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Epitranscriptome regulation

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Recent scientific breakthroughs have revealed the importance of dynamic covalent modifications of mRNAs and noncoding RNAs. These modifications play essential regulatory roles in many RNA-processing events, including splicing, transport, translation and decay. They can alter local charge, base-pairing potential, secondary structure and RNA-protein interactions, which in turn shape gene expression. Thus, the 'epitranscriptome', as this ensemble of RNA modifications is now known, bestows transcripts with information beyond RNA sequence.

As the cellular factors that deposit ('writers'), remove ('erasers') or recognize ('readers') RNA modifications are identified, their roles in cellular, developmental and disease processes are uncovered. Although research into the more recently recognized RNA modifications is in an early phase, the body of information on the functions and mechanisms of action of N^6-methyladenosine (m^6A), an abundant and well-studied RNA modification highlighted here, will pave the way to a better understanding of the importance of epitranscriptomic regulation.

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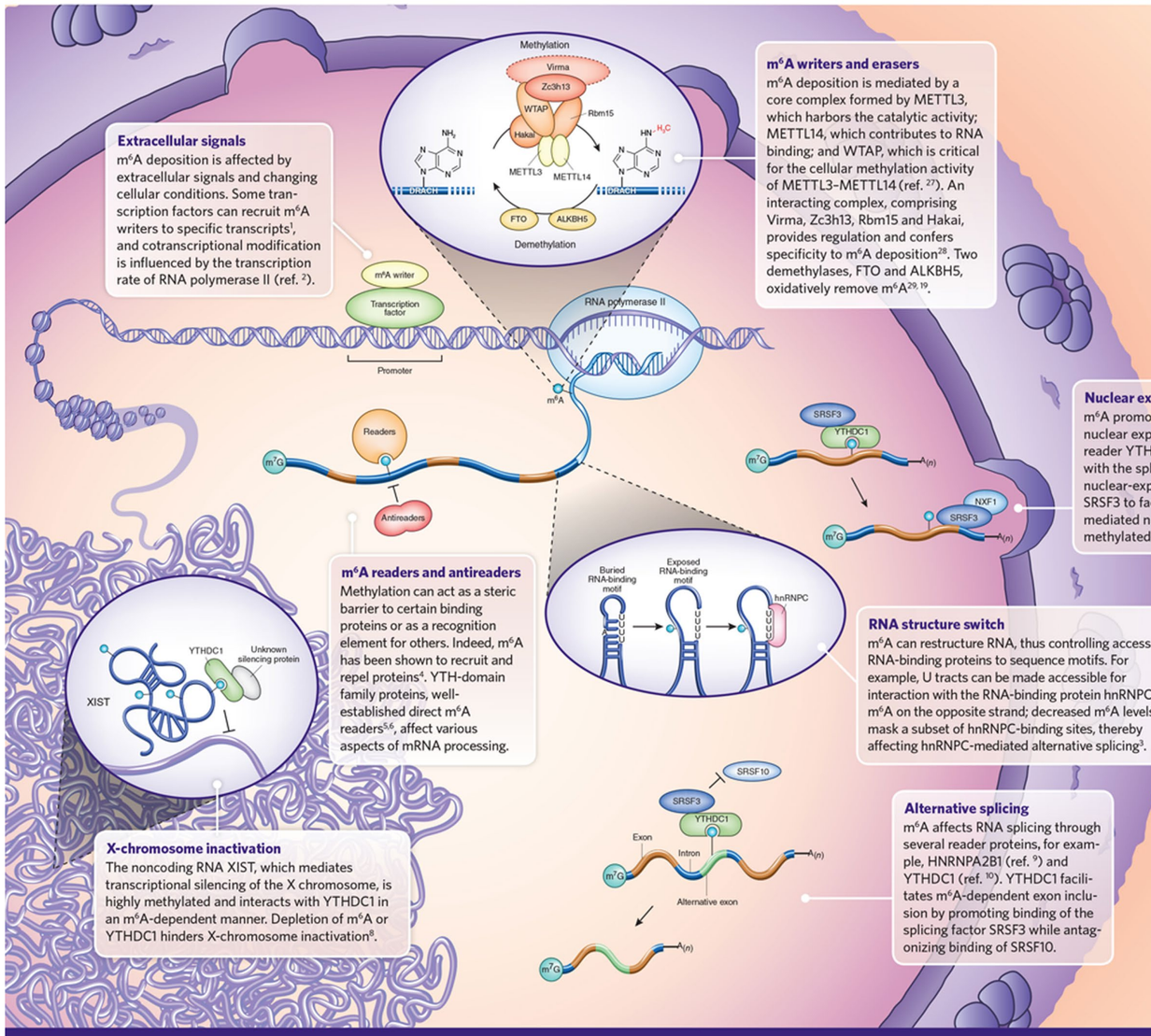


Extracellular signals
m^6A deposition is affected by extracellular signals and changing cellular conditions. Some transcription factors can recruit m^6A writers to specific transcripts[1], and cotranscriptional modification is influenced by the transcription rate of RNA polymerase II (ref. [2]).

m^6A writers and erasers
m^6A deposition is mediated by a core complex formed by METTL3, which harbors the catalytic activity; METTL14, which contributes to RNA binding; and WTAP, which is critical for the cellular methylation activity of METTL3–METTL14 (ref. [27]). An interacting complex, comprising Virma, Zc3h13, Rbm15 and Hakai, provides regulation and confers specificity to m^6A deposition[28]. Two demethylases, FTO and ALKBH5, oxidatively remove m^6A[29,19].

Nuclear export
m^6A promotes mRNA nuclear export. The m^6A reader YTHDC1 interacts with the splicing factor and nuclear-export adaptor SRSF3 to facilitate NXF1-mediated nuclear export of methylated transcripts[7].

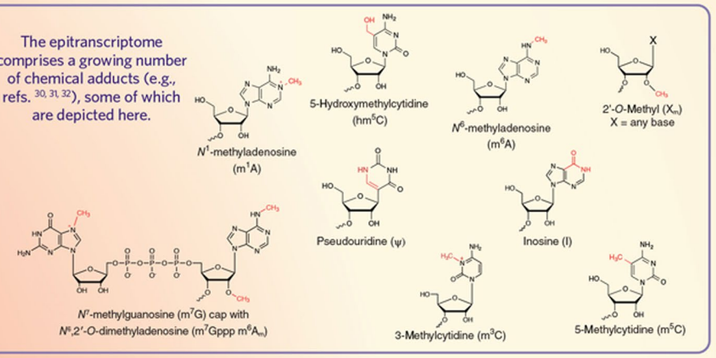
m^6A readers and antireaders
Methylation can act as a steric barrier to certain binding proteins or as a recognition element for others. Indeed, m^6A has been shown to recruit and repel proteins[4]. YTH-domain family proteins, well-established direct m^6A readers[5,6], affect various aspects of mRNA processing.

RNA structure switch
m^6A can restructure RNA, thus controlling access of RNA-binding proteins to sequence motifs. For example, U tracts can be made accessible for interaction with the RNA-binding protein hnRNPC by m^6A on the opposite strand; decreased m^6A levels mask a subset of hnRNPC-binding sites, thereby affecting hnRNPC-mediated alternative splicing[3].

X-chromosome inactivation
The noncoding RNA XIST, which mediates transcriptional silencing of the X chromosome, is highly methylated and interacts with YTHDC1 in an m^6A-dependent manner. Depletion of m^6A or YTHDC1 hinders X-chromosome inactivation[8].

Alternative splicing
m^6A affects RNA splicing through several reader proteins, for example, HNRNPA2B1 (ref. [9]) and YTHDC1 (ref. [10]). YTHDC1 facilitates m^6A-dependent exon inclusion by promoting binding of the splicing factor SRSF3 while antagonizing binding of SRSF10.

The epitranscriptome comprises a growing number of chemical adducts (e.g., refs. [30,31,32]), some of which are depicted here.



m^6A in development and disease

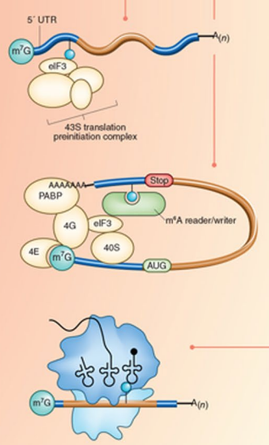
Given the broad effects of m^6A on mRNA homeostasis, it is unsurprising that epitranscriptomic regulation affects development and diseases such as cancer. A few examples are listed below:

- In embryonic stem cells (ESCs), m^6A regulates the stability of mRNAs, including those encoding core pluripotency transcription factors, and deletion of the methyltransferase METTL3 impairs ESC exit from self-renewal[17].
- During mammalian germ cell development, loss of m^6A writers, readers[18,15] and erasers[19] affects both female and male gametogenesis, as well as mouse fertility.
- METTL3 promotes the translation of specific oncogenes by directly interacting with the translation-initiation machinery; METTL3 is required for growth, survival and invasion of human lung cancer cells[20].
- m^6A influences glioblastoma tumorigenesis, partly by regulating the self-renewal of glioblastoma stem cells (GSCs) through effects on critical mRNAs. Inhibition or depletion of m^6A demethylases suppresses GSC proliferation and tumor progression[21,22].
- m^6A methylation is important for hematopoietic stem/progenitor cell specification, and aberrant methylation is linked to acute myeloid leukemia (AML) pathogenesis[23,24]. The m^6A demethylase FTO promotes oncogene-mediated leukemogenesis[25] and is highly expressed in certain AML subtypes. Promoter-bound METTL3 maintains myeloid leukemia by enhancing translation of those transcripts[26].

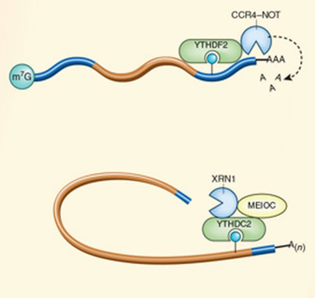
Translation
At the 5′ untranslated region (UTR) of mRNA, a region critical for ribosome recruitment, m^6A can bypass the requirement of translation initiation factor eIF4E through direct binding to eIF3, which is sufficient to recruit the 43S complex and initiate translation[11].

In addition, YTHDF1, YTHDF3 and METTL3 bind m^6A-modified transcripts and increase translation, possibly through interaction with eIF3 (refs. [12,20]).

During translation elongation, m^6A and other RNA modifications impede tRNA accommodation. The presence of m^6A within a codon disrupts cognate-tRNA selection, especially the most thermodynamically unstable steps of initial selection, thus altering translational dynamics[13].



Transcript stability
On average, m^6A-methylated transcripts are more short-lived than transcripts with unmodified adenosines. Degradation is promoted partly by the m^6A reader YTHDF2, which removes m^6A-containing transcripts from translatable pools[5]. YTHDF2 also recruits the CCR4–NOT deadenylase, which initiates mRNA deadenylation and subsequent degradation[14].



In the mouse male germ line, YTHDC2 interacts with the exoribonuclease XRN1 (ref. [15]), a protein associated with RNA decay.

In contrast to m^6A, the presence of m^6A_m at the first nucleotide adjacent to the m^7G cap increases transcript stability by conferring resistance to the mRNA-decapping enzyme DCP2 (ref. [16]).

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