Effects of N6-Methyladenosine Modification on Cancer Progression: Molecular Mechanisms and Cancer Therapy

Yong-fu Zhu1,2†, Shu-Jie Wang3†, Jie Zhou2, Ye-han Sun1, You-mou Chen1, Jia Ma1, Xing-xing Huo4* and Hang Song5*

1 The First Department of Oncology, The First Affiliated Hospital of Anhui University of Chinese Medicine, Hefei, China, 2 The Department of Acupuncture, The Third Affiliated Hospital of Zhejiang Chinese Medical University, Hangzhou, China, 3 Anhui Province Key Laboratory of Medical Physics and Technology, Institute of Health and Medical Technology, Hefei Institutes of Physical Science, Chinese Academy of Sciences, Hefei, China, 4 Experimental Center of Clinical Research, Scientific Research Department, The First Affiliated Hospital of Anhui University of Chinese Medicine, Hefei, China, 5 Department of Biochemistry and Molecular Biology, School of Integrated Chinese and Western Medicine, Anhui University of Chinese Medicine, Hefei, China

N6-methyladenosine (m6A) is a major internal epigenetic modification in eukaryotic mRNA, which is dynamic and reversible. m6A is regulated by methylases (“writers”) and demethylases (“erasers”) and is recognized and processed by m6A-binding proteins (“readers”), which further regulate RNA transport, localization, translation, and degradation. It plays a role in promoting or suppressing tumors and has the potential to become a therapeutic target for malignant tumors. In this review, we focus on the mutual regulation of m6A and coding and non-coding RNAs and introduce the molecular mechanism of m6A methylation involved in regulation and its role in cancer treatment by taking common female malignant tumors as an example.

Keywords: RNA modifications, N6-methyladenosine, female malignancies, molecular mechanisms, immunotherapy

INTRODUCTION

N6-methyladenosine (m6A) alteration is a methylation modification located on the 6th nitrogen atom of adenine. m6A is the most abundant form of epigenetic modification in eukaryotic RNA, which exists in several different types of RNA, including mRNA and non-coding RNA. At present, the research on m6A methylation and malignant tumors mainly focuses on the influence of m6A
methylation on tumor cell proliferation, invasion, resistance to radiotherapy and chemotherapy, and prognosis of patients (1). The detection rate of malignant tumors is growing year by year, and the age of onset tends to be younger, thanks to the popularization of associated cancer screening tools and an increase in people’s health awareness (2). The primary clinical treatment for malignant tumors is to select individualized surgery combined with postoperative radiotherapy and chemotherapy according to the patient’s condition (3). However, for some patients with relapsed and refractory tumors, the treatment effect is often difficult to achieve the expected (4). Therefore, one of the most pressing issues to be addressed is elucidating the etiology of malignant tumors and finding novel therapeutic medications to overcome tumor resistance. In this review, we focus on the effect of m6A methylation on the occurrence and development of malignant tumors and introduce the molecular mechanisms involved in the regulation of m6A methylation and its role in cancer treatment by taking common female malignancies as an example.

OVERVIEW OF M6A METHYLATION

The m6A methylation modification of RNA is the most common internal modification in RNA modification. m6A is the methylation modification of RNA on the 6th nitrogen atom of adenosine, and the process is dynamic and reversible. The proteins involved in the methylation and demethylation of m6A are divided into three categories, namely methyltransferases (“writers”), demethylases (“erasers”), and m6A recognition proteins (“readers”) (Figure 1). Through RNA transcription, splicing, processing, translation, and degradation, it plays a role in the formation and spread of numerous malignant cancers. (5). Furthermore, studies have shown that m6A methylation modification is closely related to the activation and inhibition of cancer-related signaling pathways, which mainly affect tumor progression by regulating related tumor biological functions (6).

m6A Readers

For m6A-modified mRNA to perform specific biological functions, a specific RNA-binding protein, methylation reader protein, is required. It mainly includes YTH m6A RNA-binding protein (YTH) domain proteins (including YTHDF1, YTHDF2, YTHDF3, YTHDC1, and YTHDC2) (9), heterogeneous nuclear ribonucleoproteins (HNRNPC, HNRNPg, and HNRNPA2B1) and eukaryotic initiation factor (eIF). The functions of these reader proteins are mainly to alter protein-RNA interactions by impairing the homologous binding of m6A to RNA-binding proteins and altering RNA secondary structure (10). Studies have found that YTHDF1 has a clear oncogenic role, and its high expression in cancer genes can accelerate the transformation of important oncogenic drivers in cancer via numerous methods, impacting cancer progression and prognosis. For example, in gastric cancer progression, mutated YTHDF1 enhances the expression of the key oncogenic factor Wnt receptor Frizzled7 (FZD7), leading to gastric cancer progression and poor prognosis (11).

m6A Writers

The m6A methyltransferase consists of methyltransferase-like 3 (METTL3), METTL14, WTAP, RBM15, ZC3H13, VIRMA, and the newly discovered METTL16, which are also called writers. Its primary function is to catalyze the m6A modification of adenylic acid on mRNA. METTL3 and METTL14 have a critical catalytic domain (12), and the METTL3-METTL14 methyltransferase complex assembly during m6A modification is a crucial factor initiating the modification. In addition, methyltransferases also include associated protein (WTAP), RBM15, ZC3H13, vir-like m6A methyltransferase associated (VIRMA), and METTL16. These methyltransferases all play an important role in forming METTL3-METTL14 complexes in different links, affecting cancer cell proliferation and migration (13–16).

m6A Erasers

m6A modification is the earliest reversible modification found among many RNA modifications, and the reversibility of its modification is due to the existence of demethylases. In addition, their encoding genes are called “erasers”. Fat mass and obesity-associated protein (FTO) is the first demethylase discovered and is a member of the Alkb protein family, which can affect the RNA-binding ability of the splicing factor serine and arginine-rich splicing factor 2 (SRSF2), thereby regulating the splicing process of pre-mRNA (17). ALKBH5, another member of the Alkb protein family, was found to have a demethylation function, which can affect the RNA binding of m6A on mRNA and has poor prognosis (11).

m6A Modification in Female Malignancies

At present, m6A RNA modification is increasingly used in cancer detection and related targeted molecules (7, 8).

THE ROLE OF M6A MODIFICATIONS IN NON-CODING RNAs

In recent years, m6A has been found to exist in various ncRNAs such as miRNAs, long non-coding RNAs (IncRNAs), circular
RNAs (circRNAs), ribosomal RNAs (rRNAs), and small nuclear RNAs (snRNAs), essential for its metabolism and function (19). Moreover, some m^6^A regulatory proteins associated with aberrant m^6^A modification of ncRNAs are also involved in cancer cells proliferation, invasion, and drug resistance, suggesting a potential link between cancer and m^6^A-ncRNA modification (20).

The Effect of m^6^A on LncRNA

LncRNAs, a type of ncRNAs of 200 or more nucleotides, can regulate gene expression at multiple levels (21). Many m^6^A-modified lncRNAs have been found recently, and they can control gene expression in a variety of ways. They can act as transcriptional regulators, acting in cis or trans, regulating the transcription of adjacent genes (22). XIST, one of the first functionally annotated lncRNAs, plays a key role in X chromosome inactivation by recruiting multiple factors (23). A study shows that in head and neck squamous cell carcinoma, METTL3- and METTL14-mediated m^6^A methylation contributes to the stability of LNCAROD, and LNCAROD overexpression promotes the malignant development of HNSCC by promoting YBX1-hspa1a interaction, thereby enhancing the stability of the YBX1 protein (24). Another study found that modifying lncRNAs with m^6^A had the opposite effect on cancers. METTL14 suppressed colorectal cancer growth and metastasis by downregulating oncogenic long non-coding RNA XIST in the METTL14-YTHDF2-lncRNA regulatory axis (25). LncRNA GAS5 binds directly to Yes-associated protein (YAP), promoting its phosphorylation and ubiquitin-mediated degradation, thereby attenuating YAP-mediated transcription of YTHDF3 and inhibiting the progression of rectal cancer (26). LncRNA-p21 acts as a tumor inhibitor in the development of esophageal squamous cell carcinoma (27). In hepatocellular carcinoma, the m^6^A modification of LINC00152 is involved in the prognosis of LIHC patients through the cytoskeleton regulation pathway (28). Furthermore, in rectal cancer, m^6^A modification of lncRNA RP11 can upregulate the translation of Zeb1 to trigger cancer cell dissemination (29); lncRNA-THOR enhances IGF2BP1-targeted mRNA expression and promotes human osteosarcoma cell survival and proliferation (30).

The Effect of m^6^A Modification on MiRNA

MiRNAs are non-coding single-stranded RNAs of 21-25 nucleotides in length that regulate gene expression at the post-transcriptional level by building RNA-induced silencing complexes (RISCs) that bind to the 3’untranslated region of...
target mRNAs (3’UTR) to regulate gene expression (31). In the nucleus, miRNAs are first transcribed into longer primary miRNAs (pri-miRNAs) and subsequently processed into precursor miRNAs (pre-miRNAs). It is then cleaved into mature single-stranded miRNAs by Dicer in the cytoplasm, and the participation and processing of such pri-miRNAs are m^6^A-dependent. METTL3 tags pre-miRNAs through m^6^A modification, enabling DGCR8 to recognize and bind its specific substrates, thereby promoting miRNA maturation and increased miRNA levels in cells (32).

The Role of m^6^A Modification of CircRNA in Cancer
Circular RNAs (circRNAs) are a class of single-stranded covalently closed RNA molecules that participate in many physiological processes, including competing with endogenous RNAs as sponge miRNAs, forming RNA-protein complexes, regulating gene transcription, and even encoding proteins (33). In most cases, abnormal m^6^A modification contributes to tumorigenesis and tumor progression. However, m^6^A modification on circRNAs can suppress innate immunity; YTHDF2 sequesters m^6^A-circRNA and is essential for suppressing innate immunity (34). Chen et al. found that m^6^A modification of circNSUN2 promoted liver metastasis of colorectal cancer by promoting cytoplasmic export and forming a circNSUN2/IGF2BP2/HMGA2 RNA-protein triple complex to stabilize HMGA2 mRNA (35).

THE ROLE OF M^6^A MODIFICATIONS IN CODING RNAs
m^6^A affects all physiological processes such as mRNA processing, nuclear export, translation, and degradation. It mainly affects mRNA stability, which is also closely related to the occurrence and development of malignant tumors. At present, the research on m^6^A and its participants in the reversible regulation process (m^6^A-modifying enzymes and m^6^A-binding proteins) and the mechanism of tumorigenesis and development has gradually become a hot spot.

Ries et al. found that m^6^A-mRNA is regulated by compartments, including mRNA stability and reduced translation. This study demonstrates that the number and distribution of m^6^A sites in cellular mRNA can modulate and influence the composition of the phase-separated transcriptome (36). Li et al. first elucidated the in vivo biological role of m^6^A modification in T cell-mediated pathogenesis. They revealed a novel mechanism for T cell homeostasis and signal-dependent induction of mRNA degradation (37). RNA methyltransferase (METTL3) acts as a translation initiation complex, thereby enhancing the translation of target mRNAs (38). In addition, at different intracellular locations, m^6^A exerts methyltransferase activity-dependent and -independent functions in gene regulation. Besides, the RNA methyltransferase METTL16 is in the nucleus, acting as an m^6^A writer, depositing m^6^A into its hundreds of a specific messenger RNA target. In the cytosol, METTL16 promotes translation in an m^6^A-independent manner (39).

THE ROLE OF M^6^A MODIFICATIONS IN COMMON FEMALE MALIGNANCIES
Common malignant tumors in women mainly include breast cancer (BC), ovarian cancer (OC), cervical cancer (CC), and endometrial cancer (EC). Despite advancements in examination methods in the prevention and treatment of common female malignant tumors, most patients are in the middle and late stages of their disease due to difficulties in early diagnosis and localization of tumors and a lack of effective efficacy evaluation and prognosis monitoring methods. Therefore, the mortality rate of common malignant tumors in women continues to increase. Genetic, epigenetic, and environmental factors drive its occurrence, development, and transfer, and epigenetic factors play an important role as a bridge between genetic and environmental factors. Epigenetics has multiple forms of expression, of which m^6^A is the most abundant form of internal modification. In this review, we take common female malignant tumors as examples to introduce the molecular mechanism of m^6^A modification in the occurrence and development of cancer and its application in cancer treatment (Table 1).

The Regulatory Role of m^6^A Methyltransferase (Writers)
m^6^A methyltransferases (“Writers”) are an essential class of catalytic enzymes. METTL3 is a key regulator that promotes m^6^A modification, and the abnormal regulation of METTL3 is also inextricably linked to tumor development. Some studies have confirmed that the overexpression of METTL3 may be an important factor in promoting the development of common malignant tumors in women. Pan et al. found that the expression of RBM15 and METTL3 in CESC (cervical squamous cell carcinoma) tissues was higher than that in normal tissues (49). In addition, Hua et al. found that METTL3 promoted the epithelial-to-mesenchymal transition of ovarian cancer cells and the proliferation, invasion, and tumor formation of ovarian cancer cells, affecting their prognosis and overall survival (50). Moreover, Ma et al. compared the expressions of METTL14, WTAP, and METTL3 in ovarian cancer and found that METTL3 independently regulates m^6^A modification and thus affects the proliferation and metastasis (51). Besides, Li et al. found that METTL3 inhibited the viability of cervical cancer cells and enhanced their sensitivity to the chemotherapeutic drug cisplatin by downregulating the expression of the receptor for advanced glycation and its product in cervical cancer tissues (40); METTL3 modulated the m^6^A modification of MALAT1. The expression of MALAT1 is upregulated, and MALAT1 can promote the expression of high mobility group A2 (HMG A2) by sponge miR-26b, thereby promoting the development of breast cancer (41). Regarding this, Wang et al. proposed that WTAP may promote the proliferation, invasion, and migration of ovarian cancer through two gene sequences of FAM76A and HBS1 (42).

The Regulatory Role of m^6^A Methylation Reader Proteins (Readers)
As m^6^A methylation reading proteins, “Readers” can recognize the information of RNA methylation modification and
TABLE 1 | Dysregulation of m6A modification in common female malignant (CFM).

| m6A regulators | Target | Regulation in CFM | Function | Mechanisms |
|---------------|--------|-------------------|----------|------------|
| METTL3        | RAGE   | Down writer       | METTL3 increases cisplatin chemosensitivity of cervical cancer cells via downregulation of the activity of RAGE. Li, R. et al. (40); Wang, J. et al. (42).|
| MALAT1        | Down writer | The m6A methyltransferase METTL3 controls epithelial-mesenchymal transition, migration and invasion of breast cancer through the MALAT1/miR-556b/CMG22 axis. Zhao, C et al. (41).|
| WTAP          | HBS1L/FAM76A Up | writer Identification of WTAP-related genes by weighted gene co-expression network analysis in ovarian cancer. Wang, J. et al. (42).|
| YTHDF1        | RANBP2 Up reader | YTHDF1 Aggravates the Progression of Cervical Cancer Through m6A-Mediated Up-Regulation of RANBP2. Wang, H. et al. (43).|
| eIF3C         | Up reader | The m6A reader YTHDF1 promotes ovarian cancer progression via augmenting eIF3C translation. Liu, T. et al. (9).|
| eIF3          | WNT    Up reader | The Immune-Related Gene ELF3 is a Novel Biomarker for the Prognosis of Ovarian Cancer. Xu, H. et al. (44).|
| FTO           | mIR-181b-3p Up eraser | FTO-Dependent N (6)-Methyladenosine Modifications Inhibit Ovarian Cancer Stem Cell Self-Renewal by Blocking CAMP Signaling. Huang, H. et al. (47).|
| BNIP3         | Up eraser | RNA N6-methyladenosine demethylase FTO promotes breast tumor progression through inhibiting BNIP3. Niu, Y. et al. (46).|
| cAMP          | Down eraser | FTO-Dependent N (6)-Methyladenosine Modifications Inhibit Ovarian Cancer Stem Cell Self-Renewal by Blocking CAMP Signaling. Huang, H. et al. (47).|
| ALKBH5        | NANOG  Up eraser | RNA demethylase ALKBH5 promotes ovarian carcinogenesis in a simulated tumour microenvironment through stimulating NF-kappaB pathway. Jiang, Y. et al. (48).|

participate in downstream mRNA translation, degradation, and miRNA processing. It mainly changes the interaction between protein and RNA by weakening the homologous binding of m6A to RNA-binding protein and changing the secondary structure of RNA.

YTHDF1 acts as an important reading element in m6A modification by recognizing m6A-containing mRNAs and promoting their translation initiation and elongation (52). Wang et al. applied online data analysis to identify RANBP2 as a critical target of YTHDF1 in cervical cancer cells, and subsequent reduction of RANBP2 decreased cervical cancer cell proliferation, migration, and invasion (43). Overexpression of YTHDF1 promoted the growth, migration, and invasion of Hela and Siha cells. At the same time, knockdown of RANBP2 reversed the effect of overexpression of YTHDF1 on cervical cancer progression, indicating that YTHDF1 promotes cervical cancer progression by regulating RANBP2 expression in an m6A-dependent manner. Some scholars have proposed that YTHDF1 directly targets eIF3C (a subunit of EIF3) and promotes ovarian cancer's occurrence, metastasis, and prognosis (51).

Heterogeneous ribonucleoproteins (hnRNPs) are a diverse family of RNA-binding proteins that function in most stages of RNA metabolism (53). Studies have shown that HNRNPC regulates target transcripts’ abundance and alternative splicing through mA binding to RNA. Other researchers have proposed that hnRNPA2B1 can inhibit the growth of ovarian cancer cells, reduce the mobility of ovarian cancer cells in vitro, and hinder the formation of xenograft tumors in vivo. In addition, hnRNPA2B1 promotes the occurrence and development of malignant phenotypes of ovarian cancer by activating the expression of Lin28B (54). Moreover, Shi et al. found that lobaplatin induced apoptosis and cell cycle arrest by downregulating hnRNP A2/B1 in cervical cancer cells, and knockdown of hnRNP A2/B1 significantly reduced tumor growth in nude mice xenografts and increased cervical cancer Cellular sensitivity to lobaplatin and irinotecan (55). Eukaryotic initiation factor 3 (eIF3) can bind to m6A-modified bases in the 5’ UTR of RNA, promoting mRNA translation (56). The high expression of eIF3 in ovarian cancer is closely related to its poor prognosis (44); Zhu et al. concluded that eIF3B is highly expressed in cervical cancer tissues and is closely related to advanced FIGO in cervical cancer patients staging, shorter overall survival and lymph node metastasis (57).

The Regulatory Role of m6A Demethylases (erasers)
Demethylase is an integral part of the reversible modification of m6A, and FTO, as the first discovered demethylase, is widely present in adult and embryonic tissues, and its expression is exceptionally high in the brain. Moreover, recent studies have shown that FTO has an important effect on glioblastoma growth and self-renewal (58).

The expression level of FTO is also elevated in cervical squamous cell carcinoma, which can enhance chemoradiotherapy resistance in vitro and in vivo by reducing m6A-regulated β-catenin expression (59). Zhao et al. believed that FTO accelerated the growth of cancer cells by promoting proliferation, inhibiting apoptosis, and activating autophagy in ovarian cancer (60). In the latest study, Huang et al. found that FTO expression in high-grade serous ovarian cancer (HGSOC) tumor cells were significantly lower than that in other tissues, and it had a significant inhibitory effect on ovarian cancer cells (47). It can be seen that FTO may have a bidirectional regulatory impact on ovarian cancer tissue, and the specific mechanism needs to be further studied. Furthermore some researchers believe that FTO primarily stimulates the oncogenic activity of breast cancer cell invasion and migration through the FTO/miR-181b-3p/ARL5B signaling pathway, promoting tumor proliferation (61). The tumor suppressor BNIP3 is a downstream target of FTO-mediated m6A modification. FTO mediates m6A demethylation in the 3’UTR of BNIP3 mRNA and induces its degradation through a YTHDF2-independent mechanism,
M6A RNA Modification and Common Female Malignancies Therapy

The Role of M6A RNA Modification Targeted Drugs in the Treatment of Common Female Malignant Tumors

Targeted therapy is at the cellular and molecular level to design corresponding therapeutic drugs for the already defined carcinogenic sites. The drug enters the body and will surround the tumor to combine and act so as to make the tumor cells die instead of normal tissue cells surrounding the tumor are affected. Early studies of targeting strategies based on M6A modulators have focused on demethylases. Besides, previous studies have shown that M6A plays an essential role in the occurrence and development of tumors. Therefore, it is of great scientific significance and clinical value to develop specific inhibitors of M6A-related proteins. As the first discovered RNA-modifying demethylase, FTO, is widely involved in various physiological processes, and its dysregulation is associated with multiple human diseases.

Due to its involvement in obesity and obesity-induced metabolic diseases and the occurrence, development and prognosis of various cancers, such as melanoma, acute myeloid leukemia, glioblastoma, lung cancer, hepatocellular carcinoma and breast cancer, studies have shown that rhein can induce apoptosis (65).

Huang et al. systematically investigated the effect of rhein on adipogenesis by transcriptional and post-transcriptional approaches and found that rhein regulates m6A methylation rearrangement and adipogenesis in an independent manner, inhibiting fat mass and obesity-related (FTO) demethylase activity (66). It is indicated that rhein can inhibit the demethylation activity of FTO on m6A on mRNA in vitro and in vivo, thereby increasing the level of m6A in cells. ALKBH5 and FTO are both m6A demethylases. Studies have found that in ovarian cancer, the core cytokine NANOG is a key target to promote the development of ovarian cancer (48). On the contrary, there is much evidence to prove the overexpression of METTL3 in tumor tissues (67), while studies targeting METTL3 have shown that it can effectively inhibit tumor growth, proliferation, and metastasis (68).

In the progression of common malignant tumors in women, drug resistance that often occurs in the later stage is also a significant difficulty in its treatment. Chemotherapy resistance, especially platinum resistance, is recognized as a major cause of poor prognosis in ovarian cancer. Bowen Li et al. found that m6A can modulate the modification of anticancer drug resistance by modulating drug-target interactions and drug-mediated cell death signaling (69). The ethyl ester form of the FTO inhibitor Medlofenac (MA2) inhibits FTO and enhances the effect of the chemotherapeutic drug temozolom by targeting the MYC-miR-155/23a cluster-MXI1 feedback circuit in gliomas anti-tumor effects (45) (Table 2).

m6A RNA Modification and Immunotherapy

The tumor microenvironment (TME) is primarily responsible for mediating immunotherapy responses in tumor progression, and bioinformatics research has shown that m6A alteration and its regulators may regulate the TME and are linked to immune checkpoint inhibition (ICB) (7, 72).

Yi et al. systematically studied head and neck squamous cell carcinoma (HNSCC) compared with adjacent normal pairs, concluded that m6A regulators were upregulated in HNSCC, and found that m6A regulators were associated with PDL in the tumor microenvironment (TIME) (73). The expression of -1 was positively correlated, which may provide a promising target for improving the responsiveness of HNSCC to immunotherapy. In addition, He et al. systematically analyzed RNA-sequencing data of 24 major m6A methylation regulators in 775 breast cancer patients from the TCGA database and classified them for overall survival in the lower RNA methylation status group (RM1). The higher methylation status (RM2) group was significantly reduced (70). Moreover, the RM2 group displayed higher expression and higher numbers of tumor-infiltrating CD8+ T cells, helper T cells, and activated NK cells. The expressions of PD-L2, TIM3, and CCR4 were lower than those of the RM1 group, so it can be considered that the regulator of m6A is closely related to the malignant degree, prognosis, and anti-tumor immune response of breast cancer and can be used as a potential target and biological target for breast cancer immunotherapy.

In addition, anti-PD-1 immunotherapy is effective initially, but its efficacy is significantly reduced later due to FTO-mediated resistance (71). However, recent studies have shown that FTO knockdown can increase tumor sensitivity to anti-PD-1 immunotherapy, thereby improving efficacy (69). Therefore, the combined use of ICB and FTO inhibitors may block the development of drug resistance in individuals who develop adaptive immunity.
more inhibitors against m6A-related proteins brings a new dawn under its mechanisms of action. Although some methylase inhibitors have been understood and lack specificity, they still need to be explored. Currently, the understanding of how m6A affects immune phenotype is still in its infancy. Therefore, the development of more inhibitors against m6A-related proteins brings a new dawn for guiding tumor-targeted therapy based on RNA epigenetics. Targeted intervention in m6A modification can promote basic research in related fields, show excellent application prospects in tumor treatment and other disease-related fields, and show important scientific significance in life sciences and new drug discovery.

**CONCLUSIONS**

With the rapid development of high-throughput sequencing technology and bioinformatics, m6A has been gradually revealed as an important epigenetic modification with reversible properties, modification-related enzyme system, and role in different disease processes. It provides infinite possibilities for subsequent tumor diagnosis and treatment. These m6A-modified molecules are expected to become effective early diagnosis and prognostic markers for tumors and potential therapeutic targets, providing new ideas for tumor diagnosis and treatment.

Since m6A research provides a new understanding of the molecular mechanisms of tumorigenesis, metastasis, immune response, and drug resistance and promotes the development of new therapeutics, the process from theory to clinical translation still needs to be explored. Currently, the understanding of how m6A modification affects immune phenotype is still in its infancy. Although some methylase inhibitors have been discovered so far and provide new targets for tumor drugs, their mechanisms of action in vitro and in vivo are not fully understood and lack specificity. Therefore, the development of more inhibitors against m6A-related proteins brings a new dawn for guiding tumor-targeted therapy based on RNA epigenetics. Targeted intervention in m6A modification can promote basic research in related fields, show excellent application prospects in tumor treatment and other disease-related fields, and show important scientific significance in life sciences and new drug discovery.

**AUTHOR CONTRIBUTIONS**

HS and X-xH conceived and designed the study, Y-Iz, S-jW, JZ, Y-hS, JM, and Y-mC collected data and aided in writing the manuscript. HS and Y-Iz edited the manuscript. All authors read and approved the final manuscript.

**FUNDING**

This study was supported by the National Natural Science Foundation of China (No. 81802103, 81803938), Project of High-Level Talents in AHUTCM (Project code: 2019rcZD001), Excellent Young Scholars Project of Natural Science Foundation of Anhui Province in China (grant No. 2108085Y29), Natural Science Research Project of Colleges and Universities in Anhui Province (No. KJ2021A0557), Opening Project of Zhejiang Provincial Preponderant and Characteristic Subject of Key University (Chinese Traditional Medicine), and Zhejiang Chinese Medical University (No.ZYXZD2019004).

**REFERENCES**

1. Han X, Liu J, Cheng G, Cui S. Gene Signatures and Prognostic Values of M6a RNA Methylation Regulators in Ovarian Cancer. *Cancer Control* (2020) 27:1073274820960460. doi: 10.1177/1073274820960460

2. Sun H, Ferlay J, Siegel RL, Laversanne M, Soerjomataram I, Jamal A, et al. Global Cancer Statistics 2020: GLOBOCAN Estimates of Incidence and Mortality Worldwide for 36 Cancers in 185 Countries. *CA Cancer J Clin* (2021) 71:209–49. doi: 10.3322/caac.21660

3. Byrd DR, Brierley JD, Baker TP, Sullivan DC, Gress DM. Current and Future Cancer Staging After Neoadjuvant Treatment for Solid Tumors. *CA Cancer J Clin* (2021) 71:140–8. doi: 10.3322/caac.21640

4. Graillon T, Tabouret E, Chinot O. Chemotherapy and Targeted Therapies for Meningiomas: What Is the Evidence? *Curr Opin Neurol* (2021) 34:857–67. doi: 10.1097/WCO.0000000000001002

5. He L, Li H, Wu A, Peng Y, Shu G, Yin G. Functions of N6-Methyladenosine and its Role in Cancer. *Mol Cancer* (2019) 18:176. doi: 10.1186/s12943-019-1109-9

6. Zhou Z, Lv J, Yu H, Han J, Yang X, Feng D, et al. Mechanism of RNA Modification N6-Methyladenosine in Human Cancer. *Mol Cancer* (2020) 19:104. doi: 10.1186/s12943-020-01216-3

7. Kuai D, Zhu S, Shi H, Yang R, Liu T, Liu H, et al. aberrant expression of M(6)A mRNA Methylation Regulators in Colorectal Adenoma and Adenocarcinoma. *Life Sci* (2021) 273:119258. doi: 10.1016/j.lfs.2021.119258

8. Zhang B, Chen Z, Tao B, Yi C, Lin Z, Li Y, et al. M(6)A Target microRNAs in Serum for Cancer Detection. *Mol Cancer* (2021) 20:170. doi: 10.1186/s12943-021-01477-6

9. Liu S, Li G, Li Q, Zhang Q, Zhuo L, Chen X, et al. The Roles and Mechanisms of YTH Domain-Containing Proteins in Cancer Development and Progression. *Am J Cancer Res* (2020) 10(4):1668–84

10. Liu N, Zhou K, Parisien M, Dai Q, Diatchenko L, Pan T. N6-Methyladenosine Alters RNA Structure to Regulate Binding of a Low-Complexity Protein. *Nucleic Acids Res* (2017) 45:6561–63. doi: 10.1093/nar/gkx141

11. Pi J, Wang W, Ji M, Wang X, Wei X, Jin J, et al. YTHDF1 Promotes Gastric Carcinogenesis by Controlling Translation of Fzd7. *Cancer Res* (2021) 81:2651–65. doi: 10.1158/0008-5472.CAN-20-0066

12. Zaccara S, Ris RJ, Jaffrey SR. Reading, Writing and Erasing mRNA Methylation. *Nat Rev Mol Cell Biol* (2019) 20:608–24. doi: 10.1038/s41580-019-0168-5

13. Wen J, Lv R, Ma H, Shen H, He C, Wang J, et al. Zc3h13 Regulates Nuclear RNA M(6)A Methylation and Mice Embryonic Stem Cell Self-Renewal. *Mol Cell* (2018) 69:1028–1036 e1026. doi: 10.1016/j.molcel.2018.02.015
14. Lan Y, Liu B, Guo H. The Role of M(6A) Modification in the Regulation of Tumor-Related IncRNAs. *Mol Ther Nucleic Acids* (2021) 24:768–79. doi: 10.1016/j.ymeth.2021.05.002

15. Mendel M, Delaney K, Pandey RR, Chen KM, Wenda JM, Vagbo CB, et al. Splice Site M(6A) Methylation Prevents Binding of U2AF35 to Inhibit RNA Splicing. *Cell* (2021) 184:3125–3142.e1325. doi: 10.1016/j.cell.2021.03.062

16. Zhu W, Wang ZJ, Wei JF, Lu C. Role of M6A Methyltransferase Component VIRMA in Multiple Human Cancers (Review). *Cancer Cell Int* (2021) 21:172. doi: 10.1186/s12935-021-01868-1

17. Su R, Dong L, Li Y, Gao M, Han L, Wunderlich M, et al. Targeting FTO Inhibits Cell Proliferation and the Metastasis of Colon Cancer by Regulating the FOXP3/miR-21/SPRY2 Axis. *Am J Transl Res* (2021) 13:10110–22.

18. Li Y, Xiao J, Tian Y, Qu Y, Chen X, et al. Molecular Characterization and Clinical Relevance of M(6A) Readers Across 33 Cancer Types. *Mol Cancer* (2019) 18:137. doi: 10.1186/s12943-019-1066-3

19. Huang W, Weng H, Chen J. M(6A) Modification in Coding and Non-Coding RNAs: Roles and Therapeutic Implications in Cancer. *Cancer Cell* (2020) 37:270–86. doi: 10.1016/j.ccell.2020.02.004

20. Kopp F, Mendell JT. Functional Classification and Experimental Dissection of Long Noncoding RNAs. *Cell* (2018) 172:393–407. doi: 10.1016/j.cell.2018.01.011

21. Furlan G, Gutiérrez Hernandez N, Huret C, Galupa R, Van Bemmel JG, Romito A, et al. The Ftx Noncoding Locus Controls X Chromosome Inactivation Independently of Its RNA Products. *Mol Cell* (2018) 70:462–472.e686. doi: 10.1016/j.molcel.2018.03.024

22. Lee JT, Bartolomei MS. X-Inactivation, Imprinting, and Long Noncoding RNAs in Health and Disease. *Cell* (2013) 152:1308–23. doi: 10.1016/j.cell.2013.02.016

23. Ban Y, Tan P, Cai J, Li J, Hu M, Zhou Y, et al. LncCAROD Is Stabilized by M6a Methylation and Promotes Cancer Progression via Forming a Ternary Complex With HSPA1A and YBX1 in Head and Neck Squamous Cell Carcinoma. *Mol Oncol* (2020) 14:1282–96. doi: 10.1016/j.molonc.2020.02.12676

24. Yang X, Zhang S, He C, Xue P, Zhang L, He Z, et al. METTL14 Suppresses G1 Arrest by P53 Pathway in Esophageal Squamous Cell Carcinoma. *Biochem Biophys Res Commun* (2022) 54:2245–55. doi: 10.1016/j.bbrc.2022.05.013

25. Yang X, Zhang S, He C, Xue P, Zhang L, He Z, et al. METTL14 Suppresses Cancer Stem Cell Maintenance and Immune Evasion. *Mol Cell* (2016) 62:335–45. doi: 10.1016/j.molcel.2016.03.021

26. Su R, Dong L, Li Y, Gao M, He PC, Liu W, et al. METTL16 Exerts an M(6A)-Independent Function to Facilitate Translation and Tumorigenesis. *Nat Biol Cell* (2022) 24:205–16. doi: 10.1038/s41556-021-00835-2

27. Li R, Song Y, Chen X, Chu M, Wang ZW, Zhu X. METTL3 Increases Cisplatin Chemosensitivity of Cervical Cancer Cells via Downregulation of the Activity of RAGE. *Mol Ther Oncolytics* (2021) 22:245–55. doi: 10.1016/j.omtn.2021.05.013

28. Zhao C, Ling X, Xia Y, Yan B, Guan Q, The M6a Methyltransferase METTL3 Controls Epithelial-Mesenchymal Transition, Migration and Invasion of Breast Cancer Through the MALAT1/miR-26b/HMGAA2 Axis. *Cancer Cell Int* (2021) 21:441. doi: 10.1186/s12935-021-02113-5

29. Wang J, Xu J, Li K, Huang Y, Dai Y, Xu C, et al. Identification of WATP-Related Genes by Weighted Gene Co-Expression Network Analysis in Ovarian Cancer. *J Ovarian Res* (2020) 13:119. doi: 10.1186/s13040-020-00710-y

30. Wang H, Luo Q, Kang J, Wei Q, Yang Y, Deng D, et al. YTHDF1 Aggravates the Progression of Cervical Cancer Through M(6A)-Mediated Up-Regulation of RNBP2. *Front Oncol* (2021) 11:650833. doi: 10.3389/fonc.2021.650833

31. Xu H, Wang H, Li G, Jin X, Chen B. The Immune-Related Gene ELF3 Is a Novel Biomarker for the Prognosis of Ovarian Cancer. *Int J Gen Med* (2021) 14:5537–48. doi: 10.2147/IJGM.S332320

32. Xiao L, Li X, Mu Z, Zhou J, Zhou P, Xie C, et al. FTO Inhibition Enhances the Antitumor Effect of Temozolomide by Targeting MYC-miR-155-23a Cluster-MX1 Feedback Circuit in Glioma. *Cancer Res* (2020) 80:3945–58. doi: 10.1158/0008-5472.CAN-20-0132

33. Niu Y, Lin Z, Wan A, Chen H, Liang H, Sun L, et al. RNA N6-Methyladenosine Demethylase FTO Promotes Breast Tumor Progression Through Inhibiting BNP3. *Mol Cancer* (2019) 18:146. doi: 10.1186/s12943-019-1004-4

34. Huang W, Yang Y, Kandpal M, Zhao G, Cardenas H, Ji Y, et al. FTO-Dependent N (6)-Methyladenosine Modifications Inhibit Ovarian Cancer Stem Cell Self-Renewal by Blocking cAMP Signaling. *Cancer Res* (2020) 80:3320–14. doi: 10.1158/0008-5472.CAN-19-19404

35. Jiang Y, Yan W, Gong M, Zhou S, Qiu J, Cheng W. RNA Demethylase ALKBH5 Promotes Ovarian Carcinogenesis in a Simulated Tumour Microenvironment Through Stimulating NF-kappaB Pathway. *J Cell Mol Med* (2020) 24:6137–48. doi: 10.1111/jcmm.15228

36. Pan J, Xu L, Pan H. Development and Validation of an M6A RNA Methylation Regulator-Based Signature for Prognostic Prediction in Cervical Squamous Cell Carcinoma. *Front Oncol* (2020) 10:1444. doi: 10.3389/fonc.2020.01444

37. Huo W, Zhao Y, Jin X, Yu D, He J, Xie D, et al. METTL3 Promotes Ovarian Carcinoma Growth and Invasion Through the Regulation of AXL Translation and Epithelial to Mesenchymal Transition. *Gynecol Oncol* (2018) 151:356–65. doi: 10.1016/j.jgyno.2018.09.015

38. Ma Z, Li Q, Liu P, Dong W, Zhuo Y. METTL13 Regulates M6a in Endometrioid Epithelial Ovarian Cancer Independently of METTL4 and WATP. *Cell Biol Int* (2020) 44:2524–31. doi: 10.1002/cbi.15228

39. Lin X, Chai G, Wu Y, Li J, Chen F, Liu J, et al. RNA M(6)A Methylation Regulates the Epithelial Mesenchymal Transition of Cancer Cells and Translation of Snail. *Nat Commun* (2019) 10:2065. doi: 10.1038/s41467-019-09885-9

40. Lerner Good JD, Tolbert BS. Idiosyncrasies of hnRNP A1-RNA Recognition: Can Binding Mode Influence Function. *Semin Cell Dev Biol* (2019) 86:151–60. doi: 10.1016/j.semcdb.2019.04.001

41. Yang Y, Wei Q, Tang Y, Yuan Yuan W, Luo Q, Zhao H, et al. Loss of Hnrnpa2B1 Inhibits Micrornanent and Promotes Apoptosis via
Huang L, Zhang J, Zhu X, Mi X, Li Q, Gao J, et al. The Phytochemical Rhein.

Heo SK, Noh EK, Kim JY, Jegal S, Jeong Y, Cheon J, et al. Rhein Augments.

Wang X, Zhang J, Wang Y. Long Noncoding RNA GAS5-AS1 Suppresses.

Zhu H, Gan X, Jiang X, Diao S, Wu H, Hu J. ALKBH5 Inhibited Autophagy of.

Gao R, Ye M, Liu B, Wei M, Ma D, Dong K. M6a Modification in Female Malignancies.

Zhu et al. m6A Modification in Female Malignancies.