REVIEW

N6-methyladenosine Steers RNA Metabolism and Regulation in Cancer

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Funding information
Guangdong Basic and Applied Basic Research Foundation, Grant/Award Number: 2021A1515010425; National Key R&D Program of China, Grant/Award Number: 2020YFA012403; Ministry of Science and Technology of the People’s Republic of China

Abstract
As one of the most studied ribonucleic acid (RNA) modifications in eukaryotes, N6-methyladenosine (m6A) has been shown to play a predominant role in controlling gene expression and influence physiological and pathological processes such as oncogenesis and tumor progression. Writer and eraser proteins, acting opposite to deposit and remove m6A epigenetic marks, respectively, shape the cellular m6A landscape, while reader proteins preferentially recognize m6A modifications and mediate fate decision of the methylated RNAs, including RNAsynthesis, splicing, exportation, translation, and stability. Therefore, RNA metabolism in cells is greatly influenced by these three classes of m6A regulators. Aberrant expression of m6A regulators has been widely reported in various types of cancer, leading to cancer initiation, progression, and drug resistance. The close links between m6A and cancer shed light on the potential use of m6A methylation and its regulators as prognostic biomarkers and drug targets for cancer therapy. Given the notable effects of m6A in reversing chemoresistance and enhancing immune therapy, it is a promising target for combined therapy. Herein, we summarize the recent discoveries on m6A and its regulators, emphasizing their influences on RNA metabolism, their dysregulation and impacts in diverse malignancies, and discuss the clinical implications of m6A modification in cancer.

KEYWORDS cancer therapy, chemoresistance, immunotherapy, m6A methylation, oncogenesis, prognostic biomarkers, RNA epigenetics, RNA metabolism

1 BACKGROUND

Analogous to deoxyribonucleic acid (DNA) and protein, RNA has more than 100 chemical modifications, which tremendously propels our understanding on gene expression control [1]. The most remarkable RNA modification is N6-methyladenosine (m6A), methylated adenosine at the N6 position, which was first discovered in the 1970s [2, 3]. Although m6A is one of the most abundant messenger RNA (mRNA) modifications in mammals, its significance was not fully acknowledged until the identification of fat mass and obesity-associated protein (FTO) as a...
The timeline of RNA epigenetics. m^6^A was first discovered in the 1970s. In 2011, FTO was identified as an m^6^A demethylase. In 2012, the antibody-based transcriptome-wide sequencing method was developed to obtain m^6^A profiling in the human transcriptome. The first FTO inhibitor was found in the same year. Association of m^6^A with cancer began to be reported in breast cancer and lung cancer in 2016, and the cancer types expanded to AML, GBM, HCC, and pancreatic cancer in 2017. Up to now, m^6^A has been found to play critical roles in most cancer types, and inhibitors against more m^6^A regulators are in development. Abbreviations: RNA, ribonucleic acid; m^6^A, N^6^-methyladenosine; FTO, fat mass and obesity-associated protein; AML, acute myeloid leukemia; GBM, glioblastoma; HCC, hepatocellular carcinoma.

FIGURE 1 The timeline of RNA epigenetics. m^6^A was first discovered in the 1970s. In 2011, FTO was identified as an m^6^A demethylase. In 2012, the antibody-based transcriptome-wide sequencing method was developed to obtain m^6^A profiling in the human transcriptome. The first FTO inhibitor was found in the same year. Association of m^6^A with cancer began to be reported in breast cancer and lung cancer in 2016, and the cancer types expanded to AML, GBM, HCC, and pancreatic cancer in 2017. Up to now, m^6^A has been found to play critical roles in most cancer types, and inhibitors against more m^6^A regulators are in development. Abbreviations: RNA, ribonucleic acid; m^6^A, N^6^-methyladenosine; FTO, fat mass and obesity-associated protein; AML, acute myeloid leukemia; GBM, glioblastoma; HCC, hepatocellular carcinoma.

2.1 m^6^A regulators

The m^6^A modification on mRNA is installed by the m^6^A methyltransferase complex (MTC, also known as m^6^A “writers”). A heterodimer consisting of methyltransferase-like 3 (METTL3) and methyltransferase-like 14 (METTL14) constitutes the core of MTC, in which METTL3 is the catalytic subunit while METTL14 mediates substrate RNA recognition and binding [6–9]. Other essential components of the MTC complex, including Willms tumor 1 associated protein (WTAP), RNA Binding Motif Protein 15 (RBM15), RNA Binding Motif Protein 15B (RBM15B), Zinc Finger CCCH-Type Containing 13 (ZC3H13), and Vir like m^6^A methyltransferase associated (VIRMA), anchor MTC to target RNAs [10–15].

The m^6^A modification is reversible and can be removed by m^6^A demethylases (also known as m^6^A “erasers”). As the first characterized RNA m^6^A demethylase, FTO also has oxidative demethylation activity towards multiple other types of DNA and RNA methylations, including m^3^T, m^3^U, m^6^Am, and m^1^A [16, 17]. Nonetheless, m^6^A is the major physiological substrate of FTO [16]. The alkB homolog 5 (ALKBH5) is the second m^6^A eraser which specifically demethylates RNA m^6^A [18].

The effect of m^6^A on gene expression is mediated by the m^6^A binding proteins, also known as m^6^A “readers”, which selectively interact with methylated RNAs and affect RNA metabolism. There are three well-known families of m^6^A readers, YT521-B homology (YTH) domain family, insulin-like growth factor 2 mRNA-binding
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The function of m^6^A and its machinery in RNA metabolism. The m^6^A modification is installed by m^6^A methyltransferases (Writers), consisting of METTL3/14, WTAP, VIRMA, RBM15/15B, and ZC3H13, and is removed by RNA demethylases (Erasers), including FTO and ALKBH5. The m^6^A reader proteins, including YTHDF1/2, YTHDF1/2/3, IGF2BP1/2/3, hnRNPA2B1/C/G, FMRP, Prrc2a and METTL3, work with m^6^A to participate in RNA synthesis, splicing, exportation, translation, and degradation. Abbreviations: m^6^A, N^6^-methyladenosine; RNA, ribonucleic acid; METTL3/14, methyltransferase-like 3/14; WTAP, Willms tumor 1 associated protein; VIRMA, Vir like m6A methyltransferase associated; RBM15/15B, RNA Binding Motif Protein 15/15B; ZC3H13, Zinc Finger CCCH-Type Containing 13; FTO, fat mass and obesity-associated protein; ALKBH5, alkB homolog 5; YTHDF1/2/3, YTH domain-containing protein 1/2/3; IGF2BP1/2/3, insulin-like growth factor 2 mRNA-binding protein 1/2/3; hnRNPA2B1/C/G, heterogeneous nuclear ribonucleoproteins Bi/C/G; FMRP, Fragile X mental retardation protein; Prrc2a, Proline-Rich Coiled-Coil 2A; carRNAs, chromosome-associated regulatory RNAs; Pol II, polymerase II; mRNAs, messenger RNAs; eIF, eukaryotic translation initiation factor.

YTH domain-containing family protein 1 (YTHDF1), YTH domain-containing family protein 2 (YTHDF2), and YTH domain-containing family protein 3 (YTHDF3), have been identified as direct m^6^A readers harboring m^6^A binding pockets [23–28]. YTHDC1 is localized in the nucleus and regulates RNA splicing and nuclear exportation [23, 24] while cytoplasmic YTHDF1, YTHDF2, YTHDF3, and YTHDC2 modulate RNA decay and translation cooperatively [25–28]. IGF2BPs, on the other hand, preferentially recognize and bind to m^6^A methylated mRNAs to promote their stability and translation [29]. Unlike these two families of m^6^A readers, the HNRNP family members, including heterogeneous nuclear ribonucleoprotein C (hnRNPC) and heterogeneous nuclear ribonucleoprotein G (hnRNPG), recognize their targets through an "m^6^A switch" mechanism in which methylated A on the opposite side of a U-tract alters the structure and accessibility of hairpin RNAs [30, 31].

In short, writers and erasers work together to modulate m^6^A dynamics and maintain its homeostasis in cells, while the activity of readers allows m^6^A to exert its influence in each step of the RNA life cycle.

2.2 m^6^A-mediated precursor mRNA (pre-mRNA) splicing

Splicing is a fundamental step of gene expression regulation by removing introns and joining exons co-transcriptionally. The alternative selection of exons results in the production of multiple mRNA variants and ultimately diverse protein products from a single gene, contributing to proteome diversity. The influence of m^6^A on alternative splicing was described by Dominissini et al. [5] and was further supported by studies showing that METTL3, WTAP, FTO and ALKBH5 all modulated alternative splicing [10, 18, 32]. The m^6^A methylated pre-mRNAs indeed undergo alternative splicing through the activity of YTHDC1, heterogeneous nuclear ribonucleoproteins A2/B1 (hnRNPA2B1), or an "m^6^A switch" mechanism. As a founding member of the YTH domain family, YTHDC1 binds methylated pre-mRNAs and promotes exon inclusion by recruiting splicing factor serine/arginine-rich splicing factor 3 (SRSF3) and repelling serine/arginine-rich splicing factor (SRSF10) [23]. Similarly, hnRNPA2B1 binds to m^6^A-bearing RNA and modulates a subset of METTL3- and m^6^A-mediated alternative splicing events [33]. In the "m^6^A switch" mechanism, m^6^A affects RNA structure and enhances the accessibility of hnRNPC and hnRNPG to the flanking U-tract, while loss of m^6^A or hnRNPC/hnRNPG can alter the splicing pattern of neighbor exons [30, 31].
2.3 | m6A-mediated RNA nuclear exportation

Fully spliced mRNAs are allowed to translocate from the nucleus to the cytoplasm, which is under tight control. The interaction of YTHDC1 and nuclear export adaptor protein SRSF3 facilitates RNA binding to nuclear export factor 1 (NXF1) which assists in nuclear translocation [24]. Fragile X mental retardation protein (FMRF), also known as FMR1, is also required for the Exportin 1 (XPO1)-mediated nuclear export of methylated mRNAs [34, 35]. The functional studies of these reader proteins support the earlier observation that accumulation of polyadenylated (polyA) RNA in the cytoplasm is associated with an increase in m6A methylation by ALKBH5 silencing [18], and the notion that m6A is a determinant for the subcellular location of mRNAs.

2.4 | m6A-modulated RNA stability

The steady level of mRNA is established by a balance between its production and degradation, thus, the stability of mRNA is of great importance on modulating mRNA metabolism and gene expression. Members of the YTH domain family play a crucial role in controlling mRNA metabolismand gene expression. Members of the YTH domain family play a crucial role in controlling the translation and decay of mRNA turnover. As the first well-defined m6A reader, YTH domain family play a crucial role in controlling mRNA metabolism and gene expression. Members of the YTH domain family play a crucial role in controlling the translation and decay of RNA half-life [29, 38-40].

2.5 | m6A-mediated RNA translation

Protein translation, a process in which the genetic codes are translated into amino acid sequences, is also tightly controlled. m6A has been widely reported to be involved in translation regulation. In the canonical cap-dependent translation, YTHDF1 facilitates cap-dependent ribosome recruitment to mRNA by forming a loop structure mediated by eukaryotic translation initiation factor 4G (eIF4G) and the interaction of YTHDF1 with eukaryotic translation initiation factor 3 (eIF3) [26]. YTHDF3 was later proven to have a coordinated translation-promoting function with YTHDF1 [27, 28]. In addition, both IGF2BP proteins and YTHDC2 couple active translation with the prevention of mRNA decay [29, 37]. Notably, METTL3 plays a methyltransferase-independent function to promote translation by interacting with eukaryotic translation initiation factor 3h (eIF3h) and forming mRNA loop machinery [41, 42]. It was also reported that m6A in 5′ untranslated region (5′UTR) of mRNAs or the body of circular RNAs (circRNAs) could promote translation in a cap-independent manner [43, 44].

2.6 | m6A-associated RNA synthesis

Although it was thought that m6A mainly affects gene expression post-transcriptionally, emerging evidence has shown that m6A carries a lot of weight in transcriptional control. Liu et al. [45] reported that METTL3 methylated chromosome-associated regulatory RNAs (carRNAs), while YTHDC1 mediated the nuclear degradation of the methylated carRNAs. Loss of m6A methylation via Mettl3 knockout in mouse embryonic stem cells increased carRNAs levels and therefore facilitated chromatin accessibility and transcription activity [45]. Moreover, m6A on mRNAs could facilitate the open state of corresponding chromatin regions through YTHDC1-mediated recruitment of histone H3 lysine 9 dimethylation (H3K9me2) demethylase lysine demethylase 3B (KDM3B), leading to the removal of the repressive H3K9me2 histone mark and the promotion of transcription [46].

In summary, m6A methylation has been widely associated with every aspect of RNA metabolism and gene expression regulation, attributing to the extensive research
on the identification of m\(^6\)A regulators and the exploration of their functions. We have been updated by rapidly expanding research in this field. For instance, the three homologs of \(YTHDF\), \(YTHDF1\), \(YTHDF2\) and \(YTHDF3\), were deemed to play distinct roles in controlling RNA decay and translation [25–28]. However, a similar function of these three proteins on RNA decay and the compensation effect among them have been revealed very recently in certain cell contexts and bioprocesses, such as the ovarian development of zebrafish and the early development of mice [47–49]. Fully studying the interaction network of m\(^6\)A regulators, including \(YTHDF\) proteins and others, under different contexts will give us a comprehensive insight into the effects of m\(^6\)A modification on RNA metabolism.

3 | ABERRANT m\(^6\)A METHYLATION IN HUMAN CANCERS

Given the importance of m\(^6\)A in controlling RNA metabolism, aberrant methylation usually causes dysregulation of gene expression, including activation of oncogenes and repression of tumor suppressors, which plays fundamental roles in the initiation, development, and progression of various cancer types (Figure 3 and Table 2).

3.1 | Acute myeloid leukemia

Acute myeloid leukemia (AML) is known for its devastating outcome and low 5-year overall survival rate (<40%) in patients aged under 60 years old. It is originated from a disordered clone of hematopoietic stem and progenitor cells (HSPCs), leading to the blockage of myeloid differentiation and the production of leukemic stem cells (LSCs) with self-renewal capacity that dominates the initiation of AML and the development of drug resistance [50]. The impact of \(FTO\) on AML when first discovered linked m\(^6\)A RNA modification to AML [51]. Specifically, the overexpression of \(FTO\) promoted oncogenesis through the demethylation of \(ANK\) and \(RARA\) mRNA transcripts [51]. The oncogenic function of \(FTO\) could be selectively inhibited by R-2-hydroxyglutarate (R-2HG) in isocitrate dehydrogenase (IDH) wild-type AML cells or by small molecular inhibitors FB23-2 and CS1/2 in a broad panel of AML cells [52,53], resulting in the suppression of cell growth, LSC maintenance and immune evasion [52, 54]. Another m\(^6\)A demethylase \(ALKBH5\) was also recently found to play oncogenic roles in AML through the \(KDM4C\) (lysine demethylase 4C)-\(ALKBH5\)-\(AXL\) (tyrosine-protein kinase receptor UFO) and \(ALKBH5\)-m\(^6\)A-\(TACC3\) (transforming acidic coiled-coil containing protein 3) axes [55, 56].

On the other hand, the m\(^6\)A methyltransferase machinery has also been linked to AML. Barbieri et al. [57] reported that \(METTL3\) was recruited to transcriptional start sites by \(CCAAT enhancer-binding protein zeta\) (CEBPZ), thus, promoted m\(^6\)A deposition in the coding region and enhanced the translation of associated mRNA transcripts which helped to maintain leukemic state. Depletion of \(METTL3\) in AML cells restrained translation of \(c\)-MYC, B-cell lymphoma 2 (BCL2), and phosphatase and tensin homolog (PTEN) through m\(^6\)A-mediated effects, leading to accelerated cell differentiation and apoptosis coupled with lower proliferative ability [58]. As another core component of the MTC, \(METTL14\) also plays a critical role in leukemogenesis. The m\(^6\)A
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Modification and stability of transcriptional activator Myb (MYB) and MYC transcripts are under tight control by the SPI1 (transcription factor PU.1)-METTL14 axis during normal hematopoiesis, while elevated expression of METTL14 leads to myeloid malignancy and enhanced self-renewal capacity of leukemia stem cells by m6A-mediated stabilization of MYB and MYC oncogenic transcripts [59].

Apart from m6A erasers and writers, the relationship between an m6A reader, YTHDF2, and leukemogenesis has also been uncovered. Paris et al. [60] reported that a decrease in Ythdf2 resulted in higher stability of the tumor necrosis factor receptor superfamily member 2 (Tnfrsf2) and more apoptosis of LSCs. They further found that depletion of YTHDF2 could promote hematopoietic stem cell (HSC) expansion, making YTHDF2 an additional promising anti-leukemia target. Although m6A writers, erasers and readers are clearly associated with AML, how these regulators cooperate in the network is to be elucidated.

### 3.2 Liver cancer

As the sixth commonly diagnosed cancer, liver carcinoma is the fourth cause of tumor-associated death globally [61]. The current existing challenge of liver cancer lies in the detection of late-stage disease, recurrence, and distant metastasis. Therefore, growing efforts are being made to further understand the underlying mechanisms of liver cancer development and progression from various aspects, including RNA epigenetics.

By examination of m6A level in paired tumor and adjacent tissues, Ma et al. [62] found that m6A levels of polyA RNAs were decreased in hepatocellular carcinoma (HCC), the most common type of primary liver cancer. Further, they found that downregulation of METTL4 was associated with metastasis and could serve as a prognostic factor in HCC. Mechanistically, METTL4 could interact with the primary microRNA (miRNA) processing protein microprocessor complex subunit DGCR8 (DGCR8) and regulate primary miRNA-126 processing in an m6A-dependent manner [62]. In contrast, METTL3 is highly expressed in HCC and promotes HCC tumorigenicity and progression by regulating the suppressor of cytokine signaling 2 (SOCS2) and Snail family transcriptional repressor 1 (Snail1) mRNAs homeostasis [63, 64]. In addition to mRNA, the dysregulation of RNA methylation on the long non-coding RNAs (lncRNAs) also contributes to the oncogenic function of METTL3. An HCC specific lncRNA, LINC00958, was stabilized by METTL3-mediated m6A modification and was found to facilitate HCC lipogenesis and progression through the sponging of miRNA-3619-5p, and thus upregulated hepatoma-derived growth factor (HDGF) expression [64]. A nanoplatform delivering LINC00958 small inter-
ferring RNA (siRNA) was then developed for anti-HCC purposes [64]. As regulatory components of MTC, WTAP guides HuR-mediated ETS Proto-Oncogene 1 (ETS1) instability in an m6A-dependent pattern [65] while VIRMA (also named KIAA1429) mediates the installation of m6A on the mRNA of DNA-binding protein inhibitor ID-2 (ID2) and the antisense lncRNA of GATA binding protein 3 (GATA3) [66, 67], thereby contributing to liver cancer development.

Besides MTC, the functions of other m6A modulators have been revealed in liver cancer as well. Declined ALKBH5 caused more m6A on LY6/PLAUR domain containing 1 (LYPD1), and the latter was recognized and stabilized by IGF2BP1, resulting in a more malignant HCC phenotype [68]. Sirtuin 1 (SIRT1)-induced FTO SUMoylation (small ubiquitin-related modifier, SUMO) leads to the degradation of FTO protein, alleviating FTO-mediated G protein subunit alpha O1 (GNAO1) demethylation and increasing its expression, which has been shown to promote hepatocarcinogenesis [69].

Controversial roles of YTHDF2 have been reported in HCC. Yang et al. [70] reported that YTHDF2 was essential for HCC cell survival. By contrast, Hou et al. [71] reported that low expression of YTHDF2 could provoke inflammation, vascular reconstruction, and metastatic progression in HCC. This function could be blocked by hypoxia-inducible factor (HIF)-2α, revealing a molecular ‘rheostat’ role of YTHDF2 in the epitranscriptome and HCC progression [71]. Coincidentally, Zhong et al. [72] found YTHDF2 could suppress HCC cell proliferation by destabilizing the epidermal growth factor receptor (EGFR) mRNA and was inhibited by the hypoxia environment of HCC. For YTHDF1, its function in promoting the translation of Snail mRNA and driving epithelial-to-mesenchymal transition (EMT) seems to be consistent with the poor prognosis associated with its high expression level in HCC patients [63, 73]. The role of another reader protein, IGF2BP1, was also described in HCC, where IGF2BP1 protected serum response factor (SRF) mRNA from miRNA-mediated decay in an m6A-dependent manner, supporting IGF2BP3 as oncogenic drivers in cancer [29, 74]. Overall, the above-mentioned research lay the foundation for treating liver cancer from the RNA epigenetics view.

### 3.3 Gastrointestinal carcinoma

Gastric cancer is the fifth most-diagnosed neoplasm globally, with approximately 1 million patients being newly diagnosed each year [75]. Considering its rapid progression and tendency to metastasis, scientists have been trying to find out intrinsic mechanisms of gastric cancer, and progress has been made in revealing the relationship between m6A regulators and the metastatic property of gastric cancer. For instance, METTL3 installs m6A on zinc finger MYM-type containing 1 (ZMYM1) to increase its stability, and ZMYM1 recruits the C-terminal-binding protein (CTBP)/lysine-specific histone demethylase 1 (LSD1)/REST corepressor 1 (CoREST) complex to repress E-cadherin (also named cadherin-1, CDH1) transcription, thus strengthening the EMT program and metastasis [76]. The activation of METTL3 transcription increases m6A modification on HDGF mRNA, facilitating the binding of IGF2BP3. Both of the secreted and nuclear HDGF contribute to this tumor-suppressive role of METTL3 in gastric cancer as also demonstrated by other researchers [78–81]. METTL14, in contrast, has a tumor-suppressive function, and the knockdown of it activates the Wnt/PI3K (phosphoinositide 3 kinase)-Akt (protein kinase B) signaling to promote tumor progression [82]. Other m6A-related proteins, including IGF2BP3 and ALKBH5, are both shown to play oncogenic roles in the development of gastric carcinoma [77, 83].

Pancreatic cancer has the lowest survival rate (9%) among all cancer types [84, 85]. Providing insights into the development of pancreatic cancer from the aspect of RNA epigenetics is also of great significance. Three studies suggested a tumor-suppressive role of ALKBH5 in pancreatic cancer. He et al. [86] reported that ALKBH5 inhibited pancreatic cancer motility by regulating the m6A level of antisense RNA 1 of KCNK15 (KCNK15-AS1) lncRNA. Tang et al. [87] found that ALKBH5 was downregulated in pancreatic ductal adenocarcinoma (PDAC) cells and its overexpression sensitized cells to chemotherapy, with Wnt inhibitory factor 1 (WIF-1) being identified as the target of ALKBH5. Recently, another research demonstrated that ALKBH5 led to demethylation of period circadian regulator 1 (PER1) mRNA and lifted PERI level in a YTHDF2-dependent manner, thereby reactivating the ATM (A-T mutated)-CHK2 (serine/threonine-protein kinase)-PS3 (tumor protein 53)/CDC25C (cell division cycle 25C) pathway [88]. In contrast to ALKBH5, other m6A regulators, including FTO, METTL3, YTHDF2 and IGF2BP2, were all shown to exhibit oncogenic roles in pancreatic cancer by promoting cell proliferation, EMT, invasion, or chemo- and radio-resistance [89–93].

As one of the most common types of carcinoma, colorectal cancer (CRC) is known for its increasing incidence globally [61]. m6A modification has been found to be involved in the pathogenesis of CRC in recent years. Reduction of METTL14 was found to be correlated with unfavorable prognosis of CRC patients. Mechanistically, less m6A modification on the oncogenic lncRNA (X inactive specific transcript) XIST or SRY-box transcription factor 4 (SOX4) mRNA due to low level of METTL14 inhibited YTHDF2 binding, preventing the decay of XIST or SOX4, and resulting in the malignant phenotype [94, 95]. In addition, the
processing of primary miRNA-375 was inhibited in CRC with decreased expression of METTL14, which contributed to CRC progression [96]. Interestingly, the demethylation of histone H3 lysine 4 trimethylation (H3K4me3) on the METTL14 promoter is responsible for the repression of METTL14 transcription in CRC [95]. In contrast, METTL3 generally contributes to tumor development in CRC [97, 98]. For example, METTL3 was reported to have not only stemness-inducing function but also tumorigenesis and metastasis-promoting activity in CRC [98]. SRY-box transcription factor 2 (SOX2), the downstream target of METTL3, was recognized and stabilized by IGF2BP2 in an m6A dependent manner [98]. In terms of m6A readers, YTHDF1 was regulated by MYC and promoted CRC development [99, 100], while YTHDF3, a well-known target of Yes-associated protein (YAP), formed a feedback loop by mediating the degradation of IncRNA growth arrest specific 5 (GAS5). The latter could facilitate YAP nuclear translocation, phosphorylation, and ubiquitin-dependent decay in CRC [101]. In addition, long intergenic noncoding RNA for IGF2BP2 stability (LINRIS) was found able to inhibit ubiquitination of IGF2BP2 at lysine 139 and prevent its degradation via the autophagy-lysosome pathway, thus promoting tumorigenesis through the LINRIS-IGF2BP2-MYC axis in CRC [102].

3.4 | Glioblastoma

Glioblastoma (GBM) is a type of commonly occurred and aggressive brain tumor, in which m6A modification has been demonstrated to play a role as well [103, 104]. METTL3 is a predictive prognostic marker in GBM and plays a role in glioma stem-like cells (GSCs) maintenance by depositing m6A modification in the SOX2 3′ untranslated region (3′UTR) region and leading to the overexpression of SOX2 [105]. Consistently, Li et al. [106] found that elevated expression of METTL3 correlated with the clinical aggressiveness of malignant gliomas. m6A modification of the splicing factor serine/arginine-rich splicing factor (SRSF) decreased upon METTL3 knockdown, leading to YTHDC1-dependent nonsense-mediated mRNA decay of the SRSF transcripts and alternative splicing isoform switches in glioblastoma [106]. However, an opposite role of METTL3 in GBM has also been reported, in which METTL3 and METTL14 were considered as tumor suppressors by targeting metalloclopeptidase domain 19 (ADAM19), suggesting more in-depth studies are remained to be done [107]. High forhead box M1 (FOXM1) level caused by ALKBH5-induced m6A reduction on FOXM1 mRNA contributes to GSC and tumorigenesis, while antisense RNA of FOXM1 (FOXM1-AS) enhances the binding of ALKBH5 and FOXM1 [108]. More recently, Dixit et al. [109] reported a dependency of GSCs on YTHDF2, which surprisingly stabilized MYC and vascular endothelial growth factor A (VEGFA) transcripts in an m6A-dependent manner, distinct from the well-recognized role of YTHDF2 in mediating mRNA decay.

3.5 | Breast cancer

As a highly heterogeneous neoplasm, breast cancer is the second cause of tumor-associated death for women globally [110]. A positive feedback loop of HBXIP (Hepatitis B X-interacting protein)/let-7g (let-7g)/METTL3/HBXIP, in which m6A modification was involved in gene expression regulation, was demonstrated to drive the aggressiveness of breast cancer [111]. In another study [112], the depletion of METTL3 induced adenylate kinase 4 (AK4) overexpression, reactive oxygen species (ROS) reduction, and less resistance of MCF-7 cells to tamoxifen. METTL14 and ALKBH5 were shown to promote breast cancer growth and invasion by regulating m6A levels of key EMT and angiogenesis-associated transcripts. Interestingly, the authors reported that METTL14 and ALKBH5 controlled each other’s expression and inhibited YTHDF3, and the latter could in turn block RNA demethylase activity, forming a writer-eraser-reader collaborative loop [113]. The role of ALKBH5 in breast cancer has also been reported by other groups [114–116]. Notably, Zhang et al. [114, 116] found that the expression of ALKBH5 could be stimulated by HIF-1α and HIF-2α upon exposure to hypoxia, which increased breast cancer stem cells by reducing m6A modification on NANOG mRNA and increased NANOG protein level. Similar to ALKBH5, FTO also promotes breast cancer progression, with BCL2 interacting protein 3 (BNIP3) and miR-181b-3p being identified as targets of FTO [117, 118]. In addition, Chang et al. [119] revealed the involvement of YTHFD3-mediated epitranscriptomic regulation in breast cancer brain metastasis. YTHFD3 overexpression was able to promote the translation of m6A-modified St6 N-acetylactosaminidase alpha-2,6-sialyltransferase 5 (St6GALNAC5), gap junction protein alpha 1 (GJA1), and EGFR, which are related to brain metastasis. Further, they found that the overexpression of YTHFD3 was the combined consequence of increased gene copy number and the autoregulation of YTHFD3 cap-independent translation by binding to m6A residues within its own 5′UTR.

3.6 | Other cancers

Although advanced detection techniques and combined treatment have been used, lung carcinoma, especially non-small cell lung cancer (NSCLC), is still the main
cause of cancer-associated death globally [61]. Most of the m^6^A modulators, including METTL3 [41, 42, 120], FTO [121, 122], ALKBH5 [123–125], YTHDF1 [126] and YTHDF2 [127], were demonstrated to be oncopgenic in NSCLC, a main subtype accounted for 80%-85% of lung cancer. It’s worth mentioning that the oncogenic function of METTL3 is attributed to its methyltransferase-dependent and -independent activities, suggested by recent studies [41, 42, 120]. As a writer, METTL3 installs m^6^A on mRNAs, such as YAP, and IncRNAs, such as metastasis-associated lung adenocarcinoma transcript 1 (MALAT1), and promotes the invasion and metastasis of NSCLC via the activation of the YAP pathway [120]. On the other hand, METTL3 functions like a reader to recognize m^6^A-modified mRNAs and promote the production of oncoproteins, such as EGFR, tafazzin (TAZ), mitogen-activated protein kinase 2 (MAPK2), DNA methyltransferase 3A (DNMT3A), and bromodomain-containing protein 4 (BRD4) [41, 42]. Whether the oncogenic roles of METTL3 rely on its writer or reader activity in other cancer types is unclear and needs to be elucidated. In contrast to METTL3, ALKBH5 inhibits tumor growth and metastasis by lessening YAP level in a YTHDFs dependent manner and impairing YAP function with the help of the miRNA-107/LATS2 (large tumor suppressor kinase 2) axis in lung cancer [125].

Diffuse large B-cell lymphoma (DLBCL) is a subtype of lymphoid malignancy with heterogenous characteristics in clinical manifestation, pathology, and biology [128]. METTL3 was found to be upregulated in DLBCL tissues and promoted DLBCL progression by depositing m^6^A methylation on pigment epithelium-derived factor (PEDF) transcript, though the detailed mechanism needs to be further studied [129]. A PIWI-interacting RNA (piRNA), piRNA-30473, was illustrated to have oncogenic activity in DLBCL through an m^6^A-dependent manner [130]. Further, it was found that piRNA-30473 could increase WTAP level, which facilitated m^6^A modifications on downstream targets, such as hexokinase 2 (HK2).

Ovarian cancer and endometrial cancer are highly aggressive gynecologic cancers [131, 132]. m^6^A regulators, including ALKBH5 [133], METTL3 [134], IGF2BP1 [74] and YTHDF1 [135], were suggested to be critical factors in promoting ovarian cancer. For instance, multi-omics analysis has been used to explore the crucial component of m^6^A-related modulators in ovarian cancer and identified a subunit of elf3, elf3C, as a direct YTHDF1 target [135]. Interestingly, the protein but not the RNA level of elf3C was increased and positively correlated with the protein level of YTHDF1 in ovarian cancer patients, suggesting that modification of elf3C mRNA could be more relevant to its role in cancer. Up to 70% of endometrial cancers exhibited m^6^A hypomethylation, possibly attributed to either a hotspot R298P (R is arginine, P is proline) mutation in METTL4 or a decline of METTL14 or METTL3 level [136]. The changes of these two key m^6^A modulators advanced endometrial tumor development via the AKT pathway.

Overall, dysregulation of m^6^A modifiers in cancer is frequently observed and plays crucial roles in cancer initiation, development, and drug resistance, through modulating/recognizing m^6^A on critical target transcripts.

### 4. FACTORS AFFECTING m^6^A IN CANCER

There is no doubt that m^6^A regulators dominate the layer of epitranscriptomic regulation; nonetheless, internal or external factors are able to regulate m^6^A incorporation in different contexts, especially in cancer. Here, we sum up factors that have an impact on m^6^A in cancer.

#### 4.1 Genetic and epigenetic factors

Noncoding RNAs (ncRNAs) are a kind of RNA traditionally regarded as molecules that are not translated but have regulatory activities in gene expression. Accumulating data have shown that m^6^A methylation affects the production and/or functions of ncRNAs, including IncRNAs, circular RNAs, and miRNAs [12, 30, 44, 64, 94, 137–140]. On the other hand, ncRNAs also play a role in m^6^A-mediated gene expression regulation. For instance, ALKBH5 acted as an oncogene in GSCs by demethylating FOXM1 mRNA. Interestingly, this process was strengthened by FOXM1-AS, a IncRNA antisense to FOXM1 [108]. The discovery of FOXM1-AS as a pivotal modulator in ALKBH5-dependent GSC proliferation emphasizes the role of ncRNA in GSC. RNA-binding regulatory peptide (RBRP) is a peptide encoded by IncRNA LINC00266-1, and its interaction with the m^6^A reader IGF2BP1 intensified the function of IGF2BP1, thus, reinforced the expression of MYC and the process of tumorigenesis. More importantly, higher RBRP level in patients was associated with shorter overall survival, confirming its oncogenic effect and the potential applications as a therapeutic target in treating cancers [141]. In colorectal cancer, the inhibition of miRNA-455-3p rescued β-catenin depletion-induced reduction of heat shock transcription factor 1 (HSF1) m^6^A modification and METTL3 interaction [142]. Taken together, ncRNAs exist as critical modulators of m^6^A-dependent gene expression control, and more of their regulatory roles and mechanisms remain to be explored.

The RNA methylation also has crosstalk with histone modifications. To be specific, m^6^A peaks are enriched in the region of histone H3 lysine 36 trimethylation (H3K36me3) and are declined with the reduction of
H3K36me3. Mechanistically, METTL14 recognizes and binds to H3K36me3, after which MTC interacts with RNA Polymerase II and further installs m^6^A to actively transcribed pre-mRNA [143]. Histone acetyltransferase P300 (EP300)-mediated histone H3 lysine 27 acetylation (H3K27ac) activates METTL3 transcription which stimulates m^6^A modification on HDGF mRNA and enhances its stability, and finally leads to tumor growth and liver metastasis in human gastric cancer [77]. Similarly, lysine demethylase 5c (KDM5C)-induced H3K4me3 demethylation in the promoter of METTL14 attenuates METTL14 transcription, resulting in reduced m^6^A deposition on SOX4 and the upregulation of the tumor suppressor SOX4 in colorectal cancer [95].

The posttranslational modifications on m^6^A regulators have also been identified and were found to play crucial roles in controlling the activity of the m^6^A machinery and therefore, the epitranscriptome. In HCC, SIRT1 activated the SUMO E3 ligase RAN binding protein 2 (RANBP2) which mediated SUMOylation and degradation of FTO, and resulted in more m^6^A on GNAO1, an anti-tumor molecule in HCC [69]. METTL3 could be activated by ATM-mediated phosphorylation at serine 43 and localized to double-strand break sites, and YTHDC1 was subsequently recruited due to METTL3-induced m^6^A deposition. Interference with this METTL3-m^6^A-YTHDC1 axis enhanced the sensitivity of cancer cells to DNA damage-based therapy [144].

To sum up, the complicated network connecting m^6^A modification and other genetic and epigenetic factors integrates comprehensive information from various sources and strengthens gene expression control more accurately.

### 4.2 Environmental exposure affects m^6^A methylation

In addition to internal factors, external exposure also has an influence on m^6^A methylation. Human carcinogens in different content elicit detrimental effects to human bodies in genotoxic or non-genotoxic ways [145]. Evidence has shown that cigarette smoke causes oncogenic mutations and epigenetic changes [146, 147]. Tobacco smoking can alter miRNA encoding genes [148–150]. Genes with aberrant levels further participate in a myriad of pathological processes, including tumorigenesis and tumor progression [151]. Cigarette smoke condensate induced hypo-methylation in METTL3 promoter caused METTL3 overexpression and subsequently more m^6^A modification which promoted maturation of miRNA-25. The latter activated the AKT-p70S6 kinase pathway and played an oncogenic role in pancreatic cancer [152].

Reduced global m^6^A level was observed in A549 lung epithelial cells in response to sodium arsenite and particulate matter, and change in m^6^A level was associated with the concentration of environmental toxicants [153]. In contrast, chronic exposure of human bronchial epithelial cells to sodium arsenite-induced malignant phenotype with increased m^6^A modification which was synergistically regulated by m^6^A modulators [154]. Dynamic m^6^A incorporation was found in chemical carcinogen-induced cellular transformation, in which the METTL3-m^6^A-CDCP1 (CUB domain-containing protein 1) axis contributed a lot to cell proliferation and progression, consistent with the effect of chemical carcinogenesis [155].

As a well-known oncogenic virus, Epstein–Barr virus (EBV) is the culprit of about 2% of all malignancies via regulating numerous host cell activities. Lang et al. [156] have observed the interplay between EBV and m^6^A decoration. EBV nuclear antigen 3C (EBNA3C), the viral-encoded latent oncoprotein, was upregulated by METTL14-mediated m^6^A modification, and could in turn activate METTL14 transcription and directly interact with METTL14 to promote its protein stability. Therefore, METTL14 appears to be an important factor in EBV-induced oncogenesis. In addition, m^6^A modification plays a role in the lifecycle and infection of the hepatitis virus which predominantly contributes to chronic liver diseases and the tumorigenesis of HCC [157, 158]. It was found that Hepatitis B virus (HBV) pregenomic RNA (pgRNA) was m^6^A modified in the RRACH motif within the epsilon domain-containing protein 1 [144].

Silencing of METTL3 and METTL14 in Huh7 hepatoma cells increased the production of infectious HCV particles and the percentage of HCV-positive cells, while depletion of FTO inhibited HCV particle production and infection. Taken together, these studies indicate the important roles of m^6^A modification during the pathogenesis, development, and progression of virus-related cancers, implicating that modulation of m^6^A modification could serve as prevention or therapeutic strategies in virus-related cancers.

### 5 Clinical implications of m^6^A in cancers

A growing body of research on m^6^A methylation reveals a new layer of epigenetic regulation in oncogenesis and provides implications for the use of m^6^A in innovative and effective diagnostic and therapeutic approaches.
5.1 Implications of m\textsuperscript{6}A in cancer diagnosis and prognosis

Effective biomarkers, along with sensitive and specific detection methods, will greatly contribute to the early diagnosis of cancers, thus, improve the survival of patients. Recently, m\textsuperscript{6}A methylation and its regulators have become emerging biomarkers for cancer diagnosis and prognosis [21, 159]. Owing to metabolic reversibility, high abundance and stability, methylated nucleosides could be accessible in biological fluids (e.g., serum and urine) or circulating cells [160, 161]. Huang et al. [161] developed a liquid chromatography-electrospray ionization tandem mass spectrometric (LC-ESI-MS/MS) method to determine m\textsuperscript{6}A level in single cells and found increased RNA m\textsuperscript{6}A methylation in circulating tumor cells from the blood of lung cancer patients. Pei et al. [162] also detected elevated m\textsuperscript{6}A level in the peripheral blood leukocyte from non-small cell lung cancer patients by flow cytometry, indicating the potential use of m\textsuperscript{6}A as a non-invasive biomarker. Furthermore, m\textsuperscript{6}A regulators, including METTL3 [77, 163-169], WTAP [170–173], FTO [174–177], IGFB2Ps [178–184], and YTHDFs [73, 185-192], have been proven to associate with favorable or unfavorable prognosis in different types of cancers (detailed in Section 3). It should be noted that prognosis is not simply associated with the expression of a certain gene but a comprehensive signature of multiple m\textsuperscript{6}A regulators in cancers, including lung cancer [163, 164], pancreatic cancer [166, 193], and HCC [194]. Despite that m\textsuperscript{6}A and its regulators exhibit powerful potential as biomarkers, it is still challenging for clinical application due to the heterogeneity of m\textsuperscript{6}A in patients and the lack of assays to detect site-specific m\textsuperscript{6}A from low-input clinical samples. Future single-cell sequencing techniques might provide powerful assistance in solving such problems.

5.2 m\textsuperscript{6}A modification and chemosensitivity

Drug resistance is the major cause of therapeutic failure and recurrence in chemotherapy. Recent studies have indicated that m\textsuperscript{6}A modification was associated with drug response and chemoresistance [21, 159]. Mutations of receptor tyrosine kinases, such as BCR-ABL (breakpoint cluster region, BCR; tyrosine-protein kinase, ABL), c-kit proto-oncogene (KIT) and fms like tyrosine kinase 3 (FLT3) frequently occur in leukemia and are effective therapeutic targets in the clinic [195–197]. The tolerance of tyrosine kinase inhibitors (TKIs), a big challenge in leukemia treatment, is mediated by m\textsuperscript{6}A demethylation resulting from elevated FTO in leukemia cells [198]. Decreased ALKBH5 was found in gemcitabine-treated patient-derived xenograft (PDX) model and predicted poor clinical outcome in PDAC, while overexpression of ALKBH5 could sensitize PDAC to chemotherapy [87]. Moreover, METTL3-induced m\textsuperscript{6}A installation was found to contribute to oxaliplatin resistance in colon cancer [199], cisplatin resistance in NSCLC [120] and tamoxifen resistance in breast cancer [112]. Therefore, silencing of METTL3 could reverse drug resistance in the above scenarios [112, 120, 199], and could enhance the sensitivity of DNA damage-based therapy in vivo and in vitro [144]. Collectively, robust evidence unveils the participation of m\textsuperscript{6}A modulators in drug resistance, shedding light on the application of these regulators as predictive markers in chemotherapy or drug targets in combination with chemotherapy.

5.3 The m\textsuperscript{6}A modification and cancer immunotherapy

Although immunotherapy has been considered as a promising treatment in defeating cancer, lacking durable effects in some groups of patients limits its efficacy [200–202]. Intriguingly, the absence of YTHDF1 in mice enhances antigen-specific cluster of differentiation 8 (CD8)-positive T-cell anti-tumor reaction due to promoted tumor antigen cross-presentation in classical dendritic cells (cDCs) [203]. As a result, the therapeutic efficacy of programmed death-ligand 1 (PD-L1) checkpoint blockade is enhanced in YTHDF1-deficient mice [203]. What’s more, the decline of FTO also improves the low response of melanoma cells to interferon-gamma and enhances the reaction to anti-PD-1 (programmed cell death protein 1, PDI) blockade in mice [204]. These studies suggest that YTHDF1 and FTO might be potential drug targets in combination with immunotherapy.

5.4 Targeting m\textsuperscript{6}A and its regulators in cancer therapy

Given the benefits of targeting m\textsuperscript{6}A methylation in cancer therapy, as discussed above, researchers never cease exploring effective inhibitors of m\textsuperscript{6}A enzymes. The most well representative one is the development of small-molecule agents targeting FTO. Initially, a natural compound named Rhein was found to bind to FTO catalytic domain and competitively inhibited the recognition of m\textsuperscript{6}A substrate [205]. An ascorbic acid analog was then designed in 2014 to inhibit the 2-oxoglutarate-dependent hydroxylase activity of FTO and elevate m\textsuperscript{6}A level [206]. Later, meclofenamic acid (MA) and an acylhydrazine compound, FTO inhibitor 12, have been identified to have inhibitory activity on FTO over ALKBH5 [207, 208].
By targeting FTO, R-2HG and FB23-2 exhibited promising inhibitory effects in the treatment of AML [52, 53]. More recently, two potent agents, CSI and CS2, have been developed by high-throughput screening from over 260,000 compounds and showed anti-tumor effects in multiple cancers by suppressing the self-renewal of cancer stem cells and immune evasion [54]. The inhibitors for m⁶A writers and readers are also of great interest to the researchers. By screening a library of 4000 analogs and derivatives of S-adenosyl-methionine (SAM), UZH1a has been found to be an effective METTL3 inhibitor and could modulate transcriptomic m⁶A signal in the MOLM13 leukemia cells; however, its in vivo effect still needs to be elucidated [209]. As a small molecule inhibitor, BTYNB was reported to disrupt the association between IGF2BP1 and target RNA, which resulted in the decrease of E2 factor (E2F)-driven cell cycle transition and inhibition of tumor progression [210]. With more and more druggable m⁶A targets being proven with proof-of-concept evidence to combat cancers, the identification of specific inhibitors and the application of these inhibitors in the clinic, especially in combination with other therapies, are of great importance and in urgent need. Instead of altering the transcriptome-wide m⁶A level, “m⁶A editing” is a CRISPR (clusters of regularly interspaced short palindromic repeats)-Cas9 (CRISPR-associated protein-9)-based method to mediate programmable RNA methylation or demethylation at a specific locus. To achieve site-specific removal of m⁶A, Liu et al. [211] engineered m⁶A ‘erasers’ by fusing catalytic dead Cas9 (dCas9) with ALKBH5 or FTO, while Li et al. [212] chose the RNA-targeting CRISPR-Cas system, dCas13, to engineer ALKBH5. Recently, dCas13 fusions with truncated METTL3 or modified METTL3-METTL14 complex have been established to direct site-specific m⁶A incorporation [213]. To date, these new techniques allow precise manipulation of a single methylation site, in the hope of evaluating the exact function of a single methylation site and targeting of single m⁶A for cancer treatment.

6 | PERSPECTIVES AND CONCLUSION

Fast-growing research in the RNA epigenetics field delineates a comprehensive picture about how m⁶A methylation is tightly controlled by its enzymes (writers and erasers) and works with reader proteins to participate in almost every step of RNA metabolism. Hypo- or hyper-methylation might lead to aberrant gene expression, abnormal cellular function, and diseases, such as cancer. The direct links between m⁶A and various cancers not only provide an insight into the mechanism of tumorigenesis but also are valuable for guiding clinical applications in fighting against cancers. Considering the broad effects of m⁶A in strengthening the anti-cancer effect of chemotherapy and immunotherapy, the combination of m⁶A-targeting agents with traditional chemotherapeutic drugs or PD-1/PD-L1 inhibitors holds great therapeutic promise. However, there is still a debate whether targeting the total abundance/level of m⁶A methylation (i.e., targeting enzymes) or targeting gene-/site-specific m⁶A methylation is a better choice, which warrants more proof-of-concept studies. Overall, m⁶A modification is a rising star in the epigenetic field and holds therapeutic promise for a broad range of cancer.

Abbreviations

RNA, ribonucleic acid; m⁶A, N⁶-methyladenosine; DNA, deoxyribonucleic acid; mRNA, messenger RNA; FTO, fat mass and obesity-associated protein; NGS, Next-generation sequencing; G, guanosine; A, adenosine; C, cytidine; U, uridine; MTC, methyltransferase complex; METTL3, methyltransferase-like 3; METTL14, methyltransferase-like 14; WTAP, Williams tumor 1 associated protein; RBM15, RNA Binding Motif Protein 15; RBM15B, RNA Binding Motif Protein 15B; ZC3H13, Zinc Finger CCCH-Type Containing 13; VIRMA, Vir like m6A methyltransferase associated; ALKBH5, alkB homolog 5; YTH, YT521-B homology; IGF2BP, insulin-like growth factor 2 mRNA-binding proteins; HNRNPs, heterogeneous nuclear ribonucleoproteins; YTHDC1, YTH domain-containing protein 1; YTHDC2, YTH domain-containing protein 2; YTHDF1, YTH domain-containing family protein 1; YTHDF2, YTH domain-containing family protein 2; YTHDF3, YTH domain-containing family protein 3; hnRNPC, heterogeneous nuclear ribonucleoprotein C; hnRNPG, heterogeneous nuclear ribonucleoprotein G; hnRNPA2B1, heterogeneous nuclear ribonucleoproteins A2/B1; SRSF3, serine/arginine-rich splicing factor 3; SRSF10, serine/arginine-rich splicing factor; NXF1, nuclear RNA export factor 1; FMRP, Fragile X mental retardation protein; XPO1, Exportin 1; polyA, polyadenylated; ELAVL1, ELAV like RNA binding protein 1; PABPC1, poly(a) binding protein cytoplasmic 1; MATR3, Matrin 3; PRRC2A, Proline Rich Coiled-Coil 2A; eIF4G, eukaryotic translation initiation factor 4G; eIF3, eukaryotic translation initiation factor 3; eIF3h, eukaryotic translation initiation factor 3h; 5'UTR, 5' untranslated region; circRNAs, circular RNAs; carRNAs, chromosome-associated regulatory RNAs; H3K9me2, histone H3 lysine 9 dimethylation; KDM3B, lysine demethylase 3B; AML, acute myeloid leukemia; HSPCs, hematopoietic stem and progenitor cells; LSCs, leukemic stem cells; ASB2, ankyrin repeat and socs box containing 2; RARA, retinoic acid receptor alpha; R-2HG, R-2-hydroxyglutarate; IDH, isocitrate dehydrogenase; KDM4C, lysine demethylase 4C; AXL, tyrosine-protein kinase receptor UFO; TACC3,
transforming acidic coiled-coil containing protein 3; CEBPZ, CCAAT enhancer-binding protein zeta; BCL2, B-cell lymphoma 2; PTK2, phosphatase and tensin homolog; SPI1, transcription factor PU.1; Tnfrsf2, tumor necrosis factor receptor superfamily member 2; HSC, hematopoietic stem cell; HCC, hepatocellular carcinoma; miRNA, microRNA; DGC-R8, microprocessor complex subunit DGC-R8; SOCS2, cytokine signaling 2; Snail1, snail family transcriptional repressor 1; IncRNAs, long non-coding RNAs; HDGF, hepatoma-derived growth factor; siRNA, small interfering RNA; ETS1, HuR-mediated ETS Proto-Oncogene 1; ID2, DNA-binding protein inhibitor ID-2; GATA3, GATA binding protein 3; LYPD1, LY6/PLAUR domain containing 1; SIRT1, Sirtuin 1; SOX4, SRY-box transcription factor 4; H3K4me3, histone H3 lysine 4 trimethylation; SOX2, SRY-box transcription factor 2; YAP, Yes-associated protein; GAS5, growth arrest specific 5; LINRIS, long intergenic noncoding RNA for IGF2BP2 stability; GBM, Glioblastoma; GSCs, glioma stem-like cells; 3’UTR, 3’ untranslated region; SRSF, serine/arginine-rich splicing factor; ADAM19, ADAM metallopeptidase domain 19; FOXM1, forkhead box M1; FOXM1-AS, antisense RNA of FOXM1; VEGFA, vascular endothelial growth factor A; HBXIP, Hepatitis B X-interacting protein; let-7g, lethal-7g; AK4, adenylate kinase 4; ROS, reactive oxygen species; BNP3, BCL2 interacting protein 3; ST6GALNAC5, ST6 N-acetylgalactosaminide alpha-2,6-sialyltransferase 5; GJA1, gap junction protein alpha 1; NSCLC, non-small cell lung cancer; MALAT1, metastasis associated lung adenocarcinoma transcript 1; TAZ, tafazzin; MAPK2, mitogen-activated protein kinase 2; DNM3A, DNA methyltransferase 3A; BRD4, bromodomain-containing protein 4; ATS2, large tumor suppressor kinase 2; DLBCL, Diffuse large B-cell lymphoma; PEDF, pigment epithelium-derived factor; piRNA, PIWI-interacting RNA; HK2, hexokinase 2; ncRNAs, noncoding RNAs; RBRP, RNA-binding regulatory peptide; HSF1, heat shock transcription factor 1; H3K36me3, histone H3 lysine 36 trimethylation; EP300, histone acetyltransferase P300; H3K27ac, histone H3 lysine 27 acetylation; KDM5C, lysine demethylase 5c; RANBP2, RAN binding protein 2; CDCP1, CUB domain containing protein 1; EBV, Epstein–Barr virus; EBN3A, EBV nuclear antigen 3C; HBV, Hepatitis B virus; pgRNA, pregenomic RNA; HCV, Hepatitis C virus; LC-ESI-MS/MS, liquid chromatography electrospray ionization tandem mass spectrometric; BCR, breakpoint cluster region; ABL, tyrosine-protein kinase; KIT, c-kit proto-oncogene; FLT3, fms like tyrosine kinase 3; TKIs, tyrosine kinase inhibitors; PDX, patient-derived xenograft; CD8, cluster of differentiation 8; cDCs, classical dendritic cells; PD-L1, programmed death-ligand 1; PD1, programmed cell death protein 1; MA, meclofenamic acid; SAM, S-adenosyl-methionine; E2F, E2 factor; CRISPR, clusters of regularly interspaced short palindromic repeats; CAS9, CRISPR-associated protein-9; dCas9, catalytic dead Cas9.

DECLARATIONS
ETHICS APPROVAL AND CONSENT TO PARTICIPATE
Not applicable.

CONSENT FOR PUBLICATION
Not applicable.

AVAILABILITY OF DATA AND MATERIALS
Not applicable.

COMPETING INTERESTS
The authors declare that they have no competing interests.

ACKNOWLEDGMENTS
This work was supported in part by the grant 2021A151S010425 (H.H.) from Guangdong Basic and Applied Basic Research Foundation, and the National Key R&D Program of China 2020YFA0112403 (H.W.) from the Ministry of Science and Technology of the People’s Republic of China.

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SHD, HYW and HLH wrote the manuscript. SHD made the figures. SHD, YTW, YDL, HYW and HLH revised and approved the final manuscript.

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