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Transcripts of vaccinia virus postreplicative genes do not contain a 5' methylguanosine cap

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Abstract

Vaccinia virus (VACV) is a prototypical poxvirus originally used for eradication of smallpox. Investigation into VACV mRNAs carried out almost half a century ago substantially contributed to the fundamental discovery of the 5' mRNA cap, a hallmark of all eukaryotic and many viral mRNAs. VACV research also facilitated the identification and understanding of the general mechanism of 5' mRNA cap synthesis. We characterized the VACV transcripts at the individual mRNA molecule level and found that vaccinia postreplicative mRNAs, containing nontemplated 5' poly(A) leaders, surprisingly lack the 5' cap structure *in vivo*. We showing that 5' cap occurrence in viral mRNAs gradually decreases in each successive gene time classes, in contrast to the reciprocal increase in 5' poly(A) leader lengths, and that these two variables are mutually negatively correlated. We also demonstrate that the initiator region element (INR) directly or indirectly influences both the frequency of 5' mRNA capping and the occurrence of 5' poly(A) leaders, including their lengths in postreplicative VACV mRNAs. Considering all the results together, we can speculate that the degree of 5' mRNA polyadenylation can directly affect the synthesis of the 5' cap by some hitherto unknown mechanism. This idea is further supported by our observation that 5' poly(A) leaders in m^7G cap-containing VACV late transcripts are significantly shorter than the 5' mRNA leaders, these lengths of which were calculated from the unbiased set of all VACV late mRNAs. Collectively, our results support the hypothesis that VACV transcription regulation ensures a gradual shift in viral mRNA translation initiation from a cap-dependent to cap-independent mechanism, which is accompanied by virus-induced modification of the host translation machinery.

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