Received:
14 January 2020

Revised:
29 April 2020

Accepted:
13 May 2020

<https://doi.org/10.1259/bjr.20200067>

Cite this article as:

Kery M, Papandreou I. Emerging strategies to target cancer metabolism and improve radiation therapy outcomes. *Br J Radiol* 2020; **93**: 20200067.

ADVANCES IN RADIATION BIOLOGY – HIGHLIGHTS FROM 16TH ICRR SPECIAL FEATURE: REVIEW ARTICLE

Emerging strategies to target cancer metabolism and improve radiation therapy outcomes

MARTIN KERY, PhD and IOANNA PAPANDREOU, PhD

Department of Radiation Oncology, Wexner Medical Center and Comprehensive Cancer Center The Ohio State University Columbus, OH, USA

Address correspondence to: Dr Ioanna Papandreou
E-mail: ioanna.papandreou@osumc.edu

ABSTRACT

Cancer-specific metabolic changes support the anabolic needs of the rapidly growing tumor, maintain a favorable redox balance, and help cells adapt to microenvironmental stresses like hypoxia and nutrient deprivation. Radiation is extensively applied in a large number of cancer treatment protocols but despite its curative potential, radiation resistance and treatment failures pose a serious problem. Metabolic control of DNA integrity and genomic stability can occur through multiple processes, encompassing cell cycle regulation, nucleotide synthesis, epigenetic regulation of gene activity, and antioxidant defenses. Given the important role of metabolic pathways in oxidative damage responses, it is necessary to assess the potential for tumor-specific radiosensitization by novel metabolism-targeted therapies. Additionally, there are opportunities to identify molecular and functional biomarkers of vulnerabilities to combination treatments, which could then inform clinical decisions. Here, we present a curated list of metabolic pathways in the context of ionizing radiation responses. Glutamine metabolism influences DNA damage responses by mechanisms such as synthesis of nucleotides for DNA repair or of glutathione for ROS detoxification. Repurposed oxygen consumption inhibitors have shown promising radiosensitizing activity against murine model tumors and are now in clinical trials. Production of 2-hydroxy glutarate by isocitrate dehydrogenase1/2 neomorphic oncogenic mutants interferes with the function of α -ketoglutarate-dependent enzymes and modulates Ataxia Telangiectasia Mutated (ATM) signaling and glutathione pools. Radiation-induced oxidative damage to membrane phospholipids promotes ferroptotic cell loss and cooperates with immunotherapies to improve tumor control. In summary, there are opportunities to enhance the efficacy of radiotherapy by exploiting cell-inherent vulnerabilities and dynamic microenvironmental components of the tumor.

INTRODUCTION

Tumor-specific metabolic changes are recognized as one of the hallmarks of cancer.¹ The mechanisms by which these changes are thought to facilitate malignant progression involve, among others, generation of building blocks for biomass expansion, adaptation to nutrient deprivation in harsh microenvironments and maintenance of redox balance. Intense efforts are underway to identify druggable tumor-specific metabolic characteristics and define the molecular or histological cancer types that can be targeted by such new treatments. As the majority of available therapies are rarely used as single modalities, it is necessary to evaluate emerging metabolic modulators in combination with standard of care treatments, including radiotherapy. In this review article, we discuss how utilization of selected metabolic substrates modifies the DNA damage response and shapes the tumor microenvironment. Emphasis is

given to metabolic pathways featured in presentations at the 16th International Congress of Radiation Research (ICRR) and we highlight paradigms of interaction between metabolic inhibitors and radiation treatment. We also discuss challenges inherent to preclinical studies, and how recent advances in cancer immunology and ferroptotic cell death mechanisms create opportunities for novel treatment paradigms. The reader is referred to excellent publications covering additional aspects of metabolic control of DNA damage and antioxidant responses.^{2–4}

TCA METABOLIC SUBSTRATES

Glutamine metabolism

Glutamine serves as an important metabolic substrate in tumors. Mitochondrially produced citrate is exported to the cytoplasm to be used for fatty acid synthesis. Consequently, anaplerotic metabolism of new carbon sources is

required to maintain the Tricarboxylic acid cycle (TCA) cycle operational. Glutamine is a crucial anaplerotic mitochondrial substrate and participates in essential cellular functions beyond energy production, including responses to genotoxic stresses and DNA damage.

Glutamine uptake is mediated by a number of membrane transporters of the solute carrier family, whose members have redundant affinities, specificities, and directionality of transport.⁵ Glutamine entry via the obligatory antiporter SCL1A5/ASCT2 has received particular attention in cancer research due to its overexpression in a wide range of tumor types and its regulation by oncogenes and tumor suppressors.⁶ Intracellularly, glutamine participates in essential cellular functions beyond energy production and protein synthesis, including purine and pyrimidine synthesis, glutathione synthesis and replenishment of the TCA cycle, which are important to genotoxic stresses and DNA damage responses. Glutamine is metabolized to glutamate by glutaminase (GLS), which is then converted to α -ketoglutarate (α KG) either by glutamate dehydrogenase (GDH) or by alanine and aspartate transaminases (ATs) and enters the TCA cycle. This anaplerotic use of glutamine-derived α KG replenishes the carbons that are leaving the cycle as citrate. Citrate export to the cytoplasm then provides the building blocks for *de novo* lipid synthesis. In addition to import of the environment, glutamine can be intracellularly produced from glutamate by the action of glutamine synthetase (GS) and support cells in conditions of glutamine depletion.

Haigis et al demonstrated that DNA damaging agents, including ionizing and UV radiation, decreased glutamine consumption in cancer models *in vitro* and *in vivo*.⁷ This decrease was dependent

of the activity on the mitochondrial localized Sirt4 enzyme and was partially mediated by inhibition of GDH. This control of metabolic repression is proposed to be important for inhibition of cell proliferation following DNA damage, and thus the maintenance of genomic integrity. In lung cancer models, inhibition of glutamine metabolism by glutaminase inhibition sensitized tumors to radiation and abolished glutathione (GSH) synthesis,⁸ so it appears that glutamine can participate in the defense against genotoxic insults as a precursor for the generation of multiple components of the DNA damage and anti-oxidative responses. Alternatively, GS, which catalyzes the formation of glutamine from glutamate, has been found to be upregulated in radioresistant cells.⁹ Genetic ablation of GS reduced nucleotide pools, slowed down proliferation and sensitized to radiation *in vitro* and *in vivo*,⁹ indicating that regulation of nucleotide biosynthesis may determine the kinetics and pathways of DNA damage repair (Figure 1).

α -Ketoglutarate: Metabolic and regulatory functions

Isocitrate dehydrogenases (IDHs) catalyze the conversion of isocitrate to α KG with concomitant generation of Reduced nicotinamide adenine dinucleotide phosphate (NADPH) (IDH1/2) or NADH (IDH3). Oncogenic mutations in IDH 1 and 2 are frequently found in tumors, such as gliomas and leukemias.¹⁰ α KG is not only a TCA cycle metabolite, but also a crucial enzymatic cofactor; therefore, aberrant α KG metabolism has extensive biochemical and biological implications beyond mitochondrial activity. IDH1/2 oncogenic properties are attributed to the ability of the mutant forms to dimerize with wild type partners, conferring the WT/MUT dimer a neomorphic activity

Figure 1. Proposed models for glutaminolytic control of radiation-induced DNA repair and survival. Glutaminase can be inhibited physiologically following DNA damage by Sirtuin 4 thus inhibiting proliferation and avoiding genomic instability, whereas small molecule inhibition of Glutaminase can impact anti-oxidant defences and radiosensitize. Conversion of glutamate to glutamine by Glutamine synthetase is associated with radiation resistance. Blockade of glutamine uptake by membrane transporters may affect multiple fates of intracellular glutamine with diverse biological outcomes

Figure 2. Oncogenic mutations in cytoplasmic or mitochondrial *IsocitrateDehydrogenases* and production of 2-Hydroxyglutarate interfere with α -ketoglutarate-dependent enzymatic functions. Inhibition of branched chain aminoacid aminotransferases decreases glutamate production and enhances the cells' reliance on Glutaminase for Glutathione synthesis. Inhibition of lysine/histone demethylases can in certain tissue contexts compromise DNA damage repair, whereas in other cases in can promote homologous recombination and genomic stability

producing the oncometabolite 2-hydroxyglutarate (2-HG). Accumulation of 2-HG inhibits the function of α KG-dependent dioxygenases, therefore impacting key processes including histone and DNA demethylation and protein hydroxylation.¹¹ Epigenetic regulation of DNA damage responses by 2-HG has been reported in IDH mutant cancers of different origins, with the functional consequences on DNA damage repair and radiation killing being context-dependent (Figure 2).

In engineered cell lines, IDH1 mutation increased basal and IR-induced DSBs, and sensitized model tumors to PARP inhibitor by suppressing homologous recombination (HR).¹² This susceptibility to DNA damage was mediated by inhibition of histone demethylases KDM4A and KDM4B by 2-HG, and was also observed *ex vivo* in mutant IDH1/2 primary glioma and Acute Myeloid Leukemia (AML) samples.¹² In agreement with suppression of DNA damage responses by 2-HG, tissue-specific expression of mutant IDH1 in the hematopoietic lineage elevated the repressive histone trimethylation mark H3K9Me3 at the Ataxia Telangiectasia Mutated (ATM) promoter and downregulated ATM levels.¹³ This deficit in DNA maintenance mechanisms led to moderate sensitivity to ionizing radiation, age-dependent accumulation of spontaneous DNA damage in hematopoietic stem cells and impaired self-renewal capacity.¹³

In contrast to hematologic malignancies, mutant IDH1 induced the expression of ATM and of other DNA Damage Response (DDR) components in murine gliomas, in the context of p53 and ATRX loss.¹⁴ Compared with wtIDH1, mtIDH1 tumors were more genetically stable, less aggressive, and more resistant to

radiation therapy in agreement with their better ability to repair DNA damage.¹⁴ Furthermore, IDH mutant patient-derived glioma spheres repaired radiation-induced DNA damage faster than wtIDH samples.¹⁵ In human lower glioma patients, IDH1 mutation confers a better prognosis,¹⁶ but its role in determining the response to radiation treatment is still debated.^{17,18}

In addition to dioxygenases, perturbation of other α KG-dependent enzymes by 2-HG can dictate metabolic preferences and modify responses to treatment. Kaelin and colleagues identified branched aminoacid transaminases BCAT1-2 as targets for 2-HG inhibition. BCATs are responsible for the initiation of branched chain aminoacid (BCAA) catabolism, by converting BCAAs to branched chain α -keto acids with concomitant generation of glutamate from α KG. Oncogenic IDH mutations inhibited BCAT activity, lowering glutamate levels and enhancing the reliance of IDH mutant cells on glutaminase for glutamate and glutathione production.¹⁹ This metabolic rewiring rendered IDH mutants sensitive to glutaminase inhibition and selectively sensitized mutant IDH tumors to glutaminase plus radiotherapy.¹⁹

The above examples highlight that 2-HG production can lead to diverse biological consequences in different biological systems. The clinical use of mtIDH inhibitors in combination with chemo/radiation should be carefully evaluated, since the functional consequences downstream of 2-HG production can be quite diverse. The reasons for the mixed preclinical results may be multiple, such as methodological variations, selective pressure during cell line establishment, or different epigenetic changes in different tissues.²⁰ Nevertheless, it has been proposed that for

some cancers exploitation of the presence of supraphysiological 2-HG, rather than inhibition of its production, may be a more useful strategy.¹⁹ This concept is now being investigated in a clinical trial, which tests the combination of a glutaminase inhibitor (CB-839) with radiation therapy and temozolomide in IDH-mutated gliomas (clinicaltrials.gov NCT03528642).

Apart from the prognostic value of IDH mutations, it is necessary to develop better mechanistic understanding of metabolic dependencies and vulnerabilities created by oncogenic alterations. This can then guide efforts to identify biomarkers for patient selection and to develop “metabolic precision therapies” with desired tumor specificity. For example, in subsets of Kras mutant adenocarcinomas, loss of the tumor suppressor LKB1 and mutations in the KEAP1 gene (Kelch-like ECH-associated protein 1) enhance glutamine utilization and sensitivity to glutaminase inhibitors.^{21,22} Another approach is the inhibition of glutamine uptake in the tumor, although the redundancy and the limited substrate specificity of the transporters may restrict the efficacy of this strategy or cause toxicity by imbalanced amino-acid trafficking. Interestingly, a small molecule inhibitor of the SLC1A5 transporter, V-9302, has shown promising anti-tumor activity in preclinical models.²³ Expression levels of SLC1A5 did not correlate with sensitivity to V-9302, therefore it will be necessary for any future clinical trials to rely on functional biomarkers such as fluorine 18-(2S,4R)-4-fluoroglutamine PET imaging to identify glutamine avid tumors²⁴

OXYGEN METABOLISM

It has long been known that oxygen is required for the fixation of the DNA damage caused by radiation therapy^{25,26} and that molecular oxygen is a very potent radiosensitizer. The seminal work by Thomlinson and Gray²⁷ laid the ground for the development of clinical strategies aiming to decrease intratumoral hypoxia and

to enhance radiation responses. Attempts to alleviate hypoxia by either increasing oxygen delivery to the tumor or by eliminating the radioresistant hypoxic fraction with hypoxic cytotoxins have had limited efficacy.²⁸ Mathematical models have estimated that decreasing oxygen demand would be significantly more effective than increasing oxygen delivery at improving tumor oxygenation.²⁹ In recent years, *in vitro* and *in vivo* models have tested the hypothesis that inhibition of mitochondrial function can correct the mismatch between oxygen supply and oxygen demand in the tumor, increase oxygenation and enhance the efficacy of radiation treatment (Figure 3). Mitochondria are the site of oxidative phosphorylation where oxygen is used as the terminal electron acceptor, they therefore represent the major oxygen sink in the cell. Significant improvement in radiation responses has been observed in transplanted model tumors after treatment with Arsenic Trioxide by a mechanism associated with decreased glutathione levels and mitochondrial depolarization³⁰ and by corticosteroids, potentially through inhibition of mitochondrial function.³¹

Repurposing of drugs already approved for other indications is an efficient strategy to expedite their clinical application as anti-cancer modifiers such as radiation sensitizers. Metformin is a well-tolerated anti-diabetic medication that, in recent years, has received attention as a possible cancer prevention and treatment modality. Metformin's blood glucose lowering action is attributed to inhibition of gluconeogenesis in the liver. At the cellular level, metformin inhibits the activity of mitochondrial complex I of the respiratory chain and of mitochondrial glycerophosphate dehydrogenase.^{32–34} *In vitro*, metformin inhibits mitochondrial oxygen consumption of tumor cells, albeit at much higher concentrations than plasma concentrations in diabetic patients.³⁵ In preclinical models, acute administration of metformin prior to irradiation was able to reduce hypoxic tumor volume and

Figure 3. Inhibitors of oxygen metabolism. Select inhibitors of mitochondrial complexes I and III are shown. By inhibiting oxygen consumption, they normalize the imbalance between oxygen and supply in the tumor and enhance the efficacy of radiation therapy

enhance radiation-induced model tumor growth delay.³⁶ In agreement with a beneficial interaction between metformin and radiation, retrospective analyses have shown improved clinical responses in prostate, rectal, and head and neck cancer patients treated with radiation while taking metformin.^{36–38} In contrast, breast cancer patients on metformin had more radiation treatment breaks due to increased locoregional toxicity,³⁹ therefore careful design of prospective studies is necessary to determine the therapeutic index of new treatment schedules, and to identify biomarkers for patient selection. Metformin is transported into the cell by a family of organic cation transporters (Oct1–3), with varying gene expression levels and activity-modifying polymorphisms across tissues and tumors, and these attributes can modify metformin's efficacy.^{40–43} Importantly, the *in vivo* anti-proliferative mechanisms of metformin and of the related biguanide phenformin are not entirely clear. Cell autonomous responses to activation of the energy sensor AMP-activated Protein Kinase (AMPK), loss of tumor suppressors p53 or LKB1,^{44–47} mitochondrial complex I mutations,⁴⁸ and systemic effects on growth factor signaling and immune cell function^{49–51} may be contributing factors in tissue-context manners. The radiosensitizing effect of metformin is thought to stem from correction of tissue hypoxia. Prospective clinical trials that incorporate quantitative estimations of oxygenation prior to metformin administration should reveal if patients with radiobiologically hypoxic tumors benefit the most from metformin's action as inhibitor of oxygen consumption *in vivo*.⁵² Current clinical trials incorporate molecular and functional markers (gene expression changes or PET imaging with hypoxic tracers) to test metformin's efficacy in alleviating hypoxia and enhancing the efficacy of chemoradiation (ClinicalTrials.gov NCT03510390, NCT04275713, NCT02394652, NCT04170959, NCT03053544). In the context of immune-oncology, the biological effectiveness of metformin in T-cell mitochondrial function and tumor responses to anti-PD-1 antibody are being investigated in melanoma and other solid cancers (ClinicalTrials.gov NCT03311308, NCT04114136).

The search for clinically viable oxygen metabolism inhibitors has intensified in recent years, and promising candidates are already moving into clinical trials. Atovaquone is an anti-parasitic agent used in the treatment of pneumocystis pneumonia and malaria, with a favorable safety profile and established pharmacokinetics.⁵³ Through a high-throughput screen for inhibitors of oxygen consumption in tumor cells, atovaquone was identified and further characterized biochemically as an Electron Transport Chain (ETC) complex III inhibitor.⁵⁴ Treatment of tumor spheroids *in vitro* and subcutaneous model tumors reduced hypoxia and enhanced the efficacy of radiation.⁵⁴

Papaverine is an FDA-approved therapeutic whose clinical potential in cancer therapy is currently being tested. Papaverine is a phosphodiesterase 10E inhibitor, traditionally prescribed as a vasodilator.^{55,56} The Denko group reported an additional activity of the molecule as a complex I inhibitor, capable of decreasing oxygen consumption *in vitro*, eliminating tumor hypoxia *in vivo*, and radiosensitizing model tumors.⁵⁷ The hypoxia-lowering effect of papaverine was dependent on its ability to block complex

I activity, as genetically manipulated cells resistant to complex I inhibition were not radiosensitized as tumors. Similarly, papaverine derivatives that maintained their mitochondrial inhibition activity, but lost their PDE10 inhibition activity, mimicked the parent molecule's tumor radiosensitization.⁵⁷

The above examples emphasize the potential of mitochondrial ETC inhibitors to manipulate oxygen metabolism and increase the effectiveness of radiation treatment. High-throughput screens have accelerated the discovery of novel agents able to inhibit mitochondrial function;^{58,59} however, other bottlenecks of the drug discovery pipeline can still delay their commercial development. By repurposing approved agents, precious time can be saved as they move into clinical trials: results from an early-phase trial testing atovaquone as a modifier of tumor hypoxia in non-small cell lung cancer (NSCLC) are expected soon (ClinicalTrials.gov identifier NCT02628080), while concurrent papaverine and SBRT are also in Phase I trial for NSCLC (ClinicalTrials.gov identifier NCT03824327). Several important questions remain to be answered in these early stages of development of oxygen metabolism radiosensitizers: their safety in the cancer therapy setting needs to be firmly established. Although repurposed drugs are considered safe, their possible interaction with other anti-cancer treatments in patients with serious comorbidities cannot be ruled out at this point. These compounds' activity as mitochondrial ETC inhibitors also raises intriguing questions regarding their mild toxicity compared to lethal poisons such as cyanide or rotenone. High *K_m* and the reversible nature of the inhibition may be partly responsible for this, and these characteristics may prove advantageous when the goal is acute and transient alleviation of hypoxia prior to irradiation. Comprehensive understanding of the biochemical targets of these inhibitors should aid the design of derivatives with desired pharmacokinetic and pharmacodynamic features and facilitate their addition into clinical protocols in logistically efficient ways. A novel mitochondrial complex I inhibitor, IACS-010759 is currently in Phase I clinical trials against advanced solid tumors and leukemias (NCT03291938 and NCT02882321).⁶⁰ It should be interesting in future, later stage trials combine this agent with radiation treatment to test its potential for radiosensitization. An additional sequela of electron transport chain inhibition may be changes in mitochondrial Reactive Oxygen Species (ROS) generation, which may impact genomic instability.⁶¹ The possible contribution of decreased ROS in the radiation-sensitizing effects of oxygen metabolism inhibitors has not been established yet and may depend on the balance between the drop of protumorigenic ROS signals and the capacity of antioxidant defense programs, such as NRF2, to detoxify these damaging oxygen species and promote survival.^{61–63}

LIPID METABOLISM

DNA damage is a well-established and crucial mechanism for cell death by ionizing radiation,⁶⁴ however, oxidative damage to other macromolecules and subcellular structures can contribute to the cytotoxic effects of radiation treatment.

Ferroptosis was recently described as a non-apoptotic form of cell death, characterized by an iron-dependent excessive

Figure 4. Proposed pathways of radiation-induced ferroptosis. Ionizing radiation induces peroxidation damage to phospholipids with polyunsaturated fatty acids. Activation of ATM signaling in tumor cells and IFN γ secreted by T cells can synergize to decrease expression of SLC7A11, deplete GSH, and trigger ferroptotic cell death

peroxidation of phospholipids containing polyunsaturated fatty acids (PUFAs) which leads to membrane destruction.⁶⁵ Lipid peroxidation can be initiated either spontaneously, when ROS directly attack the PUFA chains, or enzymatically by the activity of lipoxygenases (LOX) and the generation of lipid hydroperoxides.⁶⁶ Glutathione Peroxidase 4 (GPX4) is considered as a major enzymatic inhibitor of ferroptosis, due to its ability to reduce oxidized fatty acids and cholesterol in cell membranes using GSH as a cofactor.⁶⁷ In addition to direct GPX4 inhibitors that have recently been identified, agents that directly or indirectly deplete GSH pools also have the potential to induce ferroptosis. For example, inhibitors of glutaminase block the conversion of glutamine to glutamate, reduce GSH production and secretion, and sensitize to lung tumor cells to radiation.⁶⁸ Inhibitors of the system X_c⁻ (cystine/glutamate transporter) which imports the precursor of GSH production cystine, or inhibitors of enzymes in the GSH biosynthetic pathway have been shown to stimulate ferroptotic cell death.⁶⁹ The protective role of cysteine against radiation was recognized in the 1940s,⁷⁰ and more recently, small molecule inhibition of GSH synthesis has been shown to increase the efficacy of radiation treatment in preclinical models.^{71–74} Additionally, PUFA supplementation was known to sensitize to ionizing radiation, but the direct contribution of lipid peroxidation to loss of cell viability was not determined.^{75–77}

Two recent reports have identified radiation-induced ferroptosis contributing to cytotoxicity (Figure 4). Radiation treatment increased lipid peroxidation *in vitro* and *in vivo*, while small

molecule inducers of ferroptosis or cystine depletion improved model tumor control when combined with radiation.^{78,79} Ye et al demonstrated the effectiveness of ferroptosis inducers as radiosensitizers in *ex vivo* glioma slice cultures and a lung cancer PDX model.⁷⁹ Lang et al identified an additional component of radiation-induced ferroptosis from T-cell signaling, by utilizing syngeneic tumor models in immune-proficient mice.⁷⁸ The exact biochemical mechanisms of phospholipid damage by radiation are not clear yet, but both reports implicate glutathione depletion mediated by decreased expression of the X_c⁻ system as the ferroptosis trigger. Targeted irradiation of the cytoplasm only, but not of the nucleus, with a proton microbeam resulted in lipid peroxidation, suggesting that DNA damage is not necessary or sufficient for ferroptotic signaling.⁷⁹ Alternatively, radiotherapy-induced ferroptosis *in vivo* was attributed to two distinct mechanisms: in tumor cells, radiation suppressed in an ATM-dependent manner the expression of SLC7A11, which is a member of the heterodimeric X_c⁻ cystine-glutamate antiporter. In parallel, T cells produced interferon γ (IFN γ), which in turn activated STAT1 in tumor cells in a paracrine fashion and further downregulated SLC7A11 expression, leading to maximal activation of ferroptosis.⁷⁸ Immune checkpoint inhibitors combined with radiation led to improved model tumor control, in part via ferroptotic cell kill, whereas tumors of ferroptosis-resistant cells did not respond to radiotherapy plus immunotherapy treatment.⁷⁸

These emerging models of lipid- and iron-metabolism-dependent cytotoxicity and their potential to be targeted therapeutically

with small molecules warrant further development of ferroptosis modulators with better pharmacokinetics and biodistribution, and their careful assessment in appropriate preclinical models. One essential consideration will be their specificity for tumor-specific sensitization without exacerbating radiotherapy's effects on normal tissue. Genetic deletion of *GPX4* in mice is lethal, suggesting that its functions are essential for organismal antioxidant protection,⁸⁰ although it is possible that short-term and/or incomplete pharmacological inhibition can give an acceptable therapeutic window during localized irradiation. In support of differential sensitivity to GPX4 inhibition in various genetic contexts, clear cell renal cell carcinomas (ccRCC) are particularly sensitive to ferroptosis due to abundance of PUFAs⁸¹ mediated by pseudohypoxic signaling and overexpression of the lipolytic inhibitor HILPDA (Hypoxia Inducible Lipid Droplet Associated protein).^{82–84} Other candidate biomarkers for ferroptosis sensitivity may include genes or metabolic processes controlling iron, phospholipid, and glutamine metabolism;⁶⁶ however, the specific genetic contexts and their clinical applicability will require systematic analysis. Combination therapies of immune checkpoint inhibitors and radiation are currently being tested in a large number of clinical trials (clinicaltrials.gov). The pathways of radiation-induced tumor immunogenicity and on the other hand, the mediators of immunotherapy-induced radiosensitization are not entirely clear yet.^{85,86} Susceptibility to ferroptosis may be one of the metabolic vulnerabilities that could be exploited to improve the control of radiation-resistant tumors and elicit durable immune responses.⁷⁸ For example, subsets

of triple-negative breast cancers show elevated Xc-activity and cystine addiction,^{87,88} implying that in such tumors ferroptosis inducers may be particularly effective radiosensitizers. Anti-PD-L1 therapy recently received FDA approval for advanced triple negative breast cancer,⁸⁹ further creating opportunities to identify actionable metabolic correlatives of treatment responses.

CONCLUSIONS AND FUTURE PERSPECTIVES

The development of new and clinically successful metabolic modulators that will work as radiation sensitizers must fulfill fundamental criteria of tumor-specific sensitization and favorable pharmacological parameters such as PK/PD and biodistribution. Proof of principle paradigms of synthetic lethality between radiotherapy and unique metabolic dependencies have been reported and some are making their way into clinical trials.^{19,90} The concept of correcting tumor hypoxia has the potential to improve radiation's therapeutic ratio, as physiological areas of radiobiological hypoxia have not been found in many tissues. FDA approved drugs with off-target activities of oxygen consumption inhibition are in clinical trials and their outcome is highly anticipated.

ACKNOWLEDGMENT

This work was in part funded by NCI CA191653 (I.P.)

CONFLICT OF INTEREST DISCLOSURE

Ioanna Papandreou holds intellectual rights on a patent describing papaverine as a radiation sensitizer.

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