Tet2 at the interface between cancer and immunity

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Keeping a balance between DNA methylation and demethylation balance is central for mammalian development and cell function, particularly in the hematopoietic system. In various mammalian cells, Tet methylcytosine dioxygenase 2 (Tet2) catalyzes oxygen transfer to a methyl group of 5-methylcytosine (5mC), yielding 5-hydroxymethylcytocine (5hmC). Tet2 mutations drive tumorigenesis in several blood cancers as well as in solid cancers. Here I discuss recent studies that elucidate mechanisms and biological consequences of Tet2 dysregulation in blood cancers. I focus on recent findings concerning Tet2 involvement in lymphoid and myeloid cell development and its functional roles, which may be associated with tumorigenesis. I also discuss how Tet2 activities are modulated by microRNAs, metabolites, and other interactors, including vitamin C and 2-hydroxyglutarate (2-HG), and review the clinical relevance and potential therapeutic applications of Tet2 targeting. Finally, I propose key unanswered hypotheses regarding Tet2 in the cancer-immunity cycle.

DNA methylation/demethylation is dynamically coordinated throughout hematopoietic differentiation, and Tet proteins play crucial roles in adjusting gene expression levels through balancing DNA methylation in hematopoesis and during immune-cell activation and expansion. The first identified gene of the Tet family, Tet1, acts as a fusion regulator in cases of acute myeloid leukemia (AML) and acute lymphocytic leukemia (ALL). Two other Tet genes, Tet2 and Tet3, were later identified based on sequence homology with Tet1. In mammalian cells, all Tet family members, Tet1, Tet2, and Tet3, catalyze the successive oxidation of 5mC, yielding 5hmC, 5-formylcytosine (5fC), and 5-carboxylcytosine (5caC).

The Tet2 gene is subjected to frequent somatic mutations in an extensive range of hematopoietic cancers, including myeloid and lymphoid cancer, and several solid cancers. DNA modification by Tet2 is fundamental for gene control in both cancer cells and immune-cell subtypes. Tet2 protein and its downstream effectors-5hmC/5mC-mediated DNA modification are constitutional in immune cells, including T cells, B cells, and macrophages in both physiological and pathological conditions (Fig. 1). In blood cancer cells, Tet2 loss is the primary cause of 5mC generation, whereas loss of functional roles of all three Tets is robustly linked to solid cancer progression. Tet2 is one of the commonly mutated genes in hematopoietic cancers. In engineered mouse models, Tet2 knockout (KO), or double KO (DKO) of Tet2 and Tet3, causes myeloid or lymphoid cell spread and the progression of fully infiltrating aggressive tumors.
This review discusses Tet2-related profiles in hematopoietic cancers, different subtypes of adaptive and innate immune cells, and Tet2 regulation by various interactors. I particularly emphasize the potential applications of targeting Tet2 to influence normal and malignant cancer-immunity crosstalk.

Roles of Tet2 in hematopoiesis, immune-cell lineages, and hematopoietic malignancies

Tet2 in hematopoiesis. Tet2 epigenetically controls gene expression by modulating methylation-driven gene silencing and is expressed in diverse populations of mouse and human hematopoietic cells. Tet2 mRNA is expressed in hematopoietic cell subpopulations, including progenitors and mature immune cells, displaying detectable levels of 5hmC and 5mC. In mice, Tet2 deletion enhances the Lin–Sca-1–c-Kit+ (LSK) cell subpopulation, which exhibits boosted hematopoietic repopulation competence and biased cell differentiation toward monocyte/macrophage lineages, followed by myeloid cancer progression. In vitro tissue culture work supports that RNA interference (RNAi)-mediated Tet2 silencing in murine hematopoietic precursors can alter their differentiation toward monocyte/macrophage lineages. In mice, Tet2 is pivotal for modulating normal hematopoiesis, and negatively influences hematopoietic stem cell (HSC) homeostasis and differentiation. Tet2 loss reshapes the HSC niche and HSC proliferation/differentiation potential in vitro, whereas Tet2-KO HSCs show elevated ability to reconstitute hematopoiesis in vivo. Tet2 deletion results in reduced genomic 5hmC levels and expands the hematopoietic progenitor cell population in a cell-intrinsic manner. Tet2-KO HSCs are capable of multilineage reconstitution and show a competitive advantage over wild-type HSCs, promoting increased hematopoiesis into myeloid and lymphoid lineages. Tet2 disorder also results in DNA hypermethylation of enhancers in granulocyte–monocyte progenitor (GMP) and embryonic stem (ES) cells. Overall, Tet2 plays vital roles in modulating HSC expansion and function, presumably by controlling 5hmC levels in genes crucial for HSC self-renewal, proliferation, and differentiation, which warrants future examination of the detailed mechanisms.

Tet2 in myeloid lineages and myeloid malignancies. In humans, Tet2 mutations occur in ~10–30% of myeloid cancers, including myelodysplastic syndromes (MDS), chronic myelomonocytic leukemia (CML), and myeloproliferative neoplasm (MPN). Tet2 mutations may be caused by splice site mutations, out-of-frame insertions or deletions and base substitutions. Tet2-KO mice recapitulate characteristics of patients with myeloid cancer, indicating that Tet2 plays a functional role as a tumor suppressor to sustain hematopoietic cell homeostasis. Tet2-KO mice show a phenotype that is similar to CML, including splenomegaly and enhanced white blood cell (WBC) counts with unequal numbers of monocytes. Moreover, Tet2-KO mice develop an extensive spectrum of myeloid cancers, which occur within the first year of life in ~30% of these animals. Mechanistically, Tet2 blocks leukemic transformation by keeping enhancers free of aberrant DNA methylation that would lead to enhanced stem cell expansion and leukemogenesis. RNAi-mediated Tet2 silencing reduces 5hmC levels in cord blood CD34+ cells and promotes progenitor differentiation toward the granulomonocytic lineage. CD34+ stem cells from MPN patients with Tet2 mutations reconstitute hematopoiesis in nonobese diabetic/severe combined immunodeficiency (NOD/SCID) mice and display skewed differentiation toward myeloid lineages.

Tet2 in lymphoid lineages and lymphoid malignancies. Tet2 mutations are also common in both T-cell and B-cell lymphomas, with a 40–70% incidence observed among patients with peripheral T-cell lymphoma (PTCL), particularly in subsets of angioimmunoblastic T-cell lymphoma (AITL). Adult T-cell leukemia/lymphoma (ATLL) progression accompanies genomic 5hmC loss, suggesting that Tet2 downregulation is the vital mechanism underlying 5hmC regulation during ATLL progression. Tet2-KO mice develop B-cell tumors, and B-cell conditional Tet2 knockout (cKO) mice show B1-cell subset abnormalities and B-cell tumor development after long latency. Tet2 deletion may prompt alterations of peripheral B-cell subsets. Certain Tet2-KO cancers have AID-mediated somatic mutations. Future work is required to investigate whether and how dysregulation of Tet2 impacts on B-cell activation upon various stimuli. Young Tet2-KO mice exhibit a moderately enhanced B1a subset population in the spleen. On the other hand, aging Tet2-KO mice expand clonal CD19+B220low immunoglobulin (Ig)M+B-cell subpopulations, which are transplantable and display similarities to chronic lymphocytic leukemia (CLL), including sensitivity to ibrutinib-induced B-cell receptor
signaling blockage\textsuperscript{24}. Further investigations are needed to examine the molecular signaling pathways controlling B1a-cell development and proliferation in young adult Tet2-KO mice.

In mice, attenuated Tet2 function results in T-cell lymphoma with follicular helper T-cell-like (Tfh) features\textsuperscript{25}. Compared with control mice, middle-aged Tet2 gene trap mice (Tet2\textsuperscript{GFP}) show Tfh-like cell over-expansion in the spleen\textsuperscript{25}. Aging Tet2\textsuperscript{GFP} mice eventually generate T-cell lymphoma with Tfh-like features\textsuperscript{25}. In future research, it will be intriguing to examine how Tet2 works in Tfh cells. A mouse model with conditional Tet2-KO (cKO) in Tfh cells would be valuable for in vivo and in vitro analyses.

Both Tet1 and Tet3 are expressed in hematopoietic cells, without alterations in Tet2-KO cells, demonstrating that Tet1/Tet3 is inadequate to compensate for Tet2 deletion. It will be interesting to dissect their physiological and pathological roles in various immune-cell subsets. Tet2 and Tet3 may cooperate to inhibit abnormal hematopoiesis, including hematopoietic transformation. Further investigations are necessary to test whether and how Tet2 works together with other Tet proteins in distinct blood cancer cells. Deletion of Tet2 in hematopoietic cells or various immune-cell subsets might prompt a specific and progressive enhancer hypermethylation feature linked to altered gene expression, enhanced cell accumulation rate, and leukemia progression, which also warrants future investigation. A better understanding of Tet proteins' physiological and pathological functional roles may highlight potential novel avenues to develop epigenetic therapies to treat hematological cancers.

Intriguingly, in mice, the combined deletion of Tet1 and Tet2 apparently blocks B-cell cancer, without influencing myeloid cancers\textsuperscript{26}. Tet1 and Tet2 are simultaneously downregulated in B-cell acute lymphoblastic leukemia\textsuperscript{26}. Although double (DcKO) or triple (TcKO) conditional KO mice would be valuable, it might be challenging to conditionally KO all three Tet proteins in certain immune-cell sub-settings. Identifying Tet2-targeted genes/molecules in myelopoiesis and lymphomagenesis will be crucial for future studies aimed to uncover the molecular mechanisms through which Tet2 modulates hematopoiesis and serves as a tumor-suppressor gene in myelopoiesis and T/B-cell lymphomas. It is intriguing to hypothesize that inactivating somatic Tet2 mutations might latently remain in hematopoietic cells for a long period, possibly even years. Specifically targeting Tet2 or its downstream molecular signaling pathways might offer novel therapeutic strategies for blood cancer prevention.

**Roles of Tet2 in anticancer immune responses**

**Tet2 in B-cell and anticancer responses.** Tet2 is also essential for B-cell development and function\textsuperscript{27}. In mice, Tet2 and Tet3 KO at an early B-cell stage halts the transition of pro-B cells to pre-B cells in the bone marrow (BM), and diminishes the rearrangement of Igk locus\textsuperscript{28}. Tet2/Tet3 DKO pro-B cells exhibit enhanced CpG methylation levels at the Ig kappa 3' and distal enhancers\textsuperscript{29}. Tet2/Tet3 modulates the kappa-chains expression level, and the DNA modification status of the Ig kappa locus, both in vivo and in vitro\textsuperscript{29}. In mice, Tet2 deletion causes myeloid leukemia after a long duration. Compared to Tet2 deficiency, Tet1/Tet2 deletion results in B-cell lymphoma with delayed disease progression\textsuperscript{26}. The Tet2/Tet3 deletion in developing B cells leads to B-cell lymphoma development. These mice caused the pathological disorders within six months of age, which is earlier than in Tet1/Tet2-KO mice (\textsim 20 months)\textsuperscript{7}. Moreover, regulatory B cells (Bregs) play central roles in boosting inflammation and carcinogenesis\textsuperscript{30}, but the evidence is lacking regarding whether and how Tet2 protein functions in Bregs both in vitro and in vivo. Continued efforts to investigate the probable anticancer impact of Tet2 in Bregs will guide novel Tet2-based immune therapy. Additionally, in future studies, it would be useful to use B-cell-specific conditional (such as CD19-cKO) Tet2 cKO mice to examine whether and how Tet2 epigenetically affects antibody production in vivo.

**Tet2 in T-cell and anticancer responses.** Tet2 deletion augments CD8\textsuperscript{+} T-cell memory differentiation. Deletion of Tet2 advocates early gain of a memory CD8\textsuperscript{+} T-cell fate, in a cell-intrinsic manner, without affecting antigen-driven cell proliferation or effector behaviour\textsuperscript{31}. After secondary stimulation, Tet2-KO memory CD8\textsuperscript{+} T cells possess remarkable pathogen control capacity, both before and after re-challenge. CD8\textsuperscript{+} T cells with Tet2 conditional KO exhibit full effector functional role upon acute viral infection, and Tet2 deletion enhances memory CD8\textsuperscript{+} T-cell formation\textsuperscript{31}. CD8\textsuperscript{+} cytotoxic T lymphocytes (CTLs) are favored immune cells for treating cancer progression\textsuperscript{32}. Programmed death-1 receptor (PD-1) ligand (PD-L1) and CTL-associated antigen 4 (CTLA-4) are checkpoint receptors that can be targeted to re-activate anticancer CD8\textsuperscript{+} T-cell responses in cancer-immune therapy\textsuperscript{33}. It would be interesting to study whether Tet2 might be involved in CTLA-4 or PD-1 pathway-mediated immune checkpoint inhibition for cancer treatment. Further studies are required to explore whether and how Tet2 acts in CD8\textsuperscript{+} CTLs, and how it cooperates with PD-L1/CTLA-4 to modulate anticancer CD8\textsuperscript{+} T-cell responses.

Tet2\textsuperscript{lox/lox}\textsuperscript{CD4Cre+} mice have intact thymic and peripheral T-cell subpopulations\textsuperscript{34}. Tet2 enhances DNA demethylation and cytokine gene activation, for instance, IL17a, in CD4\textsuperscript{+} T cells. In autoimmune diseases, Tet2 modulates T-cell cytokine production in vivo, and specifically influences IL-10, IFN-g, and IL-17 expression levels\textsuperscript{34}. Tet2 depletion in CD4\textsuperscript{+} T cells is linked to reduced cytokine expression, and diminished p300 recruitment\textsuperscript{34}. Moreover, Tet2/Tet3 DKO CD4\textsuperscript{+} T cells induce pathological diseases in healthy mouse\textsuperscript{35}. Tet2 enhances typical cytokine gene expression level in Th1 and Th17 cells in vitro, and modulates Th1 and Th17 cell differentiation\textsuperscript{34}. Tet2/Tet3 loss in regulatory T cells (Tregs) contributes to effector behave phenotypes. Treg-cell-specific Tet2/Tet3 double conditional KO mice (Tet2\textsuperscript{lox/lox}\textsuperscript{CD4Cre+} DcKO mice) show severe inflammation, resulting in inflammatory diseases, and Tregs from these DcKO mice exhibit adjusted expression levels of Treg-typical cytokine genes and dysregulation of genes involved in cancer progression\textsuperscript{35}. Overall, these studies reveal that both Tet2 and Tet3 maintain Treg stability and homeostasis. Tet2/Tet3 DKO in Tregs enhances T-cell activation. Moreover, Tet2/Tet3 DKO Tregs show an altered cell surface feature and fair hypermethylation of conserved noncoding sequence 2 (CNS2)\textsuperscript{35}. Wild-type (WT) Tregs are not able to regulate autoimmunity progression in Tet2-deficient mice\textsuperscript{35}. Double Tet1/Tet2 deletion also causes Treg inactivation, differentiation, and even autoimmunity diseases\textsuperscript{35}. Notably, Tregs hamper anticancer immune surveillance in a healthy person, and prevent effective antitumor immunity in cancer patients, boosting tumor progression\textsuperscript{37}. Thus, methods for specific targeting Tet2/Tet3 in Treg cells might be valuable for modulating anticancer immune responses.

**Tet2 in iNKT cell and anticancer responses.** Tet2 and Tet3 also work together to restrict invariant natural killer T cells (iNKT cells) proliferation and cell-lineage differentiation\textsuperscript{38}. Tet2/Tet3 DKO mice exhibit impressive iNKT cell proliferation, even at earlier stages. These iNKT cells produce large amounts of cytokines, thus eliciting strong immune responses that prompt
other immune-cell subsets responses, and can activate anticancer cytotoxicity. In Tet2/Tet3 DKO mice, the enhanced iNKT cell numbers and concomitantly higher IL-4 secretion give rise to innate-like CD8+ T cells. Tet2/Tet3 deletion yields converted numbers of iNKT cells and significantly influenced gene expression levels, thus changing the identity and functional role of each cell subpopulation. Tet2/Tet3 DKO iNKT cells show enhanced proliferative abilities, which can explain their in vivo spread. Future immunotherapy methods specifically targeting Tet2/Tet3 to modulate iNKT cell activity could have interdependent consequences and employ strong anticancer immune responses.

Tet2 in innate immune-cell and anticancer responses. Tet2 is further essential for innate immune-cell functional roles. Compared to other Tets, Tet2 is the most profoundly expressed in murine macrophage differentiation. Tet2 is essential for controlling inflammation through recruiting Hdac2, inhibiting IL-6 levels. Tet2-KO macrophages and dendritic cells (DCs) produce more IL-6 in response to bacterial activation in vitro and in vivo. Thus, compared to wild-type (WT) mice, Tet2-KO mice show increased susceptibility to endotoxin-induced shock, DSS-induced colitis and Salmonella infections. Tet2 transcription is induced by LPS activation, likely in a nuclear factor κB (NF-κB)-dependent way. In vitro, Tet2 loss in BMDMs does not influence early LPS-induced gene activities but enhances arginase 1 (Arg1) mRNA level during later activation stages. Tet2 deletion does not dramatically alter alternative macrophage (M2) gene expression levels in response to IL-4 stimuli. Tet2 expression is sustained in differentiated murine macrophages, and Tet2 deletion does not negatively alter markers of macrophage differentiation. Resting Tet2-KO peritoneal macrophages reveal atypical induction of LPS-associated genes, and the Tet2 deficiency peritoneal macrophage features are linked to inflammation across the whole body.

M1 macrophages are essential for tumor cell eradication. Thus, modulating Tet2 expression levels in M1 macrophages might contribute to cancer-immune therapy. Over a 20-week time-course, certain Tet2-KO strains show strong defects in myelopoiesis, leading to features similar to human CMMML. Acute inducible Tet2/Tet3 DKO in HSCs causes the rapid emergence of aggressive myeloid leukemia. Tet2 maintains the immunosuppressive functional role of tumor-infiltrating myeloid cells, promoting the advancement of melanoma and Tet2 deletion in myeloid cells reduces melanoma tumor burden in vivo. In tumor-associated macrophages (TAMs), Tet2 expression is activated along the IL-1R/MylD88 axis. Myeloid-specific Tet2 loss causes higher numbers of tumor-infiltrating T cells. During melanoma growth, Tet2 expression is enhanced in myeloid-derived suppressor cells (MDSCs) and TAMs. Further, Tet2 preserves immunosuppressive gene expression levels in TAMs. Mice with myeloid cell-specific Tet2 deletion exhibit diminished tumor growth, and enhanced tumor-infiltrating T cells; moreover, T-cell depletion suppresses the attenuated tumor progression. Whether Tet2 plays a similar or different role in myeloid cell-mediated tumor growth in cancers other than melanoma awaits further investigation.

Here, I have summarized our current understanding of the functional roles of Tet proteins in B cells, various types of T-cell subsets, and M1/M2 macrophages in both physiological and pathological conditions. Further study is essential to investigate how all Tet proteins and 5hmC/5mC modulate DNA modification, gene expression, and transcriptional networks in various types of innate and adaptive immune cells, and to dissect the underlying molecular mechanisms and potential for cancer-immune therapy in this field.

**Fig. 2 Potential therapeutic strategies related to cancer-immunity, based on interactors, specifically targeting Tet2 activities in immune and cancer cells.** The Tet2 activities might be enhanced by vitamin C and hypoxia treatment through HIF-1a. The Tet2 activities might be halted by blocking a-KG accumulation, selective OGT inhibition, selective IDH2 inhibition, using anti-Fex2 agent, or directly suppressed by microRNAs (e.g., Let-7 and miR-22), Tet2 shRNA lentivirus, and Crispr/cas9 targeting in cancer and immune cells.
for answers. For instance, how do these interactors assist Tet2 dysregulation-mediated cancer transformation as well as anticancer immune responses? Further work is needed to demonstrate the underlying molecular mechanisms. It will also be crucial to determine the relationship between Tet2 interactors in Tet2 dysregulation of the progression of non-hematopoietic cancers and blood cancers. Additionally, it is crucial to investigate whether and how these interactors influence 5hmC quantification, which is a helpful diagnostic and/or prognostic tool in cancer treatment. Manipulation of the activity of Tet2 interactors via small molecules or other modulators would be useful for future cancer-immune therapy.

**Targeting Tet2 for potential cancer-immune therapy**

There may be several potential ways of specifically targeting Tet2 activity in immune-cell subpopulations and cancer cells (Fig. 2). In humans, functional loss of Tet2 plays an extensive potentiating role in immunotherapy against B-cell cancer, which utilizes T cells bearing the anti-CD19 chimeric antigen receptor (CAR). The insertional mutation of one copy of a Tet2 allele causes dysregulation of Tet2 activities, which apparently couples with the other Tet2 allele mutation, ultimately yielding strong anti-tumor properties to the expanded CAR-T cells. Research on Tet2 loss in immune-cell responses and tumor development unveils the significance of DNA methylation in various biological activities, and in the advancement of hematological malignancies and solid tumors. With regards to potential clinical usage, it is hard to precisely target Tet2 for treating cancer progression, due to its inactivation in various cancer cell types. However, Tet2 is still expressed to certain levels in some immune-cell subsets and may be modulated in various manner (Fig. 2). For instance, fumarate hydratase (FH) and succinate dehydrogenase (SDH) inhibitors might be developed to prevent the global effectiveness of metabolic Tet2 inactivation in CD8+ T cells. Notably, the oncometabolite 2-hydroxyxylutarate (2HG), generated by IDH mutation in immune cells, could also halt Tet2 activities in immune cells, (e.g., CD4+ T cells and M1 macrophages), which would trigger less-positive antitumor immune responses. Thus, it might also be valuable for future preclinical studies to investigate the restoration of Tet2 in these immune cells, for instance, by directly adding vitamin C or a-KG. Future work will focus on the accurate targeting of Tet2 in the context of different specific cell types, among both cancer and immune cells.

Investigations in both mouse models and human clinical samples have demonstrated that Tet2 loss is not essential to cause cancer, but rather influences cancer progression. This may be by reason for the behavior of Tet2 in various types of immune-cell subsets, and it might explain why some patients with Tet2 mutations stay healthy, whereas others develop either myeloid or lymphoid cancer. Future studies are needed to examine therapies targeting cytosine modifications, DNA methyltransferase inhibitors, and IDH inhibitors, which might be valuable for future cancer-immune therapy.

**Conclusions and current challenges**

Collectively, Tet2 is critical to the cancer-immunity crosstalk, which is essential for both cancer development and anticancer immune responses (Fig. 1). It appears that the downregulation of 5hmC depends on certain specific cell types. It is expected that elevating Tet2 activities using specific compounds that enhance or potentiate Tet2 functional roles may be broadly useful in cancer treatment and for the modulation of certain anticancer immune-cell responses. To further interpret the underlying molecular mechanisms, it is critical to elucidate the functional roles of Tet2/5hmC regulators, and the different ways of modulating the acquisition/removal of 5hmC in various cell types. This will require additional insights into how Tet2 works, and how it is modulated in various types of mammalian cells, including both cancer and immune cells. The toughest challenge might be determining how to precisely modulate Tet2 in specific cells where it plays either positive or negative roles for antitumor therapy. Future work must also explore how to utilize Tet2 interactors to maintain positive antitumor immune responses and halt negative immune responses against cancers. There is still a lot to grasp regarding the physiological and pathological roles of Tet2 in other aspects (e.g., metabolism) in both immune and cancer cells. The elucidation of these aspects and molecular mechanisms would be an exciting avenue for further study.

References

1. Wu, X. & Zhang, Y. TET-mediated active DNA demethylation: mechanism, function and beyond. Nat. Rev. Genet. 18, 517–534 (2017).
2. Lorsbach, R. B. et al. TET1, a member of a novel protein family, is fused to MLL in acute myeloid leukemia containing the t(10;11)(q22;q23). Leukemia 17, 637–641 (2003).
3. Koivunen, P. & Laukka, T. The TET enzymes. Cell. Mol. Life Sci. 75, 1339–1348 (2018).
4. Ko, M. et al. TET proteins and 5-methylcytosine oxidation in hematological cancers. Immunol. Rev. 263, 6–21 (2015).
5. Pan, F., Weeks, O., Yang, F.-C. & Xu, M. The TET2 interactors and their links to hematological malignancies. JMBB Life 67, 438–445 (2015).
6. Xu, Y.-P. et al. Tumor suppressor TET2 promotes cancer immunity and immunotherapy efficacy. J. Clin. Investig. 129, 4316–4331 (2019).
7. Tsagaratou, A., Liu, C.-W. J., Yue, X. & Rao, A. TET methylcytosine oxides in T cell and B cell development and function. Front. Immunol. 8, 220 (2017).
8. Ficz, G. & Grubben, J. G. Loss of 5-hydroxymethylcytosine in cancer: cause or consequence? Genomics 104, 352–357 (2014).
9. Lio, C.-W. J. & Rao, A. TET enzymes and 5hmC in adaptive and innate immune systems. Front. Immunol. 10, 210 (2019).
10. Solary, E., Bernard, O. A., Tefferi, A., Fuku, F. & Vainchenker, W. The ten-eleven translocation-2 (TET2) gene in hematopoiesis and hematopoietic diseases. Leukemia 28, 485–496 (2014).
11. Kohli, R. M. & Zhang, Y. TET enzymes, TDG and the dynamics of DNA demethylation. Nature 502, 472–479 (2013).
12. Feng, Y., Li, X., Cassidy, K., Zou, Z. & Zhang, X. TET2 function in hematopoietic malignancies, immune regulation, and DNA repair. Front. Oncol. 9, 210 (2019).
13. An, J. et al. Acute loss of TET2 function results in aggressive myeloid cancer in mice. Nat. Commun. 6, 10071 (2015).
14. Li, Z. et al. Deletion of Tet2 in mice leads to dysregulated hematopoietic stem cells and subsequent development of myeloid malignancies. Blood 118, 4599–4591 (2011).
15. Ko, M. et al. Ten-Eleven-Translocation 2 (TET2) negatively regulates homeostasis and differentiation of hematopoietic stem cells in mice. Proc. Natl Acad. Sci. USA 108, 14566–14571 (2011).
16. Pan, F. et al. Tet2 loss leads to hypermutagenicity in haematopoietic stem/progenitor cells. Nat. Commun. 8, 15102 (2017).
17. Moran-Crusio, K. et al. Tet2 loss leads to increased hematopoietic stem cell self-renewal and myeloid transformation. Cancer Cell 20, 11–24 (2011).
18. Pronier, E. et al. Inhibition of TET2-mediated conversion of 5-methylcytosine to 5-hydroxymethylcytosine disturbs erythroid and granulomonocytic differentiation of human hematopoietic progenitors. Blood 118, 2551–2555 (2011).
19. Tefferi, A. et al. Detection of mutant TET2 in myeloid malignancies other than myeloproliferative neoplasms: CMML, MDS, MDS/PMN and AML. Leukemia 23, 1343–1345 (2009).
20. Rasmussen, K. D. et al. Loss of TET2 in hematopoietic cells leads to DNA hypermethylation of active enhancers and induction of leukemogenesis. Genes Dev. 29, 910–922 (2015).
21. Sandell, R. F., Bodickier, R. L. & Feldman, A. L. Genetic landscape and classification of peripheral T cell lymphomas. Curr. Oncol. Rep. 19, 28 (2017).
22. Marçais, A. et al. Adult T cell leukemia aggressiveness correlates with loss of both 5-hydroxymethylcytosine and TET2 expression. Oncotarget 8, 52256–52268 (2017).
23. Moully, E. et al. B-cell tumor development in Tet2-deficient mice. Blood Adv. 2, 703–714 (2018).

24. Quivoron, C. et al. TET proteins regulate the lineage specification in pleiotropic hematopoietic abnormalities in mouse and is a recurrent event during human lymphomagenesis. Cancer Cell 20, 25–38 (2011).

25. Muto, H. et al. Reduced TET2 function leads to T-cell lymphoma with follicular helper T-cell-like features in mice. Blood Cancer J. 4, e264 (2014).

26. Zhao, Z. et al. Combined loss of Tet1 and Tet2 promotes B cell, but not myeloid malignancies, in mice. Cell Rep. 13, 1692–1704 (2013).

27. Dominguez, P. M. et al. TET2 deficiency causes germinal center hyperplasia, impairs plasma cell differentiation, and promotes B-cell lymphomagenesis. Cancer Discov. 8, 1632–1653 (2018).

28. Lio, C.-W. et al. Tet2 and Tet3 cooperate with B-lineage transcription factors to regulate DNA modification and chromatin accessibility. elife 5, e18290 (2016).

29. Orlanski, S. et al. Tissue-specific DNA demethylation is required for proper B-cell differentiation and function. Proc. Natl Acad. Sci. USA 113, 5018–5023 (2016).

30. Zhang, Y., Gallagueti, N. & Rosenblatt, J. D. Regulatory B cells in anti-tumor immunity. Int. Immunol. 27, 521–530 (2015).

31. Carty, S. A. et al. The loss of TET2 promotes CD8+ T cell memory differentiation. J. Immunol. 200, 82–91 (2018).

32. Farhood, B., Najafi, M. & Mortezaee, K. CD8+ cytotoxic T lymphocytes in cancer immunotherapy: a review. J. Cell. Physiol. 234, 8509–8521 (2019).

33. Seidel, J. A., Otsuka, A. & Kabashima, K. Anti-PD-1 and anti-CTLA-4 therapies in cancer: mechanisms of action, efficacy, and limitations. Front. Oncol. 8, 86 (2018).

34. Ichiyama, K. et al. The methylcytosine dioxygenase Tet2 promotes DNA demethylation and activation of cytokine gene expression in T cells. Immunity 42, 613–626 (2015).

35. Yue, X., Lio, C.-W. J., Samaniego-Castruita, D., Li, X. & Rao, A. Loss of TET2 and TET3 in regulatory T cells unleashes effector function. Nat. Commun. 10, 2011 (2019).

36. Yang, R. et al. Hydrogen sulfide promotes Tet1- and Tet2-mediated Foxp3 demethylation to drive regulatory T cell differentiation and maintain immune homeostasis. Immunity 43, 251–263 (2015).

37. Ohue, Y. & Nishikawa, H. Regulatory T (Treg) cells in cancer: can Treg cells be a new therapeutic target? Cancer Sci. 110, 2080–2089 (2019).

38. Tsagatou, A. et al. TET proteins regulate the lineage specification and TCR-mediated expansion of iNKT cells. Nat. Immunol. 18, 45–53 (2017).

39. Pan, W. et al. The DNA methylcytosine dioxygenase Tet2 sustains immunosuppressive function of tumor-infiltrating myeloid cells to promote melanoma progression. Immunity 47, 284–297.e5 (2017).

40. Zhang, Q. et al. Tet2 is required to resolve inflammation by recruiting Hdac2 to specifically repress IL-6. Nature 525, 389–393 (2015).

41. Jiang, S., Yan, W., Wang, S. E. & Baltimore, D. Dual mechanisms of posttranscriptional regulation of Tet2 by Let-7 microRNA in macrophages. Proc. Natl Acad. Sci. USA https://doi.org/10.1073/pnas.1811040116 (2019).

42. Coll, A. H., Snetsinger, B., Buckstein, R., Wells, R. A. & Rauh, M. J. Tet2 restrains inflammatory gene expression in macrophages. Exp. Hematol. 55, 56–70.e13 (2017).

43. Pan, X.-Q. The mechanism of the anticancer function of M1 macrophages and their use in the clinic. Chin. J. Cancer 31, 557–563 (2012).

44. Kernytsky, A. et al. IDH2 mutation-induced histone and DNA hypermethylation is progressively reversed by small-molecule inhibition. Blood 125, 296–303 (2015).

45. Scherm, M. G. et al. miRNA142-3p targets Tet2 and impairs Treg differentiation and stability in models of type 1 diabetes. Nat. Commun. 10, 5697 (2019).

46. Song, S. J. et al. The oncosgenic microRNA miR-22 targets the Tet2 tumor suppressor to promote hematopoietic stem cell self-renewal and transformation. Cell Stem Cell 13, 87–101 (2013).

47. Cheng, J. et al. An extensive network of TET2-targeting microRNAs regulates malignant hematopoiesis. Cell Rep. 5, 471–481 (2013).

48. Yang, X. & Qian, K. Protein O-GlcNAcylation: emerging mechanisms and functions. Nat. Rev. Mol. Cell Biol. 18, 452–465 (2017).

49. Chen, Q., Chen, Y., Bian, C., Fujiki, R. & Yu, X. TET2 promotes histone O-GlcNAcylation during gene transcription. Nature 493, 561–564 (2013).

50. Xiao, M. et al. Inhibition of α-KG-dependent histone and DNA demethylases by fumarate and succinate that are accumulated in mutations of FH and SDH tumor suppressors. Genes Dev. 26, 1326–1338 (2012).

51. Wang, L. et al. TET2 coactivates gene expression through demethylation of enhancers. Sci. Adv. 4, eaau986 (2018).

52. Fischer, A. P. & Miles, S. L. Silencing HIF-1α induces TET2 expression and augments ascorbic acid induced 5-hydroxymethylation of DNA in human metastatic melanoma cells. Biochem. Biophys. Res. Commun. 490, 176–181 (2017).

53. Ma, S. et al. Epigenetic regulator CXCR5 recruits DNA demethylase Tet2 to regulate TLR7/9-elicted IFN response in pDCs. J. Exp. Med. 214, 1471–1491 (2017).

54. Suzuki, T. et al. RUNX1 regulates site specificity of DNA demethylation by recruitment of DNA demethylation machineries in hematopoietic cells. Blood Adv. 1, 1699–1711 (2017).

55. Fraietta, J. A. et al. Disruption of TET2 promotes the therapeutic efficacy of CD19-targeted T cells. Nature 558, 307–312 (2018).