Trends in Immunology

CellPress OPEN ACCESS

Review

Metabolism and epigenetics at the heart of T cell function

Leticia Soriano-Baguet [1,2] and Dirk Brenner [1,2,3,*]

T cell subsets adapt and rewire their metabolism according to their functions and surrounding microenvironment. Whereas naive T cells rely on mitochondrial metabolic pathways characterized by low nutrient requirements, effector T cells induce kinetically faster pathways to generate the biomass and energy needed for proliferation and cytokine production. Recent findings support the concept that alterations in metabolism also affect the epigenetics of T cells. In this review we discuss the connections between T cell metabolism and epigenetic changes such as histone post-translational modifications (PTMs) and DNA methylation, as well as the 'extra-metabolic' roles of metabolic enzymes and molecules. These findings collectively point to a new group of potential therapeutic targets for the treatment of T cell-dependent autoimmune diseases and cancers.

Highlights

The metabolic pathways of T cells generate substrates that are essential for epigenetic enzyme function, thus connecting T cell metabolism with epigenetic modifications.

Alterations in metabolite concentrations can influence the epigenetic landscape in T cells, thereby affecting T cell differentiation, proliferation, and function.

Metabolic enzymes possess 'extra-metabolic' roles that can directly affect T cell epigenetic profiles.

Substrates and enzymes of metabolic pathways in T cells are potentially interesting targets for the treatment of T cell-related autoimmune diseases and cancers.

Significance

The metabolic adaptations that T cells undergo to differentiate and execute various functions have been described for over two decades. Only recently has the connection between T cell metabolism and epigenetic modifications emerged. The potential of this nexus to serve as a therapeutic target for the treatment of some cancers and autoimmune diseases is an exciting new development in the field.

Connection between T cell metabolism and epigenetic modifications

Nutrients and metabolites are essential for T cell functions, and ample evidence exists that T cell subsets adapt their metabolism depending on their energy requirements [1–5]. The main sources of energy in mammalian T cells are glucose, amino acids, and fatty acids, and various metabolic pathways involve the uptake of these nutrients to support T cell function and survival. The common goal of these pathways is to produce energy in the form of ATP, as well as biomass building blocks such as proteins and nucleotides [1]. **Naive T cells** (see Glossary) depend on mitochondrial pathways that require minimal nutrient uptake, such as the **tricarboxylic acid (TCA) cycle** and **oxidative phosphorylation (OXPHOS)**. After activation, **effector T cells** undergo **metabolic reprogramming** that involves shifting from mitochondrial reactions to glycolysis and **glutaminolysis**, which are pathways that draw more nutrients from the surrounding microenvironment [1]. These changes can vary by T cell subset, and the pathways activated in effector T cells are different from those seen in $CD4^+$ **regulatory T (Treg) cells** [1,2,6].

In addition to metabolic adaptation, T cell differentiation and function depend on **epigenetic modifications**, particularly modifications of **histone (H) proteins** and DNA methylation. These alterations are often stabilized through the binding of transcription factors (TFs), which are frequently activated by metabolic elements. Enzymes and substrates of various metabolic pathways have been identified that play key roles in facilitating chromatin accessibility and/or gene expression [6–10]. Thus, the proper and intersecting regulation of both metabolism and epigenetic modifications is crucial for the correct function of the adaptive immune system [11].

In this review we discuss recent studies that have uncovered connections between T cell metabolism, epigenetics, and functions. We briefly describe how alterations to metabolism induce different epigenetic modifications and thereby alter gene activation; we also offer an explanation of how this shift is linked to cellular function. Lastly, we touch on how modulating the metabolism–epigenetics connection might be envisaged in treating some T cell-driven autoimmune diseases and cancers.

[1]Experimental and Molecular Immunology, Department of Infection and Immunity, Luxembourg Institute of Health, Esch-sur-Alzette, Luxembourg
[2]Immunology and Genetics, Luxembourg Centre for Systems Biomedicine, University of Luxembourg, Esch-sur-Alzette, Luxembourg
[3]Odense Research Center for Anaphylaxis, Department of Dermatology and Allergy Center, Odense University Hospital, University of Southern Denmark, Odense, Denmark

*Correspondence:
dirk.brenner@lih.lu (D. Brenner).

Histone modifications

Chromatin is constituted by subunits called nucleosomes which are composed of DNA strands wound around a core complex of eight histone proteins: two of each of the H2A, H2B, H3, and H4 proteins. Each histone core has a globular C-terminal domain and an N-terminal tail, and the tail is the site of numerous **PTMs** such as methylation and acetylation of its lysine (K) and/or arginine residues [12].

Histone methylation

Histone methylation involves the transfer of up to three methyl groups onto lysine or arginine residues in the N-terminal tails of core histones. This process is carried out by histone methyltransferases (HMTs) which are classified as either lysine- or arginine-specific HMTs depending on the residue targeted. That being said, the majority of HMTs are lysine-specific [12]. The main donor of methyl groups used by HMTs is *S*-adenosylmethionine (SAM) which is produced via one-carbon (1C) metabolism (Figure 1, left) [13,14]. *In vitro* and *in vivo*, 1C metabolism has been shown to be essential for the proliferation of mammalian cells, notably that of $CD4^+$ and $CD8^+$ T cells [15,16].

Methyl residues on lysines are removed by histone demethylases (HDMs) [12]. Several HDM family members, including KDM6B, use α-ketoglutarate (α-KG) as a cofactor, which is a metabolite of glutaminolysis and the TCA cycle [17]. Recent studies have shown that KDM6B is essential for the differentiation and proliferation of effector $CD8^+$ T cells in mice. For example, KDM6B rapidly reverses lysine-27 trimethylation of H3 (H3K27me3) at gene loci involved in early T cell activation, such as the TF genes *Tbx21*, *Irf4*, and *Irf8*, and the effector genes *Ifng* and *Gzmb*, consequently promoting the antiviral functions of $CD8^+$ T cells [18–20]. Similarly, in human and murine $CD4^+$ T cells, demethylation of H3K27me3 by a different HDM called Jumonji domain-containing protein 3 (JMJD3) supports transcription associated with early gene activation and controls $CD4^+$ T cell differentiation, respectively [21,22]. Indeed, *JAK2* and *IL12RB2* expression in early activated human $CD4^+$ T cells has been described as being regulated by JMJD3 [21]. A murine study showed that JMJD3 ablation (*Jmjd3*$^{fl/fl}$ × *Cd4–cre* conditional knockout mice) induced Th2 and **type 17 T helper (Th17) cell** differentiation, while impairing Th1 cell differentiation in a Th1-dependent colitis disease model [22]. However, the effect of a metabolite on histone methylation can be difficult to predict. For example, 2-hydroxyglutarate (2-HG) is a metabolite produced by mutant isocitrate dehydrogenase (IDH) in a hypoxic microenvironment, and 2-HG inhibited HDM KDM4 by activating mTOR in cancer cell lines [23]. Similarly, treatment of murine $CD8^+$ T cells *in vitro* with 2-HG enhanced their proliferation, persistence, and antitumor capacity by modulating histone and DNA methylation [24]. However, a recent study used B16 melanoma and MC38 colorectal adenocarcinoma cells overexpressing wild-type IDH1 or IDH1^{R132H} gliomas in mice to show that tumor-produced 2-HG was taken up by $CD8^+$ T cells, which blocked LDH function, thus impairing their cytotoxicity and antitumor capacities *in vivo* [25].

Overall, these studies suggest that histone methylation can be modulated by manipulating either the substrates or the enzymes involved in this process. These regulatory principles are relevant in that they might become important for the treatment of some cancers.

Histone acetylation

Histone acetylation is a dynamic PTM that shows high turnover and allows chromatin to oscillate between condensed and decondensed states. Histones are acetylated by histone acetyltransferases (HATs) which transfer the acetyl group from acetyl-CoA to specific lysine residues of H3 or H4 (Figure 2). This action opens chromatin, increasing DNA accessibility and thus generally increasing gene transcription [11,26]. Acetyl groups can be removed by histone deacetylases (HDACs), triggering chromatin recondensation [11,27]. Thus, the status of histone acetylation at any one time

depends on the opposing activities of HATs and HDACs. HATs and HDACs are thus also often referred to as activators and inhibitors of transcription, respectively [27,28]. In addition to histone PTMs, HATs and HDACs can also modulate the acetylation status of non-histone proteins such as TFs and signaling molecules (e.g., STATs) [29–32].

Acetyl-CoA in histone acetylation

Cellular metabolism is connected to epigenetic modifications through acetyl-CoA, which is the main substrate of HATs and can be produced by metabolic pathways such as the TCA cycle, **fatty acid oxidation (FAO)**, and acetate metabolism (Figure 2) [33]. Several studies have demonstrated how modulation of acetyl-CoA metabolism can influence histone acetylation. For example, one report indicated that glucose transporter 3 (Glut3) is essential for the effector functions of murine pathogenic Th17 cells *in vivo* and *in vitro* [7]. The authors found that Glut3–linked glucose oxidation and ATP–citrate lyase (ACLY) are needed to generate acetyl–CoA in T cells. Independently, our group recently ablated **pyruvate dehydrogenase (PDH)**, which converts pyruvate into acetyl–CoA [8]. In both studies, Glut3 (Slc2a3$^{fl/fl}$ mice) or PDH ablation (PDH$^{fl/fl}$ CD4 cre mice) decreased or prevented the flux of glucose into the TCA cycle and glucose-derived acetyl-CoA availability was reduced. This limited histone acetylation and Th17–specific gene expression (e.g., *Il17a* and *Il17f*), compromising Th17 effector cell function [7]. Hence, we and others have demonstrated that a glucose-derived pool of citrate is crucial for histone acetylation and thus, Th17 signature gene expression [7, 8].

In another study, the effects of inhibiting the mitochondrial pyruvate carrier (MPC) on histone acetylation were examined. MPC imports pyruvate into the mitochondria where it is used to generate acetyl-CoA via PDH [34,35]. When MPC was blocked in murine CD8$^+$ T cells, increased differentiation of CD8$^+$ **memory T cells** occurred, and this was due to enhanced histone acetylation and chromatin accessibility of pro-memory genes [36]. Upon MPC ablation (*Mpc1*$^{fl/fl}$*Cd4–cre* mice) or inhibition (with a small-molecule MPC inhibitor), glutamine and fatty acid metabolism were induced to produce high amounts of acetyl-CoA, which was then used for histone acetylation to promote the transcription of memory genes such as *Sell*, *Tcf7*, and *Ccr7* [36]. In a similar vein, a different study demonstrated that, in glucose-limiting environments (such as the tumor niche) in B16 melanoma and EL4-OVA mouse tumor models, murine CD8$^+$ T cell effector functions could be activated through acetate uptake [9]. This acetate was converted into acetyl-CoA by acetyl-CoA synthetase, inducing histone acetylation and chromatin accessibility at effector gene loci such as *Ifng* [9]. Lastly, also in mouse models (including *Cd4*^{cre}*Ldha*$^{fl/fl}$ mice as well as tumor and autoimmunity models), lactate dehydrogenase (LDH) was shown to be necessary for Th1 cells to maintain the acetyl-CoA pool supporting histone acetylation, and consequently *Ifng* expression [37]. In a parallel study using *Cd4*^{cre}*Ldha*$^{fl/fl}$ mice and the experimental autoimmune encephalomyelitis (EAE) mouse model of multiple sclerosis, LDH-derived ATP production activated the PI3K–Akt–Foxo1 axis, which regulated the Th17 effector function program and thus *Il17a* expression [38]. However, LDH ablation in Th17 cells limited *Il17a* transcription [38]. This study is a prime example of the intrinsic connections between T cell metabolism and signaling that ensure the optimal functions of these cells.

Collectively, these studies suggest that acetyl-CoA might prove to be an interesting target for regulating gene expression and function in T cells and modulating the roles of these cells in T cell-dependent inflammatory diseases.

NAD$^+$ metabolism and sirtuins

The NAD$^+$/NADH ratio in a T cell is vital for regulating its redox balance, which in turn is crucial for maintaining the metabolism and functions of effector CD4$^+$ and Treg cells [7,39]. Although NAD$^+$ can be synthesized *de novo* through the **kynurenine pathway** and the **Preiss–Handler pathway**, it is most often recycled through the salvage pathway or generated as a byproduct of metabolic reactions

Glossary

Effector T cell: antigen-activated T cell that drives an immune response.
Epigenetic modification: a change in gene activity resulting in a stable phenotypic alteration; does not involve a change to the DNA sequence of a gene.
Exhausted: the status of a T cell when it loses its capacity for killing aberrant cells such as cancer cells or virus-infected cells after being active.
Fatty acid oxidation (FAO): the mitochondrial pathway that degrades fatty acids to acetyl-CoA.
Forkhead box P3 (FoxP3): immune cell transcription factor and master regulator of Treg cells.
Glutaminolysis: the process of converting glutamine into glutamate and TCA metabolites.
Glyceraldehyde 3-phosphate dehydrogenase (GAPDH): the enzyme that catalyzes the sixth step of glycolysis in which glyceraldehyde 3-phosphate is converted to 1,3-biphosphoglycerate.
Histone (H) proteins: the protein component of nucleosomes that is wrapped around by DNA strands to form chromatin.
Kynurenine pathway: the tryptophan catalytic pathway and NAD synthesis pathway.
Memory T cells: antigen-specific T cells that provide long-term memory (e.g., reoccurrence of infection).
Metabolic reprogramming: the process whereby a cell alters its metabolism to meet the augmented energy demands necessary for growth, proliferation, and function.
Methylenetetrahydrofolate dehydrogenase 2 (MTHFD2): the mitochondrial one-carbon (1C) metabolic enzyme that converts methylene tetrahydrofolate (THF) into formyl-THF.
Naive T cells: mature T cells that have exited the thymus but have not yet encountered an antigen.
Oxidative phosphorylation (OXPHOS): the process of ATP generation that is coupled to the electron transport chain; requires oxygen consumption.
3-Phosphoglycerate dehydrogenase (PHGDH): the enzyme of the *de novo* serine synthesis pathway that catalyzes the conversion of 3-phosphoglycerate to 3-phosphohydroxypyruvate.
Post-translational modification (PTM): covalent addition to or cleavage

linked to the electron transport chain (ETC) or lactate fermentation (Figure 3) [40]. In addition to its redox role, however, NAD^+ is an essential cofactor for the class III HDAC family of proteins known as sirtuins [33]. In mammalian cells there are seven sirtuins (Sirt1–7) that have specific cellular localizations and functions; these characteristics were recently reviewed elsewhere [41].

Sirtuins play a crucial role in the functional regulation of T cells both *in vitro* and *in vivo* [41]. For instance, in murine and human Treg cells, **forkhead box P3 (FoxP3)** activity and stability are induced by acetylation [30,31]. When Sirt1 deacetylates this master TF, Treg suppressive function is reduced [30,31]. Similarly, **T-bet** expression that promotes murine $CD8^+$ T cell effector differentiation and survival depends on histone acetylation, which is blocked by Sirt1 [32]. Sirt3 or Sirt5 deletion (*Sirt3*$^{-/-}$ or *Sirt5*$^{-/-}$ mice) in $CD4^+$ T cells inhibits OXPHOS or T cell receptor (TCR) signaling, respectively, thereby reducing Treg suppressive capacity; by contrast, using siRNA knockdown, Sirt4 inhibition promoted OXPHOS [42–44]. Lastly, Sirt2 impaired the transcription of glycolytic, TCA, FAO, and glutaminolytic enzymes, thus limiting the proliferation and effector functions of tumor-infiltrating $CD8^+$ T cells in humans, as well as in mouse models of solid primary and metastatic tumors (*Sirt2*$^{-/-}$ mice and non-small cell lung human and mouse models) [45]. These studies point to the important role of sirtuins in $CD4^+$ and $CD8^+$ T cell regulation, as well as the targeting potential of these proteins in therapeutics.

From another angle, because diet affects the NAD^+/NADH ratio, it is known to influence histone deacetylation (and thus gene expression). For instance, calorie restriction in mice raises NAD^+ concentrations in brown and white adipose tissue, as well as deacetylation [46]. Conversely, high-fat diets and obesity correlate with increased nicotinamide *N*-methyltransferase (NNMT) activity, which simultaneously reduces the levels of SAM and NAD^+. This decrease in methylation and increase in acetylation induces the transcription of proinflammatory genes such as *Il6*, *Tnfa*, and leptin (*Lep*) [46].

Moreover, a high-lactate environment has different effects on histone acetylation in different cell types. In murine effector $CD4^+$ T cells, high lactate reduces the NAD^+/NADH ratio, which in turn inhibits **glyceraldehyde 3-phosphate dehydrogenase (GAPDH)** and **3-phosphoglycerate dehydrogenase (PHGDH)** function *in vivo* and *in vitro* [47]. Glycolysis and glucose-dependent serine synthesis are consequently blocked, impairing T cell proliferation; these effects can be reversed upon dietary serine supplementation *in vivo* [47]. By contrast, murine Tregs *in vivo* respond to high lactate by inhibiting Myc expression and by inducing OXPHOS, which increases the cellular NAD^+/NADH ratio; Tregs are thus protected in an environment, presumably such as a tumor niche, in which effector T cells may suffer from NAD^+ depletion and lose functional activity [48].

Taken together, these studies point towards modulation of NAD^+ metabolism as a possible indirect means of altering epigenetic modifications; in turn, it might be harnessed to potentiate or inhibit the function of a particular T cell subset.

Other histone modifications

In addition to classical histone modifications such as methylation and acetylation, new histone PTMs have now surfaced. Histone lactylation is the addition of LDH-derived lactate onto lysines, a modification that facilitates the induction of homeostatic genes such as *Arg1* and *Nos2* during the so-called proinflammatory M1-like macrophage polarization [49,50].

Histones may also be phosphorylated by kinases and dephosphorylated by phosphatases. For example, phosphorylation at serine 10 of histone H3 can alter chromatin structure and regulate gene transcription; however, this process requires the interaction of chromatin with other histone PTMs such as histone acetylation [51,52].

of a modifying group on a specific amino acid of a protein.
Preiss–Handler pathway: a NAD synthesis pathway that relies on dietary nicotinic acid.
Pyruvate carboxylase (PC): the mitochondrial enzyme that carboxylates pyruvate to generate oxaloacetate.
Pyruvate dehydrogenase (PDH): that mitochondrial enzyme complex that converts pyruvate into acetyl-CoA.
Regulatory T (Treg) cells: a specialized subset of $CD4^+$ T cells that maintain immune homeostasis and tolerance by suppressing, for example, the activity of effector T cells.
T-bet: an immune cell transcription factor and master regulator of Th1 and $CD8^+$ T cells.
Tricarboxylic acid (TCA) cycle: the mitochondrial metabolic pathway that carries out acetyl-CoA oxidation for the generation of ATP and reductive equivalents.
Tumor microenvironment (TME): the cells, tissue, and molecules that surround tumor cells.
Type 17 T helper (Th17) cells: a specialized subset of $CD4^+$ T cells that are involved in host protection against extracellular bacteria and fungi; Th17 cells are known drivers of autoimmune diseases such as multiple sclerosis, rheumatoid arthritis, and psoriasis.

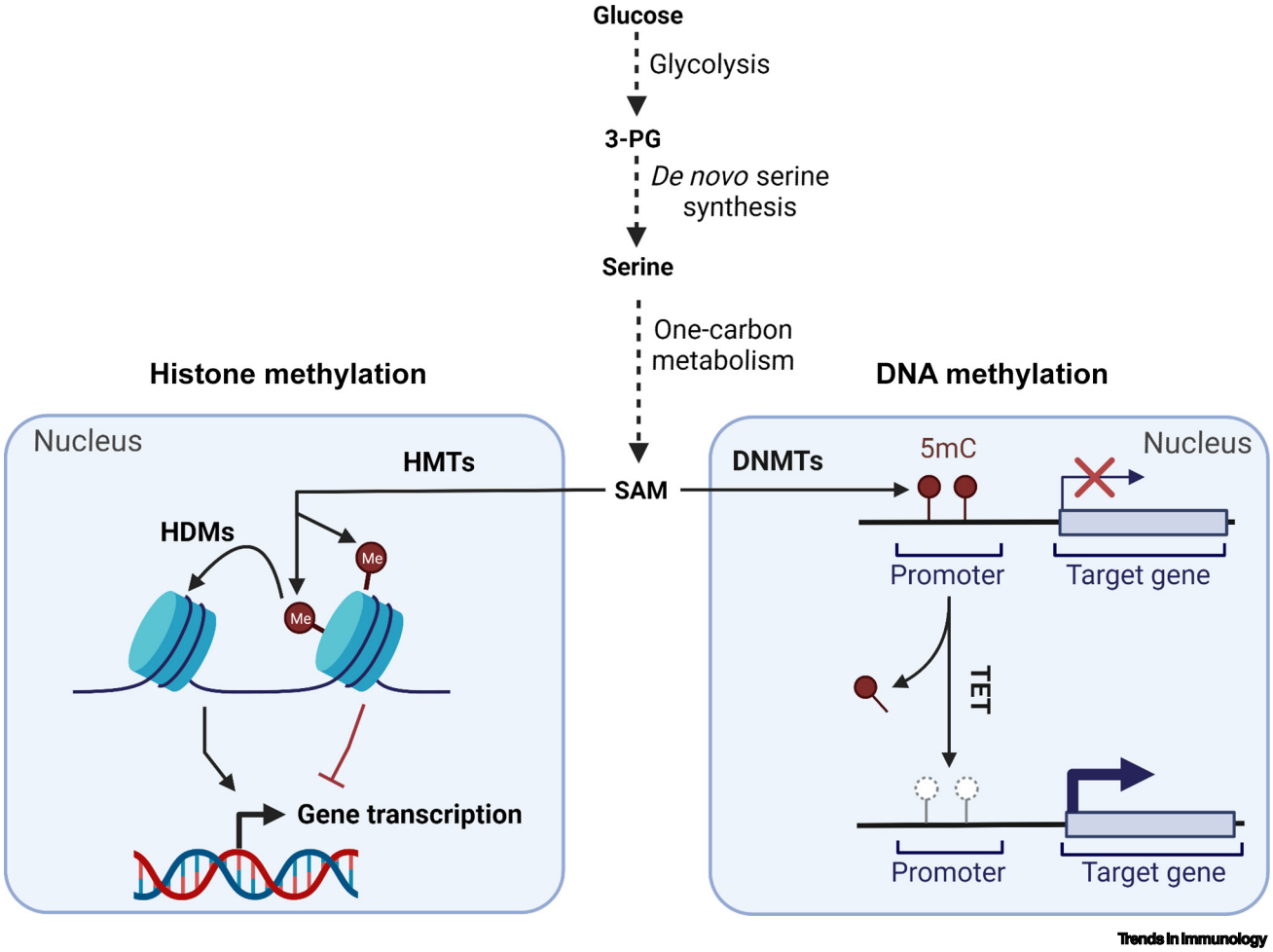

Figure 1. Histone and DNA methylation. The glucose-derived intermediate 3-phosphoglycerate (3-PG) is metabolized through the *de novo* serine synthesis pathway to generate serine [91]. The latter can then be further metabolized via one-carbon metabolism to generate *S*-adenosylmethionine (SAM), which is the main methyl donor for post-translational modifications [13,14]. On the one hand (left), SAM is used by histone methyltransferases (HMTs) for histone methylation, which represses gene transcription [12]. These methyl groups on histones can be removed by histone demethylases (HDMs) to activate transcription [12]. On the other hand (right), DNA methyltransferases (DNMTs) utilize the methyl group provided by SAM to methylate DNA at cytosine residues, generating 5-methylcytosine (5mC) [11]. When a promoter region contains 5mC residues, transcription of that gene is inhibited [11]. These methyl groups can be removed by ten-eleven translocation (TET) proteins to restore gene transcription [11]. This figure was created using BioRender.com.

Another non-conventional PTM is histone crotonylation, which is the addition of a crotonyl group to lysines by HATs [53]. The crotonyl group is a 4C chain obtained from crotonyl-CoA, which is an intermediate metabolite in lysine and tryptophan metabolism as well as in butyrate fermentation. Histone lysine crotonylation was first described as an alternative way to decompact chromatin and allow it to interact with DNA-binding factors [53–55]. A similar newly identified modification is histone methacrylation, discovered in HeLa cells by mass spectrometry [56]. Histone methacrylation is the addition of a methacryl group to lysines in a process regulated by HAT1 and Sirt2 [56]. The methacryl group is a structural isomer of the crotonyl group [54].

A different histone PTM is citrullination, which is catalyzed by peptidyl arginine deiminase (PAD) enzymes and has been connected to autoimmune diseases such as rheumatoid arthritis and systemic lupus erythematosus in humans [57]. In addition, histone succinylation, where a succinyl

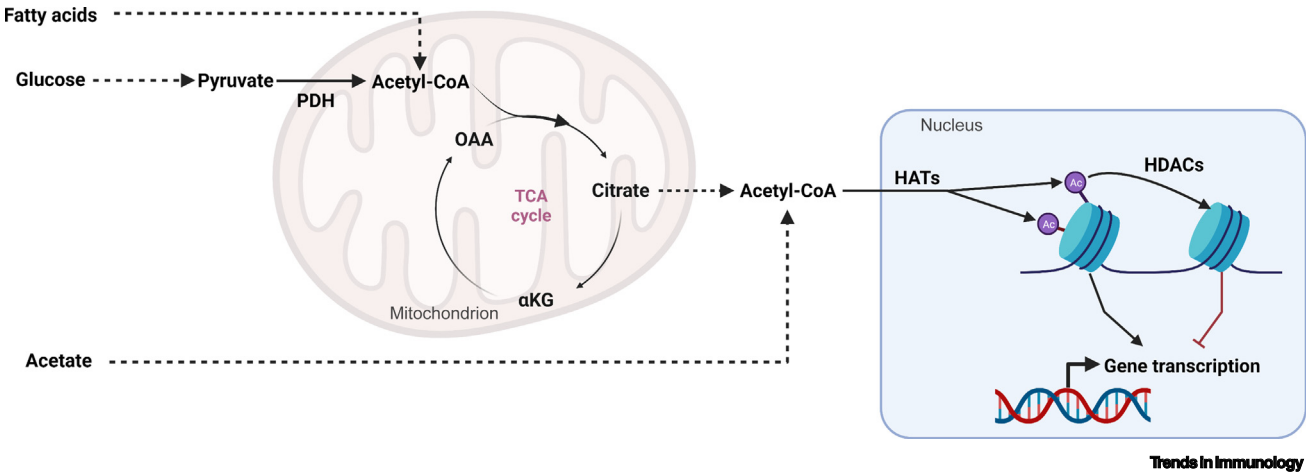

Figure 2. Histone acetylation. Histone acetylation is an epigenetic modification that relaxes chromatin condensation, opening it up for transcription [11]. Histone acetyltransferases (HATs) in the nucleus use acetyl-coenzyme A (acetyl-CoA) as their main donor of acetyl groups for histone acetylation [26]. This modification can be reversed by histone deacetylases (HDACs) [27]. The acetyl-CoA used by HATs can be derived from various pathways [92]. Glucose metabolized by glycolysis in the cytosol generates its end-product pyruvate, which is then imported into the mitochondria and converted to acetyl-CoA by pyruvate dehydrogenase (PDH) [1]. This acetyl-CoA enters the tricarboxylic acid (TCA) cycle and combines with oxaloacetate (OAA) to produce citrate [1]. Citrate can exit mitochondria and be converted back into OAA and acetyl-CoA by ATP citrate lyase in the cytosol or the nucleus [93]. This nuclear acetyl-CoA can be used by HATs for histone acetylation [26]. A similar source of acetyl-CoA is the β-oxidation of fatty acids in mitochondria [94]. Lastly, acetyl-CoA in the cytosol can be generated by acetyl-CoA synthetase from cytosolic acetate [95]. Abbreviation: α-KG, α-ketoglutarate. This figure was created using BioRender.com.

group is added to histone lysines, was recently described. Histone succinylation is mainly found at transcriptional start-sites and promotes gene transcription [58]. Finally, *N*-acetylglucosamine (GlcNAc) is added to serine or threonine residues of histones, a process known as histone GlcNAcylation. GlcNAc is generated in the hexosamine biosynthetic pathway and, depending on the residue it is attached to, the modification induces or represses gene transcription [59].

It should be noted that, as intriguing as they are, the roles of these newly identified histone PTMs in T cells remain unknown; however, they certainly merit further investigation. Taken together, these lines of research suggest that deliberate manipulation of histone modifications in T cells and other immune cell types is worth pursuing as a putative therapeutic approach for some pathologies.

DNA modifications

DNA methylation contributes to the maintenance of genome stability. DNA methyltransferases (DNMTs) use SAM as a methyl donor to add methyl groups to cytosine to generate 5-methylcytosine (5mC), which generally represses gene expression. Conversely, members of the 'ten-eleven translocation' (TET) protein family are α-KG-dependent DNA demethylases that can activate gene expression by demethylating 5mC residues to 5hmC (Figure 1, right) [11].

During the differentiation of activated $CD8^+$ and $CD4^+$ T cells into effector cells, the DNA methylation of promoters of effector genes such as *Il2*, *Tnfa*, and *Ifng* is altered and may decline progressively [60]. When $CD8^+$ effector T cells experience prolonged exposure to antigen, they may become **exhausted** and undergo *de novo* DNA methylation at loci such as *Pdcd1* (encoding the immune checkpoint inhibitor PD-1); this is a process that inhibits their effector function, and abrogates their ability to eradicate aberrant cells (i.e., cytotoxicity) [60]. Blocking *de novo* DNA methylation in mouse $CD8^+$ T cells, in combination with immune checkpoint blockade (ICB) treatment [61], can limit the 'exhaustion' phenotype such that better control of tumor growth can be achieved *in vivo*

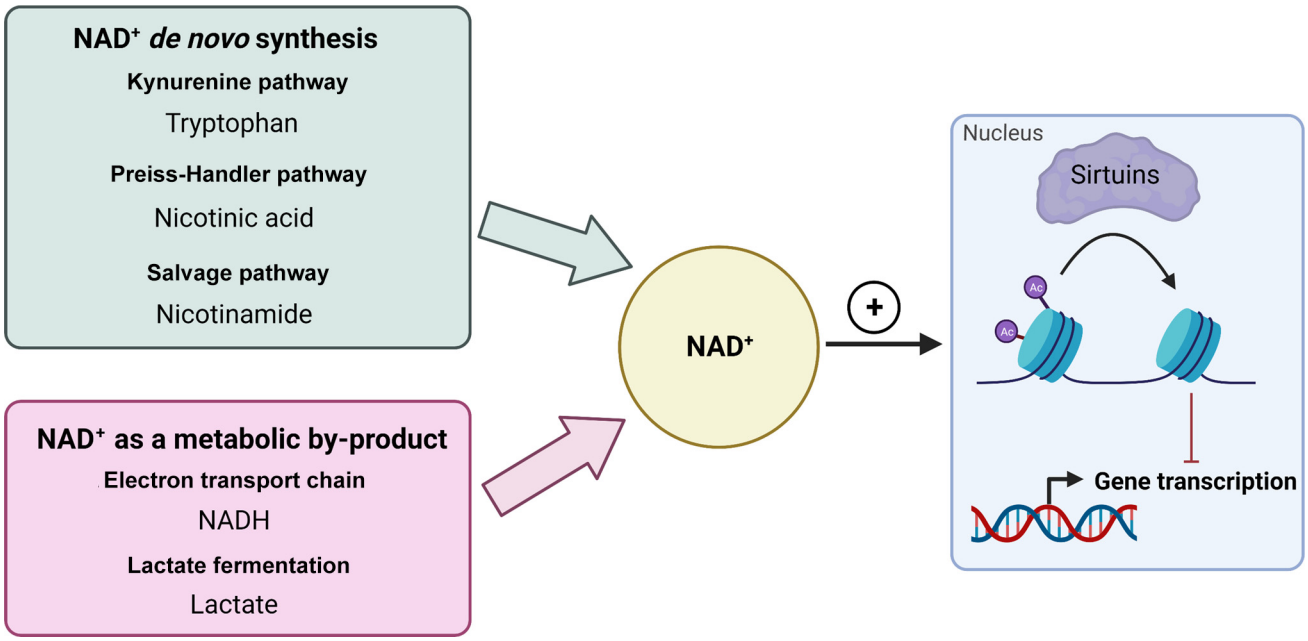

Figure 3. NAD^+ metabolism and sirtuin regulation. NAD^+ can be generated *de novo* through either the kynurenine pathway starting from tryptophan, the Preiss–Handler pathway by consuming dietary nicotinic acid, and by the salvage pathway starting from nicotinamide [40]. However, NAD^+ is also generated as a byproduct of metabolic pathways such as the electron transport chain which consumes NADH to pump protons into the mitochondrial intermembrane space and transfers electrons between complexes [40]. NAD^+ is also produced during lactate fermentation, during which NADH is oxidized [40]. NAD^+ is an essential cofactor for class III HDACs, called sirtuins, which promote histone deacetylation and limit gene transcription [41]. This figure was created using BioRender.com.

for tumor types such as the TRAMP-C2 prostate adenocarcinoma model [60]. Along the same lines, TET2-deficient murine $CD8^+$ T cells (*Tet2*$^{fl/fl}$*Cd4–cre*$^+$ P14 mice) acquire a memory phenotype earlier than wild-type $CD8^+$ T cells do, thus becoming more efficient during acute reinfection with lymphocytic choriomeningitis virus (LCMV Armstrong) [62]. These data reinforce earlier findings that manipulating DNA methylation can have profound positive effects on $CD8^+$ T cell function.

Regarding $CD4^+$ T cells, a recent study showed that naive murine $CD4^+$ T cell supplementation with exogenous cell-permeable α-KG changed their DNA methylation status and induced the cells to differentiate into Tregs *in vitro* [63]. These epigenomic modifications caused the T cells to increase their proinflammatory cytokine production (e.g., IFN-γ, TNF, GM-CSF, and IL-17A) and reduced their *Foxp3* expression, thus impairing Treg cell differentiation. As a result, chimeric antigen receptor T cells (CAR-T) treated *ex vivo* with α-KG developed increased tumor-infiltrating capacity that delayed tumor growth in fibrosarcoma-bearing mice [63].

In another study, 2-HG, which acts as a α-KG antagonist, promoted the differentiation of naive murine $CD4^+$ T cells into Th17 cells while blocking Treg generation by inducing *Foxp3* hypermethylation, thus preventing its transcription [64]. This effect was reversed upon inhibition of the conversion of glutamate into α-KG, reducing 2-HG accumulation. The *Foxp3* gene was then transcribed, inhibiting Th17 differentiation and generating Tregs [64]. Similarly, succinate and fumarate, which (like α-KG) are TCA cycle metabolites, can influence $CD4^+$ T cell differentiation because they are potent inhibitors of TET enzymes [65]. Indeed, supplementation of either of these metabolites in culture increased the methylation status and expression of hypoxia-inducible genes such as those involved in glycolysis (e.g., *Pgk1*, *Hk2*, and *Eno1*) *in vitro* [59]. Together, these studies

suggest that the concentrations of TCA metabolites are crucial for the differentiation and function of T cells.

Accordingly, increasing evidence suggests that DNA methylation status is a key determinant of T cell differentiation and function. Therefore, altering the amounts of substrates that modulate DNMTs and TET proteins in different tissues might have significant effects on T cell homeostasis as well as on disease.

Metabolites, enzymes, and gene regulation

The primary function of metabolic enzymes is the generation and conversion of metabolites. However, some of these enzymes and metabolites can carry out 'extra-metabolic' roles that have direct modulatory effects on T cell gene expression.

Serine, reactive oxygen species, and FoxP3 expression

The importance of the correct redox balance in murine effector $CD4^+$ T cells to potentiate the metabolic rewiring necessary for normal effector function was previously established [39]. Our group further showed that glutathione governs serine and 1C metabolism in murine Tregs in a manner that is crucial for *Foxp3* expression and Treg suppressive capacity both *in vivo* and *in vitro* [6]. This work uncovered a previously unknown connection between redox balance, 1C metabolism, and suppressive function that is specific to this $CD4^+$ T cell subset [6]. However, preliminary data suggest that, although reactive oxygen species (ROS) and 1C metabolism can be increased in glutathione-depleted Treg cells, methylation of the *Foxp3* promoter and Treg-specific demethylated regions (TSDRs) might remain unaltered, although this remains to be robustly validated (our unpublished observations) [6]. Collectively, these studies describe the connection between redox balance and $CD4^+$ T cell metabolism and function, and how alterations in one of these processes may affect the other. Of note, genetic ablation of complex III (RISP knockout mice) in Tregs interferes with their suppressive activity, but this is independent of FoxP3 [66]. In this situation, loss of complex III increased DNA methylation and the metabolites 2-HG and succinate relative to controls, and this is relevant because they can inhibit DNA demethylases [66].

MTHFD2 in Th17 and Treg regulation

A recent study showed that the 1C metabolism enzyme **methylenetetrahydrofolate dehydrogenase 2 (MTHFD2)** is essential for *de novo* purine synthesis and mTORC1 activity, and thus for T cell proliferation and cytokine production in mice [10]. Furthermore, in Th17 cells, MTHFD2 methylated the *Foxp3* locus, thereby limiting FoxP3 upregulation and Treg transdifferentiation [10]. Upon MTHFD2 inhibition or deletion, mTORC1 activity was reduced, inducing increased OXPHOS but decreased succinate and fumarate [10]. These TCA metabolites normally inhibit DNA and histone demethylation, and decreases in succinate/fumarate concentrations therefore reduced general methylation in the epigenome, enabling *Foxp3* expression and Treg transdifferentiation *in vitro* and *in vivo* [10]. This study pinpoints MTHFD2 as a potential therapeutic target for the treatment of inflammatory diseases such as multiple sclerosis and inflammatory bowel disease, although this possibility remains to be robustly investigated.

PKM2 in Th17 cells

The generation of pyruvate at the end of glycolysis is mediated by pyruvate kinase (PK). The M2 subtype (of four subtypes) of PK (PKM2) is required for the development of Th1 and Th17 cells in mice via a mechanism in which PKM2 dimerizes and translocates into the nucleus of a naive $CD4^+$ T cell that has experienced TCR engagement [67]. The Th1 and Th17 gene expression programs (e.g., *Tbx21* and *Rorc* expression, respectively) are subsequently induced in mice [67]. Blocking PKM2 translocation by forcing its tetramerization impairs Th1 and Th17 polarization such that

mice are protected from developing severe symptoms in the EAE model [67]. An independent study confirmed that PKM2 in mice interacts in the nucleus with STAT3, an essential TF for Th17 cell differentiation; moreover, T cell-specific PKM2 deletion (*Cd4*^{cre}*Pkm2*$^{fl/fl}$ mice) blocks Th17 differentiation and attenuates EAE symptoms [68]. PKM2 might therefore represent a potential therapeutic target for the treatment of Th17 cell-driven autoimmune diseases such as multiple sclerosis.

GAPDH and IFN-γ production

GAPDH is a crucial enzyme in glycolysis [69]. Nonetheless, when the glycolytic rate is low, GAPDH can inhibit the translation of *Ifng* mRNA in effector CD4$^+$ T cells by binding to it [70,71]. Once glycolysis is upregulated, however, GAPDH releases the mRNA, which permits IFN-γ production [70,71]. Another study demonstrated that T cell-specific overexpression of GAPDH in mice activates NF-κB via the non-canonical pathway and leads to the development of tumors mimicking human angioimmunoblastic T cell lymphoma [72]. Thus, GAPDH expression requires tight regulation to enable the proper function of T cells.

β-Hydroxybutyrate and memory CD8$^+$ T cell formation

β-Hydroxybutyrate (βOHB) is a ketone body generated from acetyl-CoA by the ketogenic pathway in the liver during fasting or calorie restriction in mammals [73]. In mice, βOHB inhibits class I HDACs and thereby increases global histone acetylation, specifically inducing the transcription of antioxidative genes essential for CD8$^+$ memory T cell formation and maintenance *in vivo* and *in vitro* [74,75]. This may represent an alternative route for potential modulation of CD8$^+$ T cell memory which might be relevant for recurrent infections [74,75].

Lactate and CD8$^+$ T cell effector functions

In the **tumor microenvironment (TME)**, LDH-derived lactate is significantly increased and can have different effects on antitumor CD8$^+$ T cells [76]. Lactate limits CD8$^+$ T cell function by impairing upregulation of the TF NFAT [77]. Moreover, in mice, high lactate concentrations in the TME can also drive CD8$^+$ T cell metabolism towards the mitochondrial oxidation of pyruvate by PDH; this reduces the antitumor cytotoxic function of these cells *in vivo* when mice are subcutaneously injected with B16F10 melanoma cells [78]. However, when PDH is blocked *in vivo*, **pyruvate carboxylase (PC)** activity in CD8$^+$ T cells is induced, driving succinate secretion and activation of succinate receptor 1 (SUCNR1) and downstream targets; these events promote the antitumor function of CD8$^+$ T cells [78]. However, increased lactate also impairs HDAC activity, resulting in augmented histone acetylation at the *Tcf7* locus and enhancing Tcf-1 expression in murine CD8$^+$ T cells [79]. In response, stem cell-like CD8$^+$ T cells are generated which exhibit more efficient antitumor activity against MC38 tumors *in vivo* than terminally differentiated CD8$^+$ T cells [79]. These studies suggest that lactate is an interesting target in cancer therapy, but they also indicate that we should be cautious in manipulating this pathway.

Arginine and polyamines in T cell differentiation and function

Recently, metabolic pathways of amino acids as potential targets, especially arginine, and also polyamines, have been investigated in inflammatory diseases. For instance, *in vitro* treatment of murine and human CD4$^+$ T cells with L-arginine induces their survival and promotes the antitumor activity of CD8$^+$ T cells in a melanoma mouse model *in vivo* [80]. In human and murine CD4$^+$ T cell cultures, the arginine-derived polyamine spermidine can promote Treg differentiation, and this has been confirmed *in vivo* in a T cell adoptive transfer-induced colitis model [81]. Moreover, arginine metabolism is essential for removal of ammonia through the urea and citrulline cycle, which then fosters the development of memory CD8$^+$ T cells *in vivo* in mice, as well as *in vitro* [82].

Diet and microbiome influence T cell function

As previously stated, serine supplementation and calorie restrictions impact on the metabolism and function of T cells, which highlights the role of diet in immunomodulation [6,45,46]. Furthermore, changes in diet have effects on the gut microbiome which can impact on the mucosal immune system [83]. Thus, the connection between the microbiome and T cell activity is important for the function of the immune system, as recently described elsewhere [84–87]. In particular, the signaling function of short-chain fatty acids (SCFAs) such as acetate, propionate, butyrate, and pentanoate has gained significant attention in recent years. These SCFAs are produced by the gut microbiota, and their therapeutic potential in cancer and autoimmunity is intensively being investigated [88,89].

Collectively, studies such as these indicate that metabolic enzymes and molecules play important 'extra-metabolic' roles, and their modulation in T cells can affect the epigenetic landscape as well as the differentiation and function of these T cell subsets. Targeting metabolic enzymes or metabolites (by either potentiating or inhibiting their activity) could pave the road for developing putative strategies to modulate T cell differentiation and function in disease-relevant scenarios. However, this will require in-depth investigations, especially in the human context.

Concluding remarks

We have described recent studies that further our knowledge on the interconnection between T cell metabolism and epigenetic modifications. We perceive three target areas in which molecular perturbations might open new avenues for candidate treatments of T cell-related diseases such as autoimmune diseases and cancer: histone modifications, DNA methylation, and metabolism/metabolic enzyme pathways. In Table 1 (Key table), we summarize the specific metabolites and enzymes discussed in this review.

Regarding histone modifications, we have described the concept that acetyl-CoA metabolism is essential for the acetylation of histones and other proteins. Modulating acetyl-CoA pools by supplementing acetate or increasing the glycolytic function of T cells might be used to alter T cell function in a specific cellular compartment. An elevated concentration of acetyl-CoA provides an increased substrate for HATs, and HAT activity promotes effector T cell gene expression and function [90]. Such an approach is currently under investigation as a potential immunotherapeutic treatment for different cancers [7,9,36,37,79]. Conversely, limiting glucose flux into the TCA cycle reduces histone acetylation in Th17 cells, and this might be beneficial for therapies involving Th17 cell-associated autoimmune diseases such as multiple sclerosis [7, 8]. It will also be interesting to determine whether additional histone PTMs, such as lysine lactylation, crotonylation, methacrylation, as well as serine phosphorylation, citrullination, succinylation, and GlcNAcylation [49,52,53,56–59], might be exploited in T cells for therapeutic benefit (see Outstanding questions).

Manipulation of DNA methylation is a double-edged sword because this process is not only crucial for genome stability but can also contribute to T cell exhaustion, as well as to other T cell processes. Finding ways to limit DNA methylation at particular loci while preserving genomic stability might prolong some antitumor responses [60]. To combat particular infectious diseases, one approach might be to block specific HDMs to promote the memory $CD8^+$ T cell responses necessary to thwart pathogen reinfection [62].

In addition, as described already, modulating the concentrations of particular metabolic enzymes and/or TCA cycle metabolites can affect both the metabolism and epigenetic landscape of T cells. For instance, targeting the glutamate pathway to inhibit its conversion to α-KG can induce FoxP3

Outstanding questions

What are the various roles of distinct histone modifications in T cells? Not much is known about non-classical histone modifications, and even less about their possible impact on T cells and immunity.

What are the signaling pathways that specifically foster such histone modifications, and how are these pathways connected to metabolism? Understanding these might further increase our knowledge about the complex regulation of epigenetics and how it is interconnected with metabolism and cellular function.

How is the specificity of epigenetic changes regulated in T cells? Why are only specific gene groups targeted by several epigenetic changes? Epigenetic changes are cell type-specific or even T cell subtype-specific, although metabolic shifts are similar, for example, an increase in acetyl-CoA due to increased glycolysis. Detailed knowledge of such mechanisms would be important for targeting the functionality of different T cell populations in health and disease.

How do different epigenetic modifications operate/cross-operate at the histone and DNA levels? Such interactions might further increase the complexity of gene regulation and specificity. In this context, non-classical modifications should also be considered because these might be crucial for the epigenetic specificity of T cell subsets.

Key table

Table 1. Modulation points that alter the epigenome and function of T cells[a,b]

Modulation point	T cell subset	Mechanism	Effect on function	Refs
Modulation of methylation				
α-Ketoglutarate	Effector $CD4^+$ T cells	Activates HDM and demethylation	Proinflammatory effector function	[63]
JMJD3	Effector $CD4^+$ T cells	Regulates demethylation	Promotes Th1 cell differentiation and impairs Th2 and Th17 cell differentiation	[22]
2-HG	$CD8^+$ T cells	Inhibits HDM	Increases proliferation, persistence, and antitumor function	[24]
	$CD8^+$ T cells	Inhibits LDH	Impairs cytotoxicity and antitumor function	[25]
	$CD4^+$ T cells	Promotes *Foxp3* locus hypermethylation	Induces Th17 differentiation	[64]
Succinate and fumarate	Effector $CD4^+$ T cells	Inhibit TET proteins	Induce glycolytic gene expression	[65]
Modulation of acetylation				
Glut3	Th17 cells	Increases the acetyl CoA pool and histone acetylation	Increases effector function	[7]
PDH	Th17 cells	Generates a citrate pool for proliferation and histone acetylation	Increases effector functions	[8]
MPC	$CD8^+$ T cells	Decreases the roles of glutamine/fatty acids in acetyl-CoA generation	Suppresses memory differentiation	[36]
Acetate	$CD8^+$ T cells	Increases the acetyl-CoA pool and histone acetylation	Increased effector function	[9]
LDH	Th1 and Th17 cells	Increases the acetyl-CoA pool and histone acetylation	Increases effector function	[37,38]
Metabolites and enzymes modulating epigenetics and T cell function				
Glutathione	Effector $CD4^+$ T cells	Redox balance	Functional	[39]
	Tregs	Regulates serine metabolism	Functional	[6]
MTHFD2	Th17 cells	Increases succinate/fumarate	Limits Treg transdifferentiation	[10]
PKM2	Th17 cells	Translocates to the nucleus and binds to STAT3	Promotes Th17 function	[67,68]
Lactate	Effector $CD4^+$ T cells	Blocks glycolysis and serine synthesis by altering the NAD^+/NADH ratio	Impairs effector proliferation	[47]
	Tregs		Promotes Treg function	[48]
	$CD8^+$ T cells	Impairs NFAT upregulation and induces PDH function	Blocks cytotoxicity	[78]
	$CD8^+$ T cells	Impairs HDAC activity	Impairs effector proliferation	[79]
GAPDH	Effector $CD4^+$ T cells	Glycolytic enzyme and *Ifng* mRNA binding	Induces IFN-γ production upon increased glycolysis	[70,71]
			Overexpression in T cells causes T cell lymphoma	[72]
βOHB	$CD8^+$ T cells	Inhibits class I HDACs	Induces memory formation	[74,75]
Arginine	Effector $CD4^+$ T cells	Shift from glycolysis to OXPHOS and allows ammonia removal	Induces survival	[80]
	$CD8^+$ T cells		Promotes antitumor function and memory development	[80,82]
Spermidine	$CD4^+$ T cells	Promotes autophagy signaling	Promotes Treg development	[81]

[a]Mainly derived from murine studies.
[b]Abbreviations: FoxP3, forkhead box P3; GAPDH, glyceraldehyde-3-phosphate dehydrogenase; Glut3, glucose transporter 3; HDAC, histone deacetylase; HDM, histone demethylase; 2-HG, 2-hydroxyglutarate; IFN-γ, interferon-γ; JMJD3, Jumonji domain-containing protein 3; LDH, lactate dehydrogenase; MPC, mitochondrial pyruvate carrier; MTHFD2, methylenetetrahydrofolate dehydrogenase 2; NFAT, nuclear factor of activated T cells; βOHB, β-hydroxybutyrate; OXPHOS, oxidative phosphorylation; PDH, pyruvate dehydrogenase; PKM2, pyruvate kinase isoform M2; STAT3, signal transducer and activator of transcription 3; TET, ten-eleven translocation; Th, T helper cell; Treg, regulatory T cell.

expression, thus limiting Th17 differentiation in favor of Treg polarization [64]. Similarly, MTHFD2 inhibition can block Th17 differentiation and induce Tregs [10]. Finally, inhibition of PKM2 might contribute to the treatment of proinflammatory diseases such as multiple sclerosis [67,68].

Clearly, a thorough investigation of these signaling pathways is warranted. Nevertheless, we anticipate that the targeted application of such approaches might contribute to mitigating T cell-associated autoimmune and inflammatory diseases such as multiple sclerosis and cancers.

Two related potential targets are NAD^+ and serine metabolism. High lactate causes a change in the NAD^+/NADH ratio that impairs the function of effector T cells, whereas it promotes the suppressive capacity of Tregs [47,48]. Thus, blocking high lactate concentrations might help to prolong antitumor T cell responses against some cancers. Similarly, serine supplementation can protect effector T cell function but inhibit FoxP3 expression and Treg-mediated suppression, which might be beneficial for some cancer therapeutic strategies [6,47]. Antitumor activity might also be augmented by inhibiting PDH, thus increasing succinate in $CD8^+$ T cells [78]. These studies demonstrate that metabolic manipulations have different regulatory effects on different T cell subsets, which might enable precision-targeted therapies.

Achieving a fine balance in the regulation of these enzymes is necessary. For example, although low glycolytic activity inhibits IFN-γ production through GAPDH, GAPDH overexpression promotes T cell lymphoma generation in mice [70,72].

Noteworthy, most results described herein were obtained using mouse models. This basic research is crucial and represents the first step in translating some of these fundamental findings into the clinic. Nonetheless, studies at the interface between metabolism and epigenetics should ultimately provide insights into potential treatments for T cell-associated disorders in the years to come.

Acknowledgments

We apologize to researchers whose work could not be referenced here owing to space limitations. We thank Anouk Ewen for the design of the figures using BioRender.com. L.S-B. and D.B. are supported by the Fonds National de la Recherche Luxembourg (FNR; PRIDE/11012546/NEXTIMMUNE). D.B. is supported by FNR CORE grants (C21/BM/15796788 and C18/BM/12691266).

Declaration of interests

The authors declare no conflicts of interest.

References

1. Buck, M.D. *et al.* (2015) T cell metabolism drives immunity. *J. Exp. Med.* 212, 1345–1360
2. Franchina, D.G. *et al.* (2018) Reactive oxygen species: involvement in T cell signaling and metabolism. *Trends Immunol.* 39, 489–502
3. Guerra, L. *et al.* (2020) Metabolic modulation of immunity: a new concept in cancer immunotherapy. *Cell Rep.* 32, 107848
4. Kurniawan, H. *et al.* (2020) Regulatory T cell metabolism at the intersection between autoimmune diseases and cancer. *Eur. J. Immunol.* 50, 1626–1642
5. Rangel Rivera, G.O. *et al.* (2021) Fundamentals of T cell metabolism and strategies to enhance cancer immunotherapy. *Front. Immunol.* 12, 645242
6. Kurniawan, H. *et al.* (2020) Glutathione restricts serine metabolism to preserve regulatory T cell function. *Cell Metab.* 31, 920–936
7. Hochrein, S.M. *et al.* (2022) The glucose transporter GLUT3 controls T helper 17 cell responses through glycolytic-epigenetic reprogramming. *Cell Metab.* 34, 516–532.e11
8. Soriano-Baguet, L. *et al.* (2023) Pyruvate dehydrogenase fuels a critical citrate pool that is essential for Th17 cell effector function. *Cell Rep.* 42 (2)
9. Qiu, J. *et al.* (2019) Acetate promotes T cell effector function during glucose restriction. *Cell Rep* 27, 2063–2074
10. Sugiura, A. *et al.* (2022) MTHFD2 is a metabolic checkpoint controlling effector and regulatory T cell fate and function. *Immunity* 55, 65–81
11. Dutta, A. *et al.* (2021) New insights into epigenetic regulation of T cell differentiation. *Cells* 10, 3459
12. Rhodes, C.T. and Lin, C.-H.A. (2023) Role of the histone methyltransferases Ezh2 and Suv4-20h1/Suv4-20h2 in neurogenesis. *Neural Regen. Res.* 18, 469–473
13. Ducker, G.S. and Rabinowitz, J.D. (2017) One-carbon metabolism in health and disease. *Cell Metab.* 25, 27–42
14. Kurniawan, H. *et al.* (2021) The emerging role of one-carbon metabolism in T cells. *Curr. Opin. Biotechnol.* 68, 193–201
15. Ma, E.H. *et al.* (2017) Serine is an essential metabolite for effector T cell expansion. *Cell Metab.* 25, 345–357

16. Meiser, J. *et al.* (2016) Serine one-carbon catabolism with formate overflow. *Sci. Adv.* 2, e1601273
17. Tran, K.A. *et al.* (2019) The role of α-ketoglutarate-dependent proteins in pluripotency acquisition and maintenance. *J. Biol. Chem.* 294, 5408–5419
18. Li, J. *et al.* (2021) KDM6B-dependent chromatin remodeling underpins effective virus-specific $CD8^+$ T cell differentiation. *Cell Rep.* 34, 108839
19. Xu, T. *et al.* (2021) Kdm6b regulates the generation of effector $CD8^+$ T cells by inducing chromatin accessibility in effector-associated genes. *J. Immunol.* 206, 2170–2183
20. Russ, B.E. *et al.* (2017) Regulation of H3K4me3 at transcriptional enhancers characterizes acquisition of virus-specific $CD8^+$ T cell-lineage-specific function. *Cell Rep.* 21, 3624–3636
21. LaMere, S.A. *et al.* (2017) H3K27 methylation dynamics during CD4 T cell activation: regulation of JAK/STAT and IL12RB2 expression by JMJD3. *J. Immunol.* 199, 3158–3175
22. Li, Q. *et al.* (2014) Critical role of histone demethylase Jmjd3 in the regulation of $CD4^+$ T-cell differentiation. *Nat. Commun.* 5, 5780
23. Carbonneau, M. *et al.* (2016) The oncometabolite 2-hydroxyglutarate activates the mTOR signalling pathway. *Nat. Commun.* 7, 12700
24. Tyrakis, P.A. *et al.* (2016) The immunometabolite S-2-hydroxyglutarate regulates $CD8^+$ T-lymphocyte fate. *Nature* 540, 236–241
25. Notarangelo, G. *et al.* (2022) Oncometabolite D-2HG alters T cell metabolism to impair $CD8^+$ T cell function. *Science* 377, 1519–1529
26. Sun, X.-J. *et al.* (2015) The role of histone acetyltransferases in normal and malignant hematopoiesis. *Front. Oncol.* 5, 108
27. Ellmeier, W. and Seiser, C. (2018) Histone deacetylase function in $CD4^+$ T cells. *Nat. Rev. Immunol.* 18, 617–634
28. Liotti, A. *et al.* (2022) Epigenetics: an opportunity to shape innate and adaptive immune responses. *Immunology* 167, 451–470
29. Glozak, M.A. *et al.* (2005) Acetylation and deacetylation of non-histone proteins. *Gene* 363, 15–23
30. Kwon, H.-S. *et al.* (2012) Three novel acetylation sites in the Foxp3 transcription factor regulate the suppressive activity of regulatory T cells. *J. Immunol.* 188, 2712–2721
31. van Loosdregt, J. *et al.* (2010) Regulation of Treg functionality by acetylation-mediated Foxp3 protein stabilization. *Blood* 115, 965–974
32. Kuroda, S. *et al.* (2011) Basic leucine zipper transcription factor, ATF-like (BATF) regulates epigenetically and energetically effector CD8 T-cell differentiation via Sirt1 expression. *Proc. Natl. Acad. Sci. U. S. A.* 108, 14885–14889
33. Moller, S.H. *et al.* (2022) Metabolic programs tailor T cell immunity in viral infection, cancer, and aging. *Cell Metab.* 34, 378–395
34. Bricker, D.K. *et al.* (2012) A mitochondrial pyruvate carrier required for pyruvate uptake in yeast, *Drosophila*, and humans. *Science* 337, 96–100
35. Herzig, S. *et al.* (2012) Identification and functional expression of the mitochondrial pyruvate carrier. *Science* 337, 93–96
36. Wenes, M. *et al.* (2022) The mitochondrial pyruvate carrier regulates memory T cell differentiation and antitumor function. *Cell Metab.* 34, 731–746
37. Peng, M. *et al.* (2016) Aerobic glycolysis promotes T helper 1 cell differentiation through an epigenetic mechanism. *Science* 354, 481–484
38. Xu, K. *et al.* (2021) Glycolytic ATP fuels phosphoinositide 3-kinase signaling to support effector T helper 17 cell responses. *Immunity* 54, 976–987
39. Mak, T.W. *et al.* (2017) Glutathione primes T cell metabolism for inflammation. *Immunity* 46, 675–689
40. Xie, N. *et al.* (2020) NAD^+ metabolism: pathophysiologic mechanisms and therapeutic potential. *Signal. Transduct. Target. Ther.* 5, 227
41. Hamaidi, I. and Kim, S. (2022) Sirtuins are crucial regulators of T cell metabolism and functions. *Exp. Mol. Med.* 54, 207–215
42. Beier, U.H. *et al.* (2015) Essential role of mitochondrial energy metabolism in $Foxp3^+$ T-regulatory cell function and allograft survival. *FASEB J.* 29, 2315–2326
43. Lin, W. *et al.* (2019) Sirtuin4 suppresses the anti-neuroinflammatory activity of infiltrating regulatory T cells in the traumatically injured spinal cord. *Immunology* 158, 362–374
44. Wang, K. *et al.* (2020) SIRT5 contributes to colorectal cancer growth by regulating T cell activity. *J Immunol Res* 2020, 3792409
45. Hamaidi, I. *et al.* (2020) Sirt2 inhibition enhances metabolic fitness and effector functions of tumor-reactive T cells. *Cell Metab.* 32, 420–436
46. Wei, X. *et al.* (2020) Depot-specific regulation of NAD^+/SIRTs metabolism identified in adipose tissue of mice in response to high-fat diet feeding or calorie restriction. *J. Nutr. Biochem.* 80, 108377
47. Quinn, W.J. *et al.* (2020) Lactate limits T cell proliferation via the NAD(H) redox state. *Cell Rep.* 33, 108500
48. Angelin, A. *et al.* (2017) Foxp3 reprograms T cell metabolism to function in low-glucose, high-lactate environments. *Cell Metab.* 25, 1282–1293
49. Zhang, D. *et al.* (2019) Metabolic regulation of gene expression by histone lactylation. *Nature* 574, 575–580
50. Chen, A.-N. *et al.* (2021) Lactylation, a novel metabolic reprogramming code: current status and prospects. *Front. Immunol.* 12, 688910
51. Wang, Z. *et al.* (2016) Histone posttranslational modifications of $CD4^+$ T cell in autoimmune diseases. *Int. J. Mol. Sci.* 17, 1547
52. Alaskhar Alhamwe, B. *et al.* (2018) Histone modifications and their role in epigenetics of atopy and allergic diseases. *Allergy Asthma Clin. Immunol.* 14, 39
53. Tan, M. *et al.* (2011) Identification of 67 histone marks and histone lysine crotonylation as a new type of histone modification. *Cell* 146, 1016–1028
54. Ntorla, A. and Burgoyne, J.R. (2021) The regulation and function of histone crotonylation. *Front. Cell Dev. Biol.* 9, 624914
55. Jiang, G. *et al.* (2021) Protein lysine crotonylation: past, present, perspective. *Cell Death Dis.* 12, 703
56. Delaney, K. *et al.* (2021) Histone lysine methacrylation is a dynamic post-translational modification regulated by HAT1 and SIRT2. *Cell Discov.* 7, 122
57. Zhu, D. *et al.* (2021) Histone citrullination: a new target for tumors. *Mol. Cancer* 20, 90
58. Liu, J. *et al.* (2021) Histone succinylation and its function on the nucleosome. *J. Cell. Mol. Med.* 25, 7101–7109
59. Dehennaut, V. *et al.* (2014) O-GlcNAcylation, an epigenetic mark. Focus on the histone code, TET family proteins, and Polycomb group proteins. *Front. Endocrinol.* 5, 155
60. Ghoneim, H.E. *et al.* (2017) De novo epigenetic programs inhibit PD-1 blockade-mediated T cell rejuvenation. *Cell* 170, 142–157
61. Korman, A.J. *et al.* (2022) The foundations of immune checkpoint blockade and the ipilimumab approval decennial. *Nat. Rev. Drug Discov.* 21, 509–528
62. Carty, S.A. *et al.* (2018) The loss of TET2 promotes $CD8^+$ T cell memory differentiation. *J. Immunol.* 200, 82–91
63. Matias, M.I. *et al.* (2021) Regulatory T cell differentiation is controlled by αKG-induced alterations in mitochondrial metabolism and lipid homeostasis. *Cell Rep.* 37, 109911
64. Xu, T. *et al.* (2017) Metabolic control of TH17 and induced Treg cell balance by an epigenetic mechanism. *Nature* 548, 228–233
65. Laukka, T. *et al.* (2016) Fumarate and succinate regulate expression of hypoxia-inducible genes via TET enzymes. *J. Biol. Chem.* 291, 4256–4265
66. Weinberg, S.E. *et al.* (2019) Mitochondrial complex III is essential for suppressive function of regulatory T cells. *Nature* 565, 495–499
67. Angiari, S. *et al.* (2020) Pharmacological activation of pyruvate kinase M2 inhibits $CD4^+$ T cell pathogenicity and suppresses autoimmunity. *Cell Metab.* 31, 391–405
68. Damasceno, L.E.A. *et al.* (2020) PKM2 promotes Th17 cell differentiation and autoimmune inflammation by fine-tuning STAT3 activation. *J. Exp. Med.* 217, e20190613
69. Almeida, L. *et al.* (2016) Metabolic pathways in T cell activation and lineage differentiation. *Semin. Immunol.* 28, 514–524
70. Chang, C.-H. *et al.* (2013) Posttranscriptional control of T cell effector function by aerobic glycolysis. *Cell* 153, 1239–1251
71. Shyer, J.A. *et al.* (2020) Metabolic signaling in T cells. *Cell Res.* 30, 649–659
72. Mondragón, L. *et al.* (2019) GAPDH overexpression in the T cell lineage promotes angioimmunoblastic T cell lymphoma through an NF-κB-dependent mechanism. *Cancer Cell* 36, 268–287
73. Newman, J.C. and Verdin, E. (2017) β-Hydroxybutyrate. *Annu. Rev. Nutr.* 37, 51–76
74. Shimazu, T. *et al.* (2013) Suppression of oxidative stress by β-hydroxybutyrate, an endogenous histone deacetylase inhibitor. *Science* 339, 211–214

75. Zhang, H. *et al.* (2020) Ketogenesis-generated β-hydroxybutyrate is an epigenetic regulator of CD8$^+$ T-cell memory development. *Nat. Cell Biol.* 22, 18–25
76. Apostolova, P. and Pearce, E.L. (2022) Lactic acid and lactate: revisiting the physiological roles in the tumor microenvironment. *Trends Immunol.* 43, 969–977
77. Brand, A. *et al.* (2016) LDHA-associated lactic acid production blunts tumor immunosurveillance by T and NK cells. *Cell Metab.* 24, 657–671
78. Elia, I. *et al.* (2022) Tumor cells dictate anti-tumor immune responses by altering pyruvate utilization and succinate signaling in CD8$^+$ T cells. *Cell Metab.* 34, 1137–1150
79. Feng, Q. *et al.* (2022) Lactate increases stemness of CD8$^+$ T cells to augment anti-tumor immunity. *Nat. Commun.* 13, 4981
80. Geiger, R. *et al.* (2016) L-arginine modulates T cell metabolism and enhances survival and anti-tumor activity. *Cell* 167, 829–842
81. Carriche, G.M. *et al.* (2021) Regulating T-cell differentiation through the polyamine spermidine. *J. Allergy Clin. Immunol.* 147, 335–348
82. Tang, K. *et al.* (2023) Ammonia detoxification promotes CD8$^+$ T cell memory development by urea and citrulline cycles. *Nat. Immunol.* 24, 162–173
83. Leeming, E.R. *et al.* (2019) Effect of diet on the gut microbiota: rethinking intervention duration. *Nutrients* 11, 2862
84. Ivanov, I.I. *et al.* (2022) T cell responses to the microbiota. *Annu. Rev. Immunol.* 40, 559–587
85. Collins, N. (2020) Dietary regulation of memory T cells. *Int. J. Mol. Sci.* 21, 4363
86. Tan, J. *et al.* (2022) Your regulatory T cells are what you eat: how diet and gut microbiota affect regulatory T cell development. *Front. Nutr.* 9, 878382
87. Arroyo Hornero, R. *et al.* (2020) The impact of dietary components on regulatory T cells and disease. *Front. Immunol.* 11, 253
88. Kim, C.H. *et al.* (2014) Gut microbiota-derived short-chain fatty acids, T cells, and inflammation. *Immune Netw.* 14, 277–288
89. Schiweck, C. *et al.* (2022) Regulation of CD4$^+$ and CD8$^+$ T cell biology by short-chain fatty acids and its relevance for autoimmune pathology. *Int. J. Mol. Sci.* 23, 8272
90. Yerinde, C. *et al.* (2019) Metabolic control of epigenetics and its role in CD8$^+$ T cell differentiation and function. *Front. Immunol.* 10, 2718
91. Wu, Q. *et al.* (2020) Serine and metabolism regulation: a novel mechanism in antitumor immunity and senescence. *Aging Dis.* 11, 1640–1653
92. Pietrocola, F. *et al.* (2015) Acetyl coenzyme A: a central metabolite and second messenger. *Cell Metab.* 21, 805–821
93. Wellen, K.E. *et al.* (2009) ATP-citrate lyase links cellular metabolism to histone acetylation. *Science* 324, 1076–1080
94. Lochner, M. *et al.* (2015) Fatty acid metabolism in the regulation of T cell function. *Trends Immunol.* 36, 81–91
95. Moffett, J.R. *et al.* (2020) Acetate revisited: a key biomolecule at the nexus of metabolism, epigenetics and oncogenesis – part 1: acetyl-CoA, acetogenesis and acyl-CoA short-chain synthetases. *Front. Physiol.* 11, 580167