

PEARLS

RNA modifications go viral

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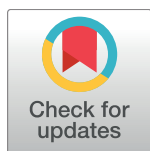
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Introduction

Viral life cycles are often coordinated by precise mechanisms that act on their RNA. For example, the microRNA miR-122 interacts with the viral RNA genome of hepatitis C virus (HCV) and is required for HCV replication [1]. In the past year, several groups have reported a new RNA regulatory control to viral infection—the posttranscriptional RNA modification N⁶-methyladenosine (m⁶A). This reversible RNA modification is the most prevalent internal modification of the more than 60 known chemical modifications in eukaryotic RNA. The deposition of m⁶A on RNA is controlled by cellular m⁶A machinery comprising methyltransferase and demethylase enzymes, as well as m⁶A-specific binding proteins (recently reviewed in [2]; Fig 1). By affecting mRNA and noncoding RNA structure, localization, and function, m⁶A plays an important role in many fundamental biological processes [2].

A role for m⁶A in viral infection has been hypothesized since the 1970s, when m⁶A was found on RNA of several viruses [3–7]. Recently, advances in sequencing-based strategies used to profile m⁶A have expanded the known repertoire of viruses with m⁶A in their RNA to include human immunodeficiency virus 1 (HIV-1) and RNA viruses in the family *Flaviviridae*,



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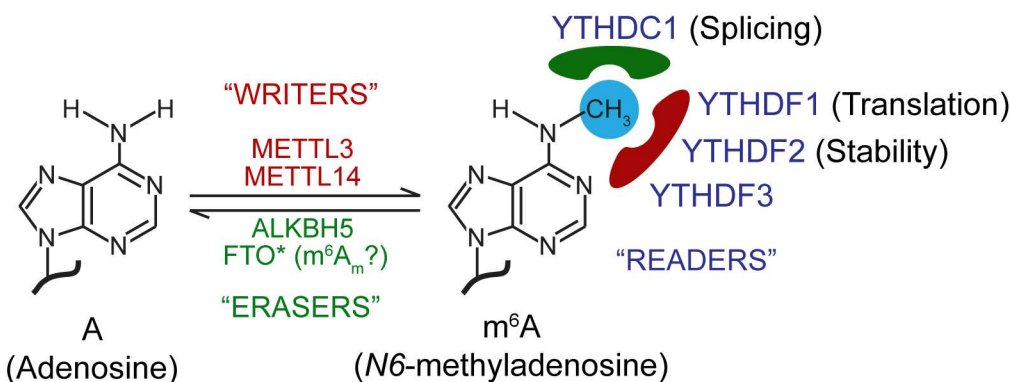


Fig 1. The cellular m⁶A machinery. N⁶-methyladenosine (m⁶A) is a reversible RNA modification that occurs in cellular and viral RNA. The deposition of m⁶A at the consensus motif DRAmCH (where D = G/A/U, R = G > A, and H = U/C/A) is governed by a cellular methyltransferase complex composed of the “writers” METTL3 and METTL14, and other noncatalytic cofactors. m⁶A modification can be reversed by the “erasers” FTO and ALKBH5. *We note that FTO has recently been found to have greater specificity for the m⁶Am modifications present in mRNA cap structures than for m⁶A [34]. “Reader” m⁶A-specific RNA binding proteins, including the cytoplasmic YTHDF1, YTHDF2, YTHDF3, and nuclear YTHDC1 control the function of m⁶A on RNA. YTHDF1 promotes translation of cellular m⁶A-mRNAs, while YTHDF2 targets them for degradation. YTHDC1 regulates the splicing of m⁶A-modified pre-mRNA. The role of m⁶A and the m⁶A machinery in RNA function and biological processes is further reviewed in [2].

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Table 1. List of viruses known to contain m⁶A in their RNA.

Virus	Summary of knowledge	References
DNA viruses		
Simian virus 40	<ul style="list-style-type: none"> • Viral late transcripts have ~3 internal m⁶A residues. • Blocking m⁶A with cycloleucine impairs nuclear processing and export of late viral mRNAs. 	[3, 17, 28]
Adenovirus-2	<ul style="list-style-type: none"> • Viral RNAs contain internal m⁶A. • Prior to splicing, viral RNA is modified by m⁶A. • m⁶A is retained in the viral mRNA after nuclear export. 	[4, 29]
Herpes simplex virus	<ul style="list-style-type: none"> • Viral mRNAs contain internal m⁶A. 	[5]
Retroviruses		
HIV-1	<ul style="list-style-type: none"> • Viral mRNA and genomes contain m⁶A, which is concentrated at 3' regions. • m⁶A sites at the Rev-response element RNA structure alter nuclear export of viral RNA. • m⁶A-binding YTHDF proteins bind to viral RNA, promote viral replication, and may suppress genomic RNA reverse transcription following infection. 	[8, 9, 10]
Rous sarcoma virus	<ul style="list-style-type: none"> • Genomic RNA has ~10–15 m⁶A residues per molecule mostly in the 3' terminal third of the genomic RNA. • Blocking m⁶A by cycloleucine reduces formation of the mature, spliced <i>Env</i> mRNA. 	[6, 13, 14, 19, 21, 22, 26, 27]
Feline leukemia virus	<ul style="list-style-type: none"> • Genomic RNA contains internal m⁶A modification. 	[15]
Moloney murine leukemia virus	<ul style="list-style-type: none"> • Genomic RNA contains internal m⁶A modification. 	[16]
(+)-stranded RNA virus		
HCV	<ul style="list-style-type: none"> • The viral RNA genome has multiple internal m⁶A sites. • m⁶A suppresses viral particle production, but does not affect viral RNA replication. • YTHDF proteins suppress viral particle production, and relocalize to viral assembly sites around lipid droplets. • Mutation of one cluster of m⁶A sites increases viral particle production. 	[11]
ZIKV	<ul style="list-style-type: none"> • The viral RNA genome has multiple internal m⁶A sites, with differences in m⁶A modification patterns in 3 strains. • m⁶A and YTHDF proteins suppress viral infection. 	[11, 12]
Dengue, Yellow fever, and West Nile virus	<ul style="list-style-type: none"> • The viral RNA genome has multiple internal m⁶A sites. 	[11]
(-)-stranded RNA viruses		
Influenza A virus	<ul style="list-style-type: none"> • Viral mRNAs and genomic RNA segments have internal m⁶A. • m⁶A is unequally distributed on viral mRNAs. 	[7, 18]

YTHDF, YTH domain family.

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such as HCV and Zika virus (ZIKV; [Table 1](#)) [8–12]. In this article, we will review the emerging role for m⁶A in regulating viral infection.

A historical perspective on m⁶A in viral RNA

Early work on RNA modifications in the 1970s often used viral systems to characterize RNA modifications, including the mRNA “cap” structures. Such early studies, which used chromatographic analysis of radiolabeled and enzymatically digested RNA, uncovered a high degree of internal m⁶A modification in cellular mRNA and viral RNAs from the DNA viruses simian virus 40 (SV40), adenovirus-2, and herpes simplex virus 1 [3–5]. m⁶A was also found in the viral genomic RNA of multiple retroviruses and in the mRNA from influenza A virus, a negative-stranded RNA virus [6, 7, 13–16]. Interestingly, all these viruses have a nuclear stage in their life cycle, which led the field to believe that the nucleus was the primary site of m⁶A modification of RNA.

Subsequent experiments mapped the m⁶A sites in viral RNAs, revealing an interesting heterogeneity in viral m⁶A patterns. Rous sarcoma virus genomic RNA contained 10–15 m⁶A modifications per molecule, all localized to the 3' half of genomic RNA, while m⁶A in SV40 and adenovirus-2 mRNA was near spliced regions [4, 14, 17]. Furthermore, the number of m⁶A residues on individual segments of influenza A virus mRNAs varied greatly between segments [18]. These viral m⁶A-mapping studies revealed a putative consensus motif for m⁶A: GA^mC and AA^mC, which was also later confirmed in cellular mRNA [17, 19, 20]. Indeed mutation of GAC to GAU in a cluster of two such motifs in Rous sarcoma virus prevented m⁶A modification at these sites [21, 22]. Modern sequencing techniques to detect m⁶A based on enrichment of m⁶A-modified RNA fragments using an m⁶A-specific antibody (m⁶A-seq), as well as biochemical analyses of the specificity of the m⁶A methyltransferase complex, have validated the early findings on viral RNA. The consensus motif for m⁶A is now known to be DR^mCH (where D = G/A/U, R = G > A, and H = U/C/A) [23–25].

Early research on m⁶A in viral infection pointed to the modification regulating viral RNA splicing. The m⁶A-methylation inhibitor cycloleucine reduced splicing of the Rous sarcoma virus *Env* mRNA and impaired the proper nuclear processing and export of SV40 late mRNA [26–28]. Furthermore, m⁶A was proposed to regulate the splicing of adenovirus-2 late transcripts [29]. Indeed, m⁶A has now been shown to regulate mRNA splicing, highlighting the value of these early viral studies in uncovering m⁶A function [30].

Recent advances in m⁶A in viral RNA

The recent identification of the cellular m⁶A machinery (see Fig 1) now allows for mechanistic studies on the function of this RNA modification during viral infection [2]. Recently, three groups have found a proviral role for m⁶A in HIV-1 infection [8–10]. Interestingly, these studies found that the function of individual m⁶A sites in HIV-1 RNA can be varied, ranging from regulating HIV-1 RNA nuclear export to enhancing viral gene expression [8, 9]. Furthermore, the m⁶A-binding cytosolic YTH domain family (YTHDF) proteins were found to bind to HIV-1 RNA at m⁶A sites [9, 10] but have varied roles in regulating HIV-1 infection, from promoting viral transcript abundance and translation to suppressing viral reverse transcription [9, 10]. Given that m⁶A regulates splicing during infection by other retroviruses, HIV-1 mRNA splicing may also be affected by m⁶A and by YTHDC1, a nuclear YTH domain containing m⁶A-binding protein involved in cellular mRNA splicing [30]. While this work has uncovered m⁶A and the m⁶A machinery as important regulators of HIV-1 infection, one general limitation of experiments involving the knockdown of the cellular m⁶A machinery is that such depletion could affect the expression of pro- or anti-viral host factors, leading to an indirect effect on viral infection. Experiments involving viruses that contain m⁶A-abrogating mutations will be invaluable in pinpointing the direct role of this modification on viral RNA during infection.

We, and others, have recently found a role for m⁶A in regulating RNA viruses of the *Flaviviridae* family. Using m⁶A-seq, we mapped several regions modified by m⁶A across the RNA genomes of the *Flaviviridae* members HCV, ZIKV, dengue virus, yellow fever virus, and West Nile virus [11]. Concurrently, another group also identified m⁶A in ZIKV RNA [12]. These viral RNAs are the first examples of exclusively cytoplasmic RNA species that contain m⁶A, indicating that the cellular m⁶A methyltransferases may be active in the cytoplasm under some cases. Indeed, the m⁶A methyltransferases are present in the cytoplasm as well as the nucleus [11]. Perhaps they are targeted to viral RNAs by cellular factors yet to be defined that modulate the specificity and localization of the methyltransferase complex. We also tested if m⁶A had any role in *Flaviviridae* infection, and found that m⁶A suppressed the packaging of HCV RNA

into infectious viral particles. A conserved cluster of four m⁶A sites in the HCV E1 gene was the primary driver of this phenotype, such that abrogation of m⁶A in this region by mutation altered RNA–protein interactions required for viral assembly [11]. Similar to our work, Lichinchi et al. found that m⁶A also limited ZIKV infection, suggesting that m⁶A negatively regulates *Flaviviridae* infection [11, 12]. Both of these studies mapped the *Flaviviridae* m⁶A sites at a single time point of infection, catching only a snapshot of the overall m⁶A profile on the viral genomes. As current m⁶A-mapping technologies do not allow us to easily determine the m⁶A occupancy of any individual site or whether it occurs on the same viral RNA species, it is likely that these viral genomes will have divergent m⁶A sites and occupancies at different stages of their life cycles. For example, we found that virion-associated RNA had less overall m⁶A than intracellular replicating HCV RNA [11]. By expanding these studies to capture the viral m⁶A sites over a time course of infection or on specific viral RNA species, we could identify new controls governed by m⁶A that regulate specific aspects of viral replication, including viral RNA stability, translation, replication, packaging, or even immune evasion (see below). Furthermore, as m⁶A destabilizes RNA secondary structure [2], it could directly alter *cis*-regulatory structural elements in RNA virus genomes.

The presence of RNA modifications on viral RNAs may prevent detection by host pattern recognition receptors that trigger antiviral innate immunity. Indeed, two studies have shown that internal m⁶A modification of in vitro synthesized RNAs ameliorates innate immune activation by the known RNA-sensing pattern recognition receptors TLR3 and RIG-I [31, 32]. Therefore, m⁶A-modification of the pathogen-associated molecular patterns within viral RNA may be an evolutionary adaptation for immune evasion. Identifying m⁶A modification in viral RNA pathogen-associated molecular patterns during infection will be critical in proving that m⁶A serves as a shield on viral RNA to prevent induction of antiviral signaling pathways.

Epitranscriptomic changes to host mRNA during viral infection

Viral infection induces broad changes in the host transcriptome and proteome. Therefore, it is not surprising that viral infection can also alter the m⁶A-epitranscriptome in host mRNA. Indeed, both HIV-1 and ZIKV impact the host m⁶A-epitranscriptome with changes to the specific transcripts containing m⁶A and to the overall m⁶A-topology [8, 12]. Specifically, during viral infection, the level of m⁶A increases at the 5'UTR of mRNAs, with a concomitant decrease in m⁶A modification at 3'UTRs. A similar increase in m⁶A at 5'UTRs has been reported in heat shock-related transcripts during heat shock, which promotes the translation of these mRNAs [33]. Interestingly, during viral infection, many m⁶A-altered transcripts are related to viral replication and immune responses [8, 12]. Therefore, m⁶A modification to specific mRNAs could be virally induced to promote infection, or by the host to restrict infection, allowing for an additional layer of gene expression regulation. Future studies on viral- or host-mediated epitranscriptomic changes and identification of the factors that regulate these altered epitranscriptomes will be essential to understanding how viral infection alters host gene expression.

Conclusions and future perspectives

As important posttranscriptional modulators of RNA function, m⁶A and other RNA modifications likely regulate infection by all classes of viruses. Recent scientific and technological advances have now set the stage for the systematic exploration of many outstanding questions regarding the role of m⁶A during viral infection. Going forward, perturbing the host m⁶A machinery and mutating m⁶A motifs in viral RNAs will be invaluable techniques used to study the function of m⁶A on viral RNA structure, localization, splicing, stability, translation, and

immune evasion. Furthermore, understanding viral- or host-induced changes in the cellular m⁶A epitranscriptome will be crucial in understanding gene regulation during viral infection. Indeed, as for many fundamental biological systems, viral infection may prove to be a useful model for understanding how m⁶A affects cellular RNA expression and function. Therefore, we expect that virology and its exciting discoveries will be at the heart of the renaissance of m⁶A and RNA modification research.

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References

- Jopling CL, Yi M, Lancaster AM, Lemon SM, Sarnow P. Modulation of hepatitis C virus RNA abundance by a liver-specific MicroRNA. *Science*. 2005; 309(5740):1577–81. doi: [10.1126/science.1113329](https://doi.org/10.1126/science.1113329) PMID: [16141076](https://pubmed.ncbi.nlm.nih.gov/16141076/)
- Zhao BS, Roundtree IA, He C. Post-transcriptional gene regulation by mRNA modifications. *Nat Rev Mol Cell Biol*. 2017; 18(1):31–42. doi: [10.1038/nrm.2016.132](https://doi.org/10.1038/nrm.2016.132) PMID: [27808276](https://pubmed.ncbi.nlm.nih.gov/27808276/)
- Lavi S, Shatkin AJ. Methylated simian virus 40-specific RNA from nuclei and cytoplasm of infected BSC-1 cells. *Proc Natl Acad Sci U S A*. 1975; 72(6):2012–6. PMID: [166375](https://pubmed.ncbi.nlm.nih.gov/166375/)
- Sommer S, Salditt-Georgieff M, Bachenheimer S, Darnell JE, Furuichi Y, Morgan M, et al. The methylation of adenovirus-specific nuclear and cytoplasmic RNA. *Nucleic Acids Res*. 1976; 3(3):749–65. PMID: [1272797](https://pubmed.ncbi.nlm.nih.gov/1272797/)
- Moss B, Gershowitz A, Stringer JR, Holland LE, Wagner EK. 5'-Terminal and internal methylated nucleosides in herpes simplex virus type 1 mRNA. *J Virol*. 1977; 23(2):234–9. PMID: [196108](https://pubmed.ncbi.nlm.nih.gov/196108/)
- Furuichi Y, Shatkin AJ, Stavnezer E, Bishop JM. Blocked, methylated 5'-terminal sequence in avian sarcoma virus RNA. *Nature*. 1975; 257(5527):618–20. PMID: [170541](https://pubmed.ncbi.nlm.nih.gov/170541/)
- Krug RM, Morgan MA, Shatkin AJ. Influenza viral mRNA contains internal N6-methyladenosine and 5'-terminal 7-methylguanosine in cap structures. *J Virol*. 1976; 20(1):45–53. PMID: [1086370](https://pubmed.ncbi.nlm.nih.gov/1086370/)
- Lichinchi G, Gao S, Saletore Y, Gonzalez GM, Bansal V, Wang Y, et al. Dynamics of the human and viral m(6)A RNA methylomes during HIV-1 infection of T cells. *Nature microbiology*. 2016; 1(4):16011.
- Kennedy EM, Bogerd HP, Kornepati AV, Kang D, Ghoshal D, Marshall JB, et al. Posttranscriptional m(6)A Editing of HIV-1 mRNAs Enhances Viral Gene Expression. *Cell Host Microbe*. 2016; 19(5):675–85. doi: [10.1016/j.chom.2016.04.002](https://doi.org/10.1016/j.chom.2016.04.002) PMID: [27117054](https://pubmed.ncbi.nlm.nih.gov/27117054/)
- Tirumuru N, Zhao BS, Lu W, Lu Z, He C, Wu L. N(6)-methyladenosine of HIV-1 RNA regulates viral infection and HIV-1 Gag protein expression. *Elife*. 2016; 5.
- Gokhale NS, McIntyre AB, McFadden MJ, Roder AE, Kennedy EM, Gandara JA, et al. N6-Methyladenosine in Flaviviridae Viral RNA Genomes Regulates Infection. *Cell Host Microbe*. 2016; 20(5):654–65. doi: [10.1016/j.chom.2016.09.015](https://doi.org/10.1016/j.chom.2016.09.015) PMID: [27773535](https://pubmed.ncbi.nlm.nih.gov/27773535/)
- Lichinchi G, Zhao BS, Wu Y, Lu Z, Qin Y, He C, et al. Dynamics of Human and Viral RNA Methylation during Zika Virus Infection. *Cell Host Microbe*. 2016; 20(5):666–73. doi: [10.1016/j.chom.2016.10.002](https://doi.org/10.1016/j.chom.2016.10.002) PMID: [27773536](https://pubmed.ncbi.nlm.nih.gov/27773536/)
- Stoltzfus CM, Dimock K. Evidence of methylation of B77 avian sarcoma virus genome RNA subunits. *J Virol*. 1976; 18(2):586–95. PMID: [178899](https://pubmed.ncbi.nlm.nih.gov/178899/)
- Beemon K, Keith J. Localization of N6-methyladenosine in the Rous sarcoma virus genome. *J Mol Biol*. 1977; 113(1):165–79. PMID: [196091](https://pubmed.ncbi.nlm.nih.gov/196091/)
- Thomason AR, Brian DA, Velicer LF, Rottman FM. Methylation of high-molecular-weight subunit RNA of feline leukemia virus. *J Virol*. 1976; 20(1):123–32. PMID: [185408](https://pubmed.ncbi.nlm.nih.gov/185408/)
- Bondurant M, Hashimoto S, Green M. Methylation pattern of genomic RNA from Moloney murine leukemia virus. *J Virol*. 1976; 19(3):998–1005. PMID: [972437](https://pubmed.ncbi.nlm.nih.gov/972437/)
- Canaani D, Kahana C, Lavi S, Groner Y. Identification and mapping of N6-methyladenosine containing sequences in simian virus 40 RNA. *Nucleic Acids Res*. 1979; 6(8):2879–99. PMID: [223130](https://pubmed.ncbi.nlm.nih.gov/223130/)
- Narayan P, Ayers DF, Rottman FM, Maroney PA, Nilsen TW. Unequal distribution of N6-methyladenosine in influenza virus mRNAs. *Mol Cell Biol*. 1987; 7(4):1572–5. PMID: [3600638](https://pubmed.ncbi.nlm.nih.gov/3600638/)
- Dimock K, Stoltzfus CM. Sequence specificity of internal methylation in B77 avian sarcoma virus RNA subunits. *Biochemistry*. 1977; 16(3):471–8. PMID: [189800](https://pubmed.ncbi.nlm.nih.gov/189800/)

20. Wei CM, Gershowitz A, Moss B. 5'-Terminal and internal methylated nucleotide sequences in HeLa cell mRNA. *Biochemistry*. 1976; 15(2):397–401. PMID: [174715](#)
21. Kane SE, Beemon K. Precise localization of m6A in Rous sarcoma virus RNA reveals clustering of methylation sites: implications for RNA processing. *Mol Cell Biol*. 1985; 5(9):2298–306. PMID: [3016525](#)
22. Kane SE, Beemon K. Inhibition of methylation at two internal N6-methyladenosine sites caused by GAC to GAU mutations. *J Biol Chem*. 1987; 262(7):3422–7. PMID: [3029112](#)
23. Meyer KD, Saletore Y, Zumbo P, Elemento O, Mason CE, Jaffrey SR. Comprehensive analysis of mRNA methylation reveals enrichment in 3' UTRs and near stop codons. *Cell*. 2012; 149(7):1635–46. doi: [10.1016/j.cell.2012.05.003](#) PMID: [22608085](#)
24. Dominissini D, Moshitch-Moshkovitz S, Schwartz S, Salmon-Divon M, Ungar L, Osenberg S, et al. Topology of the human and mouse m6A RNA methylomes revealed by m6A-seq. *Nature*. 2012; 485(7397):201–6. doi: [10.1038/nature11112](#) PMID: [22575960](#)
25. Liu J, Yue Y, Han D, Wang X, Fu Y, Zhang L, et al. A METTL3-METTL14 complex mediates mammalian nuclear RNA N6-adenosine methylation. *Nat Chem Biol*. 2014; 10(2):93–5. doi: [10.1038/nchembio.1432](#) PMID: [24316715](#)
26. Dimock K, Stoltzfus CM. Cycloleucine blocks 5'-terminal and internal methylations of avian sarcoma virus genome RNA. *Biochemistry*. 1978; 17(17):3627–32. PMID: [210799](#)
27. Stoltzfus CM, Dane RW. Accumulation of spliced avian retrovirus mRNA is inhibited in S-adenosyl-methionine-depleted chicken embryo fibroblasts. *J Virol*. 1982; 42(3):918–31. PMID: [6285005](#)
28. Finkel D, Groner Y. Methylations of adenosine residues (m6A) in pre-mRNA are important for formation of late simian virus 40 mRNAs. *Virology*. 1983; 131(2):409–25. PMID: [6318439](#)
29. Chen-Kiang S, Nevins JR, Darnell JE Jr. N-6-methyl-adenosine in adenovirus type 2 nuclear RNA is conserved in the formation of messenger RNA. *J Mol Biol*. 1979; 135(3):733–52. PMID: [537090](#)
30. Xiao W, Adhikari S, Dahal U, Chen YS, Hao YJ, Sun BF, et al. Nuclear m(6)A Reader YTHDC1 Regulates mRNA Splicing. *Mol Cell*. 2016; 61(4):507–19. doi: [10.1016/j.molcel.2016.01.012](#) PMID: [26876937](#)
31. Kariko K, Buckstein M, Ni H, Weissman D. Suppression of RNA recognition by Toll-like receptors: the impact of nucleoside modification and the evolutionary origin of RNA. *Immunity*. 2005; 23(2):165–75. doi: [10.1016/j.immuni.2005.06.008](#) PMID: [16111635](#)
32. Durbin AF, Wang C, Marcotrigiano J, Gehrke L. RNAs Containing Modified Nucleotides Fail To Trigger RIG-I Conformational Changes for Innate Immune Signaling. *mBio*. 2016; 7(5).
33. Zhou J, Wan J, Gao X, Zhang X, Jaffrey SR, Qian SB. Dynamic m(6)A mRNA methylation directs translational control of heat shock response. *Nature*. 2015; 526(7574):591–4. doi: [10.1038/nature15377](#) PMID: [26458103](#)
34. Mauer J, Luo X, Blanjoie A, Jiao X, Grozhik AV, Patil DP, et al. Reversible methylation of m6Am in the 5' cap controls mRNA stability. *Nature*. 2016.