ANNUAL REVIEWS

Annual Review of Cancer Biology

Cancer Dependencies: PRMT5 and MAT2A in MTAP/p16-Deleted Cancers

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Annu. Rev. Cancer Biol. 2021. 5:371–90

First published as a Review in Advance on December 22, 2020

The *Annual Review of Cancer Biology* is online at cancerbio.annualreviews.org

https://doi.org/10.1146/annurev-cancerbio-030419-033444

Keywords

MTAP, p16, PRMT5, MAT2A, synthetic lethal

Abstract

Discovery of targeted therapies that selectively exploit the genetic inactivation of specific tumor suppressors remains a major challenge. This includes the prevalent deletion of the *CDKN2A/MTAP* locus, which was first reported nearly 40 years ago. The more recent advent of RNA interference and functional genomic screening technologies led to the identification of hidden collateral lethalities occurring with passenger deletions of *MTAP* in cancer cells. In particular, small-molecule inhibition of the type II arginine methyltransferase PRMT5 and the *S*-adenosylmethionine-producing enzyme MAT2A each presents a precision medicine approach for the treatment of patients whose tumors have homozygous loss of *MTAP*. In this review, we highlight key aspects of MTAP, PRMT5, and MAT2A biology to provide a conceptual framework for developing novel therapeutic strategies in tumors with *MTAP* deletion and to summarize ongoing efforts to drug PRMT5 and MAT2A.

MTAP: PASSENGER CODELETION WITH BENEFITS?

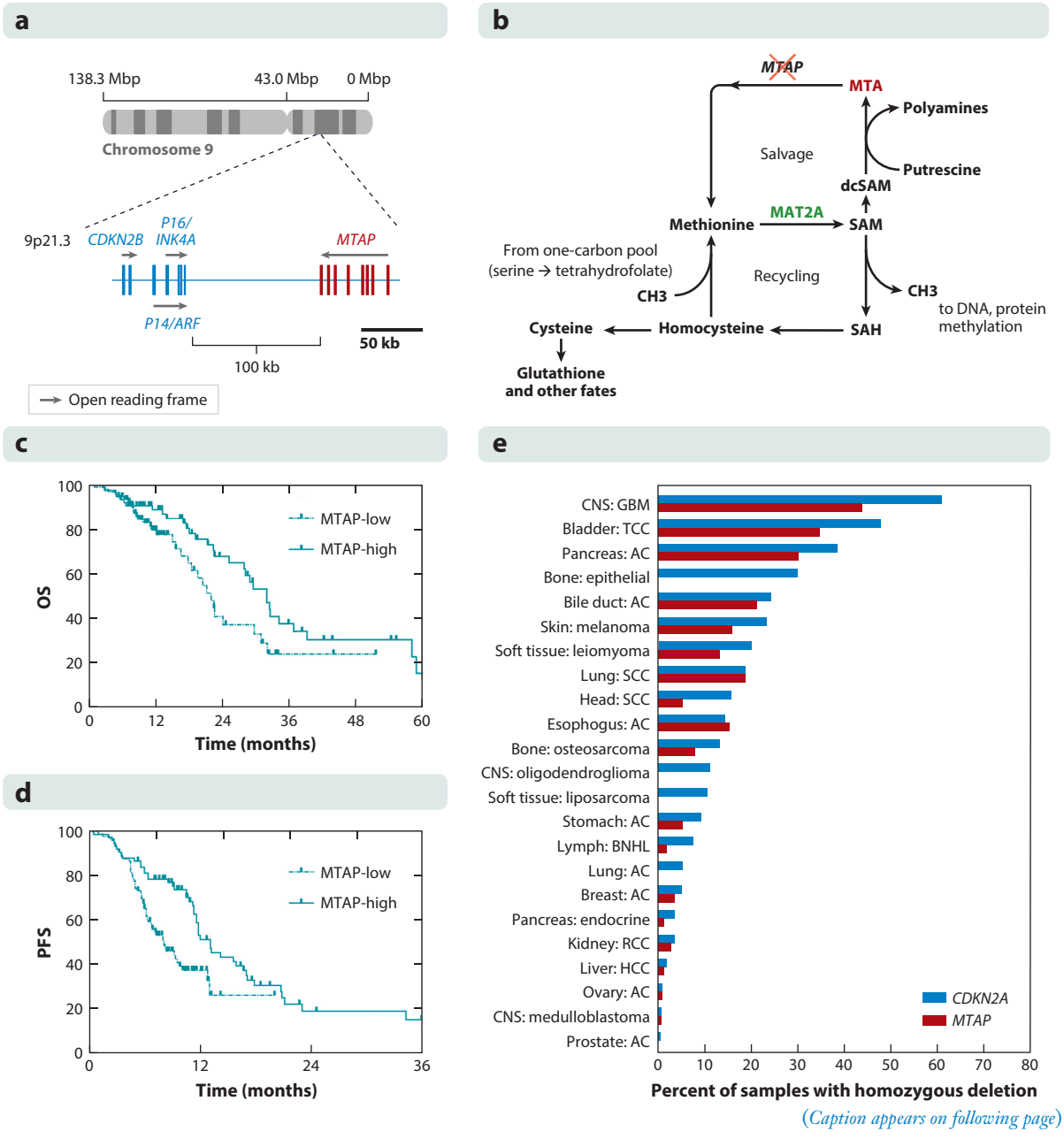
Targeted therapies that block the activity of amplified or gain-of-function mutated oncogenes have become critical components of cancer therapy (Bollag et al. 2010, Hochhaus et al. 2017, Shaw et al. 2014, Slamon et al. 2001, Zhang et al. 2012), but there remains a lack of therapies that selectively exploit loss-of-function mutations or deletions in tumor suppressors. High-resolution analyses of somatic copy number alterations (Beroukhim et al. 2010) and recent pan-cancer analysis of whole genomes (PCAWG Consort. 2020) have identified deletion of the 9p21.3 genomic locus, which encompasses several genes including the tumor suppressor cyclin-dependent kinase inhibitor 2A/B (*CDKN2A/B*), as one of the most prevalent biallelic somatic copy number alterations in a wide variety of human cancers, with approximately 15% of all cancers impacted. Moreover, analysis of somatic evolution of mutational processes in oncogenesis has demonstrated that 9p21 deletions appear to be an early and clonal event in several tumor types (Gerstung et al. 2020). This suggests a lower risk for tumor heterogeneity and thus an increased potential for efficacious targeted therapies for patients with this genetic alteration.

The 5′-deoxy-5′-methylthioadenosine phosphorylase (*MTAP*) gene, which resides within 100 kb of the *CDKN2A* tumor suppressor on chromosome 9p21, is homozygously codeleted in 80–90% of cancer cell lines with *CDKN2A* deletion (Zhang et al. 1996) (**Figure 1**). Although cases have been described where MTAP loss was detected in the absence of CDKN2A deletion in primary non-small-cell lung cancer (NSCLC) samples and cell lines (Schmid et al. 1998), in the recent pan-cancer studies (PCAWG Consort. 2020), *MTAP* loss in the absence of concordant loss of *CDKN2A* appears to be a rare event, suggesting that *CDKN2A* is the primary deletion target on the 9p21 region that is selected for in human cancers.

MTAP is a critical enzyme in the methionine (Met) salvage pathway that cleaves the byproduct of polyamine synthesis, 5′-methylthioadenosine (MTA), to adenine and MTR-1-P (5-methythioribose-1-phosphate), leading to the eventual regeneration of Met (Zappia et al. 1988) (**Figure 1*b***). Several highly potent cell-permeable transition-state analog inhibitors of MTAP have been identified and tested (Basu et al. 2007, 2011; Longshaw et al. 2010; Singh et al. 2004), providing experimental tools that recapitulate genetic loss-of-function phenotypes (Marjon et al. 2016). Because MTAP is the only enzyme in mammalian cells known to catalyze the degradation of MTA, MTAP loss leads to accumulation of MTA in cancer cells (Kryukov et al. 2016, Marjon et al. 2016, Mavrakis et al. 2016). MTA in turn has been reported to behave as a competitive inhibitor of methyltransferase enzymes (Enouf et al. 1979, Williams-Ashman et al. 1982), which may explain some of the biological consequences of MTAP genetic loss discussed below. However, recent work suggests that MTA accumulation in primary human glioblastoma in vivo may be moderate compared to tissue culture conditions, likely due to the metabolism of MTA by MTAP-competent stromal cells (Barekatain et al. 2019). Further work is required to more fully characterize the impact of MTAP loss on MTA levels in a variety of tumor types.

It remains incompletely understood whether *MTAP* is simply a passenger codeletion or has unique tumor-suppressor functions. This later notion is supported by work in the MCF7 breast adenocarcinoma cell line demonstrating inhibition of anchorage-independent growth in vitro and tumor growth in vivo upon reconstitution of wild-type MTAP into this *MTAP*-deleted model (Christopher et al. 2002). Critically, although homozygous deletion of *MTAP* is embryonic lethal, mice heterozygous for a germline deletion of *MTAP* survive to birth but succumb to early death due to the onset of T cell lymphoma (Kadariya et al. 2009). Further work has demonstrated that when mice heterozygous for germline loss of MTAP are crossed with the Eμ-Myc mouse model, this results in the accelerated onset of B cell lymphoma (Kadariya et al. 2013). Finally, convincing evidence for a tumor-suppressive role of MTAP is derived from its role in diaphyseal medullary

stenosis with malignant fibrous histiocytoma, a syndrome associated with bone dysplasia and cancer, where germline mutations occur in *MTAP* and result in exon skipping and dysregulated alternative splicing of MTAP isoforms, some of which lack enzymatic activity (Camacho-Vanegas et al. 2012). In addition to its enzymatic function, MTAP has been proposed to have a nonenzymatic role in suppressing tumor-related phenotypes in the HT1080 fibrosarcoma model, as its



(*Caption appears on following page*)

Figure 1 (*Figure appears on preceding page*)

Cell biology of *MTAP*, a genomic neighbor of *CDKN2A* frequently lost in human cancers that has a key role in MTA catabolism. (*a*) Chromosome 9 and the 9p21.3 region containing the *MTAP* gene close to the *CDKN2A* gene and its two coding sequences, *P16/INK4A* and *P14/ARF*. (*b*) The methionine recycling and salvage pathways highlighting MTAP and MAT2A. The red X indicates deletion of the *MTAP* gene and corresponding loss of MTAP protein, the green text denotes the metabolic enzyme MAT2A, and the red text highlights MTA, which accumulates upon *MTAP* deletion. Panels *a*,*b* adapted with permission from Marjon et al. (2016); copyright 2016 Elsevier. (*c*) OS curve of advanced lung AC patients stratified by *MTAP* expression. The median OS was 22 months in the MTAP-low group versus 32 months in the MTAP-high group ($p = 0.044$). (*d*) PFS curve of advanced lung AC patients stratified by *MTAP* expression. The median PFS in the MTAP-low group was 8.1 months, compared to 13.1 months in the MTAP-high group ($p = 0.002$). Panels *c*,*d* adapted from Jing et al. (2020); CC-BY 4.0. (*e*) Frequency of *CDKN2A* and *MTAP* homozygous deletions according to pan-cancer whole genome sequencing analysis. Panel *e* data are from PCAWG Consort. (2020). Abbreviations: AC, adenocarcinoma; BNHL, B cell non-Hodgkin lymphoma; CNS, central nervous system; dcSAM, decarboxylated SAM; GBM, glioblastoma multiforme; HCC, hepatocellular carcinoma; MTA, 5′-methylthioadenosine; OS, overall survival; PFS, progression-free survival; RCC, renal cell carcinoma; SAH, *S*-adenosylhomocysteine; SAM, *S*-adenosylmethionine; SCC, squamous cell carcinoma; TCC, transitional cell carcinoma.

catalytic-dead mutant D220A was capable of reversing soft agar colony formation, cell migration in a wound-healing assay, and metalloproteinase activity (Tang et al. 2014).

Regardless of whether MTAP is a tumor suppressor, low *MTAP* expression as analyzed by immunohistochemistry has been linked to poor prognosis (shorter overall survival and disease progression-free survival), tumor recurrence, and a higher risk of metastatic disease in several histological tumor types (Hansen et al. 2019, He et al. 2015, Jing et al. 2020, Roy et al. 2016, Su et al. 2014, Xu et al. 2019) (**Figure 1*c*,*d***). Furthermore, according to multivariate survival analysis, it is an independent prognostic factor with a greater significance than CDKN2A (Hansen et al. 2019, Su et al. 2014).

Despite the prevalence of this genetic lesion and the initial discovery of *MTAP* deletions in human cancer nearly 40 years ago (Kamatani et al. 1981, 1982), very few approaches have been described that selectively target *CDKN2A/MTAP*-deleted tumors. Inhibitors of CDK4/CDK6 have been reported to selectively block the growth of *CDKN2A*-deleted cells in vitro (Cen et al. 2012, Young et al. 2014), but this therapeutic approach has not yielded efficacy in clinical studies to date.

A metabolic strategy to drug *MTAP*-deleted cancers has been previously proposed (Bertino et al. 2011, Lubin & Lubin 2009). These earlier efforts focused on the hypothesis that purine biosynthesis is a synthetic lethal vulnerability in *MTAP*-deleted cancers. The cornerstone of this hypothesis is the lack of the adenine salvage in *MTAP*-deficient tumors, which allows for more efficient conversion of chemotherapeutic purine/pyrimidine (uracil) analogs to toxic nucleotides via the activity of APRT (adenine phosphoribosyltransferase) in the absence of competition for the cellular PRPP (phosphoribosyl-1-pyrophosphate) pool by adenine. In addition, *MTAP*-null cancers were proposed to be more sensitive to inhibitors of de novo purine biosynthesis, such as methotrexate and L-alanosine (Chen et al. 1997, Hori et al. 1996), once again due to lack of MTAP and adenine salvage. This later notion was not supported by subsequent work (Ruefli-Brasse et al. 2011). L-alanosine was even tested in the clinic and, when dosed up to its maximum tolerated dose, did not produce significant clinical benefit in patients with *MTAP*-deficient tumors (Kindler et al. 2009). MTA coadministration in fact did provide a selective growth advantage to MTAP-proficient cells, as shown using *MTAP* isogenic in vitro cell line models (Ruefli-Brasse et al. 2011). However, this later approach did not translate to the in vivo setting, as the growth of *MTAP*-null T cell acute lymphoblastic leukemia CEM cells was rescued by MTA in vivo (Ruefli-Brasse et al. 2011). The rescue was attributed to ample adenine salvage pathway activity in normal stromal cells.

Elaboration of MTAP-selective anticancer targets remained an intriguing and unsolved scientific question (Muller et al. 2015) until recent advances in functional genomic approaches,

combined with a deeper understanding of the unique metabolic environment of *MTAP*-deleted cancers and its impact on biochemical activity of a specific family of methyltransferases, officially put this genetic lesion on the map as a classic and therapeutically promising collateral vulnerability.

IDENTIFICATION OF NOVEL SYNTHETIC LETHAL VULNERABILITIES IN *MTAP*-DELETED CANCERS IN THE ERA OF FUNCTIONAL GENOMICS AND CRISPR

The discovery and implementation of RNA interference (Boutros & Ahringer 2008, Elbashir et al. 2001) and CRISPR (clustered regularly interspaced short palindromic repeats)-Cas9 (Cong et al. 2013; Jinek et al. 2012, 2013; Mali et al. 2013) have opened a new chapter in oncology drug target discovery, as they enable molecular barcode–based pooled depletion screening to identify essential genes in cancer cells. Three major large-scale studies, Project DRIVE, Project Achilles, and Project Score, have been performed using these approaches to identify essential genes from nearly genome-wide libraries across a large number of human cancer cell lines with broad histologic tumor origins and variable genetic spectra (Behan et al. 2019, McDonald et al. 2017, Tsherniak et al. 2017). In addition to identifying genes that are broadly required across all (or most) cancer cell lines, bioinformatic analyses of these large-scale data sets have identified selective vulnerabilities—genes that are essential only in molecularly defined subsets of cancers.

Project DRIVE and Project Achilles simultaneously identified for the first time PRMT5 (protein arginine methyltransferase 5), a type II arginine methyltransferase that generates symmetric arginine dimethylation marks on proteins, as a synthetic lethal vulnerability in cells with *MTAP* deletions (Kryukov et al. 2016, Mavrakis et al. 2016). Importantly, a similar observation was made using a short-hairpin RNA (shRNA) depletion screen in an HCT116 *MTAP* isogenic cell line pair, further strengthening confidence that PRMT5 and MTAP have a synthetic lethal interaction (Marjon et al. 2016). Not only PRMT5 but also its binding partners, such as the PRMT5 regulator WDR77/MEP50, the methylosome subunit pICIn, and RIOK1 (RIO kinase 1), all scored as differentially required for the growth of *MTAP*-deficient cancer cell lines in these studies.

The findings that PRMT5 is a synthetic lethal vulnerability with *MTAP* loss and that MTA levels are characteristically elevated in *MTAP*-deficient cancer cell lines have prompted further biochemical characterization of the sensitivity of PRMT5 to inhibition by MTA as a possible reason behind this newly uncovered vulnerability. In fact, Novartis, the Broad Institute, and Agios Pharmaceuticals have all reported on the uniquely high sensitivity of PRMT5 to inhibition by MTA in biochemical assays in vitro (Kryukov et al. 2016, Marjon et al. 2016, Mavrakis et al. 2016). Mavrakis et al. (2016) reported a crystal structure of MTA-bound PRMT5:WDR77 in complex with a H4 peptide, which potentially provides further explanation. Upon MTA binding, a conserved Glu^{435} in a PRMT5 substrate binding pocket was observed to shift from coordinating Arg residues of the peptide substrate to instead forming a split hydrogen bond with Lys^{333} and a salt bridge with Tyr^{334} residues that are unique to PRMT5.

The interaction between MTAP and the *S*-adenosylmethionine (SAM)-producing enzyme MAT2A was another synthetic lethal interaction that emerged from these functional genomic studies (Marjon et al. 2016, Mavrakis et al. 2016). In fact, in our work in the HCT116 *MTAP* isogenic pair, MAT2A was identified as the top hit. The MAT2A and MTAP synthetic lethality was further validated by doxycycline-inducible shRNA-mediated knockdown of MAT2A and shRNA-resistant complementary DNA rescue experiments in HCT116 $MTAP^{-/-}$ cells and in additional endogenous *MTAP*-deleted models in vitro and in vivo (Marjon et al. 2016). To explain this synthetic lethality, we measured changes in symmetric arginine dimethylation marks [symmetric dimethylarginine (SDMA)] in the HCT116 isogenic pair upon MAT2A knockdown and

observed a significant reduction in SDMA signal upon MAT2A knockdown in *MTAP*$^{-/-}$ cells but not *MTAP* wild-type cells (Marjon et al. 2016). Critically, MTAP pharmacologic inhibition synergized with MAT2A knockdown to reduce SDMA marks in parental HCT116 cells (Marjon et al. 2016). To explain this further, we compared SAM Michaelis-Menten constant (K_M) values for the *N*-methyltransferases using an in vitro biochemical panel analysis and observed that PRMT5 exhibited the lowest affinity for SAM (Marjon et al. 2016). This feature of PRMT5, especially in the context of the metabolically altered, high-MTA environment of *MTAP*-deficient cells, results in a second hit on PRMT5 activity upon reduction in intracellular SAM levels due to modulation of MAT2A and provides a mechanistic basis for MAT2A synthetic lethality in *MTAP*-deficient cancers (Marjon et al. 2016). Thus, recent functional genomics work has revealed two potential precision medicine approaches for treatment of *MTAP*-deficient cancers: via direct targeting of PRMT5 or via indirect modulation of PRMT5 activity by targeting MAT2A.

PRMT5 BIOLOGY AND ITS ROLES IN CANCER

Arginine methylation is a highly coordinated multistep process mediated by nine members of the PRMT family in mammalian cells. Each PRMT recognizes the side chain of arginine residues characterized by two positively charged guanidino groups that can be subject to methylation. Methylation reduces the number of arginine hydrogen bond donors, which alters protein-protein, protein-RNA, and protein-DNA interactions. Methylation reactions are catalyzed by PRMTs through the transfer of a methyl group from SAM to a guanidine nitrogen of arginine, resulting in the production of ω-NG-MMA (monomethylated arginine) and SAH (*S*-adenosylhomocysteine) (Di Lorenzo & Bedford 2011, Radzisheuskaya et al. 2019). Additionally, MMA can undergo a secondary methylation by PRMTs to produce either ω-N^G,N^G-dimethyl-L-arginine [asymmetric dimethylarginine (ADMA)] or ω-N^G,N'^G-dimethyl-L-arginine (SDMA). Based on the methylation generated, PRMTs can be divided into three major groups (Bedford & Clarke 2009): type I (PRMT1, 2, 3, 4, 6 and 8), which catalyzes the formation of MMA and ADMA; type II (PRMT5 and PRMT9), which catalyzes the formation of MMA and SDMA; and type III (PRMT7), which is limited to the production of MMA only (Bedford & Clarke 2009, Feng et al. 2013, Stopa et al. 2015).

PRMT5 is the primary type II arginine methyltransferase responsible for most of the symmetric arginine methylation activity within the cell (Wang et al. 2018). It was initially identified in a yeast two-hybrid screen as JBP1 (Jak-binding protein 1), a protein that possesses methyltransferase activity toward nuclear histone proteins (Fabbrizio et al. 2002). Particularly, PRMT5 mediates the symmetric methylation of arginines in histone H2A (H2AR3me2s), H3 (H3R2me2s, H3R8me2s), and H4 (H4R3me2s), which are associated with transcriptional regulation of gene expression (Ancelin et al. 2006, Girardot et al. 2014, Migliori et al. 2012, Pal et al. 2004, Tee et al. 2010) (**Figure 2*a***). For example, PRMT5 methylation of H4R3 at the transcription start site of the cyclin E1 promoter is found to suppress cyclin E transcription and substantially decrease cell proliferation (Fabbrizio et al. 2002). In a separate study, microarray analysis has revealed that *PRMT5* expression stimulates cell proliferation and anchorage-independent growth through the repression of 227 tumor-suppressor and cell cycle–regulatory genes. Moreover, ectopic expression of *PRMT5* led to an increase of histone H3R8 methylation, which correlated with decreased expression of *AT7* and *NM23* tumor-suppressor genes and induced cell transformation. In addition, PRMT5 was found to methylate histone H3R2 (H3R2me2s), which is enriched in actively transcribed euchromatin regions of the nucleus (Migliori et al. 2012). Multiple studies have demonstrated that histone H4R3me2s is specifically recognized by DNA methyltransferase DNMT3A, which is recruited through its ADD (ATRX-DNMT3-DNMT3L) domain and

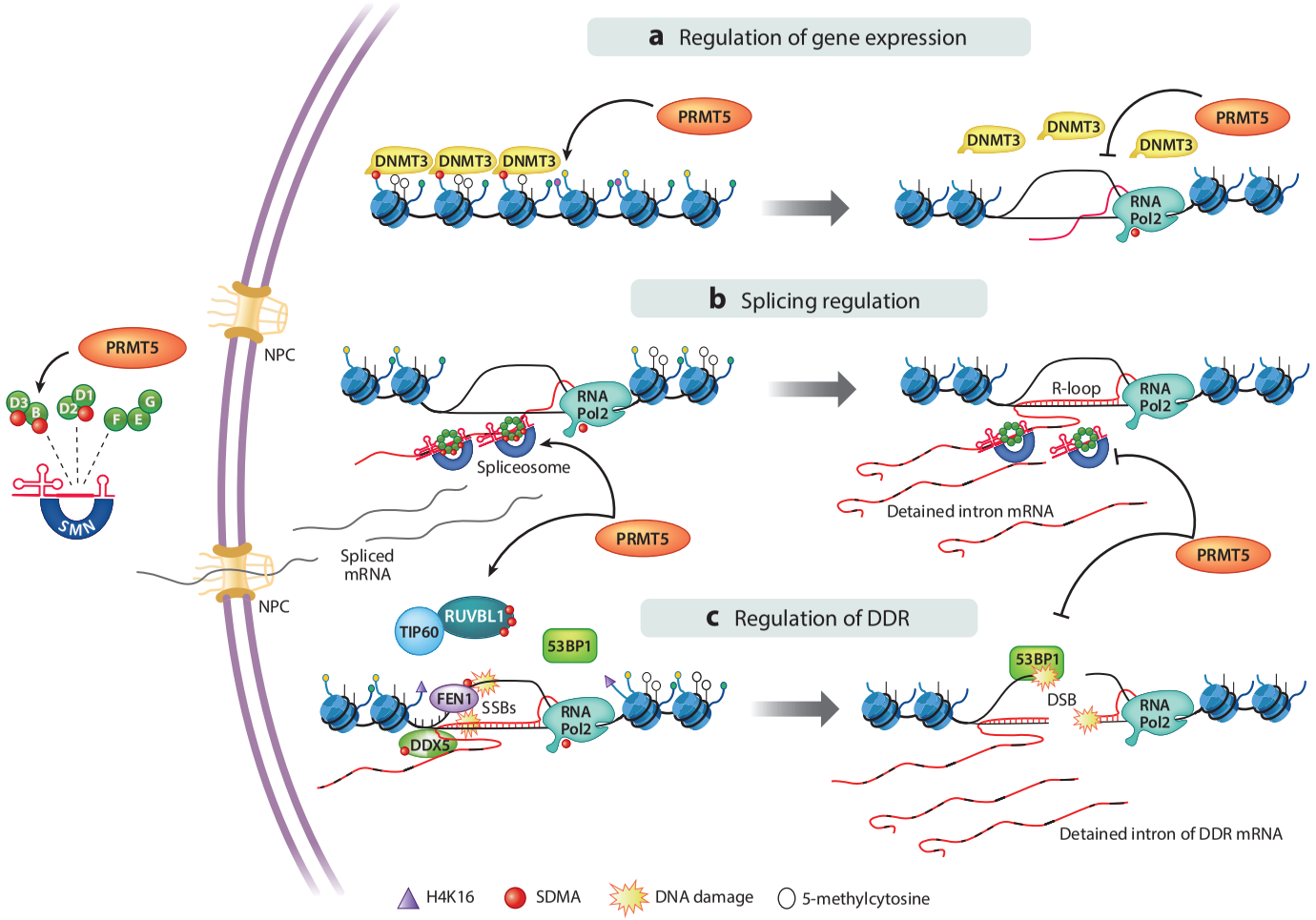


Figure 2

A model representing the role of PRMT5 in the regulation of gene expression, splicing, and DDR. (*a*) PRMT5-mediated methylation of histone H4 promotes the recruitment of DNMT3 to G/C-rich gene-promotor regions. Loss of H4R3me2s signal impedes the chromatin binding of DNMT3 and increases gene expression. (*b*) PRMT5 function is critical for the assembly of spliceosomal machinery. Loss of PRMT5 function induces aberrant splicing, formation of R-loops, and increased detained intron–containing mRNAs. (*c*) SDMA methylation of multiple DDR and transcription factors, including RUVBL1, FEN1, DDX5, and RNA Pol2, is essential for the maintenance of genome integrity. Inhibition of PRMT5 function decreases the efficiency of DNA repair pathways and leads to increased DNA damage. Abbreviations: DDR, DNA damage response; DSB, double-strand DNA break; NPC, nuclear pore complex; SDMA, symmetric dimethylarginine; SMN, survival of motor neuron protein; SSB, single-strand DNA break.

regulates gene suppression (Ancelin et al. 2006). Loss of H4R3me2s at gene promotors impedes chromatin binding of DNMT3A, leading to decreased DNA methylation and subsequent activation of target genes (Zhao et al. 2009).

In addition to histone methylation, PRMT5 regulates gene expression through methylation of transcription factors and modulation of cell growth signaling cascades (Zhu & Rui 2019). Arginine methylation modulates the transcriptional activity and DNA-binding affinity of E2F-1, which is essential for cell proliferation and apoptosis (Cho et al. 2012, Zheng et al. 2013). PRMT5 methylation of epidermal growth factor receptor (EGFR) at R1175 positively modulates EGF-induced phosphorylation of EGFR at tyrosine Y1175 in breast cancer cells (Hsu et al. 2011). Loss of EGFR R1175me2s enhanced EGF-stimulated activation of ERK signaling by reducing the ability of DHP1 to bind EGFR, leading to decreased proliferation and cell migration. Multiple studies have demonstrated that loss of PRMT5 activity induces cellular stress, which leads to activation of the p53 tumor-suppressor gene. In an attempt to assess whether PRMT5 can directly modulate p53 activity, Jansson et al. (2008) studied the interaction of PRMT5 and p53 in control untreated conditions and upon genotoxic stress. The results of this study demonstrated that PRMT5 can directly bind and methylate p53 at R333, R335, and R337 residues. Moreover, expression of a three-lysine p53 mutant (p53KKK) demonstrated reduced nuclear localization and failed to induce G1 cell cycle arrest. These data suggest that PRMT5-mediated methylation of p53 can directly influence its biochemical properties and thus regulate the transcriptional outcome of the p53 response.

While PRMT5 methylates a variety of cellular substrates, it has become clear that PRMT5-mediated methylation of RNA splicing machinery represents a particularly critical function of PRMT5 (**Figure 2*b***). PRMT5-mediated SDMA methyl marks are a physiological ligand for Tudor domain–containing proteins such as SMN (survival of motor neurons), which plays a critical role in the biogenesis of spliceosomal small nuclear (sn) ribonucleoproteins (RNPs) and the spliceosome. Spliceosomal RNPs are the key components of splicing machinery composed of one or two small RNAs (U1, U2, U4, U5, U6) and multiple RNA-binding proteins, including Arg/Gly-rich Sm proteins (SmB/B′, SmD1, SmD2, SmD3, SmE, and SmF), which are assembled in a timely, coordinated manner (Friesen et al. 2001, 2002). During spliceosomal assembly, the SMN protein forms a large complex through interactions with a snRNA and the symmetrically dimethylated tails of Sm proteins. Although Sm proteins can self-assemble in vitro on any RNA with a short Sm site sequence, in vivo experiments have demonstrated that Sm cores assemble on the proper snRNA only through the interaction with the Tudor domain of the SMN protein in an ATP-dependent manner. Moreover, PRMT5-mediated methylation of SmD1, SmD3, and SmB-B′ enhances their transfer onto the SMN complex and facilitates the assembly of newly produced snRNPs (Brahms et al. 2000, Meister et al. 2001). Loss of PRMT5 function leads to aberrant splicing and formation of transcripts with detained introns and skipped exons (Braun et al. 2017). These alternative splicing events decrease the total pool of fully spliced mRNA for multiple genes and decrease the abundance of their corresponding proteins.

Accumulating evidence in the literature demonstrates that PRMT5-mediated methylation of replication and DNA damage response (DDR) factors is critical for maintenance of genomic integrity (Clarke et al. 2017, Hamard et al. 2018, Owens et al. 2020) (**Figure 2*c***). In particular, methylation of *POLR2A* at R1810 is essential for the recruitment of the SMN complex to RNAP II transcription sites and stimulates the interaction with DNA-RNA helicase senataxin (Zhao et al. 2016). Loss of PRMT5 induces premature termination of transcription, which is associated with accumulation of RNA-DNA structures called R-loops (Zhao et al. 2016). Accumulation of unresolved R-loops is often accompanied by formation of single-strand DNA breaks and double-strand DNA breaks (DSB) (Skourti-Stathaki & Proudfoot 2014). Moreover, Mersaoui et al. (2019) recently demonstrated the importance of PRMT5-mediated methylation of DDX5 helicase in

resolution of R-loops. DDX5 methylation promotes the recruitment of XRN2 to transcription termination paused sites of multiple transcriptionally active genes. Loss of DDX5 results in accumulation of R-loops at transcription termination sites, resulting in an increase of DSBs. Another regulator of R-loop formation is FEN1 (Flap endonuclease1) (Cristini et al. 2019). PRMT5-mediated methylation of FEN1 at R192 promotes the binding to DNA clamp PCNA (proliferating cell nuclear antigen) (Guo et al. 2010). Mutations that disrupt *FEN1* methylation impair its ability to localize at the sites of DNA replication or DNA damage and lead to a high frequency of genome-wide mutations. PRMT5 has also been reported to symmetrically methylate the AAA+ ATPase RUVBL1 at R205 and promote TIP60-mediated histone H4K16 acetylation (Clarke et al. 2017). Loss of RUVBL1 R205me2s induces increased retention of 53BP1 at the sites of DNA damage and inhibition of homologous recombination repair (HRR).

PRMT5 is also known to impact the efficiency of DDR by regulating the splicing of multiple DDR-regulating genes (Hamard et al. 2018, Zhu & Rui 2019). Specifically, PRMT5 was reported to prevent DDR genes from exon skipping and intron retention in hematopoietic stem cells (Tan et al. 2019). Depletion of PRMT5 induced increased intron retention and decreased expression of *FANCA*, *FANCG*, *RTEL1*, and *RAD51* genes involved in regulation of interstrand cross-link DNA repair, leading to accumulation of reactive oxygen species (ROS). Loss of PRMT5 induced aberrant splicing of *TIP60* and *KMT5C*, leading to stabilization of 53BP1. Recent studies by Owens et al. demonstrated that PRMT5 cooperates with plCln to activate a subset of DNA repair genes that regulate both HRR and NHEJ (nonhomologous end joining) independently of MEP50 (Owens et al. 2020). Loss of either PRMT5 or plCln hindered DNA repair and decreased the expression of multiple genes involved in DDR, including *RAD51*, *RAD51AP1*, *RAD51D*, *BRCA1*, *BRCA2*, *NHEJ1*, and *PRKDC*. In addition, Bezzi et al. (2013) demonstrated that loss of PRMT5 function in a mouse knockout model induces aberrant splicing and skipping of exon-6 in the p53 negative regulator *MDM4*. The authors showed that exon-6 skipping leads to loss of the p53-interacting domain in MDM4, which stabilizes *TP53* expression. Taken together, all these results indicate that PRMT5 is an integral regulator of gene expression and coordinates DDR to maintain the genomic integrity of tumor cells.

MAT2A AND ITS ROLE IN CANCER

Methionine adenosyltransferase (MAT), EC (Enzyme Commission) number 2.5.1.6, is the enzyme principally responsible for the production of SAM from ATP and L-Met. SAM is the universal biological donor of methyl groups for all intracellular transmethylation reactions. SAM can also donate its amino-propyl group during polyamine biosynthesis and its sulfur for glutathione biosynthesis via cysteine, and it participates in 5′-deoxyadenosyl 5′-radical-mediated biochemical transformations (Lu & Mato 2012) (**Figure 1*b***).

There are three protein isoforms of the MAT catalytic alpha subunit (MATI, MATII, and MATIII) encoded by two distinct genes (*MAT1A* and *MAT2A*) in mammals (Kotb et al. 1997). *MAT2A* encodes the MATII (referred to as MAT2A in this review) catalytic homodimer, a ubiquitously expressed enzyme that generates SAM in all normal and cancer cells, while *MAT1A* encodes two liver-specific protein isoforms, MATI (a dimer of MAT1A catalytic subunits) and MATIII (a tetramer of MAT1A catalytic subunits). MAT1A and MAT2A catalytic subunits are highly homologous and share 85% sequence similarity (Murray et al. 2016). MAT2B, an accessory protein encoded by the *MAT2B* gene, interacts with MAT2A and is a negative regulator of MAT2A catalytic activity (Halim et al. 1999, Kotb & Kredich 1990, Kotb et al. 1997, LeGros et al. 1997). MAT enzymes demonstrate differences in their kinetic properties and in their affinity for the Met substrate [K_M for Met for is ∼1.8 mM for MAT1A and ∼19 μM for MAT2A (Agios

Pharmaceuticals, unpublished data)]. Both isoforms are subject to MAT2B-dependent inhibition by its product SAM, with MAT2A demonstrating stronger sensitivity to SAM inhibition (Halim et al. 1999, LeGros et al. 1997, Quinlan et al. 2017, Sullivan & Hoffman 1983).

The multiple fates of SAM in cellular metabolism and its impact on critical biological processes, such as methylation- and polyamine-regulated epigenetic and transcriptional regulation of gene expression, regulation of translation via methylation of guanosine mRNA caps (Cowling 2009) and eIF5A hypusination (Pegg 2016), and glutathione-dependent ROS buffering, suggest that MAT2A represents a key regulatory node capable of affecting growth, proliferation, differentiation, and viability of normal and transformed cells.

In fact, dysregulated expression of hepatic MAT enzymes has been linked with malignant transformation in the liver, with MAT1A being downregulated during tumorigenesis while MAT2A becomes the predominant isoform (Cai et al. 1996, 1998; Frau et al. 2013). The role of MAT isoform switch in liver tumorigenesis is further supported by *MAT1A*-knockout mouse model studies where upregulation of MAT2A has been observed in the liver of knockout animals, leading to the development of liver steatosis and eventually hepatocellular carcinoma (Lu et al. 2001, Martinez-Chantar et al. 2002). The downregulation of MAT1 was proposed to have a critical impact on the key aspects of its downstream biology such as induction of oxidative stress, disruption of lipid homeostasis, induction of genomic instability, dysregulation of ERK and LKB1/AMPK signaling, and expansion of progenitor cells, while upregulation of MAT2A is thought to confer cancer cell proliferation and survival advantages (for reviews, see Frau et al. 2013, Lu & Mato 2012). In addition, MAT enzymes have also been proposed to play a role in cancer biology via impact on the levels of polyamine metabolites produced using SAM and via effects on downstream polyamine-regulated biology (for additional information, see Casero et al. 2018, Hesterberg et al. 2018).

Although the role of MAT is most well studied in liver cancer, there is continuously emerging evidence of MAT2A or MAT2B dysregulation in other cancers, including T cell leukemia (Jani et al. 2009) and colon carcinoma (Chen et al. 2007, Wang et al. 2016), as well as gastric (T. Zhang et al. 2013), breast (Phuong et al. 2016), pancreatic, and prostate cancer (Tomasi et al. 2017) (reviewed in Maldonado et al. 2018). Furthermore, two recent studies have suggested a role for SAM biosynthesis in cancer stem cell biology. Wang et al. (2019) observed that TIC (tumor-initiating cell)-enriched lines derived from resected primary NSCLC adenocarcinoma samples and grown as nonadherent tumor spheres are addicted to exogenous Met and demonstrate dependency on MAT2A activity for growth in vitro and in vivo. Similar observations were made by Strekalova et al. (2019) when evaluating effects of Met restriction and MAT2A suppression in human triple-negative breast carcinoma (TNBC) cell lines that were cultured as cancer stem cell–enriched mammospheres and in murine models of metastatic TNBC.

CURRENT EFFORTS TO DRUG PRMT5 AND MAT2A

Likely inspired by the findings described above, several academic institutions and pharmaceutical and biotechnology companies are currently developing PRMT5 inhibitors, including four that have entered clinical development (**https://clinicaltrials.gov/** identifiers NCT02783300, NCT03573310, NCT03854227, NCT03886831) (see **Table 1**).

The first highly potent, orally bioavailable PRMT5 inhibitor to be developed was EPZ015666 (Chan-Penebre et al. 2015). EPZ015666 inhibits PRMT5 in a SAM-uncompetitive and peptide substrate–competitive manner, with IC_{50} (half-maximal inhibitory concentration) values in the low nanomolar range (22 ± 14 nM) in biochemical assays. It selectively inhibited PRMT5 and showed no activity at concentrations up to 50 μM against 20 other protein methyltransferases. In addition,

Table 1 Summary of the current preclinical and clinical stage inhibitors of MAT2A and PRMT5

Inhibitor	Target	Origin/developer	IC_{50} (nM)	Stage of development
AG-270	MAT2A	Agios	10 nM[a]	Clinical stage (NCT03435250)
IDB-361	MAT2A	Ideaya	3.5 nM[b]	Preclinical development
PF-9366	MAT2A	Pfizer, Quinlan et al. 2017	420 nM[a]	Preclinical development
Cycloleucine	MAT2A	Lombardini & Sufrin 1983	140–190 μM[a]	Academic study
Stilbene derivatives	MAT2A	W. Zhang et al. 2013	2.1–4.9 μM[a]	Academic study
GSK-3326595	PRMT5	GlaxoSmithKline, Li et al. 2019	6.2 nM[a]	Clinical stage (NCT02783300)
JNJ-64619178	PRMT5	Johnson & Johnson, Li et al. 2019	0.14 nM[a]	Clinical stage (NCT03573310)
PF-06939999	PRMT5	Pfizer	ND	Clinical stage (NCT03854227)
Ctx series	PRMT5	Ctx Pty Ltd., Li et al. 2019	19–533 nM[a]	Preclinical development
PRT-543	PRMT5	Prelude Therapeutics	ND	Clinical stage (NCT03886831)
LLY-283	PRMT5	Eli Lilly, Bonday et al. 2018	22 nM[a]	Preclinical development
Compound-9	PRMT5	Lin et al. 2019	11 nM[a]	Academic study
C_4	PRMT5	Zhu et al. 2019	0.72 μM[a]	Academic study
4b14	PRMT5	Zhu et al. 2018	2.71 μM[a]	Academic study
Compound-4 and -10	PRMT5	Tao et al. 2019	8.1 and 6.5 μM[a]	Academic study
CMP5 and HLCL series	PRMT5	Baiocchi et al. 2014	~5 μM[a]	Academic study
Sinefungin	PRMT5	Tao et al. 2019	8.6 μM[a]	Academic study

[a]IC_{50} values are based on a published biochemical assay.
[b]IC_{50} values are based on a published cell-based assay.
Abbreviations: IC_{50}, half-maximal inhibitory concentration; ND, not documented.

EPZ015666 treatment demonstrated dose-dependent reduction of SmD3me2s expression in a panel of mantle cell lymphoma cell lines and antiproliferative activity in Z-138 and Maver-1 cell lines in in vitro growth assays. These results were supported by in vivo studies, which demonstrated that EPZ015666 induces tumor stasis with more than 90% tumor growth inhibition (TGI) in the Z-138 models and more than 70% TGI in Maver-1 xenograft models. These initial findings encouraged further structural optimization of the EPZ015666 scaffold to better understand the mechanism of efficacy and improve its physicochemical characteristics.

Crystallographic analysis of the PRMT5:MEP50-SAM-EPZ015666 complex prompted the development of a series of compounds that led to the discovery of the clinical candidate GSK-3326595 (Gerhart et al. 2018). This compound demonstrated improved cellular potency and pharmacokinetic properties, with more than 4,000-fold selectivity for PRMT5:MEP50 over any other protein methyltransferase tested. The antiproliferative activity of GSK-3326595 was profiled in a panel of 240 cancer cell lines of diverse tumor origin. The result of this analysis revealed that breast, acute myeloid leukemia, and multiple myeloma cell lines were among the most sensitive to GSK-3326595 treatment. Furthermore, mechanism of efficacy studies showed that GSK-3326595 induced inhibition of total SDMA in a dose-dependent manner that correlated with efficacy in vitro and in vivo. Inhibition of PRMT5 after treatment with GSK-3326595 induced expression of p53 and p21/CDKN1 in p53-proficient Z-138 cell lines in vitro, which was validated in vivo by immunohistochemistry. Moreover, GSK-3326595 treatment induced alternative splicing that

was characterized by an isoform switch of MDM4 and, together with the activation of p53 transcription, established a response to inhibition of PRMT5 activity. All of these data suggested that patients with an intact p53 pathway may benefit from treatment with GSK-3326595.

A mechanistically distinct, SAM-competitive PRMT5 inhibitor approach led to the successful development of several other selective PRMT5 inhibitors such as LLY-283, JNJ-64619178 (Li et al. 2019), and PF-06939999. To date, only LLY-283 has been fully described in the literature (Bonday et al. 2018). LLY-283 is an analog of SAM, in which the methyl moiety is replaced by a phenyl group. It is a highly potent inhibitor of PRMT5 with IC_{50} values in the low nanomolar range, like that of GSK-3326595. LLY-283 also displays more than 100-fold PRMT5 selectivity over other methyltransferases. Cell-based assays revealed LLY-283 to be a potent inhibitor of SmBB′-Rme2s methylation in breast MCM7 (IC_{50}: 20 nM) and lung A549 (IC_{50}: 40 nM) cell lines. Notably, treatment with LLY-283 has also been shown to induce splicing changes and isoform switching in MDM4, which led to p53 pathway activation similar to that seen with GSK-3326595. These findings suggest that both the GSK-3326595 and LLY-283 compounds share similar mechanisms of efficacy despite their different mechanisms for inhibiting PRMT5.

Several other efforts to discover small-molecule inhibitors of PRMT5 have been described, including covalent approaches that take advantage of the active site cysteine C449 that is unique to PRMT5 (Alinari et al. 2015; Baiocchi et al. 2014; Lin et al. 2019; Tao et al. 2019; Zhu et al. 2018, 2019).

Interestingly, the SAM-uncompetitive PRMT5 inhibitor EPZ01566 was tested for MTAP-selective growth inhibition and failed to display robust selectivity for cancers with deletion of *MTAP* (Kryukov et al. 2016, Marjon et al. 2016, Mavrakis et al. 2016). This difference between genetic ablation of PRMT5 and pharmacologic inhibition of PRMT5 likely occurs because the mechanism of action of the SAM-uncompetitive PRMT5 inhibitor fails to leverage the MTA-high metabolic state of *MTAP*-deficient cells. Mavrakis et al. (2016) postulated that SAM-competitive inhibition of PRMT5 would lead to greater selective growth inhibition of *MTAP*-deficient cells. We have noted that two inhibitors of a single enzyme can only be synergistic if they bind to separate binding sites and their interaction is not mutually exclusive (Breitinger 2012); thus, we have postulated that small molecules that bind synergistically to the PRMT5:MTA complex would trap the enzyme in its inactive state. Since MTA levels are higher in *MTAP*-deficient cancer cells, an inhibitor that binds synergistically with MTA would be expected to selectively inhibit PRMT5 in *MTAP*-deficient cells. To date, there are no publications that describe PRMT5 inhibitors that display a high degree of selectivity for *MTAP*-deficient cancers, and thus such *MTAP*-selective PRMT5 inhibitor strategies currently remain hypothetical. The compelling genetic validation of PRMT5 as an *MTAP*-selective target will likely motivate continued drug discovery efforts to explore alternate modes of inhibition of PRMT5, like those noted above.

Despite its clear implication in cancer biology, MAT2A has only recently emerged as a validated therapeutic target in *MTAP*-deleted cancers (Marjon et al. 2016). Although substrate-competitive Met analog inhibitors of MAT2A such as cycloleucine were identified by Lombardini and colleagues in the 1970s and 1980s (Lombardini et al. 1970, Lombardini & Sufrin 1983), their low potency and structural simplicity raise concerns about their specificity, especially in the cell-based assay context, due to the potential impact on cellular amino acid metabolism. More recent efforts identified stilbene derivatives as inhibitors of MAT2A (Sviripa et al. 2014, W. Zhang et al. 2013), but these compounds carry redox reactivity flags and are unlikely to specifically inhibit MAT2A. Recently, a high-throughput screening campaign conducted by Pfizer identified a moderately potent allosteric MAT2A inhibitor, PF-9366 (Quinlan et al. 2017). PF-9366 demonstrates the potential to drug MAT2A via an allosteric mechanism, namely by binding to the MAT2B regulatory pocket. However, PF-9366 treatment in cells induced cellular adaptation, including upregulation

of MAT2A itself (Pendleton et al. 2017, Vazquez-Chantada et al. 2010, Yang et al. 2015), which blunted cellular potency and led to inadequate antiproliferative effects.

Konteatis and colleagues have discovered and developed small-molecule inhibitors of MAT2A that are highly potent, cell permeable, selective, and orally bioavailable, including AGI-25696 and AG-270 (Z. Konteatis, J. Travins, S. Gross, K. Marjon, A. Barnett, et al., manuscript in review). These MAT2A inhibitors were discovered via fragment screening approaches followed by iterative structure-guided design, enabling more than 10,000-fold improvement of the potency. This family of MAT2A inhibitors are allosteric, substrate noncompetitive, and inhibit MAT2A activity via enhanced enzyme product (SAM) inhibition, as they bind to the same allosteric site as the regulatory MAT2B subunit, similarly to what was reported for PF-9366 (Quinlan et al. 2017). Furthermore, small-molecule inhibition of MAT2A potently reduces SAM levels in cells by blocking de novo SAM biosynthesis and leads to *MTAP*-genotype selective antiproliferative activity in vitro and in vivo, as well as *MTAP*-selective effects on PRMT5 methyl marks. Thus, targeting of MAT2A in *MTAP*-deficient cancers represents a successful application of synthetic lethality and a novel therapeutic approach for the substantial subset of patients with loss of the *CDKN2A/MTAP* locus.

Based on these discoveries, AG-270, a MAT2A inhibitor, has entered clinical development and is under investigation in a phase I trial that is currently enrolling patients with *MTAP*-deleted solid tumors and lymphomas (NCT03435250).

CONCLUDING REMARKS

Identifying and translating novel synthetic lethal vulnerabilities into precision medicine approaches has been greatly enabled by the implementation of functional genomic approaches and continuous progress in their development. The insight to target PRMT5 and MAT2A in *MTAP*-deficient tumors emerged from these efforts, which is one among a wave of clinically applicable synthetic lethalities to follow the success of PARP [poly(ADP-ribose) polymerase] inhibitors in *BRCA1*- and *BRCA2*-mutant cancers (Bryant et al. 2005, Farmer et al. 2005, Tutt et al. 2005). However, in order to recapitulate the phenotypes observed using genetic tools for target modulation with pharmacologic small-molecule agents, it is critical to develop an in-depth understanding of the underlying target biology and biochemistry in order to identify appropriate drugging strategies. Only one of the two recently discovered synthetic lethal vulnerabilities in *MTAP*-deficient cancers, MAT2A, has thus far been translated into an *MTAP*-null selective small-molecule therapeutic agent according to preclinical studies. Current small-molecule inhibitors of PRMT5 have not yet yielded *MTAP*-null selectivity, perhaps owing to their mechanism of action. Further efforts are necessary to identify alternative means of drugging PRMT5 that would take advantage of the high-MTA environment of *MTAP*-deficient cancers. Both approaches have yet to achieve proof-of-concept in the clinic and may require the identification of rational combination strategies to provide greater antitumor activity. In addition, an understanding of potential effects on the tumor microenvironment and immune cells following targeting of MAT2A and PRMT5 (Benci et al. 2019, Henrich et al. 2016, Inoue et al. 2018, Roy et al. 2020) may produce further insights into ways of optimizing the efficacy of MAT2A and PRMT5 inhibitors and the potential for combinations with existing and emerging immune therapies.

DISCLOSURE STATEMENT

The authors are, or were at the time of this work, employees and shareholders of Agios Pharmaceuticals, which has developed the MAT2A inhibitor AG-270, which is currently under investigation in a phase I clinical trial.

ACKNOWLEDGMENTS

The authors would like to acknowledge Chi-Chao Chen for providing CDKN2A and MTAP deletion rates across cancers, and Stefan Gross for insights regarding MAT2A enzymology.

LITERATURE CITED

Alinari L, Mahasenan KV, Yan F, Karkhanis V, Chung JH, et al. 2015. Selective inhibition of protein arginine methyltransferase 5 blocks initiation and maintenance of B-cell transformation. *Blood* 125:2530–43

Ancelin K, Lange UC, Hajkova P, Schneider R, Bannister AJ, et al. 2006. Blimp1 associates with Prmt5 and directs histone arginine methylation in mouse germ cells. *Nat. Cell Biol.* 8:623–30

Baiocchi R, Li C, Lai H, Sif S. 2014. *Inhibitors of PRMT5 and methods of their use*. Int. Patent 2014/145214 A2

Barekatain Y, Yan VC, Ackroyd JJ, Poral AH, Tran T, et al. 2019. Methylthioadenosine is not dramatically elevated in *MTAP*-homozygous deleted primary glioblastomas. bioRxiv 769885. **https://doi.org/10.1101/769885**

Basu I, Cordovano G, Das I, Belbin TJ, Guha C, Schramm VL. 2007. A transition state analogue of 5′-methylthioadenosine phosphorylase induces apoptosis in head and neck cancers. *J. Biol. Chem.* 282:21477–86

Basu I, Locker J, Cassera MB, Belbin TJ, Merino EF, et al. 2011. Growth and metastases of human lung cancer are inhibited in mouse xenografts by a transition state analogue of 5′-methylthioadenosine phosphorylase. *J. Biol. Chem.* 286:4902–11

Bedford MT, Clarke SG. 2009. Protein arginine methylation in mammals: who, what, and why. *Mol. Cell* 33:1–13

Behan FM, Iorio F, Picco G, Goncalves E, Beaver CM, et al. 2019. Prioritization of cancer therapeutic targets using CRISPR-Cas9 screens. *Nature* 568:511–16

Benci JL, Johnson LR, Choa R, Xu Y, Qiu J, et al. 2019. Opposing functions of interferon coordinate adaptive and innate immune responses to cancer immune checkpoint blockade. *Cell* 178:933–48.e14

Beroukhim R, Mermel CH, Porter D, Wei G, Raychaudhuri S, et al. 2010. The landscape of somatic copy-number alteration across human cancers. *Nature* 463:899–905

Bertino JR, Waud WR, Parker WB, Lubin M. 2011. Targeting tumors that lack methylthioadenosine phosphorylase (MTAP) activity: current strategies. *Cancer Biol. Ther.* 11:627–32

Bezzi M, Teo SX, Muller J, Mok WC, Sahu SK, et al. 2013. Regulation of constitutive and alternative splicing by PRMT5 reveals a role for *Mdm4* pre-mRNA in sensing defects in the spliceosomal machinery. *Genes Dev.* 27:1903–16

Bollag G, Hirth P, Tsai J, Zhang J, Ibrahim PN, et al. 2010. Clinical efficacy of a RAF inhibitor needs broad target blockade in *BRAF*-mutant melanoma. *Nature* 467:596–99

Bonday ZQ, Cortez GS, Grogan MJ, Antonysamy S, Weichert K, et al. 2018. LLY-283, a potent and selective inhibitor of arginine methyltransferase 5, PRMT5, with antitumor activity. *ACS Med. Chem. Lett.* 9:612–17

Boutros M, Ahringer J. 2008. The art and design of genetic screens: RNA interference. *Nat. Rev. Genet.* 9:554–66

Brahms H, Raymackers J, Union A, de Keyser F, Meheus L, Luhrmann R. 2000. The C-terminal RG dipeptide repeats of the spliceosomal Sm proteins D1 and D3 contain symmetrical dimethylarginines, which form a major B-cell epitope for anti-Sm autoantibodies. *J. Biol. Chem.* 275:17122–29

Braun CJ, Stanciu M, Boutz PL, Patterson JC, Calligaris D, et al. 2017. Coordinated splicing of regulatory detained introns within oncogenic transcripts creates an exploitable vulnerability in malignant glioma. *Cancer Cell* 32:411–26.e11

Breitinger H-G. 2012. Drug synergy—mechanisms and methods of analysis. *InTech*. **https://doi.org/10.5772/30922**

Bryant HE, Schultz N, Thomas HD, Parker KM, Flower D, et al. 2005. Specific killing of BRCA2-deficient tumours with inhibitors of poly(ADP-ribose) polymerase. *Nature* 434:913–17

Cai J, Mao Z, Hwang JJ, Lu SC. 1998. Differential expression of methionine adenosyltransferase genes influences the rate of growth of human hepatocellular carcinoma cells. *Cancer Res.* 58:1444–50

Cai J, Sun WM, Hwang JJ, Stain SC, Lu SC. 1996. Changes in *S*-adenosylmethionine synthetase in human liver cancer: molecular characterization and significance. *Hepatology* 24:1090–97

Camacho-Vanegas O, Camacho SC, Till J, Miranda-Lorenzo I, Terzo E, et al. 2012. Primate genome gain and loss: a bone dysplasia, muscular dystrophy, and bone cancer syndrome resulting from mutated retroviral-derived MTAP transcripts. *Am. J. Hum. Genet.* 90:614–27

Casero RA Jr., Murray Stewart T, Pegg AE. 2018. Polyamine metabolism and cancer: treatments, challenges and opportunities. *Nat. Rev. Cancer* 18:681–95

Cen L, Carlson BL, Schroeder MA, Ostrem JL, Kitange GJ, et al. 2012. p16-Cdk4-Rb axis controls sensitivity to a cyclin-dependent kinase inhibitor PD0332991 in glioblastoma xenograft cells. *Neuro-Oncology* 14:870–81

Chan-Penebre E, Kuplast KG, Majer CR, Boriack-Sjodin PA, Wigle TJ, et al. 2015. A selective inhibitor of PRMT5 with *in vivo* and *in vitro* potency in MCL models. *Nat. Chem. Biol.* 11:432–37

Chen H, Xia M, Lin M, Yang H, Kuhlenkamp J, et al. 2007. Role of methionine adenosyltransferase 2A and S-adenosylmethionine in mitogen-induced growth of human colon cancer cells. *Gastroenterology* 133:207–18

Chen ZH, Olopade OI, Savarese TM. 1997. Expression of methylthioadenosine phosphorylase cDNA in *p16*$^{-}$, *MTAP*$^{-}$ malignant cells: restoration of methylthioadenosine phosphorylase-dependent salvage pathways and alterations of sensitivity to inhibitors of purine *de novo* synthesis. *Mol. Pharmacol.* 52:903–11

Cho EC, Zheng S, Munro S, Liu G, Carr SM, et al. 2012. Arginine methylation controls growth regulation by E2F-1. *EMBO J.* 31:1785–97

Christopher SA, Diegelman P, Porter CW, Kruger WD. 2002. Methylthioadenosine phosphorylase, a gene frequently codeleted with p16$^{cdkN2a/ARF}$, acts as a tumor suppressor in a breast cancer cell line. *Cancer Res.* 62:6639–44

Clarke TL, Sanchez-Bailon MP, Chiang K, Reynolds JJ, Herrero-Ruiz J, et al. 2017. PRMT5-dependent methylation of the TIP60 coactivator RUVBL1 is a key regulator of homologous recombination. *Mol. Cell* 65:900–16.e7

Cong L, Ran FA, Cox D, Lin S, Barretto R, et al. 2013. Multiplex genome engineering using CRISPR/Cas systems. *Science* 339:819–23

Cowling VH. 2009. Regulation of mRNA cap methylation. *Biochem. J.* 425:295–302

Cristini A, Ricci G, Britton S, Salimbeni S, Huang SN, et al. 2019. Dual processing of R-loops and topoisomerase I induces transcription-dependent DNA double-strand breaks. *Cell Rep.* 28:3167–81.e6

Di Lorenzo A, Bedford MT. 2011. Histone arginine methylation. *FEBS Lett.* 585:2024–31

Elbashir SM, Harborth J, Lendeckel W, Yalcin A, Weber K, Tuschl T. 2001. Duplexes of 21-nucleotide RNAs mediate RNA interference in cultured mammalian cells. *Nature* 411:494–98

Enouf J, Lawrence F, Tempete C, Robert-Gero M, Lederer E. 1979. Relationship between inhibition of protein methylase I and inhibition of Rous sarcoma virus-induced cell transformation. *Cancer Res.* 39:4497–502

Fabbrizio E, El Messaoudi S, Polanowska J, Paul C, Cook JR, et al. 2002. Negative regulation of transcription by the type II arginine methyltransferase PRMT5. *EMBO Rep.* 3:641–45

Farmer H, McCabe N, Lord CJ, Tutt AN, Johnson DA, et al. 2005. Targeting the DNA repair defect in *BRCA* mutant cells as a therapeutic strategy. *Nature* 434:917–21

Feng Y, Maity R, Whitelegge JP, Hadjikyriacou A, Li Z, et al. 2013. Mammalian protein arginine methyltransferase 7 (PRMT7) specifically targets RXR sites in lysine- and arginine-rich regions. *J. Biol. Chem.* 288:37010–25

Frau M, Feo F, Pascale RM. 2013. Pleiotropic effects of methionine adenosyltransferases deregulation as determinants of liver cancer progression and prognosis. *J. Hepatol.* 59:830–41

Friesen WJ, Paushkin S, Wyce A, Massenet S, Pesiridis GS, et al. 2001. The methylosome, a 20S complex containing JBP1 and pICln, produces dimethylarginine-modified Sm proteins. *Mol. Cell Biol.* 21:8289–300

Friesen WJ, Wyce A, Paushkin S, Abel L, Rappsilber J, et al. 2002. A novel WD repeat protein component of the methylosome binds Sm proteins. *J. Biol. Chem.* 277:8243–47

Gerhart SV, Kellner WA, Thompson C, Pappalardi MB, Zhang XP, et al. 2018. Activation of the p53-MDM4 regulatory axis defines the anti-tumour response to PRMT5 inhibition through its role in regulating cellular splicing. *Sci. Rep.* 8:9711

Gerstung M, Jolly C, Leshchiner I, Dentro SC, Gonzalez S, et al. 2020. The evolutionary history of 2,658 cancers. *Nature* 578:122–28

Girardot M, Hirasawa R, Kacem S, Fritsch L, Pontis J, et al. 2014. PRMT5-mediated histone H4 arginine-3 symmetrical dimethylation marks chromatin at G + C-rich regions of the mouse genome. *Nucleic Acids Res.* 42:235–48

Guo Z, Zheng L, Xu H, Dai H, Zhou M, et al. 2010. Methylation of FEN1 suppresses nearby phosphorylation and facilitates PCNA binding. *Nat. Chem. Biol.* 6:766–73

Halim AB, LeGros L, Geller A, Kotb M. 1999. Expression and functional interaction of the catalytic and regulatory subunits of human methionine adenosyltransferase in mammalian cells. *J. Biol. Chem.* 274:29720–25

Hamard PJ, Santiago GE, Liu F, Karl DL, Martinez C, et al. 2018. PRMT5 regulates DNA repair by controlling the alternative splicing of histone-modifying enzymes. *Cell Rep.* 24:2643–57

Hansen LJ, Sun R, Yang R, Singh SX, Chen LH, et al. 2019. MTAP loss promotes stemness in glioblastoma and confers unique susceptibility to purine starvation. *Cancer Res.* 79:3383–94

He HL, Lee YE, Shiue YL, Lee SW, Chen TJ, Li CF. 2015. Characterization and prognostic significance of methylthioadenosine phosphorylase deficiency in nasopharyngeal carcinoma. *Medicine* 94:e2271

Henrich FC, Singer K, Poller K, Bernhardt L, Strobl CD, et al. 2016. Suppressive effects of tumor cell-derived 5′-deoxy-5′-methylthioadenosine on human T cells. *OncoImmunology* 5:e1184802

Hesterberg RS, Cleveland JL, Epling-Burnette PK. 2018. Role of polyamines in immune cell functions. *Med. Sci.* 6(1):22

Hochhaus A, Larson RA, Guilhot F, Radich JP, Branford S, et al. 2017. Long-term outcomes of imatinib treatment for chronic myeloid leukemia. *N. Engl. J. Med.* 376:917–27

Hori H, Tran P, Carrera CJ, Hori Y, Rosenbach MD, et al. 1996. Methylthioadenosine phosphorylase cDNA transfection alters sensitivity to depletion of purine and methionine in A549 lung cancer cells. *Cancer Res.* 56:5653–58

Hsu JM, Chen CT, Chou CK, Kuo HP, Li LY, et al. 2011. Crosstalk between Arg 1175 methylation and Tyr 1173 phosphorylation negatively modulates EGFR-mediated ERK activation. *Nat. Cell Biol.* 13:174–81

Inoue M, Okamoto K, Terashima A, Nitta T, Muro R, et al. 2018. Arginine methylation controls the strength of γc-family cytokine signaling in T cell maintenance. *Nat. Immunol.* 19:1265–76

Jani TS, Gobejishvili L, Hote PT, Barve AS, Joshi-Barve S, et al. 2009. Inhibition of methionine adenosyltransferase II induces FasL expression, Fas-DISC formation and caspase-8-dependent apoptotic death in T leukemic cells. *Cell Res.* 19:358–69

Jansson M, Durant ST, Cho EC, Sheahan S, Edelmann M, et al. 2008. Arginine methylation regulates the p53 response. *Nat. Cell Biol.* 10:1431–39

Jinek M, Chylinski K, Fonfara I, Hauer M, Doudna JA, Charpentier E. 2012. A programmable dual-RNA-guided DNA endonuclease in adaptive bacterial immunity. *Science* 337:816–21

Jinek M, East A, Cheng A, Lin S, Ma E, Doudna J. 2013. RNA-programmed genome editing in human cells. *eLife* 2:e00471

Jing W, Zhu H, Liu W, Zhai X, Tian H, Yu J. 2020. MTAP-deficiency could predict better treatment response in advanced lung adenocarcinoma patients initially treated with pemetrexed-platinum chemotherapy and bevacizumab. *Sci. Rep.* 10:843

Kadariya Y, Tang B, Wang L, Al-Saleem T, Hayakawa K, et al. 2013. Germline mutations in *Mtap* cooperate with *Myc* to accelerate tumorigenesis in mice. *PLOS ONE* 8:e67635

Kadariya Y, Yin B, Tang B, Shinton SA, Quinlivan EP, et al. 2009. Mice heterozygous for germ-line mutations in methylthioadenosine phosphorylase (*MTAP*) die prematurely of T-cell lymphoma. *Cancer Res.* 69:5961–69

Kamatani N, Nelson-Rees WA, Carson DA. 1981. Selective killing of human malignant cell lines deficient in methylthioadenosine phosphorylase, a purine metabolic enzyme. *PNAS* 78:1219–23

Kamatani N, Yu AL, Carson DA. 1982. Deficiency of methylthioadenosine phosphorylase in human leukemic cells in vivo. *Blood* 60:1387–91

Kindler HL, Burris HA 3rd, Sandler AB, Oliff IA. 2009. A phase II multicenter study of L-alanosine, a potent inhibitor of adenine biosynthesis, in patients with MTAP-deficient cancer. *Investig. New Drugs* 27:75–81
Kotb M, Kredich NM. 1990. Regulation of human lymphocyte S-adenosylmethionine synthetase by product inhibition. *Biochim. Biophys. Acta* 1039:253–60
Kotb M, Mudd SH, Mato JM, Geller AM, Kredich NM, et al. 1997. Consensus nomenclature for the mammalian methionine adenosyltransferase genes and gene products. *Trends Genet.* 13:51–52
Kryukov GV, Wilson FH, Ruth JR, Paulk J, Tsherniak A, et al. 2016. *MTAP* deletion confers enhanced dependency on the PRMT5 arginine methyltransferase in cancer cells. *Science* 351:1214–18
LeGros HL Jr., Geller AM, Kotb M. 1997. Differential regulation of methionine adenosyltransferase in superantigen and mitogen stimulated human T lymphocytes. *J. Biol. Chem.* 272:16040–47
Li X, Wang C, Jiang H, Luo C. 2019. A patent review of arginine methyltransferase inhibitors (2010–2018). *Expert Opin. Ther. Pat.* 29:97–114
Lin H, Wang M, Zhang YW, Tong S, Leal RA, et al. 2019. Discovery of potent and selective covalent protein arginine methyltransferase 5 (PRMT5) inhibitors. *ACS Med. Chem. Lett.* 10:1033–38
Lombardini JB, Coulter AW, Talalay P. 1970. Analogues of methionine as substrates and inhibitors of the methionine adenosyltransferase reaction: deductions concerning the conformation of methionine. *Mol. Pharmacol.* 6:481–99
Lombardini JB, Sufrin JR. 1983. Chemotherapeutic potential of methionine analogue inhibitors of tumor-derived methionine adenosyltransferases. *Biochem. Pharmacol.* 32:489–95
Longshaw AI, Adanitsch F, Gutierrez JA, Evans GB, Tyler PC, Schramm VL. 2010. Design and synthesis of potent "sulfur-free" transition state analogue inhibitors of 5′-methylthioadenosine nucleosidase and 5′-methylthioadenosine phosphorylase. *J. Med. Chem.* 53:6730–46
Lu SC, Alvarez L, Huang ZZ, Chen L, An W, et al. 2001. Methionine adenosyltransferase 1A knockout mice are predisposed to liver injury and exhibit increased expression of genes involved in proliferation. *PNAS* 98:5560–65
Lu SC, Mato JM. 2012. *S*-adenosylmethionine in liver health, injury, and cancer. *Physiol. Rev.* 92:1515–42
Lubin M, Lubin A. 2009. Selective killing of tumors deficient in methylthioadenosine phosphorylase: a novel strategy. *PLOS ONE* 4:e5735
Maldonado LY, Arsene D, Mato JM, Lu SC. 2018. Methionine adenosyltransferases in cancers: mechanisms of dysregulation and implications for therapy. *Exp. Biol. Med.* 243:107–17
Mali P, Yang L, Esvelt KM, Aach J, Guell M, et al. 2013. RNA-guided human genome engineering via Cas9. *Science* 339:823–26
Marjon K, Cameron MJ, Quang P, Clasquin MF, Mandley E, et al. 2016. *MTAP* deletions in cancer create vulnerability to targeting of the MAT2A/PRMT5/RIOK1 axis. *Cell Rep.* 15:574–87
Martinez-Chantar ML, Corrales FJ, Martinez-Cruz LA, Garcia-Trevijano ER, Huang ZZ, et al. 2002. Spontaneous oxidative stress and liver tumors in mice lacking methionine adenosyltransferase 1A. *FASEB J.* 16:1292–94
Mavrakis KJ, McDonald ER 3rd, Schlabach MR, Billy E, Hoffman GR, et al. 2016. Disordered methionine metabolism in MTAP/CDKN2A-deleted cancers leads to dependence on PRMT5. *Science* 351:1208–13
McDonald ER 3rd, de Weck A, Schlabach MR, Billy E, Mavrakis KJ, et al. 2017. Project DRIVE: a compendium of cancer dependencies and synthetic lethal relationships uncovered by large-scale, deep RNAi screening. *Cell* 170:577–92.e10
Meister G, Eggert C, Buhler D, Brahms H, Kambach C, Fischer U. 2001. Methylation of Sm proteins by a complex containing PRMT5 and the putative U snRNP assembly factor pICln. *Curr. Biol.* 11:1990–94
Mersaoui SY, Yu Z, Coulombe Y, Karam M, Busatto FF, et al. 2019. Arginine methylation of the DDX5 helicase RGG/RG motif by PRMT5 regulates resolution of RNA:DNA hybrids. *EMBO J.* 38:e100986
Migliori V, Muller J, Phalke S, Low D, Bezzi M, et al. 2012. Symmetric dimethylation of H3R2 is a newly identified histone mark that supports euchromatin maintenance. *Nat. Struct. Mol. Biol.* 19:136–44
Muller FL, Aquilanti EA, DePinho RA. 2015. Collateral lethality: a new therapeutic strategy in oncology. *Trends Cancer* 1:161–73
Murray B, Antonyuk SV, Marina A, Lu SC, Mato JM, et al. 2016. Crystallography captures catalytic steps in human methionine adenosyltransferase enzymes. *PNAS* 113:2104–9

Owens JL, Beketova E, Liu S, Tinsley SL, Asberry AM, et al. 2020. PRMT5 cooperates with pICln to function as a master epigenetic activator of DNA double-strand break repair genes. *iScience* 23:100750

Pal S, Vishwanath SN, Erdjument-Bromage H, Tempst P, Sif S. 2004. Human SWI/SNF-associated PRMT5 methylates histone H3 arginine 8 and negatively regulates expression of *ST7* and *NM23* tumor suppressor genes. *Mol. Cell Biol.* 24:9630–45

PCAWG (Pan-Cancer Atlas Whole Genomes) Consort. 2020. Pan-cancer analysis of whole genomes. *Nature* 578:82–93

Pegg AE. 2016. Functions of polyamines in mammals. *J. Biol. Chem.* 291:14904–12

Pendleton KE, Chen B, Liu K, Hunter OV, Xie Y, et al. 2017. The U6 snRNA m^6A methyltransferase METTL16 regulates SAM synthetase intron retention. *Cell* 169:824–35.e14

Phuong NT, Kim SK, Im JH, Yang JW, Choi MC, et al. 2016. Induction of methionine adenosyltransferase 2A in tamoxifen-resistant breast cancer cells. *Oncotarget* 7:13902–16

Quinlan CL, Kaiser SE, Bolanos B, Nowlin D, Grantner R, et al. 2017. Targeting *S*-adenosylmethionine biosynthesis with a novel allosteric inhibitor of Mat2A. *Nat. Chem. Biol.* 13:785–92

Radzisheuskaya A, Shliaha PV, Grinev V, Lorenzini E, Kovalchuk S, et al. 2019. PRMT5 methylome profiling uncovers a direct link to splicing regulation in acute myeloid leukemia. *Nat. Struct. Mol. Biol.* 26:999–1012

Roy DG, Chen J, Mamane V, Ma EH, Muhire BM, et al. 2020. Methionine metabolism shapes T helper cell responses through regulation of epigenetic reprogramming. *Cell Metab.* 31:250–66.e9

Roy DM, Walsh LA, Desrichard A, Huse JT, Wu W, et al. 2016. Integrated genomics for pinpointing survival loci within arm-level somatic copy number alterations. *Cancer Cell* 29:737–50

Ruefli-Brasse A, Sakamoto D, Orf J, Rong MQ, Shi T, et al. 2011. Methylthioadenosine (MTA) rescues methylthioadenosine phosphorylase (MTAP)-deficient tumors from purine synthesis inhibition *in vivo* via non-autonomous adenine supply. *J. Cancer Ther.* 2:523–34

Schmid M, Malicki D, Nobori T, Rosenbach MD, Campbell K, et al. 1998. Homozygous deletions of methylthioadenosine phosphorylase (*MTAP*) are more frequent than $p16^{INK4A}$ (*CDKN2*) homozygous deletions in primary non-small cell lung cancers (NSCLC). *Oncogene* 17:2669–75

Shaw AT, Kim DW, Mehra R, Tan DS, Felip E, et al. 2014. Ceritinib in ALK-rearranged non-small-cell lung cancer. *N. Engl. J. Med.* 370:1189–97

Singh V, Shi W, Evans GB, Tyler PC, Furneaux RH, et al. 2004. Picomolar transition state analogue inhibitors of human 5′-methylthioadenosine phosphorylase and X-ray structure with MT-immucillin-A. *Biochemistry* 43:9–18

Skourti-Stathaki K, Proudfoot NJ. 2014. A double-edged sword: R loops as threats to genome integrity and powerful regulators of gene expression. *Genes Dev.* 28:1384–96

Slamon DJ, Leyland-Jones B, Shak S, Fuchs H, Paton V, et al. 2001. Use of chemotherapy plus a monoclonal antibody against HER2 for metastatic breast cancer that overexpresses HER2. *N. Engl. J. Med.* 344:783–92

Stopa N, Krebs JE, Shechter D. 2015. The PRMT5 arginine methyltransferase: many roles in development, cancer and beyond. *Cell Mol. Life Sci.* 72:2041–59

Strekalova E, Malin D, Weisenhorn EMM, Russell JD, Hoelper D, et al. 2019. *S*-adenosylmethionine biosynthesis is a targetable metabolic vulnerability of cancer stem cells. *Breast Cancer Res. Treat.* 175:39–50

Su CY, Chang YC, Chan YC, Lin TC, Huang MS, et al. 2014. MTAP is an independent prognosis marker and the concordant loss of MTAP and p16 expression predicts short survival in non-small cell lung cancer patients. *Eur. J. Surg. Oncol.* 40:1143–50

Sullivan DM, Hoffman JL. 1983. Fractionation and kinetic properties of rat liver and kidney methionine adenosyltransferase isozymes. *Biochemistry* 22:1636–41

Sviripa VM, Zhang W, Balia AG, Tsodikov OV, Nickell JR, et al. 2014. 2′,6′-Dihalostyrylanilines, pyridines, and pyrimidines for the inhibition of the catalytic subunit of methionine S-adenosyltransferase-2. *J. Med. Chem.* 57:6083–91

Tan DQ, Li Y, Yang C, Li J, Tan SH, et al. 2019. PRMT5 modulates splicing for genome integrity and preserves proteostasis of hematopoietic stem cells. *Cell Rep.* 26:2316–28.e6

Tang B, Kadariya Y, Chen Y, Slifker M, Kruger WD. 2014. Expression of MTAP inhibits tumor-related phenotypes in HT1080 cells via a mechanism unrelated to its enzymatic function. *G3* 5:35–44

Tao H, Yan X, Zhu K, Zhang H. 2019. Discovery of novel PRMT5 inhibitors by virtual screening and biological evaluations. *Chem. Pharm. Bull.* 67:382–88

Tee WW, Pardo M, Theunissen TW, Yu L, Choudhary JS, et al. 2010. Prmt5 is essential for early mouse development and acts in the cytoplasm to maintain ES cell pluripotency. *Genes Dev.* 24:2772–77

Tomasi ML, Cossu C, Spissu Y, Floris A, Ryoo M, et al. 2017. S-adenosylmethionine and methylthioadenosine inhibit cancer metastasis by targeting microRNA 34a/b-methionine adenosyltransferase 2A/2B axis. *Oncotarget* 8:78851–69

Tsherniak A, Vazquez F, Montgomery PG, Weir BA, Kryukov G, et al. 2017. Defining a cancer dependency map. *Cell* 170:564–76.e16

Tutt AN, Lord CJ, McCabe N, Farmer H, Turner N, et al. 2005. Exploiting the DNA repair defect in BRCA mutant cells in the design of new therapeutic strategies for cancer. *Cold Spring Harb. Symp. Quant. Biol.* 70:139–48

Vazquez-Chantada M, Fernandez-Ramos D, Embade N, Martinez-Lopez N, Varela-Rey M, et al. 2010. HuR/methyl-HuR and AUF1 regulate the MAT expressed during liver proliferation, differentiation, and carcinogenesis. *Gastroenterology* 138:1943–53

Wang J, Zhu ZH, Yang HB, Zhang Y, Zhao XN, et al. 2016. Cullin 3 targets methionine adenosyltransferase IIα for ubiquitylation-mediated degradation and regulates colorectal cancer cell proliferation. *FEBS J.* 283:2390–402

Wang Y, Hu W, Yuan Y. 2018. Protein arginine methyltransferase 5 (PRMT5) as an anticancer target and its inhibitor discovery. *J. Med. Chem.* 61:9429–41

Wang Z, Yip LY, Lee JHJ, Wu Z, Chew HY, et al. 2019. Methionine is a metabolic dependency of tumor-initiating cells. *Nat. Med.* 25:825–37

Williams-Ashman HG, Seidenfeld J, Galletti P. 1982. Trends in the biochemical pharmacology of 5′-deoxy-5′-methylthioadenosine. *Biochem. Pharmacol.* 31:277–88

Xu J, Chang WH, Fong LWR, Weiss RH, Yu SL, Chen CH. 2019. Targeting the insulin-like growth factor-1 receptor in MTAP-deficient renal cell carcinoma. *Signal. Transduct. Target Ther.* 4:2

Yang HB, Xu YY, Zhao XN, Zou SW, Zhang Y, et al. 2015. Acetylation of MAT IIα represses tumour cell growth and is decreased in human hepatocellular cancer. *Nat. Commun.* 6:6973

Young RJ, Waldeck K, Martin C, Foo JH, Cameron DP, et al. 2014. Loss of *CDKN2A* expression is a frequent event in primary invasive melanoma and correlates with sensitivity to the CDK4/6 inhibitor PD0332991 in melanoma cell lines. *Pigment Cell Melanoma Res.* 27:590–600

Zappia V, Della Ragione F, Pontoni G, Gragnaniello V, Carteni-Farina M. 1988. Human 5′-deoxy-5′-methylthioadenosine phosphorylase: kinetic studies and catalytic mechanism. *Adv. Exp. Med. Biol.* 250:165–77

Zhang H, Chen ZH, Savarese TM. 1996. Codeletion of the genes for p16INK4, methylthioadenosine phosphorylase, interferon-α_1, interferon-β_1, and other 9p21 markers in human malignant cell lines. *Cancer Genet. Cytogenet.* 86:22–28

Zhang L, Ma S, Song X, Han B, Cheng Y, et al. 2012. Gefitinib versus placebo as maintenance therapy in patients with locally advanced or metastatic non-small-cell lung cancer (INFORM; C-TONG 0804): a multicentre, double-blind randomised phase 3 trial. *Lancet Oncol.* 13:466–75

Zhang T, Zheng Z, Liu Y, Zhang J, Zhao Y, et al. 2013. Overexpression of methionine adenosyltransferase II alpha (MAT2A) in gastric cancer and induction of cell cycle arrest and apoptosis in SGC-7901 cells by shRNA-mediated silencing of MAT2A gene. *Acta Histochem.* 115:48–55

Zhang W, Sviripa V, Chen X, Shi J, Yu T, et al. 2013. Fluorinated *N,N*-dialkylaminostilbenes repress colon cancer by targeting methionine *S*-adenosyltransferase 2A. *ACS Chem. Biol.* 8:796–803

Zhao DY, Gish G, Braunschweig U, Li Y, Ni Z, et al. 2016. SMN and symmetric arginine dimethylation of RNA polymerase II C-terminal domain control termination. *Nature* 529:48–53

Zhao Q, Rank G, Tan YT, Li H, Moritz RL, et al. 2009. PRMT5-mediated methylation of histone H4R3 recruits DNMT3A, coupling histone and DNA methylation in gene silencing. *Nat. Struct. Mol. Biol.* 16:304–11

Zheng S, Moehlenbrink J, Lu YC, Zalmas LP, Sagum CA, et al. 2013. Arginine methylation-dependent reader-writer interplay governs growth control by E2F-1. *Mol. Cell* 52:37–51

Zhu F, Rui L. 2019. PRMT5 in gene regulation and hematologic malignancies. *Genes Dis.* 6:247–57
Zhu K, Shao J, Tao H, Yan X, Luo C, et al. 2019. Rational design, synthesis and biological evaluation of novel *triazole* derivatives as potent and selective PRMT5 inhibitors with antitumor activity. *J. Comput.-Aided Mol. Des.* 33:775–85
Zhu K, Song JL, Tao HR, Cheng ZQ, Jiang CS, Zhang H. 2018. Discovery of new potent protein arginine methyltransferase 5 (PRMT5) inhibitors by assembly of key pharmacophores from known inhibitors. *Bioorg. Med. Chem. Lett.* 28:3693–99

Annual Review of Cancer Biology

Volume 5, 2021

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Errata

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