ENHANCING RECOMBINANT PROTEIN AND VIRAL VECTOR PRODUCTION IN MAMMALIAN CELLS BY TARGETING THE YTHDF READERS OF N^6-METHYLADENOSINE IN MRNA

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N^6–methyl adenosine (m6A) is the most abundant internal modification on eukaryotic mRNA and has been implicated in a wide range of fundamental cellular processes. This modification is regulated and interpreted by a set of writer, eraser, and reader proteins. To date, there have been no reports on the potential of mRNA epigenetic regulators to influence recombinant protein expression in mammalian cells. In this study we evaluated the potential of manipulating the expression of the m6A YTH domain-containing readers, YTHDF1, 2, and 3 to improve recombinant protein yield based on their role in regulating mRNA stability and promoting translation. Using siRNA-mediated gene depletion, cDNA over-expression and methylation-specific RNA immunoprecipitation, we demonstrate that (i) knock-down of YTHDF2 enhances (~2-fold) the levels of recombinant protein derived from GFP and EPO transgenes in CHO cells; (ii) the effects of YTHDF2 depletion on transgene expression is m6A-mediated and (iii) YTHDF2 depletion or over-expression of YTHDF1 increases viral protein expression and yield of infectious lentiviral particles (~2-3 fold) in HEK293 cells. We conclude that various transgenes can be subjected to regulation by m6A regulators in mammalian cell lines and that these findings demonstrate the utility of epi-transcriptomic-based approaches to host cell line engineering for improved recombinant protein and viral vector production.