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Emerging Roles for SIRT5 in Metabolism and Cancer

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Abstract

Significance: Developing evidence in the literature suggests that sirtuin 5 (SIRT5) may be involved in metabolic reprogramming, an emerging hallmark of cancer by which neoplastic cells reconfigure their metabolism to support the anabolic demands of rapid cell division. SIRT5 is one of the seven members of the nicotinamide adenine dinucleotide-dependent sirtuin family of lysine deacetylases. It removes succinyl, malonyl, and glutaryl groups from protein targets within the mitochondrial matrix and other subcellular compartments. SIRT5 substrates include a number of proteins integral to metabolism.
Recent Advances: New work has begun to elucidate the roles of SIRT5 in glycolysis, tricarboxylic acid cycle, fatty acid oxidation, nitrogen metabolism, pentose phosphate pathway, antioxidant defense, and apoptosis.
Critical Issues: In this study, we summarize biological functions of SIRT5 reported in normal tissues and in cancer and discuss potential mechanisms whereby SIRT5 may impact tumorigenesis, particularly focusing on its reported roles in metabolic reprogramming. Finally, we review current efforts to target SIRT5 pharmacologically.
Future Directions: The biological significance of SIRT5 has been elucidated in the context of only an extremely small fraction of its targets and interactors. There is no doubt that further studies in this area will provide a wealth of insights into functions of SIRT5 and its targets in normal and neoplastic cells. *Antioxid. Redox Signal.* 28, 677–690.

Keywords: SIRT5, sirtuins, desuccinylase, demalonylase, deglutarylase, metabolic reprogramming

Introduction

SIRTUINS MODULATE CELLULAR metabolism and other processes in response to fluctuations in cellular energy demands. Originally identified in yeast, the silent information regulator 2 genes (sirtuins) are highly conserved proteins present in species ranging from bacteria to humans. The seven mammalian sirtuin proteins (SIRT1–7) are nicotinamide adenine dinucleotide (NAD^+)-dependent lysine deacetylases implicated in regulating diverse aspects of cell biology, including metabolism, genome stability, and gene expression, among others (11). Sirtuin activity is regulated, in part, by alterations in cellular NAD^+ levels; NAD^+ levels undergo modest fluctuations under normal conditions and are reported to change dramatically under conditions of nutrient deprivation, obesity, and aging (4).

Despite their conserved catalytic domains, sirtuins are generally nonredundant in their functions, in part, due to differences in their subcellular localizations, biochemical targets, and enzymatic activities. Although sirtuins were originally characterized as deacetylases, a substantial body of literature has demonstrated that all sirtuins are capable of removing alternative lysine modifications, a topic discussed in depth subsequently. SIRT1, 6, and 7 are predominantly nuclear under most conditions, and SIRT2 generally resides in the cytosol, but undergoes nuclear translocation during mitosis (11, 99). These nonmitochondrial sirtuins participate in diverse cellular processes and have been extensively discussed in other recent reviews (12, 24, 35, 37, 40, 42, 46, 54).

SIRT3, SIRT4, and SIRT5 are primarily present in the mitochondrial matrix and possess distinct biochemical activities and substrate specificities (42). SIRT3 represents the major deacetylase activity in mitochondria, playing prominent roles in promoting fatty acid oxidation (FAO), tricarboxylic acid (TCA) cycle activity, and the mitochondrial unfolded protein response, among other processes, while suppressing levels of reactive oxygen species (ROS) (33, 42, 53). Among its

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multiple roles in ROS management, SIRT3 deacetylates and activates superoxide dismutase (SOD)2 and isocitrate dehydrogenase (IDH)2, an enzyme involved in regenerating the antioxidant glutathione (GSH). SIRT3 is implicated in tumor suppression, and elevated SIRT3 activity partially explains ROS suppression associated with the longevity-promoting intervention calorie restriction (31, 78, 89).

SIRT4 is a very weak deacetylase that inhibits the pyruvate dehydrogenase complex (PDC) by removing lipoamide modifications from the PDC E2 subunit (60). SIRT4 also deacetylates and inhibits malonyl-coenzyme A (CoA) decarboxylase, thereby reducing FAO (44). SIRT4 was reported to adenosine diphosphate (ADP)-ribosylate glutamate dehydrogenase (GDH) to block glutaminolysis, a role thought to be important for SIRT4 function in suppressing tumorigenesis (28, 39, 66). Very recently, SIRT4 was shown to remove multiple derivatives of methylglutaryl (methylglutaryl, hydroxymethylglutaryl, and 3-methylglutaryl groups) from lysine residues to regulate leucine metabolism, thereby inhibiting insulin secretion (1).

Like SIRT4, SIRT5 also possesses very weak deacetylase activity. Instead, SIRT5 targets lysine succinylation, malonylation, and glutarylation (Ksucc, Kmal, and Kglu) groups on its target proteins (16, 42, 70, 73, 74, 80, 94). The published roles of SIRT5 implicate this enzyme in the regulation of mitochondrial metabolism and other cellular processes, some of them extramitochondrial. Emerging literature highlights an involvement of SIRT5 in oncogenesis, the major focus of our discussion.

General phenotypes of malignancy have been conceptualized in the form of established hallmarks, such as uncontrolled cell cycle progression, genome instability, and metastatic potential, and the more recently described emerging hallmarks, including altered metabolism. Sirtuins act as major regulators of many of these hallmarks (11, 29). Roles for SIRT1, SIRT3, and SIRT6 in malignancy have been extensively characterized, and an emerging literature has begun to elucidate roles for SIRT4 and SIRT7 in cancer. Functions of SIRT5 in cancer have been less well defined (11, 42). In this study, we review the enzymatic activities and functions of SIRT5 in normal cell metabolism and in cancer metabolic reprogramming.

A Brief Primer on Sirtuin Biochemistry

Sirtuin family members share a core catalytic domain, but vary in their flanking N- and C-terminal sequences (11). Sirtuin core domains differ at specific amino acids—differences that help confer distinct enzymatic activities. Structural analysis reveals that the sirtuin catalytic domain comprises two subdomains, a highly conserved Rossmann fold typically found in NAD^+-consuming enzymes and a more variable zinc-binding finger (20, 65). These two subdomains are connected by several homologous, but structurally variant, loops that collectively form a binding pocket for an acylated lysine and NAD^+ (117). This binding pocket aligns the acyl-lysine and the ribose moieties of NAD^+ with key amino acids responsible for catalyzing the sirtuin reaction (18). Although not directly involved in catalysis, the zinc finger is integral for maintaining proper enzyme structure and function (10).

Sirtuin-mediated deacetylation is initiated by the cleavage and release of nicotinamide (NAM) derived from NAD^+ and the formation of an intermediate between ADP-ribose and the acyl moiety. Upon resolution of this intermediate, the newly deacetylated lysine and 2′-*O*-acyl-ADP-ribose are released (117). A product of the sirtuin reaction, NAM, inhibits sirtuin catalytic activity in a noncompetitive manner (5). NAM is recycled back to NAD^+ *via* a salvage pathway, the major source of NAD^+ in mammalian cells (81).

SIRT5 Possesses Unique Biochemical Activities

SIRT5 remains a somewhat mysterious protein, in part, because SIRT5 enzymatic activity is unique compared with that of other sirtuin proteins. SIRT5 was initially characterized as a lysine deacetylase, although it possesses very weak deacetylase activity (71). In 2011, the crystal structure of SIRT5 bound to a substrate peptide was solved, revealing that SIRT5 possesses a larger binding cavity compared with other sirtuins, with three unique active site amino acid residues, Ala86, Tyr102, and Arg105 (16, 74). It was established that SIRT5 efficiently removes acidic acyl groups—succinyl, malonyl, and glutaryl moieties—from lysine residues (16, 42, 70, 73, 74, 80, 94). Similar to phosphorylation, succinylation, malonylation, or glutarylation of a lysine residue (Ksucc, Kmal, and Kglu) renders it negatively charged at physiologic pH (74, 94).

Recently, it was reported that SIRT7, like SIRT5, is capable of desuccinylating histones (16, 47, 73, 74). However, since SIRT7 is not thought to localize to the mitochondria and since cellular Ksucc, Kmal, and Kglu levels globally increase in response to loss of SIRT5, current evidence supports the view that SIRT5 function is nonredundant with other sirtuins in the mitochondria and likely elsewhere in the cell as well (16, 47, 70, 73, 84, 94, 112).

Ksucc, Kmal, and Kglu are structurally quite similar (74). Succinyl-, malonyl-, and glutaryl-CoA, the precursors to Ksucc, Kmal, and Kglu, respectively, are derived from metabolites succinate, malonate, and glutarate (34). Succinyl-CoA is a 4-carbon mitochondrial TCA cycle intermediate (11a). Malonyl-CoA is a three-carbon product of acetyl-CoA carboxylation in the cytosol, involved in fatty acid synthesis (96). Mitochondrial malonyl-CoA is generated by ACSF3 and the major source of mitochondrial protein lysine malonylation (6). Glutaryl-CoA, a product of tryptophan and lysine catabolism, comprises four carbons and is predominantly mitochondrial (79). How these moieties become conjugated to lysines remains incompletely defined and a subject of active investigation.

Acetylation of nuclear and cytosolic lysines is catalyzed by lysine acetyltransferase (KAT) enzymes (ketoacyl coenzyme A thiolase) that employ acetyl-CoA as the source of acetyl groups. In addition to acetyl-CoA, the human KAT p300 has been found to accommodate propionyl-CoA as well as larger moieties such as crotonyl-, butyryl-, hydroxyisobutyryl-, succinyl-, and glutaryl-CoA (83, 84, 94). It is highly likely that acylation of many SIRT5 substrates occurs in a nonenzymatic manner. Acyl-CoAs possess a highly reactive thioester bond and function as acyl carrier species. Under chemical conditions in the mitochondrial matrix, the acyl groups can be conjugated to lysine side chains in an enzyme-independent mechanism, as in the case of mitochondrial lysine succinylation (38). Importantly, succinyl-CoA was recently found to exhibit much greater lysine acylation propensity than acetyl-CoA (101).

Enzyme-independent acylation was recently shown to occur within the cytosol as well (41, 102); acylation of glyceraldehyde-3-phosphate dehydrogenase (GAPDH) and pyruvate kinase muscle isozyme M2 (PKM2) by malonyl-

CoA was found to be catalytically independent (41). Consistently, SIRT5 was shown to target GAPDH and PKM2 (59, 70, 104, 106). Thus, many lysine modifications targeted by SIRT5 may simply occur as a passive by-product of cellular metabolism, rather than as an actively enzyme-catalyzed event.

SIRT5 possesses a well-defined mitochondrial localization sequence (MLS) and the majority of SIRT5 proteins localize to the mitochondrial matrix (61, 64). However, a significant fraction of SIRT5 is cytosolic and a modest portion is even nuclear (64, 73). Mechanisms of SIRT5 localization to non-mitochondrial compartments have not been clearly elucidated. In humans, alternative splicing of the *Sirt5* mRNA yields two dominant isoforms, $SIRT5^{iso1}$ and $SIRT5^{iso2}$, both of which contain an N-terminal MLS, but differ at their C-termini (61). Several other SIRT5 isoforms are reported in the NCBI database (14, 22). Based on studies using ectopically expressed green fluorescent protein (GFP)-tagged SIRT5, one report found that while both SIRT5 isoforms localized to mitochondria, only $SIRT5^{iso1}$ was present in the cytoplasm. Most studies of SIRT5 have focused on elucidating functions of $SIRT5^{iso1}$, the only reported form of SIRT5 present in mice (14, 61, 73).

Unbiased mass spectrometry-based proteomic surveys have proven extremely valuable in defining cellular substrates of, and potential functions for, SIRT5. This work has been greatly facilitated by the availability of a viable SIRT5-deficient mouse model (53, 73, 113). These mice showed greatly increased tissue Ksucc, Kmal, and Kglu levels, but little change in Kac, confirming the *in vitro* characterization of SIRT5 activity (73, 74, 94, 113). Collectively, these studies revealed that SIRT5 targets hundreds of protein substrates, typically on multiple lysines per substrate protein. A large majority of these modified proteins localize to the mitochondrial matrix, with many involved in cellular metabolism, particularly in the TCA cycle, urea cycle, and FAO (70, 73, 74, 80, 94). However, in the cytosol, SIRT5 was shown to act on multiple glycolytic enzymes as well as proteins associated with the 40S and 60S ribosomal subunits, to name a few (61, 73). Ksucc, Kmal, and Kglu sites were also identified on histones, suggesting a potential role for SIRT5 in transcriptional regulation (73, 74, 94, 107).

Ksucc and Kglu levels are highest in mitochondria, where succinyl-CoA, as a TCA cycle intermediate, and glutaryl-CoA are particularly abundant. By contrast, Kmal is especially prominent in the cytosol (94, 107). Pathway analysis of Ksucc has identified potential roles for SIRT5 in regulating glycolysis, keto acid breakdown, redox homeostasis, cardioblast proliferation, acid thiol ligation, and protein translation (16, 73, 80, 94). Similarly, examining Kmal enrichment revealed proteins that modulate glycolysis and gluconeogenesis, retinoid X receptor inhibition, and estrogen biosynthesis (16, 70), and Kglu sites predominantly occurred on proteins involved in aerobic respiration, redox homeostasis, and the TCA cycle (16, 94). All three marks are enriched on proteins involved in FAO (1, 16, 70, 73, 74, 94). The diversity of SIRT5 targets and interacting partners indicates that SIRT5 is functionally important in multiple subcellular compartments, and not only in the mitochondrial matrix.

SIRT5 Function in Organismal Homeostasis

SIRT5 is expressed in most tissues, with the highest SIRT5 levels in the heart and brain (61, 68). Despite the widespread expression of SIRT5 and unique enzymatic activities, germline SIRT5 deficiency is well tolerated in mice under basal conditions (53, 85, 113). One study showed that SIRT5-deficient animals on the C57BL/6 background were born at a sub-Mendelian ratio (113), although this effect had not been observed in previous work using 129 background mice (53). *Sirt5* knockouts (KOs) show perturbed FAO and mildly depressed ketogenesis (80) and elevated blood ammonia levels following a prolonged fast (113). *Sirt5* KOs showed marked cardiac protein hypersuccinylation and develop mild cardiac dysfunction and hypertrophy with age under basal conditions (85). SIRT5 protects from myocardial and cerebral injury in response to ischemia (7, 67). In humans, the presence of a single-nucleotide polymorphism in the SIRT5 promoter correlates with reduced *Sirt5* mRNA expression and gene expression changes, suggestive of aging in different brain regions (25).

SIRT5 Expression in Cancer

In humans, the *Sirt5* gene is found on the highly unstable cytogenetic band on chromosome 6p23 (58, 93). Likewise, gain or loss of the *Sirt5* gene locus occurs frequently across a range of cancers, although often in the context of nonfocal genomic events resulting in alterations in many flanking genes in addition to SIRT5 (Fig. 1) (23, 58, 69, 93). *Sirt5* mRNA expression is detectable across a wide variety of cancers (Fig. 2).

A number of studies have shown that in specific tissues, *Sirt5* mRNA expression tends to be elevated in tumors relative to the corresponding normal tissue type, an issue discussed in more depth later in the review, suggesting a potential tumor-promoting role for SIRT5 in certain contexts (36, 50, 56, 91). For example, *Sirt5* mRNA and protein expression levels are frequently elevated in human lung cancers and serve as a predictor of recurrence, rendering SIRT5 a candidate biomarker for poor survival in non small cell lung cancer (NSCLC) (50, 56). Additionally, tumor tissues from patients suffering from Waldenstrom's macroglobulinemia, a B cell malignancy, show elevated *Sirt5* mRNA levels compared with B-lymphocytes from healthy donors (91). Likewise, *Sirt5* mRNA expression was markedly increased in grade III estrogen receptor-negative/progesterone receptor-positive invasive breast tumors (36).

In contrast, *Sirt5* mRNA expression was significantly reduced in endometrial carcinoma, another hormone-responsive cancer, compared with benign endometria. *Sirt5* mRNA levels were also lower in head and neck squamous cell carcinoma compared with corresponding noncancerous tissue (3, 43). For the remainder of this review, we discuss known functions of SIRT5 in normal and malignant tissues, focusing on SIRT5 interactors and targets that may be most relevant in cancer.

SIRT5 in Normal and Cancer Metabolism

Cancer cells reprogram their metabolism to support the anabolic demands of cell growth (29). Malignant cells require amplified production of nucleosides, amino acids, and lipids for synthesis of RNA, DNA, and membranes. Additionally, these cells require mechanisms for efficient ROS elimination (11). A particularly notable aspect of metabolic reprogramming involves rewiring of glucose and glutamine metabolism. For example, glycolysis becomes a major source of ATP synthesis in many cancer cells and provides

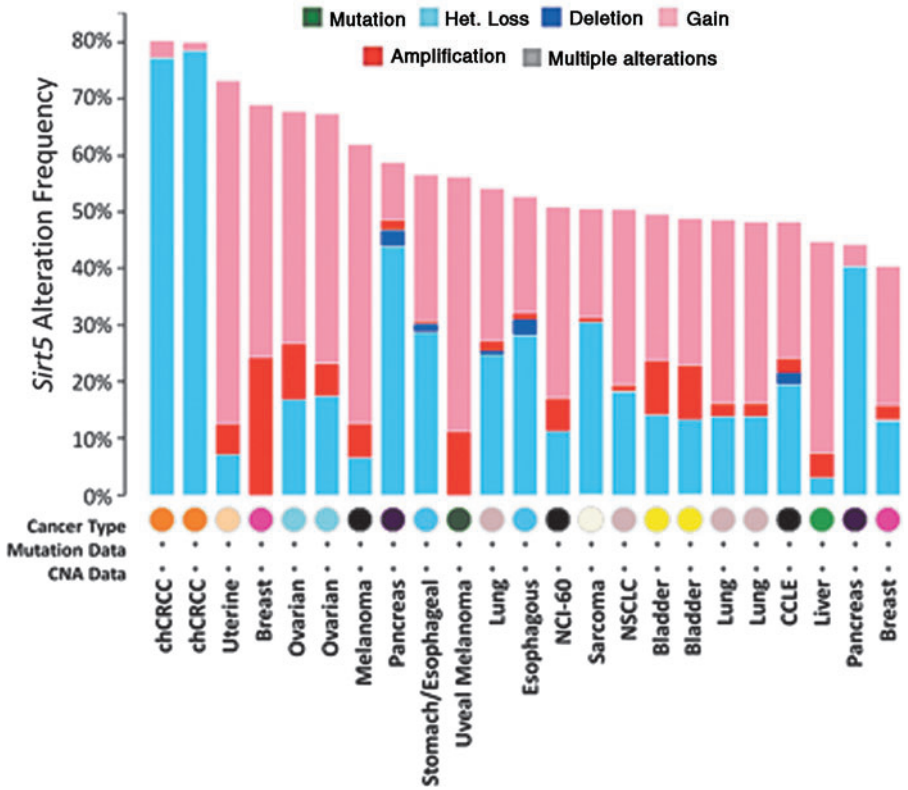

FIG. 1. *SIRT5* gene alteration across multiple cancer types. The frequency of *SIRT5* gene alteration was measured across cancer types in The Cancer Genome Atlas (cBioportal) on April 15, 2017. The following genetic alterations were queried: copy number gain (low-level gene gain), amplification (high-level gene amplification), het loss (heterozygous deletion), and deletion (homozygous deletion). Cancer types with alteration frequency greater than 40% are shown (69).

precursors for numerous anabolic processes and for antioxidant defense (57).

SIRT5 Promotes Glycolysis and Regulates Carbon Flux into Mitochondria

Glucose is transported into the cell by glucose transport proteins, particularly GLUT1 (57, 88). SIRT5 depletion in H1299 NSCLC cells led to a decrease in *GLUT1* mRNA and protein levels (57). Glycolysis ultimately results in the conversion of glucose to either pyruvate (*via* pyruvate kinase, including the PKM2 isoform) or lactate (*via* lactate dehydrogenase, LDHA, or LDHB) (11a). SIRT5 was shown to demalonylate GAPDH and other enzymes in the glycolytic cascade in mouse liver, resulting in increased GAPDH activity and elevated glycolytic flux (70). An independent

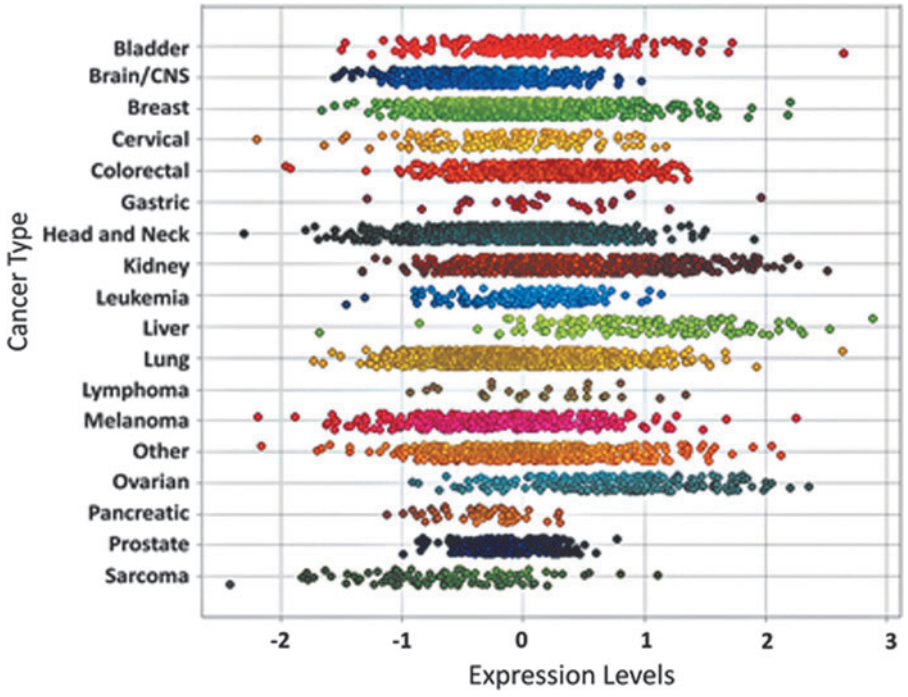

FIG. 2. *SIRT5* mRNA expression across multiple cancer types. SIRT5 expression across >30,000 cancer samples using the Oncomine powertools Oncomine Gene Expression Browser (82). Normalized linear expression plotted for all cancer samples profiled by Affymetrix U133A, U133A 2.0, and U133 Plus 2.0 arrays in the Oncomine database. Samples stratified by cancer type as indicated. Courtesy of Dr. Scott Tomlins, Department of Pathology at the University of Michigan, Ann Arbor, MI.

proteomic survey in 293T cells confirmed that SIRT5 physically interacts with GAPDH (59).

In the final step of glycolysis, PKM2 catalyzes conversion of phosphoenolpyruvate to pyruvate, and PKM2 suppression impairs conversion of glucose into pyruvate (13). One report found that SIRT5 desuccinylates PKM2 at a single lysine residue in A549 NSCLC and 293T cells, suppressing PKM2 activity. SIRT5-dependent inhibition of PKM2 in these cells led to reduced glycolytic flux, reduced mitochondrial respiration, and enhancement of cell proliferation and protection against oxidative insult. Additionally, the interaction between PKM2 and SIRT5 was enhanced by increased cellular ROS levels. This suggests a potential feedback mechanism, whereby ROS induces SIRT5 to restrict carbon entry into the TCA cycle, thereby minimizing further ROS production (106, 118). Yet another report found that desuccinylation of PKM2 by SIRT5 maintains its cytosolic localization in a proinflammatory environment (104), further demonstrating the context-dependent impact of SIRT5 activity.

Elevated lactic acid levels foster a microenvironment that suppresses antitumoral immune response (8, 63). SIRT5 depletion in H1299 NSCLC cells correlates with reduced *LDHA* mRNA and protein levels through mechanisms that remain to be elucidated (57). LDHA consumes pyruvate to produce lactate, suppressing conversion of pyruvate to acetyl-CoA by PDC, which would otherwise fuel the TCA cycle. SIRT5 desuccinylates the PDC E1α subunit at multiple sites, suppressing overall PDC enzymatic activity in 293T cells. SIRT5-deficient liver mitochondria show increased pyruvate-dependent respiration (73). However, conflicting results from HepG2 cells and mouse cardiac tissue suggest that the impact of SIRT5 on PDC and mitochondrial respiration may be context dependent (9, 85).

Although it is a very weak deacetylase, cytosolic SIRT5 has been reported to inhibit PDC through deacetylation and deactivation of signal transduction and activator of transcription 3 (STAT3) in A549 lung cancer cells (71, 108). Upon phosphorylation by interleukin-6-stimulated Janus kinases, cytosolic STAT3 undergoes rapid nuclear translocation to bind and activate the promoters of hundreds of progrowth and prosurvival genes. In contrast, acetylation promotes mitochondrial translocation of STAT3, which has been shown to drive RAS-dependent tumorigenesis (111). Mitochondrial STAT3 binds and activates PDC, correlating with elevated mitochondrial membrane potential (MMP) (86, 115) and ATP synthesis in PC3 prostate cancer cells (100, 108). In PC3 cells, deacetylation of cytosolic STAT3 by SIRT5 inhibited its mitochondrial translocation and interaction with PDC, thereby reducing pyruvate conversion into acetyl-CoA, lowering the MMP, and inhibiting ATP production. Two of the three Kac sites of STAT3, K685 and K707, are also conserved in STAT1 and STAT5b, indicating that SIRT5 may regulate the mitochondrial activity of multiple STAT family members (108).

In summary, SIRT5 regulates several aspects of glucose metabolism across multiple subcellular compartments. In the cytosol, it promotes glycolysis *via* effects on GAPDH and likely other glycolytic enzymes and it suppresses PKM2 activity in noninflammatory conditions (70, 106). Similarly, SIRT5 inhibits pyruvate entry into the TCA cycle by deacetylating cytosolic STAT3 and inhibiting its mitochondrial translocation, thereby indirectly reducing PDC function (45). Mitochondrial SIRT5 also directly desuccinylates and inhibits PDC (73). Overall, these roles for SIRT5 in glucose metabolism, on balance, favor metabolic reprogramming and a tumor-promoting cellular metabolism (Fig. 3).

SIRT5 Regulation of the TCA Cycle and Electron Transport Chain

SIRT5 also regulates multiple TCA cycle enzymes. SIRT5 inhibits the conversion of succinate to fumarate by desuccinylating succinate dehydrogenase subunit A (SDHA) in 293T cells (73). SDHA, also known as Complex II of the electron transport chain, catalyzes conversion of succinate to fumarate, coupled to reduction of NAD^+ to NADH and the generation of ubiquinol from ubiquinone (11a). SIRT5-depleted 293T cells, or SIRT5-deficient liver mitochondria, show increased succinate-dependent respiration (73). Contradictory results have been reported using distinct approaches for measuring Complex II activity and mitochondrial respiration (115). SIRT5 also desuccinylates IDH2, increasing enzyme activity and upregulating conversion of isocitrate to alpha-ketoglutarate (α-KG) in 293T cells. SIRT5 depletion induces

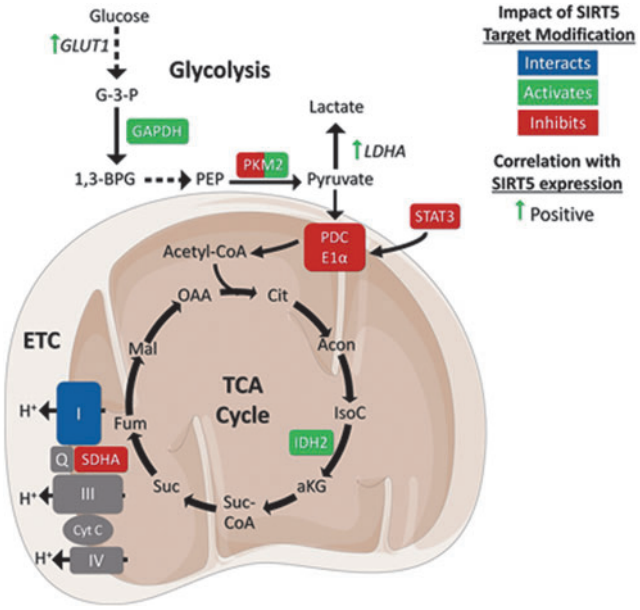

FIG. 3. SIRT5 regulates glycolysis, the TCA cycle, and the ETC. SIRT5 impacts activities and expression levels of enzymes involved in glycolysis, the TCA cycle, and the ETC. See text for details. Figures were produced using material from Servier Medical Art (www.servier.com). αKG, alpha ketoglutarate; Acetyl-CoA, acetyl-coenzyme A; Acon, aconitate; Cit, citrate; Cyt C, cytochrome C; ETC, electron transport chain; Fum, fumarate; G-3-P, glucose 3 phosphate; GAPDH, glyceraldehyde phosphate dehydrogenase; *GLUT1*, glucose transporter 1; IDH2, isocitrate dehydrogenase 2; I–IV, complex I–IV; IMM, inner mitochondrial membrane; IsoC, isocitrate; *LDHA*, lactate dehydrogenase A; Mal, malate; MM, mitochondrial matrix; OAA, oxaloacetate; PDCE1α, pyruvate dehydrogenase complex E1α subunit; PEP, phosphoenolpyruvate; PKM2, pyruvate kinase muscle isozyme 2; Q, Quinolone; SDH/complex II, succinate dehydrogenase; STAT3, signal transducer and activator of transcription 3; Suc, succinate; Suc-CoA, succinyl-coenzyme A; TCA cycle, tricarboxylic acid cycle.

increased IDH2 succinylation and impaired IDH2 activity (118). SIRT5 was also found to interact with the complex I subunit NDUFA4 (Fig. 3) (59), although the functional significance of this interaction has not been elucidated. Cytochrome *c* (Cyt C) was found to be deacetylated by SIRT5 *in vitro*, a topic we discuss subsequently (86).

In recent years, mutations in IDH1 and IDH2 have been identified as driver events in a number of cancer types, particularly including gliomas and acute myeloid leukemia (AML) (15). These mutations cause IDH enzymes to produce a novel oncometabolite, (R)-2-hydroxyglutarate (2-HG). A large literature documents the ability of 2-HG to inhibit α-KG-dependent dioxygenases, particularly those involved in DNA and histone demethylation, in turn leading to widespread epigenetic dysregulation and promoting tumorigenesis.

Li *et al.* identified a novel role for 2-HG in inhibiting the TCA cycle enzymes, SDH and fumarate hydratase, resulting in increased cellular succinate levels and widespread increased protein succinylation, predominantly not only on mitochondrial proteins but also in the cytosol and the nucleus. Functionally, this hypersuccinylation was associated with mitochondrial dysfunction and accumulation of the antiapoptotic protein B cell lymphoma-2 (BCL-2). In cells expressing mutant IDH1, enforced SIRT5 expression relieved 2-HG-induced protein hypersuccinylation, reversing mitochondrial dysfunction and apoptotic resistance. Xenograft tumor formation by cells expressing mutant IDH was impaired by SIRT5 overexpression (45). These findings imply a tumor suppressive role for SIRT5 specifically in the context of mutant IDH1.

Taking these findings altogether, SIRT5 controls the production of several intermediates in the TCA cycle and electron transport. SIRT5 desuccinylates and inhibits SHDA, restricting TCA cycling by decreased production of fumarate as well as electron transport (73). By contrast, SIRT5 promotes the production of TCA cycle intermediate α-KG by activating IDH2; however, in glioma cells bearing the IDH1 R123H mutant, SIRT5 relieves the oncogenic effects of 2-HG accumulation (45, 118).

SIRT5 Promotes FAO

FAO is initiated when fatty acids of varying lengths are converted into fatty acyl-CoA intermediates, which are then transported into mitochondria by way of the carnitine shuttle. For very long fatty acyl-CoA (VLCFA) substrates, very long-chain acyl-CoA dehydrogenase (VLCAD) converts these intermediates to enoyl-CoA. Enoyl-CoA hydratase (ECHA) is a subunit of the mitochondrial trifunctional (MTF) complex that catalyzes conversion of enoyl-CoA to ketoacyl- and acetyl-CoA to break down fatty acids for energy (11a). SIRT5, together with SIRT3, promotes FAO by deacetylating VLCAD in mice, increasing its activity and enhancing conversion of acyl-CoA to enoyl-CoA (Fig. 4).

SIRT5-mediated desuccinylation works alongside SIRT3-mediated deacetylation to upregulate VLCAD function by stabilizing its association with flavin adenine dinucleotide, a redox cofactor required for VLCAD activity. Notably, SIRT3 and SIRT5 target overlapping, highly evolutionarily conserved lysine residues, K298/K299 and K507. Indeed, K298/K299 lie within the active site of VLCAD, and K507 occurs within the inner mitochondrial transmembrane domain, potentially affecting VLCAD localization to this organelle. Cooperative deacetylation of VLCAD by SIRT3 and SIRT5 is required for VLCAD-cardiolipin substrate binding, increasing FAO flux through the regulation of mitochondrial membrane remodeling (95, 115).

SIRT5 also desuccinylates and activates ECHA, thereby promoting the final step in lipid metabolism. Consistently, SIRT5-deficient myocardium shows impaired FAO (7). Overall, SIRT5 promotes robust ATP generation through enhancing the breakdown of very long-chain fatty acid molecules, an event that can yield upward of 70 ATP molecules per VLCFA to support tissue energy demands (11a).

SIRT5 Regulates Nitrogen Metabolism

Glutaminolysis involves the conversion of glutamine to glutamate and ammonia by glutaminase enzymes (GLS1 and GLS2) and subsequently of glutamate to α-KG by GDH. A by-product of this pathway is ammonia, which is highly neurotoxic if it accumulates to an appreciable degree (11a). SIRT5 desuccinylates and is proposed to inhibit GLS2, thereby reducing production of glutamate and ammonia from glutamine (Fig. 5). In MDA-MB-231 breast cancer cells, SIRT5 was shown to interact with GLS2. Indeed, inhibition of SIRT5 by either genetic KD or a novel SIRT5 inhibitor, MC3482, correlated with increased GLS2 succinylation and elevated cellular glutamate and ammonia levels in MDA-MB-231 cells and mouse myoblast C2C12 cell lines. Overexpression of SIRT5 reversed these phenotypes. Ectopic expression of SIRT5 reduced expression of autophagy and mitophagy proteins; consistently, ammonia is known to promote expression of factors involved in cellular mitophagy

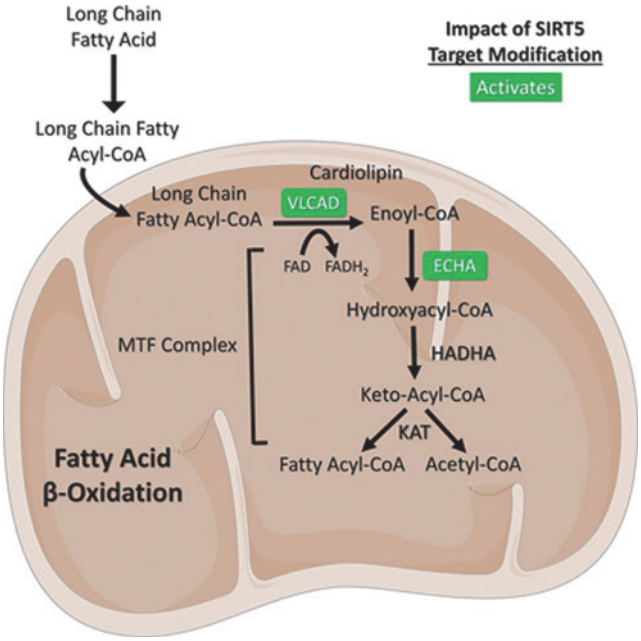

FIG. 4. SIRT5 promotes fatty acid *β*-oxidation. SIRT5 desuccinylates VLCAD and ECHA to activate their enzymatic activities. See text for details. Figures were produced using material from Servier Medical Art (www.servier.com). ECHA, enoyl-coenzyme A hydratase; HADH, hydroxyacyl-coenzyme A dehydrogenase; KAT, lysine acetyltransferase; MTF, mitochondrial trifunctional; VLCAD, very long chain acyl-coenzyme A dehydrogenase.

and autophagy (55, 76). Importantly, however, the impact of SIRT5 on GLS2 biochemical activity has not been directly measured (76).

Ammonia is neutralized *via* conversion to urea in the urea cycle, a process that occurs primarily in the liver and kidney. Carbamoyl phosphate synthetase 1 (CPS1) converts ammonia to carbamoyl phosphate to initiate formation of urea for excretion and represents the rate-limiting step in urea biosynthesis (11a). SIRT5 was originally reported to activate urea cycle function *via* deacetylation of CPS1. SIRT5 associates with CPS1 in mitochondria, and CPS1 activity is decreased in *Sirt5* KO liver tissues (68). Multiple studies have since determined that SIRT5 also desuccinylates and deglutarylates CPS1 to activate it (94, 113). These findings demonstrate that SIRT5 regulates cellular ammonia levels, both at the source of ammonia production, by potentially inhibiting GLS2, and by promoting the detoxifying conversion of ammonia to urea (Fig. 5).

SIRT5 Activates the Pentose Phosphate Pathway and Antioxidant Defense

Hyperactive glutaminolysis drives rapid production of glutamate and aspartate, which, in turn, promotes amino acid and nucleic acid anabolism (11a, 68). The sugar backbone of DNA, ribose-5-phosphate, is generated by the pentose phosphate pathway (PPP). The PPP is initiated through conversion of glucose-6-phosphate (G6P) from glycolysis to 6-phosphogluconolactone by G6P dehydrogenase, an enzyme activated by SIRT5-mediated deglutarylation (Fig. 6) (118). The activity of the enzymes in the PPP is coupled to reduction of two molecules of NADP+ into NADPH, a major reducing equivalent within the cell (11a).

NADPH production by the PPP plays a major role in suppressing accumulation of ROS, toxic by-products of oxygen metabolism that can react with and damage cellular macromolecules (11a). NADPH is required for regeneration of the reduced form of GSH, a potent cellular ROS scavenger, from its oxidized form by glutathione reductase (GSR) (2). SIRT5 plays a role in ROS elimination by stimulating NADPH production *via* the PPP to regenerate GSH (118). Likewise, manipulation of SIRT5 expression revealed a positive correlation between SIRT5 and *GSR* mRNA levels in NSCLC cells (56). Furthermore, *Sirt5* KO mouse embryonic fibroblasts (MEFs) exhibited reduced GSH levels and NADPH production and increased sensitivity to oxidative stress (Fig. 6) (118). Elevated GSH is also implicated in conferring a multidrug resistance (MDR) phenotype in cancers (see Regulation of Apoptotic Resistance by SIRT5) (2, 40a).

In addition to its function in the PPP, SIRT5 possesses other roles in ROS management, *via* desuccinylation and activation of SOD1, a key antioxidant enzyme in ROS detoxification. NSCLC cells expressing a succinylation mimetic mutant form of SOD1 exhibited reduced SOD1 activity and cell viability. Conversely, cells co-overexpressing WT SOD1 and SIRT5 exhibited augmented SOD1 function and decreased levels of cellular ROS (Fig. 6), with enhanced xenograft formation ability (50). Consistently, an independent study in Regulation of Apoptotic Resistance by SIRT5 human neuroblastoma cells showed that SIRT5 overexpression was protective against hydrogen peroxide-induced damage (48). SIRT5 protein expression declined upon induction of oxidative stress in H92c rat cardiomyocytes, and SIRT5 depletion sensitized cells to oxidative stress-induced death (51). Mechanisms of SIRT5 downregulation in this context have not been elucidated. Although SIRT3 had previously been considered to play a dominant role in cellular antioxidant defense among the sirtuins, these recent studies identify SIRT5 as a major factor in this process as well.

Regulation of Apoptotic Resistance by SIRT5

Several studies have demonstrated that SIRT5 promotes apoptotic resistance in normal and cancer cells. Like several other sirtuins (92, 98, 103), SIRT5 is reported to deacetylate

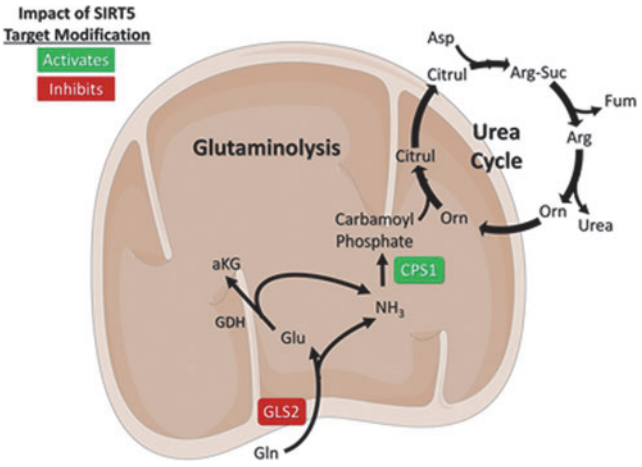

FIG. 5. SIRT5 downregulates glutaminolysis and promotes ammonia detoxification. SIRT5 modifies CPS1 to increase its activity. SIRT5 suppresses ammonia production *via* proposed regulation of GLS2. See text for details. Figures were produced using material from Servier Medical Art (www.servier.com). αKG, alpha ketoglutarate; Arg, arginine; Arg-Suc, arginosuccinate; Asp, aspartate; Citrul, citrulline; CPS1, carbamoyl phosphate synthetase 1; Fum, fumarate; GDH, glutamate dehydrogenase; Gln, glutamine; GLS2, glutaminase 2; Glu, glutamate; NH_3, ammonia; Orn, ornithine.

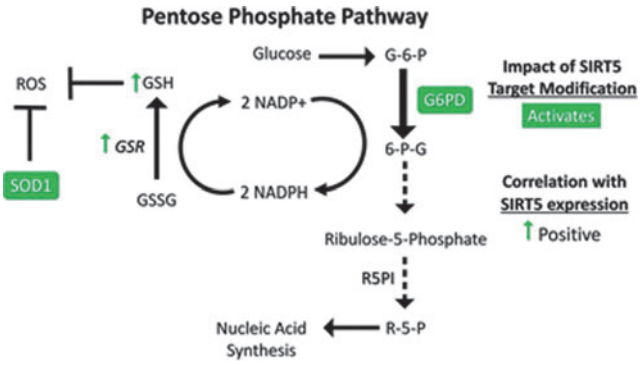

FIG. 6. SIRT5 promotes ROS detoxification *via* SOD1 and the pentose phosphate pathway. SIRT5 desuccinylates SOD1 and deglutarylates G6PD, activating both enzymes. Figures were produced using material from Servier Medical Art (www.servier.com). 6-G-P, 6-phosphogluconate; G-6-P, glyceraldehyde-6-phosphate; G6PD, glucose-6-phosphate dehydrogenase; GSH, glutathione; GSR, glutathione reductase; GSSG, oxidized glutathione; NADP/NADPH, nicotinamide adenine dinucleotide phosphate; R-5-P, ribose 5 phosphate; R5PI, ribulose 5 phosphate isomerase; ROS, reactive oxygen species; SOD1, superoxide dismutase 1.

forkhead box O3A (FOXO3A) (105) despite the weak deacetylase activity of SIRT5 (71). FOXO3A is a transcription factor that promotes expression of genes involved in antioxidant defense and translocates to the nucleus upon deacetylation by SIRT5. Notably, this effect relieves elevated ROS levels and proapoptotic signaling induced upon treatment of NSCLC cells with cigarette smoke extract (105).

The antiapoptotic role of SIRT5 in rat cardiomyocytes in response to oxidative stress mentioned previously likely relates to the observation that SIRT5 directly interacts with the B cell lymphoma-XL (BCL-XL) antiapoptotic protein. While the specific impact of SIRT5 on BCL-XL remains unclear, this interaction may help explain the prosurvival functions of SIRT5 described in many contexts (Fig. 7) (51).

Survival of photoreceptor cells in mice was also found to be SIRT5 and NAD^+ dependent (49). Similarly, SIRT5 was also shown to protect MEFs from mitophagic mitochondrial fragmentation by way of regulating the localization, expression, and dynamics of the profragmentation dynamin-related protein 1. Mitochondrial fragmentation initiates the mitophagic degradation of the organelle and can initiate the apoptotic cascade (26). This link between SIRT5 and mitophagy is consistent with the finding that experimental manipulation of SIRT5 levels in cultured breast cancer cells or mouse myoblasts affects expression of mitophagy-related genes (76). Although the precise mechanism of SIRT5 activity remains to be elucidated in these settings, collectively, these data reveal a role for SIRT5 as an antimitophagy factor.

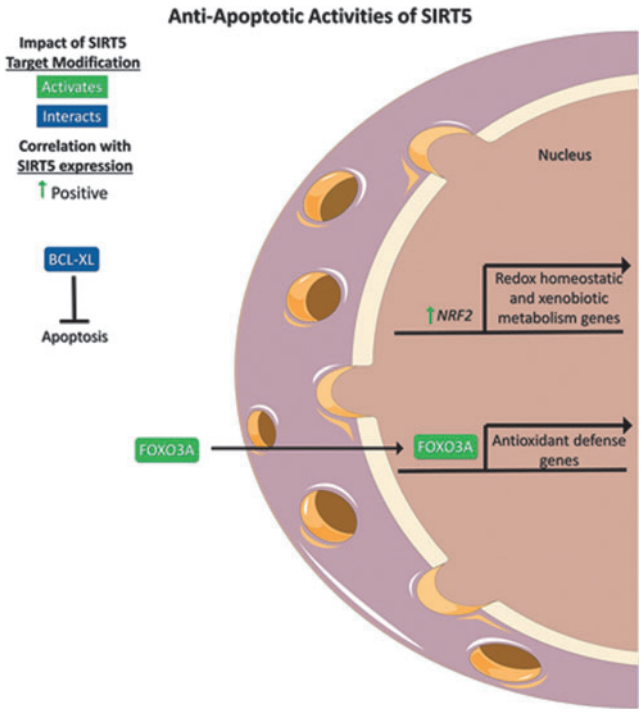

FIG. 7. SIRT5 promotes apoptotic resistance. SIRT5 expression positively correlates with the expression of NRF2, a regulator of redox homeostatic and xenobiotic metabolism genes. SIRT5 deacetylates and promotes the nuclear translocation of FOXO3A, a transcriptional regulator of antioxidant defense genes. SIRT5 also interacts with BCL-XL, which inhibits apoptosis. Figures were produced using material from Servier Medical Art (www.servier.com). BCL-XL, B cell lymphoma-XL; FOXO3A, forkhead box O3A; NRF2, nuclear factor erythroid-2-related factor 2.

Primary or secondary resistance to genotoxic chemotherapy is a major impediment to cancer treatment and cure. *In vitro*, SIRT5 depletion reduces cell viability and sensitizes multiple NSCLC cell lines to genotoxic chemotherapeutics, including cisplatin (CDDP), fluorouracil, and bleomycin. In cultured NSCLC cells, experimental manipulation of SIRT5 levels led to corresponding changes in nuclear factor erythroid-2-related factor 2 (NRF2) expression through mechanisms yet to be defined. NRF2 regulates expression of genes involved in cellular redox homeostasis and xenobiotic metabolism, including chemotherapy drugs. A significant positive correlation was identified between SIRT5 and *NRF2 (NRE2L2)* mRNA levels in NSCLC patients (Fig. 7). In these clinical samples, SIRT5 levels also correlated with expression of a variety of ATP-dependent drug efflux pump transcripts, which promote an MDR phenotype, conferred by increased drug expulsion (56).

SIRT5 is also a negative regulator of the tumor suppressor SUN domain-containing protein 2 (SUN2), a protein that enhances CDDP sensitivity of lung cancer cells. Most NSCLC cells express low levels of SUN2 in comparison with normal lung tissue, and elevated levels of SUN2 protein within the inner nuclear membrane of lung cancer are associated with improved patient survival. SIRT5 suppresses proapoptotic activity of SUN2 and desensitizes NSCLC cells to chemotherapeutics, although the mechanism of SIRT5 activity against SUN2 has not yet been identified (50). Overall, SIRT5 plays roles in resistance to multiple chemotherapeutics, especially CDDP.

Although many reports have identified prosurvival functions of SIRT5, in other contexts, SIRT5 may facilitate cell death. One report found that the impact of SIRT5 on apoptosis was localization dependent in both normal human neurons and neuroblastoma cells. Nuclear and cytosolic SIRT5 supported cell survival, whereas mitochondrial SIRT5 enhanced cell death. Importantly, the fraction of SIRT5 localized to mitochondria was greater in neuroblastoma cells than in neurons (75).

As mentioned previously, SIRT5 overexpression in IDH mutant cells relieved accumulation of the antiapoptotic protein BCL-2 that normally accompanies excess 2-HG production, thereby resensitizing IDH mutant cells to cell death signaling (45). Cyt C, the major initiator of the intrinsic apoptotic cascade, was shown to be deacetylated and stimulated by SIRT5 *in vitro*, although a role for SIRT5 in deacetylating Cyt C *in vivo* could not be demonstrated (86). SIRT5 was also found to promote necroptosis, a cell death pathway characterized by the formation of the RIPK3 complex. SIRT5 depletion protected L929 cells from tumor necrosis factor-induced necroptosis (77). Collectively, these findings support a proapoptotic role for SIRT5 in both normal and cancer cells.

Pharmacologic Targeting of SIRT5

SIRT5 is largely dispensable in nonmalignant tissues and cells (53, 85, 113), yet it plays a progrowth and prosurvival role in several cancer types (48, 51, 56, 57, 105), rendering it a potentially attractive drug target. Although many SIRT5 inhibitors have been described, these agents have not been evaluated for their clinical anticancer activity in cells or in whole animals. The majority of these inhibitors lack

selectivity for SIRT5; the SIRT5 inhibitors, suramin, thiobarbiturates, NAM, cambinol, GW5074, and Nε-carboxyethylthiourea-lysine, have all demonstrated *in vitro* inhibition of other sirtuins at comparable potencies (21, 30, 32, 62, 87, 90, 97, 109, 114). By contrast, a SIRT5 target covalent inhibitor linked to a cyclic pentapeptide inhibited SIRT5 *in vitro* deacetylase activity at a 50% inhibitory concentration (IC50) of 7.5 μM and SIRT1/2/3/6 at IC_{50} values between 200 and 1000 μM (52). Moreover, MC3482 demonstrated $\sim$40% inhibition of SIRT5 desuccinylase activity in MDA-MB-231 cells at a concentration of 50 μM, but was found to have no significant impact on SIRT1 or SIRT3 activities. The effects of MC3482 treatment on the remaining sirtuins have not been reported, and the mechanism for SIRT5 selectivity of MC3482 has yet to be defined (76).

Collectively, these studies have provided insights to support the development of new, more selective, and potent SIRT5 inhibitors. Current efforts to achieve SIRT5-selective molecules aim at targeting the substrate-binding pocket, which contains the SIRT5-specific residues Tyr102, Arg105, and Ala86. Large-scale screening methods are underway to identify highly selective compounds (110). Recently, a microdroplet-based approach has been applied to the identification of SIRT5 inhibitors; this technique allows the use of a longer, near-native peptide substrate derived from the SDHA protein sequence, enhancing throughput and potentially reducing the false discovery rate (27). SIRT5 inhibitors may eventually prove to be effective anticancer therapeutics, either as single agents or to sensitize cancer cells to established genotoxic therapies, although potential cardiac toxicity represents a significant concern given data from the *Sirt5* KO mouse model (85, 113).

Discussion

Although substantial literature now links many sirtuins to cancer, roles for SIRT5 in neoplasia have only begun to be addressed. There are several challenges to the study of SIRT5 in many contexts, including cancer. First, SIRT5 loss of function is associated with relatively mild phenotypes under basal conditions (53, 85, 113). Additionally, known functions of SIRT5 are pleiotropic and very context dependent (36, 45, 50, 56, 75, 77, 91, 104, 106). The fact that SIRT5 targets at least three distinct lysine modifications implies that this sirtuin likely exerts differing biological effects in different cell types, or even in the same cell type in differing physiological states, depending on the presence of these modifications. Finally, the effects of SIRT5 and its target modifications on hundreds of reported targets of SIRT5 remain to be defined (16, 42, 70, 73, 74, 80, 94). Still, a growing body of work has begun to elucidate the prosurvival and protumorigenic roles for SIRT5 in cancer cells, including promotion of metabolic reprogramming, antioxidant defense, and resistance to chemotherapy-induced cell death.

In the context of metabolic reprogramming, SIRT5 stimulates glycolysis and restricts TCA cycling and electron transport (57, 70, 73, 106, 108, 118). SIRT5 reduces cellular ROS by promoting NADPH production to support GSH regeneration, and by stimulating SOD1 antioxidant activity, thereby promoting cell survival (50, 106, 118). Conversely, specifically in the setting of IDH mutant cancer cells, SIRT5 appears to play a tumor suppressive role by reversing BCL-2 upregulation and mitigating epigenetic dysregulation in response to elevated 2-HG levels (45). Thus, SIRT5 likely impacts cancer biology in a context-specific manner, in some situations, promoting cancer cell survival and proliferation (51, 56, 57) and, in others, functioning to restrict cancer growth (45, 77). Such Janus-faced activity is similar to roles described for other sirtuins in malignancy (24).

Clearly, much remains to be learned about the biology of SIRT5. In this regard, in addition to direct substrates of SIRT5, several key cellular regulators have shown to be impacted by SIRT5 *via* mechanisms that remain undefined. In particular, expression levels of *Glut1, Ldha, Bcl-xl, Bcl-2, Sun2, Nrf2* (45, 50, 56, 106, 118), promitophagy genes (*Bnip3, Pink1,* and *Park2*), proautophagy genes (*Map1lc3b, Gabarap,* and *Gabarapl2*) (76), pro-MDR genes (*Gclc, Gclm, Ho-1, Gsr,* and *Gpx2*), and proxenobiotic genes (*Nqo-1, Txn, Txnrd1, Mrp1/2,* and *Atp7a*) (56) are all altered by manipulation of SIRT5 levels. Whether SIRT5 regulates expression of these genes *via* direct chromatin modification, or alternatively whether these effects represent indirect consequences of changes in mitochondrial and cellular physiology brought about by alterations in SIRT5 activity, represents an important open question in this area.

Hundreds of potential SIRT5 substrates have been identified, and likewise hundreds were suggested to associate with endogenous SIRT5 in immunoprecipitation/mass spectrometry studies (59). The biological significance of SIRT5 has been elucidated in the context of only an extremely small fraction of these targets and interactors. Among this handful of targets, it seems that no overall conclusions can be made at this point regarding the general activating or inhibitory effects of desuccinylation, demalonylation, or deglutarylation on SIRT5 targets. Similarly, given the current state of this field, it is challenging to correlate specific SIRT5 target modifications and the pro- or antitumorigenic functions of SIRT5 as a general matter. There is no doubt that further studies in this area will provide a wealth of insights into functions of SIRT5 and its targets in normal and neoplastic cells.

A key question revolves around potential cooperativity, redundancy, or antagonism of SIRT5 with other sirtuins, especially SIRT3 and SIRT4, and relationships between the Kac and target modifications of SIRT5, Ksucc, Kmal, and Kglu. In this regard, mitochondrial sirtuins share numerous substrates in common (60, 116). This is relevant since all three mitochondrial sirtuins depend on the presence of mitochondrial NAD^+ for activity, thus raising the general question of how the activities of sirtuins may be differentially regulated. The observation that sirtuins show differing affinities for NAD^+, and differing sensitivity to inhibition by NAM, may offer a partial answer to this question (19). Adding to the complexity of potential cross-talk among sirtuins, distinct lysine modifications may exert additive or even opposing effects on substrate function, especially since Kac is quite chemically distinct from major target modifications of SIRT5.

SIRT5 may represent an attractive pharmacological target for treatment of specific cancer types, with a potentially wide therapeutic window. In specific cancer types, including uterine, breast, cutaneous and uveal melanoma, lung, and lymphoma, the *SIRT5* gene frequently shows increased copy number; likewise, *SIRT5* mRNA and protein expression is elevated in multiple tumor types (Figs. 1 and 2). Development

of SIRT5 inhibitors suitable for *in vivo* studies represents a significant area of current investigation in many laboratories.

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Author Disclosure Statement

No competing financial interests exist.

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Abbreviations Used

1,3-BPG = 1,3-bisphosphoglycerate
293T = human epithelial kidney 293 SV40 large T antigen cell line
2-HG = (R)-2-hydroxyglutarate
A549 = human nonsmall cell lung adenocarcinoma cell line
Acon = aconitate
Ala86 = alanine 86
Arg = arginine
Arg105 = arginine 105
Arg-Suc = arginosuccinate
Asp = aspartate
BCL-2 = B cell lymphoma-2
BCL-XL = B cell lymphoma-XL
C2C12 = mouse myoblast cell line
CDDP = cisplatin
Cit = citrate
Citrul = citrulline
CoA = coenzyme A
CPS1 = carbamoyl phosphate synthetase 1
Cyt C = cytochrome C
ECHA = enoyl-coenzyme A hydratase
ETC = electron transport chain
FAO = fatty acid oxidation
FOXO3A = forkhead box O3A
Fum = fumarate
G-3-P = glucose-3-phosphate
G6P = glucose-6-phosphate
G6PD = glucose-6-phosphate dehydrogenase
GAPDH = glyceraldehyde phosphate dehydrogenase
GDH = glutamate dehydrogenase
Gln = glutamine
GLS = glutaminase
Glu = glutamate
GLUT1 = glucose transporter 1
GSH = glutathione
GSR = glutathione reductase
GSSG = oxidized glutathione
H92c = rat embryonic cardiomyocyte cell line
HADH = hydroxyacyl-coenzyme A dehydrogenase
IC_{50} = 50% inhibitory concentration
IDH = isocitrate dehydrogenase
IMM = inner mitochondrial membrane
IsoC = isocitrate
Kac = acetyllysine
KAT = lysine acetyltransferase
Kglu = glutaryllysine
Kmal = malonyllysine
KO = knockout
Ksucc = succinyllysine
L929 = mouse fibroblast cells
LDHA/B = lactase dehydrogenase A/B
Mal = malate
MC3482 = Sirt5 inhibitor
MDA-MB = 231 human metastatic breast cancer cell line
MDR = multidrug resistance
MEFs = mouse endothelial fibroblasts
MLS = mitochondrial localization sequence
MMP = mitochondrial membrane potential
MTF = mitochondrial trifunctional
NAD^+ = nicotinamide adenine dinucleotide
NADH = reduced nicotinamide adenine dinucleotide
NADPH = reduced form of $NADP^+$
NAM = nicotinamide
NDUFA4 = complex I subunit
NH_3/NH_4^+ = ammonia
NRF2 = nuclear factor erythroid-2-related factor 2
NSCLC = nonsmall cell lung cancer
OAA = oxaloacetate
Orn = ornithine
PC3 = human prostate cancer cell line
PDC = pyruvate dehydrogenase complex
PDCE1α = pyruvate dehydrogenase complex E1α subunit
PEP = phosphoenolpyruvate
PKM2 = pyruvate kinase muscle isozyme M2
PPP = pentose phosphate pathway
Q = quinolone
R-5-P = ribose 5 phosphate
R5PI = ribulose 5 phosphate isomerase
ROS = reactive oxygen species
SDHA = succinate dehydrogenase subunit A
SIRT1–7 = seven mammalian sirtuin proteins
SIRT5 = sirtuin 5
$SIRT5^{iso1/iso2}$ = SIRT5 isoform 1/2
Sirtuins = silent information regulator 2
SOD = superoxide dismutase
STAT3 = signal transducer and activator of transcription
Suc = succinate
Suc-CoA = succinyl coenzyme A
SUN2 = SUN domain-containing protein 2
TCA = tricarboxylic acid cycle
Tyr102 = tyrosine 102
VLCAD = very long chain acyl-CoA dehydrogenase
VLCFA = very long fatty acyl-CoA
WT = wild-type
α-KG = alpha ketoglutarate