistics, 2013. CA Cancer J Clin. 2013;63(1):11–30.
12. Seyfried TN. Cancer as a metabolic disease: on the origin, management, and prevention of cancer. Hoboken: Wiley; 2012.
13. Martincorena I, Campbell PJ. Somatic mutation in cancer and normal cells. Science. 2015;349(6255):1483–9.
14. Seyfried TN. Cancer as a mitochondrial metabolic disease. Front Cell Dev Biol. 2015;3:43.
15. Alexandrov LB, Nik-Zainal S, Wedge DC, Aparicio SA, Behjati S, Biankin AV, Bignell GR, Bolli N, Borg A, Borresen-Dale AL, et al. Signatures of mutational processes in human cancer. Nature. 2013;500(7463):415–21.
16. Vogelstein B, Papadopoulos N, Velculescu VE, Zhou S, Diaz Jr LA, Kinzler KW. Cancer genome landscapes. Science. 2013;339(6127):1546–58.
17. Mazzocca A, Ferraro G, Misciagna G, Carr BI. A systemic evolutionary approach to cancer: Hepatocarcinogenesis as a paradigm. Med Hypotheses. 2016;93:132–7.
18. Bizzarri M, Cucina A. SMT and TOFT: Why and How they are opposite and incompatible paradigms. Acta Biotheor. 2016;64(3):221–39.
19. Baker SG. A cancer theory kerfuffle can lead to new lines of research. J Natl Cancer Inst. 2015;107(2).
20. Wishart DS. Is cancer a genetic disease or a metabolic disease? EBioMedicine. 2015;2(6):478–9.
21. Baker SG, Kramer BS. Paradoxes in carcinogenesis: new opportunities for research directions. BMC Cancer. 2007;7:151.
22. Burgio E, Migliore L. Towards a systemic paradigm in carcinogenesis: linking epigenetics and genetics. Mol Biol Rep. 2015;42(4):777–90.
23. Soto AM, Sonnenschein C. Is systems biology a promising approach to resolve controversies in cancer research? Cancer Cell Int. 2012;12(1):12.
24. Braun AC. On the origin of the cancer cells. Am Sci. 1970;58(3):307–20.
25. Nik-Zainal S, Davies H, Staaf J, Ramakrishna M, Glodzik D, Zou X, Martincorena I, Alexandrov LB, Martin S, Wedge DC, et al. Landscape of somatic mutations in 560 breast cancer whole-genome sequences. Nature. 2016;534(7605):47–54.
26. Stratton MR. Exploring the genomes of cancer cells: progress and promise. Science. 2011;331(6024):1553–8.
27. Cooke SL, Shlien A, Marshall J, Pipinikas CP, Martincorena I, Tubio JM, Li Y, Menzies A, Mudie L, Ramakrishna M, et al. Processed pseudogenes acquired somatically during cancer development. Nat Commun. 2014;5:3644.
28. Bartesaghi S, Graziano V, Galavotti S, Henriquez NV, Betts J, Saxena J, Minieri V, Deli A, Karlsson A, Martins LM, et al. Inhibition of oxidative metabolism leads to p53 genetic inactivation and transformation in neural stem cells. Proc Natl Acad Sci U S A. 2015;112(4):1059–64.
29. Pacini N, Borziani F. Oncostatic-Cytoprotective Effect of Melatonin and Other Bioactive Molecules: A Common Target in Mitochondrial Respiration. Int J Mol Sci. 2016;17(3):341.
30. Kim A. Mitochondria in cancer energy metabolism: culprits or bystanders? Toxicol Res. 2015;31(4):323–30.
31. Warburg O. On the origin of cancer cells. Science. 1956;123(3191):309–14.
32. Warburg O. On the respiratory impairment in cancer cells. Science. 1956; 124:269–70.
33. Putignani L, Raffa S, Pescosolido R, Aimati L, Signore F, Torrisi MR, Grammatico P. Alteration of expression levels of the oxidative phosphorylation system (OXPHOS) in breast cancer cell mitochondria. Breast Cancer Res Treat. 2008;110(3):439–52.
34. Dienel GA, Cruz NF. Aerobic glycolysis during brain activation: adrenergic regulation and influence of norepinephrine on astrocytic metabolism. J Neurochem. 2016;138(1):14–52.
35. Racker E. History of the Pasteur effect and its pathobiology. Mol Cell Biochem. 1974;5(1–2):17–23.
36. Warburg O. The Metabolism of Tumours. New York: Richard R. Smith; 1931.
37. Seyfried TN. The Warburg dispute. In: Cancer as a Metabolic Disease: On the Origin, Management, and Prevention of Cancer. edn. Hoboken: Wiley; 2012. p. 107–17.
38. Zu XL, Guppy M. Cancer metabolism: facts, fantasy, and fiction. Biochem Biophys Res Commun. 2004;313(3):459–65.
39. Koppenol WH, Bounds PL, Dang CV. Otto Warburg's contributions to current concepts of cancer metabolism. Nat Rev Cancer. 2011;11(5): 325–37.
40. Poff AM, Ari C, Seyfried TN, D'Agostino DP. The ketogenic diet and hyperbaric oxygen therapy prolong survival in mice with systemic metastatic cancer. PLoS One. 2013;8(6):e65522.
41. Kiebish MA, Han X, Cheng H, Seyfried TN. In vitro growth environment produces lipidomic and electron transport chain abnormalities in mitochondria from non-tumorigenic astrocytes and brain tumours. ASN Neuro. 2009;1(3):e00011.
42. Diaz-Ruiz R, Rigoulet M, Devin A. The Warburg and Crabtree effects: On the origin of cancer cell energy metabolism and of yeast glucose repression. Biochim Biophys Acta. 2011;1807(6):568–76.
43. Leznev EI, Popova II, Lavrovskaja VP, Evtodienko YV. Comparison of oxygen consumption rates in minimally transformed BALB/3 T3 and virus-transformed 3T3B-SV40 cells. Biochemistry (Mosc). 2013;78(8):904–8.
44. Hall A, Meyle KD, Lange MK, Klima M, Sanderhoff M, Dahl C, Abildgaard C, Thorup K, Moghimi SM, Jensen PB, et al. Dysfunctional oxidative phosphorylation makes malignant melanoma cells addicted to glycolysis driven by the V600EBRAF oncogene. Oncotarget. 2013; 4(4):584–99.
45. Seyfried TN. Is respiration normal in cancer cells? In: Cancer as a Metabolic Disease: On the Origin, Management, and Prevention of Cancer. edn. Hoboken: Wiley; 2012. p. 119–32.
46. Hochachka PW, Somero GN. Biochemical Adaptation: Mechanism and Process in Physiological Evolution. New York: Oxford Press; 2002.
47. Ramanathan A, Wang C, Schreiber SL. Perturbational profiling of a cell-line model of tumorigenesis by using metabolic measurements. Proc Natl Acad Sci U S A. 2005;102(17):5992–7.
48. Arcos JC, Tison MJ, Gosch HH, Fabian JA. Sequential alterations in mitochondrial inner and outer membrane electron transport and in respiratory control during feeding of amino azo dyes; stability of phosphorylation. Correlation with swelling-contraction changes and tumorigenesis threshold. Cancer Res. 1969;29(6):1298–305.
49. Suarez RK, Lighton JR, Brown GS, Mathieu-Costello O. Mitochondrial respiration in hummingbird flight muscles. Proc Natl Acad Sci U S A. 1991; 88(11):4870–3.
50. Burk D, Schade AL. On respiratory impairment in cancer cells. Science. 1956; 124(3215):270–2.
51. Smith AE, Kenyon DH. A unifying concept of carcinogenesis and its therapeutic implications. Oncology. 1973;27(5):459–79.
52. Colowick SP. The status of Warburg's theory of glycolysis and respiration in tumors. Q Rev Biol. 1961;36:273–6.
53. Hu Y, Lu W, Chen G, Wang P, Chen Z, Zhou Y, Ogasawara M, Trachootham D, Feng L, Pelicano H, et al. K-ras (G12V) transformation leads to mitochondrial dysfunction and a metabolic switch from oxidative phosphorylation to glycolysis. Cell Res. 2012;22(2):399–412.
54. Cuezva JM, Chen G, Alonso AM, Isidoro A, Misek DE, Hanash SM, Beer DG. The bioenergetic signature of lung adenocarcinomas is a molecular marker of cancer diagnosis and prognosis. Carcinogenesis. 2004;25(7):1157–63.
55. Ferreira LM. Cancer metabolism: the Warburg effect today. Exp Mol Pathol. 2010;89(3):372–80.
56. Seyfried TN, Shelton LM. Cancer as a metabolic disease. Nutr Metab (Lond). 2010;7(1):7.
57. Poff AM, Ari C, Arnold P, Seyfried TN, D'Agostino DP. Ketone supplementation decreases tumor cell viability and prolongs survival of mice with metastatic cancer. Int J Cancer. 2014;135(7):1711–20.
58. Pedersen PL. Warburg, me and Hexokinase 2: Multiple discoveries of key molecular events underlying one of cancers' most common phenotypes, the "Warburg Effect", i.e., elevated glycolysis in the presence of oxygen. J Bioenerg Biomembr. 2007;39(3):211–22.
59. Warburg O. Revidsed Lindau Lectures: The prime cause of cancer and prevention - Parts 1 & 2. In: Lindau BD, editor. Meeting of the Nobel-

Laureates. Lake Constance: K. Triltsch; 1969. p. 1–9. http://www.hopeforcancer.com/OxyPlus.htm.
60. Racker E. Bioenergetics and the problem of tumor growth. Am Sci. 1972; 60(1):56–63.
61. Weinhouse S. The Warburg hypothesis fifty years later. Z Krebsforsch Klin Onkol Cancer Res Clin Oncol. 1976;87(2):115–26.
62. Marin-Valencia I, Yang C, Mashimo T, Cho S, Baek H, Yang XL, Rajagopalan KN, Maddie M, Vemireddy V, Zhao Z, et al. Analysis of tumor metabolism reveals mitochondrial glucose oxidation in genetically diverse human glioblastomas in the mouse brain in vivo. Cell Metab. 2012;15(6):827–37.
63. Seyfried TN. Respiratory dysfunction in cancer cells. In: Cancer as a Metabolic Disease: On the Origin, Management, and Prevention of Cancer. edn. Hoboken: Wiley; 2012. p. 73–105.
64. Lichtor T, Dohrmann GJ. Respiratory patterns in human brain tumors. Neurosurgery. 1986;19(6):896–9.
65. Seyfried TN, Mukherjee P. Targeting energy metabolism in brain cancer: review and hypothesis. Nutr Metab (Lond). 2005;2:30.
66. Vander Heiden MG, Cantley LC, Thompson CB. Understanding the Warburg effect: the metabolic requirements of cell proliferation. Science. 2009; 324(5930):1029–33.
67. Cuezva JM, Krajewska M, de Heredia ML, Krajewski S, Santamaria G, Kim H, Zapata JM, Marusawa H, Chamorro M, Reed JC. The bioenergetic signature of cancer: a marker of tumor progression. Cancer Res. 2002;62(22):6674–81.
68. Pedersen PL. Tumor mitochondria and the bioenergetics of cancer cells. Prog Exp Tumor Res. 1978;22:190–274.
69. Morton R, Cunningham C, Jester R, Waite M, Miller N, Morris HP. Alteration of mitochondrial function and lipid composition in Morris 7777 hepatoma. Cancer Res. 1976;36(9 pt.1):3246–54.
70. Schild L, Lendeckel U, Gardemann A, Wiswedel I, Schmidt CA, Wolke C, Walther R, Grabarczyk P, Busemann C. Composition of molecular cardiolipin species correlates with proliferation of lymphocytes. Exp Biol Med. 2012; 237(4):372–9.
71. Sapandowski A, Stope M, Evert K, Evert M, Zimmermann U, Peter D, Page I, Burchardt M, Schild L. Cardiolipin composition correlates with prostate cancer cell proliferation. Mol Cell Biochem. 2015;410(1–2):175–85.
72. Canuto RA, Biocca ME, Muzio G, Dianzani MU. Fatty acid composition of phospholipids in mitochondria and microsomes during diethylnitrosamine carcinogenesis in rat liver. Cell Biochem Funct. 1989;7(1):11–9.
73. Kiebish MA, Han X, Cheng H, Chuang JH, Seyfried TN. Cardiolipin and electron transport chain abnormalities in mouse brain tumor mitochondria: lipidomic evidence supporting the Warburg theory of cancer. J Lipid Res. 2008;49(12):2545–56.
74. Peyta L, Jarnouen K, Pinault M, Guimaraes C, de Barros JP P, Chevalier S, Dumas JF, Maillot F, Hatch GM, Loyer P, et al. Reduced cardiolipin content decreases respiratory chain capacities and increases ATP synthesis yield in the human HepaRG cells. Biochim Biophys Acta. 2016;4:443–53.
75. Kiebish MA, Han X, Cheng H, Seyfried TN. Mitochondrial lipidome and electron transport chain alterations in non-metastatic and metastatic murine brain tumors. J Neurochem. 2008;104 Suppl 1:37–8.
76. Claypool SM, Koehler CM. The complexity of cardiolipin in health and disease. Trends Biochem Sci. 2012;37(1):32–41.
77. Ren M, Phoon CK, Schlame M. Metabolism and function of mitochondrial cardiolipin. Prog Lipid Res. 2014;55:1–16.
78. Chinopoulos C. Which way does the citric acid cycle turn during hypoxia? The critical role of alpha-ketoglutarate dehydrogenase complex. J Neurosci Res. 2013;91(8):1030–43.
79. Peiris-Pages M, Martinez-Outschoorn UE, Pestell RG, Sotgia F, Lisanti MP. Cancer stem cell metabolism. Breast Cancer Res. 2016;18(1):55.
80. Deighton RF, Le Bihan T, Martin SF, Gerth AM, McCulloch M, Edgar JM, Kerr LE, Whittle IR, McCulloch J. Interactions among mitochondrial proteins altered in glioblastoma. J Neuro-Oncol. 2014;118(2):247–56.
81. Arismendi-Morillo GJ, Castellano-Ramirez AV. Ultrastructural mitochondrial pathology in human astrocytic tumors: potentials implications pro-therapeutics strategies. J Electron Microsc (Tokyo). 2008;57(1):33–9.
82. Schmitt S, Schulz S, Schropp EM, Eberhagen C, Simmons A, Beisker W, Aichler M, Zischka H. Why to compare absolute numbers of mitochondria. Mitochondrion. 2014;19 Pt A:113–23.
83. Verschoor ML, Ungard R, Harbottle A, Jakupciak JP, Parr RL, Singh G. Mitochondria and cancer: past, present, and future. Biomed Res Int. 2013; 2013:612369.
84. Srinivasan S, Guha M, Dong DW, Whelan KA, Ruthel G, Uchikado Y, Natsugoe S, Nakagawa H, Avadhani NG. Disruption of cytochrome c oxidase function induces the Warburg effect and metabolic reprogramming. Oncogene. 2015;35:1585–95.
85. Sriskanthadevan S, Jeyaraju DV, Chung TE, Prabha S, Xu W, Skrtic M, Jhas B, Hurren R, Gronda M, Wang X, et al. AML cells have low spare reserve capacity in their respiratory chain that renders them susceptible to oxidative metabolic stress. Blood. 2015;125(13):2120–30.
86. Levine AJ, Puzio-Kuter AM. The control of the metabolic switch in cancers by oncogenes and tumor suppressor genes. Science. 2010;330(6009):1340–4.
87. Kaipparettu BA, Ma Y, Park JH, Lee TL, Zhang Y, Yotnda P, Creighton CJ, Chan WY, Wong LJ. Crosstalk from non-cancerous mitochondria can inhibit tumor properties of metastatic cells by suppressing oncogenic pathways. PLoS One. 2013;8(5):e61747.
88. Seyfried TN. Mitochondria: The ultimate tumor suppressor. In: Cancer as a Metabolic Disease: On the Origin, Management, and Prevention of Cancer. edn. Hoboken: Wiley; 2012. p. 195–205.
89. Kloc M, Li XC, Ghobrial RM. Are Macrophages Responsible for Cancer Metastasis? J Immuno Biol. 2016;1:1.
90. Pawelek JM, Chakraborty AK. Fusion of tumour cells with bone marrow-derived cells: a unifying explanation for metastasis. Nat Rev Cancer. 2008; 8(5):377–86.
91. Bastida-Ruiz D, Van Hoesen K, Cohen M: The Dark Side of Cell Fusion. Int J Mol Sci. 2016, 17 (5). doi: 10.3390/ijms17050638
92. Seyfried TN. Mitochondrial respiratory dysfunction and the extrachromosomal origin of cancer. In: Cancer as a Metabolic Disease: On the Origin, Management, and Prevention of Cancer. edn. Hoboken: Wiley; 2012. p. 253–9.
93. Nemeth B, Doczi J, Csete D, Kacso G, Ravasz D, Adams D, Kiss G, Nagy AM, Horvath G, Tretter L, et al. Abolition of mitochondrial substrate-level phosphorylation by itaconic acid produced by LPS-induced Irg1 expression in cells of murine macrophage lineage. FASEB J. 2016;30(1):286–300.
94. Seyfried TN. Is mitochondrial glutamine fermentation a missing link in the metabolic theory of cancer? In: Cancer as a Metabolic Disease: On the Origin, Management, and Prevention of Cancer. edn. Hoboken: Wiley; 2012. p. 133–44.
95. Chinopoulos C, Gerencser AA, Mandi M, Mathe K, Torocsik B, Doczi J, Turiak L, Kiss G, Konrad C, Vajda S, et al. Forward operation of adenine nucleotide translocase during F0F1-ATPase reversal: critical role of matrix substrate-level phosphorylation. FASEB J. 2010;24(7):2405–16.
96. Phillips D, Aponte AM, French SA, Chess DJ, Balaban RS. Succinyl-CoA synthetase is a phosphate target for the activation of mitochondrial metabolism. Biochemistry. 2009;48(30):7140–9.
97. Schwimmer C, Lefebvre-Legendre L, Rak M, Devin A, Slonimski PP, di Rago JP, Rigoulet M. Increasing mitochondrial substrate-level phosphorylation can rescue respiratory growth of an ATP synthase-deficient yeast. J Biol Chem. 2005;280(35):30751–9.
98. Kiss G, Konrad C, Pour-Ghaz I, Mansour JJ, Nemeth B, Starkov AA, Adam-Vizi V, Chinopoulos C. Mitochondrial diaphorases as NAD (+) donors to segments of the citric acid cycle that support substrate-level phosphorylation yielding ATP during respiratory inhibition. FASEB J. 2014; 28(4):1682–97.
99. Newsholme EA, Board M. Application of metabolic-control logic to fuel utilization and its significance in tumor cells. Adv Enzyme Regul. 1991; 31:225–46.
100. DeBerardinis RJ, Cheng T. Q's next: the diverse functions of glutamine in metabolism, cell biology and cancer. Oncogene. 2010;29(3):313–24.
101. Yuneva M. Finding an "Achilles' heel" of cancer: the role of glucose and glutamine metabolism in the survival of transformed cells. Cell Cycle. 2008; 7(14):2083–9.
102. Medina MA. Glutamine and cancer. J Nutr. 2001;131(9 Suppl):2539–2542S. discussion 2550S-2531S.
103. Huang W, Choi W, Chen Y, Zhang Q, Deng H, He W, Shi Y. A proposed role for glutamine in cancer cell growth through acid resistance. Cell Res. 2013; 23(5):724–7.
104. Nakashima RA, Paggi MG, Pedersen PL. Contributions of glycolysis and oxidative phosphorylation to adenosine 5′-triphosphate production in AS-30D hepatoma cells. Cancer Res. 1984;44(12 Pt 1):5702–6.
105. Ta NL, Seyfried TN. Influence of Serum and Hypoxia on Incorporation of [(14) C]-D-Glucose or [(14) C]-L-Glutamine into Lipids and Lactate in Murine Glioblastoma Cells. Lipids. 2015;50(12):1167–84.

106. Portais JC, Voisin P, Merle M, Canioni P. Glucose and glutamine metabolism in C6 glioma cells studied by carbon 13 NMR. Biochimie. 1996;78(3):155–64.
107. Scott DA, Richardson AD, Filipp FV, Knutzen CA, Chiang GG, Ronai ZA, Osterman AL, Smith JW. Comparative metabolic flux profiling of melanoma cell lines: beyond the Warburg effect. J Biol Chem. 2011;286(49):42626–34.
108. Shelton LM, Huysentruyt LC, Seyfried TN. Glutamine targeting inhibits systemic metastasis in the VM-M3 murine tumor model. Int J Cancer. 2010; 127(10):2478–85.
109. Pisarenko OI, Solomatina ES, Ivanov VE, Studneva IM, Kapelko VI, Smirnov VN. On the mechanism of enhanced ATP formation in hypoxic myocardium caused by glutamic acid. Basic Res Cardiol. 1985;80(2):126–34.
110. Weinberg JM, Venkatachalam MA, Roeser NF, Nissim I. Mitochondrial dysfunction during hypoxia/reoxygenation and its correction by anaerobic metabolism of citric acid cycle intermediates. Proc Natl Acad Sci U S A. 2000;97(6):2826–31.
111. Tannahill GM, Curtis AM, Adamik J, Palsson-McDermott EM, McGettrick AF, Goel G, Frezza C, Bernard NJ, Kelly B, Foley NH, et al. Succinate is an inflammatory signal that induces IL-1beta through HIF-1alpha. Nature. 2013; 496(7444):238–42.
112. Hochachka PW, Owen TG, Allen JF, Whittow GC. Multiple end products of anaerobiosis in diving vertebrates. Comp Biochem Physiol B. 1975;50(1):17–22.
113. King A, Selak MA, Gottlieb E. Succinate dehydrogenase and fumarate hydratase: linking mitochondrial dysfunction and cancer. Oncogene. 2006; 25(34):4675–82.
114. Marsh J, Mukherjee P, Seyfried TN. Akt-dependent proapoptotic effects of dietary restriction on late-stage management of a phosphatase and tensin homologue/tuberous sclerosis complex 2-deficient mouse astrocytoma. Clin Cancer Res. 2008;14(23):7751–62.
115. Semenza GL. HIF-1 mediates the Warburg effect in clear cell renal carcinoma. J Bioenerg Biomembr. 2007;39(3):231–4.
116. Zhang H, Gao P, Fukuda R, Kumar G, Krishnamachary B, Zeller KI, Dang CV, Semenza GL. HIF-1 Inhibits Mitochondrial Biogenesis and Cellular Respiration in VHL-Deficient Renal Cell Carcinoma by Repression of C-MYC Activity. Cancer Cell. 2007;11(5):407–20.
117. Comerford SA, Huang Z, Du X, Wang Y, Cai L, Witkiewicz AK, Walters H, Tantawy MN, Fu A, Manning HC, et al. Acetate dependence of tumors. Cell. 2014;159(7):1591–602.
118. Hosios AM, Vander Heiden MG. Acetate metabolism in cancer cells. Cancer & metabolism. 2014;2(1):27.
119. Ballard FJ. Supply and utilization of acetate in mammals. Am J Clin Nutr. 1972;25(8):773–9.
120. Jaworski DM, Namboodiri AM, Moffett JR. Acetate as a Metabolic and Epigenetic Modifier of Cancer Therapy. J Cell Biochem. 2015;117:574–88.
121. Huysentruyt LC, Seyfried TN. Perspectives on the mesenchymal origin of metastatic cancer. Cancer Metastasis Rev. 2010;29(4):695–707.
122. Pawelek JM. Tumour-cell fusion as a source of myeloid traits in cancer. Lancet Oncol. 2005;6(12):988–93.
123. Ruff MR, Pert CB. Small cell carcinoma of the lung: macrophage-specific antigens suggest hemopoietic stem cell origin. Science. 1984;225(4666): 1034–6.
124. Powell AE, Anderson EC, Davies PS, Silk AD, Pelz C, Impey S, Wong MH. Fusion between Intestinal epithelial cells and macrophages in a cancer context results in nuclear reprogramming. Cancer Res. 2011;71(4):1497–505.
125. Yeh MH, Chang YH, Tsai YC, Chen SL, Huang TS, Chiu JF, Ch'ang HJ. Bone marrow derived macrophages fuse with intestine stromal cells and contribute to chronic fibrosis after radiation. Radiother Oncol. 2016; 119(2):250–8.
126. Abodief WT, Dey P, Al-Hattab O. Cell cannibalism in ductal carcinoma of breast. Cytopathology. 2006;17(5):304–5.
127. Fais S. Cannibalism: a way to feed on metastatic tumors. Cancer Lett. 2007; 258(2):155–64.
128. Lugini L, Matarrese P, Tinari A, Lozupone F, Federici C, Iessi E, Gentile M, Luciani F, Parmiani G, Rivoltini L, et al. Cannibalism of live lymphocytes by human metastatic but not primary melanoma cells. Cancer Res. 2006;66(7): 3629–38.
129. Matarrese P, Ciarlo L, Tinari A, Piacentini M, Malorni W. Xeno-cannibalism as an exacerbation of self-cannibalism: a possible fruitful survival strategy for cancer cells. Curr Pharm Des. 2008;14(3):245–52.
130. Gupta K, Dey P. Cell cannibalism: diagnostic marker of malignancy. Diagn Cytopathol. 2003;28(2):86–7.
131. Kojima S, Sekine H, Fukui I, Ohshima H. Clinical significance of "cannibalism" in urinary cytology of bladder cancer. Acta Cytol. 1998;42(6):1365–9.
132. Shelton LM. Targeting energy metabolism in brain cancer. Chestnut Hill: Boston College; 2010.
133. Kamphorst JJ, Nofal M, Commisso C, Hackett SR, Lu W, Grabocka E, Vander Heiden MG, Miller G, Drebin JA, Bar-Sagi D, et al. Human pancreatic cancer tumors are nutrient poor and tumor cells actively scavenge extracellular protein. Cancer Res. 2015;75(3):544–53.
134. Lu J, Sharma LK, Bai Y. Implications of mitochondrial DNA mutations and mitochondrial dysfunction in tumorigenesis. Cell Res. 2009;19(7):802–15.
135. Yang D, Wang MT, Tang Y, Chen Y, Jiang H, Jones TT, Rao K, Brewer GJ, Singh KK, Nie D. Impairment of mitochondrial respiration in mouse fibroblasts by oncogenic H-RAS (Q61L). Cancer Biol Ther. 2010;9(2):122–33.
136. Smiraglia DJ, Kulawiec M, Bistulfi GL, Gupta SG, Singh KK. A novel role for mitochondria in regulating epigenetic modification in the nucleus. Cancer Biol Ther. 2008;7(8):1182–90.
137. Delsite RL, Rasmussen LJ, Rasmussen AK, Kalen A, Goswami PC, Singh KK. Mitochondrial impairment is accompanied by impaired oxidative DNA repair in the nucleus. Mutagenesis. 2003;18(6):497–503.
138. Kulawiec M, Safina A, Desouki MM, Still I, Matsui SI, Bakin A, Singh KK. Tumorigenic transformation of human breast epithelial cells induced by mitochondrial DNA depletion. Cancer Biol Ther. 2008;7(11):1732–43.
139. Rasmussen AK, Chatterjee A, Rasmussen LJ, Singh KK. Mitochondria-mediated nuclear mutator phenotype in Saccharomyces cerevisiae. Nucleic Acids Res. 2003;31(14):3909–17.
140. Chandra D, Singh KK. Genetic insights into OXPHOS defect and its role in cancer. Biochim Biophys Acta. 2011;1807(6):620–5.
141. Veatch JR, McMurray MA, Nelson ZW, Gottschling DE. Mitochondrial dysfunction leads to nuclear genome instability via an iron-sulfur cluster defect. Cell. 2009;137(7):1247–58.
142. Samper E, Nicholls DG, Melov S. Mitochondrial oxidative stress causes chromosomal instability of mouse embryonic fibroblasts. Aging Cell. 2003; 2(5):277–85.
143. Seoane M, Mosquera-Miguel A, Gonzalez T, Fraga M, Salas A, Costoya JA. The Mitochondrial Genome Is a "Genetic Sanctuary" during the Oncogenic Process. PLoS One. 2011;6(8):e23327.
144. Minocherhomji S, Tollefsbol TO, Singh KK. Mitochondrial regulation of epigenetics and its role in human diseases. Epigenetics. 2012;7(4):326–34.
145. Veech RL. The therapeutic implications of ketone bodies: the effects of ketone bodies in pathological conditions: ketosis, ketogenic diet, redox states, insulin resistance, and mitochondrial metabolism. Prostaglandins Leukot Essent Fatty Acids. 2004;70(3):309–19.
146. Sabharwal SS, Schumacker PT. Mitochondrial ROS in cancer: initiators, amplifiers or an Achilles' heel? Nat Rev Cancer. 2014;14(11):709–21.
147. Klaunig JE, Kamendulis LM, Hocevar BA. Oxidative stress and oxidative damage in carcinogenesis. Toxicol Pathol. 2010;38(1):96–109.
148. Szent-Gyorgyi A. The living state and cancer. Proc Natl Acad Sci U S A. 1977; 74(7):2844–7.
149. Cairns J. The origin of human cancers. Nature. 1981;289(5796):353–7.
150. Mukherjee S. The Emperor of All Maladies: A Biography of Cancer (pages 285, 303, 333, 342). New York: Scribner; 2010.
151. Potts R. Environmental hypotheses of hominin evolution. Am J Phys Anthropol. 1998;Suppl 27:93–136.
152. Potts R. Humanity's Descent: The Consequences of Ecological Instability. New York: William Morrow & Co., Inc.; 1996.
153. Potts R. Complexity of Adaptibility in Human Evolution. In: Goodman M, Moffat AS, editors. Probing Human Origins. edn. Cambridge: American Academy of Arts & Sciences; 2002. p. 33–57.
154. Seyfried TN. Nothing in cancer biology makes sense except in the light of evolution. In: Cancer as a Metabolic Disease: On the Origin, Management, and Prevention of Cancer. edn. Hoboken: Wiley; 2012. p. 261–75.
155. Darwin C. On the Origin of Species by Means of Natural Selection, or on the Preservation of Favored Races in the Struggle for Life. London: John Murry; 1859.
156. Moiseeva O, Bourdeau V, Roux A, Deschenes-Simard X, Ferbeyre G. Mitochondrial dysfunction contributes to oncogene-induced senescence. Mol Cell Biol. 2009;29(16):4495–507.
157. de Groof AJ, te Lindert MM, van Dommelen MM, Wu M, Willemse M, Smift AL, Winer M, Oerlemans F, Pluk H, Fransen JA, et al. Increased OXPHOS activity precedes rise in glycolytic rate in H-RasV12/E1A transformed fibroblasts that develop a Warburg phenotype. Mol Cancer. 2009;8:54.

158. Matoba S, Kang JG, Patino WD, Wragg A, Boehm M, Gavrilova O, Hurley PJ, Bunz F, Hwang PM. p53 regulates mitochondrial respiration. Science. 2006; 312(5780):1650–3.
159. Galmiche A, Fueller J. RAF kinases and mitochondria. Biochim Biophys Acta. 2007;1773(8):1256–62.
160. Kerr EM, Gaude E, Turrell FK, Frezza C, Martins CP. Mutant Kras copy number defines metabolic reprogramming and therapeutic susceptibilities. Nature. 2016;531(7592):110–3.
161. Grabacka M, Pierzchalska M, Reiss K. Peroxisome Proliferator Activated Receptor alpha Ligands As Anti-Cancer Drugs Targeting Mitochondrial Metabolism. Curr Pharm Biotechnol. 2013;14:342–56.
162. Eales KL, Hollinshead KE, Tennant DA. Hypoxia and metabolic adaptation of cancer cells. Oncogenesis. 2016;5:e190.
163. Xu RH, Pelicano H, Zhou Y, Carew JS, Feng L, Bhalla KN, Keating MJ, Huang P. Inhibition of glycolysis in cancer cells: a novel strategy to overcome drug resistance associated with mitochondrial respiratory defect and hypoxia. Cancer Res. 2005;65(2):613–21.
164. Hensley CT, Wasti AT, DeBerardinis RJ. Glutamine and cancer: cell biology, physiology, and clinical opportunities. J Clin Invest. 2013;123(9):3678–84.
165. Rozhok AI, DeGregori J. Toward an evolutionary model of cancer: Considering the mechanisms that govern the fate of somatic mutations. Proc Natl Acad Sci U S A. 2015;112(29):8914–21.
166. Mukherjee P, Mulrooney TJ, Marsh J, Blair D, Chiles TC, Seyfried TN. Differential effects of energy stress on AMPK phosphorylation and apoptosis in experimental brain tumor and normal brain. Mol Cancer. 2008;7:37.
167. Mukherjee P, Sotnikov AV, Mangian HJ, Zhou JR, Visek WJ, Clinton SK. Energy intake and prostate tumor growth, angiogenesis, and vascular endothelial growth factor expression. J Natl Cancer Inst. 1999;91(6):512–23.
168. Nebeling LC, Miraldi F, Shurin SB, Lerner E. Effects of a ketogenic diet on tumor metabolism and nutritional status in pediatric oncology patients: two case reports. J Am Coll Nutr. 1995;14(2):202–8.
169. Zuccoli G, Marcello N, Pisanello A, Servadei F, Vaccaro S, Mukherjee P, Seyfried TN. Metabolic management of glioblastoma multiforme using standard therapy together with a restricted ketogenic diet: Case Report. Nutr Metab (Lond). 2010;7(1):33.
170. Mukherjee P, El-Abbadi MM, Kasperzyk JL, Ranes MK, Seyfried TN. Dietary restriction reduces angiogenesis and growth in an orthotopic mouse brain tumour model. Br J Cancer. 2002;86(10):1615–21.
171. Mukherjee P, Abate LE, Seyfried TN. Antiangiogenic and proapoptotic effects of dietary restriction on experimental mouse and human brain tumors. Clin Cancer Res. 2004;10(16):5622–9.
172. Seyfried TN, Sanderson TM, El-Abbadi MM, McGowan R, Mukherjee P. Role of glucose and ketone bodies in the metabolic control of experimental brain cancer. Br J Cancer. 2003;89(7):1375–82.
173. Seyfried TN, Mukherjee P. Anti-Angiogenic and Pro-Apoptotic Effects of Dietary Restriction in Experimental Brain Cancer: Role of Glucose and Ketone Bodies. In: Meadows GG, editor. Integration/Interaction of Oncologic Growth. Volume 15. 2nd ed. New York: Kluwer; 2005. p. 259–70.
174. Zhou W, Mukherjee P, Kiebish MA, Markis WT, Mantis JG, Seyfried TN. The calorically restricted ketogenic diet, an effective alternative therapy for malignant brain cancer. Nutr Metab (Lond). 2007;4:5.
175. Soto AM, Sonnenschein C. The somatic mutation theory of cancer: growing problems with the paradigm? Bioessays. 2004;26(10):1097–107.
176. Nowell PC. The clonal evolution of tumor cell populations. Science. 1976; 194(4260):23–8.
177. Cahill Jr GF. Fuel metabolism in starvation. Annu Rev Nutr. 2006;26:1–22.
178. Magee BA, Potezny N, Rofe AM, Conyers RA. The inhibition of malignant cell growth by ketone bodies. Aust J Exp Biol Med Sci. 1979;57(5):529–39.
179. Skinner R, Trujillo A, Ma X, Beierle EA. Ketone bodies inhibit the viability of human neuroblastoma cells. J Pediatr Surg. 2009;44(1):212–6. discussion 216.
180. Maurer GD, Brucker DP, Baehr O, Harter PN, Hattingen E, Walenta S, Mueller-Klieser W, Steinbach JP, Rieger J. Differential utilization of ketone bodies by neurons and glioma cell lines: a rationale for ketogenic diet as experimental glioma therapy. BMC Cancer. 2011;11(1):315.
181. Chang HT, Olson LK, Schwartz KA. Ketolytic and glycolytic enzymatic expression profiles in malignant gliomas: implication for ketogenic diet therapy. Nutr Metab. 2013;10(1):47.
182. Mulrooney TJ, Marsh J, Urits I, Seyfried TN, Mukherjee P. Influence of Caloric Restriction on Constitutive Expression of NF-kappaB in an Experimental Mouse Astrocytoma. PLoS One. 2011;6(3):e18085.
183. Abdelwahab MG, Fenton KE, Preul MC, Rho JM, Lynch A, Stafford P, Scheck AC. The ketogenic diet is an effective adjuvant to radiation therapy for the treatment of malignant glioma. PLoS One. 2012;7(5):e36197.
184. Martuscello RT, Vedam-Mai V, McCarthy DJ, Schmoll ME, Jundi MA, Louviere CD, Griffith BG, Skinner CL, Suslov O, Deleyrolle LP, et al. A Supplemented High-Fat Low-Carbohydrate Diet for the Treatment of Glioblastoma. Clin Cancer Res. 2015;22:2482–95.
185. Vincent M. Cancer: a de-repression of a default survival program common to all cells?: a life-history perspective on the nature of cancer. BioEssays. 2012;34(1):72–82.
186. Cervantes-Madrid D, Romero Y, Duenas-Gonzalez A. Reviving Lonidamine and 6-Diazo-5-oxo-L-norleucine to Be Used in Combination for Metabolic Cancer Therapy. Biomed Res Int. 2015;2015:690492.
187. Freeman JM, Kossoff EH. Ketosis and the ketogenic diet, 2010: advances in treating epilepsy and other disorders. Adv Pediatr. 2010;57(1):315–29.
188. Kossoff EH, Hartman AL. Ketogenic diets: new advances for metabolism-based therapies. Curr Opin Neurol. 2012;25(2):173.
189. Meidenbauer JJ, Mukherjee P, Seyfried TN. The glucose ketone index calculator: a simple tool to monitor therapeutic efficacy for metabolic management of brain cancer. Nutr Metab (Lond). 2015;12:12.
190. Poff AM, Ward N, Seyfried TN, Arnold P, D'Agostino DP. Non-Toxic Metabolic Management of Metastatic Cancer in VM Mice: Novel Combination of Ketogenic Diet, Ketone Supplementation, and Hyperbaric Oxygen Therapy. PLoS One. 2015;10(6):e0127407.
191. Burt ME, Gorschboth CM, Brennan MF. A controlled, prospective, randomized trial evaluating the metabolic effects of enteral and parenteral nutrition in the cancer patient. Cancer. 1982;49(6):1092–105.
192. Campbell TC. Dietary protein, growth factors, and cancer. Am J Clin Nutr. 2007;85(6):1667.
193. Lu Z, Xie J, Wu G, Shen J, Collins R, Chen W, Kang X, Luo M, Zou Y, Huang LJ, et al. Fasting selectively blocks development of acute lymphoblastic leukemia via leptin-receptor upregulation. Nature. 2017;23:79–90.
194. Jiang YS, Wang FR. Caloric restriction reduces edema and prolongs survival in a mouse glioma model. J Neuro-Oncol. 2013;114(1):25–32.
195. Tisdale MJ, Brennan RA. A comparison of long-chain triglycerides and medium-chain triglycerides on weight loss and tumour size in a cachexia model. Br J Cancer. 1988;58(5):580–3.
196. Tisdale MJ, Brennan RA, Fearon KC. Reduction of weight loss and tumour size in a cachexia model by a high fat diet. Br J Cancer. 1987;56(1):39–43.
197. Lussier DM, Woolf EC, Johnson JL, Brooks KS, Blattman JN, Scheck AC. Enhanced immunity in a mouse model of malignant glioma is mediated by a therapeutic ketogenic diet. BMC Cancer. 2016;16:310.
198. Shukla SK, Gebregiworgis T, Purohit V, Chaika NV, Gunda V, Radhakrishnan P, Mehla K, Pipinos II, Powers R, Yu F, et al. Metabolic reprogramming induced by ketone bodies diminishes pancreatic cancer cachexia. Cancer metabolism. 2014;2:18.
199. Morscher RJ, Aminzadeh-Gohari S, Feichtinger RG, Mayr JA, Lang R, Neureiter D, Sperl W, Kofler B. Inhibition of Neuroblastoma Tumor Growth by Ketogenic Diet and/or Calorie Restriction in a CD1-Nu Mouse Model. PLoS One. 2015;10(6):e0129802.
200. Morscher RJ, Aminzadeh-Gohari S, Hauser-Kronberger C, Feichtinger RG, Sperl W, Kofler B. Combination of metronomic cyclophosphamide and dietary intervention inhibits neuroblastoma growth in a CD1-nu mouse model. Oncotarget. 2016;7(13):17060–73.
201. Allen BG, Bhatia SK, Buatti JM, Brandt KE, Lindholm KE, Button AM, Szweda LI, Smith BJ, Spitz DR, Fath MA. Ketogenic diets enhance oxidative stress and radio-chemo-therapy responses in lung cancer xenografts. Clin Cancer Res. 2013;19(14):3905–13.
202. Mavropoulos JC, Buschemeyer 3rd WC, Tewari AK, Rokhfeld D, Pollak M, Zhao Y, Febbo PG, Cohen P, Hwang D, Devi G, et al. The effects of varying dietary carbohydrate and fat content on survival in a murine LNCaP prostate cancer xenograft model. Cancer Prev Res (Phila). 2009; 2(6):557–65.
203. Kim HS, Masko EM, Poulton SL, Kennedy KM, Pizzo SV, Dewhirst MW, Freedland SJ. Carbohydrate restriction and lactate transporter inhibition in a mouse xenograft model of human prostate cancer. BJU Int. 2012;110(7): 1062–9.
204. Lv M, Zhu X, Wang H, Wang F, Guan W. Roles of caloric restriction, ketogenic diet and intermittent fasting during initiation, progression and metastasis of cancer in animal models: a systematic review and meta-analysis. PLoS One. 2014;9(12):e115147.

205. Zhuang Y, Chan DK, Haugrud AB, Miskimins WK. Mechanisms by which low glucose enhances the cytotoxicity of metformin to cancer cells both in vitro and in vivo. PLoS One. 2014;9(9):e108444.
206. Hao GW, Chen YS, He DM, Wang HY, Wu GH, Zhang B. Growth of human colon cancer cells in nude mice is delayed by ketogenic diet with or without omega-3 fatty acids and medium-chain triglycerides. Asian Pac J Cancer Prev. 2015;16(5):2061–8.
207. Maroon JC, Seyfried TN, Donohue JP, Bost J. The role of metabolic therapy in treating glioblastoma multiforme. Surg Neurol Int. 2015;6:61.
208. Rieger J, Bahr O, Maurer GD, Hattingen E, Franz K, Brucker D, Walenta S, Kammerer U, Coy JF, Weller M, et al. ERGO: a pilot study of ketogenic diet in recurrent glioblastoma. Int J Oncol. 2014;44(6):1843–52.
209. Klement RJ. Calorie or carbohydrate restriction? The ketogenic diet as another option for supportive cancer treatment. Oncologist. 2013;18(9):1056.
210. Klement RJ. Restricting carbohydrates to fight head and neck cancer-is this realistic? Cancer biol med. 2014;11(3):145–61.
211. Tan-Shalaby JL, Carrick J, Edinger K, Genovese D, Liman AD, Passero VA, Shah RB. Modified Atkins diet in advanced malignancies - final results of a safety and feasibility trial within the Veterans Affairs Pittsburgh Healthcare System. Nutr Metab (Lond). 2016;13:52.
212. Schmidt M, Pfetzer N, Schwab M, Strauss I, Kammerer U. Effects of a ketogenic diet on the quality of life in 16 patients with advanced cancer: A pilot trial. Nutr Metab. 2011;8(1):54.
213. Champ CE, Palmer JD, Volek JS, Werner-Wasik M, Andrews DW, Evans JJ, Glass J, Kim L, Shi W. Targeting metabolism with a ketogenic diet during the treatment of glioblastoma multiforme. J Neuro-Oncol. 2014;117(1):125–31.
214. Champ CE, Mishra MV, Showalter TN, Ohri N, Dicker AP, Simone NL. Dietary recommendations during and after cancer treatment: consistently inconsistent? Nutr Cancer. 2013;65(3):430–9.
215. Fine EJ, Segal-Isaacson CJ, Feinman RD, Herszkopf S, Romano MC, Tomuta N, Bontempo AF, Negassa A, Sparano JA. Targeting insulin inhibition as a metabolic therapy in advanced cancer: a pilot safety and feasibility dietary trial in 10 patients. Nutrition. 2012;28(10):1028–35.
216. Schwartz K, Chang HT, Nikolai M, Pernicone J, Rhee S, Olson K, Kurniali PC, Hord NG, Noel M. Treatment of glioma patients with ketogenic diets: report of two cases treated with an IRB-approved energy-restricted ketogenic diet protocol and review of the literature. Cancer metab. 2015;3:3.
217. Klement RJ, Sweeney RA. Impact of a ketogenic diet intervention during radiotherapy on body composition: I. Initial clinical experience with six prospectively studied patients. BMC Res Notes. 2016;9:143.
218. Freeman JM, Kossoff EH, Freeman JB, Kelly MT. The Ketogenic Diet: A Treatment for Children and Others with Epilepsy. 4th ed. New York: Demos; 2007.
219. Mantis JG, Centeno NA, Todorova MT, McGowan R, Seyfried TN. Management of multifactorial idiopathic epilepsy in EL mice with caloric restriction and the ketogenic diet: role of glucose and ketone bodies. Nutr Metab (Lond). 2004;1(1):11.
220. Cahill Jr GF, Veech RL. Ketoacids? Good medicine? Trans Am Clin Climatol Assoc. 2003;114:149–61. discussion 162–143.
221. Fein EJ, Feinman RD. Insulin, carbohydrate restriction, metabolic syndrome and cancer. Expert Rev Endocrinol Metab. 2015;10:15–24.
222. Sato K, Kashiwaya Y, Keon CA, Tsuchiya N, King MT, Radda GK, Chance B, Clarke K, Veech RL. Insulin, ketone bodies, and mitochondrial energy transduction. Faseb J. 1995;9(8):651–8.
223. VanItallie TB, Nufert TH. Ketones: metabolism's ugly duckling. Nutr Rev. 2003; 61(10):327–41.
224. Veech RL, Chance B, Kashiwaya Y, Lardy HA, Cahill Jr GF. Ketone bodies, potential therapeutic uses. IUBMB Life. 2001;51(4):241–7.
225. Chance B, Sies H, Boveris A. Hydroperoxide metabolism in mammalian organs. Physiol Rev. 1979;59(3):527–605.
226. Fine EJ, Miller A, Quadros EV, Sequeira JM, Feinman RD. Acetoacetate reduces growth and ATP concentration in cancer cell lines which over-express uncoupling protein 2. Cancer Cell Int. 2009;9:14.
227. Ciraolo ST, Previs SF, Fernandez CA, Agarwal KC, David F, Koshy J, Lucas D, Tammaro A, Stevens MP, Tserng KY, et al. Model of extreme hypoglycemia in dogs made ketotic with (R, S)-1,3-butanediol acetoacetate esters. Am J Phys. 1995;269(1 Pt 1):E67–75.
228. Chance B, editor. Energy-Linked Functions of Mitochondria. New York: Academic; 1963.
229. Shimazu T, Hirschey MD, Newman J, He W, Shirakawa K, Le Moan N, Grueter CA, Lim H, Saunders LR, Stevens RD, et al. Suppression of oxidative stress by beta-hydroxybutyrate, an endogenous histone deacetylase inhibitor. Science. 2013;339(6116):211–4.
230. West AC, Johnstone RW. New and emerging HDAC inhibitors for cancer treatment. J Clin Invest. 2014;124(1):30–9.
231. Youm YH, Nguyen KY, Grant RW, Goldberg EL, Bodogai M, Kim D, D'Agostino D, Planavsky N, Lupfer C, Kanneganti TD, et al. The ketone metabolite beta-hydroxybutyrate blocks NLRP3 inflammasome-mediated inflammatory disease. Nat Med. 2015;21(3):263–9.
232. Kossoff EH, Zupec-Kania BA, Amark PE, Ballaban-Gil KR, Christina Bergqvist AG, Blackford R, Buchhalter JR, Caraballo RH, Helen Cross J, Dahlin MG, et al. Optimal clinical management of children receiving the ketogenic diet: recommendations of the International Ketogenic Diet Study Group. Epilepsia. 2009;50(2):304–17.
233. Jang HJ, Boo HJ, Lee HJ, Min HY, Lee HY. Chronic Stress Facilitates Lung Tumorigenesis by Promoting Exocytosis of IGF2 in Lung Epithelial Cells. Cancer Res. 2016;76(22):6607–19.
234. Feng Z, Liu L, Zhang C, Zheng T, Wang J, Lin M, Zhao Y, Wang X, Levine AJ, Hu W. Chronic restraint stress attenuates p53 function and promotes tumorigenesis. Proc Natl Acad Sci U S A. 2012;109(18):7013–8.
235. Rush SE, Sharma M. Mindfulness-Based Stress Reduction as a Stress Management Intervention for Cancer Care: A Systematic Review. J Evid Based Complementary Altern Med. 2014;19:271–86.
236. Lopes-Junior LC, Bomfim EO, Nascimento LC, Nunes MD, Pereira-da-Silva G, Lima RA. Non-pharmacological interventions to manage fatigue and psychological stress in children and adolescents with cancer: an integrative review. Eur J Cancer Care (Engl). 2016;25(6):921–35.
237. Bradt J, Dileo C, Magill L, Teague A. Music interventions for improving psychological and physical outcomes in cancer patients. Cochrane Database Syst Rev. 2016;8:CD006911.
238. Levin GT, Greenwood KM, Singh F, Tsoi D, Newton RU. Exercise Improves Physical Function and Mental Health of Brain Cancer Survivors: Two Exploratory Case Studies. Integr Cancer Ther. 2016;15(2):190–6.
239. Ari C, Kovacs Z, Juhasz G, Murdun C, Goldhagen CR, Koutnik AM, Poff AM, Kesl SL, D'Agostino DP. Exogenous Ketone Supplements Reduce Anxiety-Related Behavior in Sprague–Dawley and Wistar Albino Glaxo/Rijswijk Rats. Front Mol Neurosci. 2016;9:137.
240. Meynet O, Ricci JE. Caloric restriction and cancer: molecular mechanisms and clinical implications. Trends Mol Med. 2014;20(8):419–27.
241. De Lorenzo MS, Baljinnyam E, Vatner DE, Abarzua P, Vatner SF, Rabson AB. Caloric restriction reduces growth of mammary tumors and metastases. Carcinogenesis. 2011;32(9):1381–7.
242. Longo VD, Mattson MP. Fasting: molecular mechanisms and clinical applications. Cell Metab. 2014;19(2):181–92.
243. Al-Wahab Z, Tebbe C, Chhina J, Dar SA, Morris RT, Ali-Fehmi R, Giri S, Munkarah AR, Rattan R. Dietary energy balance modulates ovarian cancer progression and metastasis. Oncotarget. 2014;5(15):6063–75.
244. Safdie FM, Dorff T, Quinn D, Fontana L, Wei M, Lee C, Cohen P, Longo VD. Fasting and cancer treatment in humans: A case series report. Aging (Albany NY). 2009;1(12):988–1007.
245. Raffaghello L, Lee C, Safdie FM, Wei M, Madia F, Bianchi G, Longo VD. Starvation-dependent differential stress resistance protects normal but not cancer cells against high-dose chemotherapy. Proc Natl Acad Sci U S A. 2008;105(24):8215–20.
246. Raffaghello L, Safdie F, Bianchi G, Dorff T, Fontana L, Longo VD. Fasting and differential chemotherapy protection in patients. Cell Cycle. 2010;9(22):4474–6.
247. Marsh J, Mukherjee P, Seyfried TN. Drug/diet synergy for managing malignant astrocytoma in mice: 2-deoxy-D-glucose and the restricted ketogenic diet. Nutr Metab (Lond). 2008;5:33.
248. Williams DS, Cash A, Hamadani L, Diemer T. Oxaloacetate supplementation increases lifespan in Caenorhabditis elegans through an AMPK/FOXO-dependent pathway. Aging Cell. 2009;8(6):765–8.
249. Farah IO. Differential modulation of intracellular energetics in A549 and MRC-5 cells. Biomed Sci Instrum. 2007;43:110–5.
250. Pelicano H, Martin DS, Xu RH, Huang P. Glycolysis inhibition for anticancer treatment. Oncogene. 2006;25(34):4633–46.
251. Pitter KL, Tamagno I, Alikhanyan K, Hosni-Ahmed A, Pattwell SS, Donnola S, Dai C, Ozawa T, Chang M, Chan TA, et al. Corticosteroids compromise survival in glioblastoma. Brain. 2016;139(Pt 5):1458–71.
252. Seyfried TN, Flores R, Poff AM, D'Agostino DP, Mukherjee P. Metabolic therapy: a new paradigm for managing malignant brain cancer. Cancer Lett. 2015;356(2 Pt A):289–300.

253. Seyfried TN, Shelton LM, Mukherjee P. Does the existing standard of care increase glioblastoma energy metabolism? Lancet Oncol. 2010;11(9):811–3.
254. Moen I, Stuhr LE. Hyperbaric oxygen therapy and cancer–a review. Target Oncol. 2012;7(4):233–42.
255. Kohshi K, Beppu T, Tanaka K, Ogawa K, Inoue O, Kukita I, Clarke RE. Potential roles of hyperbaric oxygenation in the treatments of brain tumors. UHM. 2013;40(4):351–62.
256. Poff AM, Kernagis D, D'Agostino DP. Hyperbaric Environment: Oxygen and Cellular Damage versus Protection. Comp Physiology. 2017; 7(January 2017):213–34.
257. D'Agostino DP, Colomb Jr DG, Dean JB. Effects of hyperbaric gases on membrane nanostructure and function in neurons. J Appl Physiol. 2009; 106(3):996–1003.
258. Ma Y, Chapman J, Levine M, Polireddy K, Drisko J, Chen Q. High-dose parenteral ascorbate enhanced chemosensitivity of ovarian cancer and reduced toxicity of chemotherapy. Sci Transl Med. 2014;6(222):222ra218.
259. Michelakis ED, Sutendra G, Dromparis P, Webster L, Haromy A, Niven E, Maguire C, Gammer TL, Mackey JR, Fulton D, et al. Metabolic modulation of glioblastoma with dichloroacetate. Sci Transl Med. 2010;2(31):31ra34.
260. Cox PJ, Kirk T, Ashmore T, Willerton K, Evans R, Smith A, Murray AJ, Stubbs B, West J, McLure SW, et al. Nutritional Ketosis Alters Fuel Preference and Thereby Endurance Performance in Athletes. Cell Metab. 2016;24(2):256–68.
261. Murray AJ, Knight NS, Cole MA, Cochlin LE, Carter E, Tchabanenko K, Pichulik T, Gulston MK, Atherton HJ, Schroeder MA, et al. Novel ketone diet enhances physical and cognitive performance. FASEB J. 2016;30(12): 4021–32.
262. Wise DR, DeBerardinis RJ, Mancuso A, Sayed N, Zhang XY, Pfeiffer HK, Nissim I, Daikhin E, Yudkoff M, McMahon SB, et al. Myc regulates a transcriptional program that stimulates mitochondrial glutaminolysis and leads to glutamine addiction. Proc Natl Acad Sci U S A. 2008;105(48):18782–7.
263. Reitzer LJ, Wice BM, Kennell D. Evidence that glutamine, not sugar, is the major energy source for cultured HeLa cells. J Biol Chem. 1979; 254(8):2669–76.
264. Dang CV. Glutaminolysis: supplying carbon or nitrogen or both for cancer cells? Cell Cycle. 2010;9(19):3884–6.
265. Venneti S, Dunphy MP, Zhang H, Pitter KL, Zanzonico P, Campos C, Carlin SD, La Rocca G, Lyashchenko S, Ploessl K, et al. Glutamine-based PET imaging facilitates enhanced metabolic evaluation of gliomas in vivo. Sci Transl Med. 2015;7(274):274ra217.
266. Mueller C, Al-Batran S, Jaeger E, Schmidt B, Bausch M, Unger C, Sethuraman N. A phase IIa study of PEGylated glutaminase (PEG-PGA) plus 6-diazo-5-oxo-L-norleucine (DON) in patients with advanced refractory solid tumors. J Clin Oncol. 2008;26:2533. In: ASCO.
267. Chakrabarti G, Moore ZR, Luo X, Ilcheva M, Ali A, Padanad M, Zhou Y, Xie Y, Burma S, Scaglioni PP, et al. Targeting glutamine metabolism sensitizes pancreatic cancer to PARP-driven metabolic catastrophe induced by ss-lapachone. Cancer & metabolism. 2015;3:12.
268. Mates JM, Segura JA, Campos-Sandoval JA, Lobo C, Alonso L, Alonso FJ, Marquez J. Glutamine homeostasis and mitochondrial dynamics. Int J Biochem Cell Biol. 2009;41(10):2051–61.
269. Michalak KP, Mackowska-Kedziora A, Sobolewski B, Wozniak P. Key Roles of Glutamine Pathways in Reprogramming the Cancer Metabolism. Oxid Med Cell Longev. 2015;2015:964321.
270. Huysentruyt LC, Mukherjee P, Banerjee D, Shelton LM, Seyfried TN. Metastatic cancer cells with macrophage properties: evidence from a new murine tumor model. Int J Cancer. 2008;123(1):73–84.
271. Shelton LM, Mukherjee P, Huysentruyt LC, Urits I, Rosenberg JA, Seyfried TN. A novel pre-clinical in vivo mouse model for malignant brain tumor growth and invasion. J Neurooncol. 2010;99(2):165–76.
272. Huysentruyt LC, Shelton LM, Seyfried TN. Influence of methotrexate and cisplatin on tumor progression and survival in the VM mouse model of systemic metastatic cancer. Int J Cancer. 2010;126(1):65–72.
273. Hamilton JD, Rapp M, Schneiderhan T, Sabel M, Hayman A, Scherer A, Kropil P, Budach W, Gerber P, Kretschmar U, et al. Glioblastoma multiforme metastasis outside the CNS: three case reports and possible mechanisms of escape. J Clin Oncol. 2014;32(22):e80–84.
274. Hoffman HJ, Duffner PK. Extraneural metastases of central nervous system tumors. Cancer. 1985;56(7 Suppl):1778–82.
275. Xu M, Wang Y, Xu J, Yao Y, Yu WX, Zhong P. Extensive Therapies for Extraneural Metastases from Glioblastoma, as Confirmed with the OncoScan Assay. World Neurosurg. 2016;90:698 e697–11.
276. Yasuhara T, Tamiya T, Meguro T, Ichikawa T, Sato Y, Date I, Nakashima H, Ohmoto T. Glioblastoma with metastasis to the spleen–case report. Neurol Med Chir (Tokyo). 2003;43(9):452–6.
277. Kalokhe G, Grimm SA, Chandler JP, Helenowski I, Rademaker A, Raizer JJ. Metastatic glioblastoma: case presentations and a review of the literature. J Neurooncol. 2012;107(1):21–7.
278. Huysentruyt LC, Akgoc Z, Seyfried TN. Hypothesis: are neoplastic macrophages/microglia present in glioblastoma multiforme? ASN neuro. 2011;3(4):AN20110011.
279. Newsholme P. Why is L-glutamine metabolism important to cells of the immune system in health, postinjury, surgery or infection? J Nutr. 2001; 131(9 Supp):2515–2522S. discussion 2523S-2514S.
280. Shelton LM, Huysentruyt LC, Mukherjee P, Seyfried TN. Calorie restriction as an anti-invasive therapy for malignant brain cancer in the VM mouse. ASN neuro. 2010;2(3):e00038.
281. Seyfried TN. Metabolic management of cancer. In: Cancer as a Metabolic Disease: On the Origin, Management, and Prevention of Cancer. edn. Hoboken: Wiley; 2012. p. 291–354.
282. Arismendi-Morillo G. Electron microscopy morphology of the mitochondrial network in human cancer. Int J Biochem Cell Biol. 2009;41(10):2062–8.
283. Cogliati S, Frezza C, Soriano ME, Varanita T, Quintana-Cabrera R, Corrado M, Cipolat S, Costa V, Casarin A, Gomes LC, et al. Mitochondrial cristae shape determines respiratory chain supercomplexes assembly and respiratory efficiency. Cell. 2013;155(1):160–71.