

IN BRIEF

Epitranscriptomics and Flowering: mRNA Methylation/Demethylation Regulates Flowering Time ^{OPEN}

DNA modifications such as methylation serve as epigenetic markers that influence gene expression and function. The importance of similar modifications in RNA is gaining appreciation, leading to the emergence of epitranscriptomic analyses. RNA modifications appear to be a general feature of eukaryotes and are best understood in mammals, where reversible mRNA methylation is now known to regulate processes ranging from cell fate transition to stress responses (reviewed in Zhao et al., 2017). Methylation of mRNA—and other types of RNA—is also present in plants. In a recent epitranscriptomics study, David et al. (2017) cataloged over 1000 sites of cytosine methylation (m^5C) in the *Arabidopsis thaliana* transcriptome. In mammals, N^6 -methyladenosine (m^6A) is the most common modification of mRNA (see figure). The m^6A mark is reversible: It can be placed (or “written”) by a methylation complex made of several subunits and “erased” by demethylases. So-called “readers” interpret the marks and transduce them into downstream effects via alterations in RNA splicing, maturation, translation, and degradation.

The first plant homolog of an m^6A eraser was recently reported: *Arabidopsis* ALKBH9B,

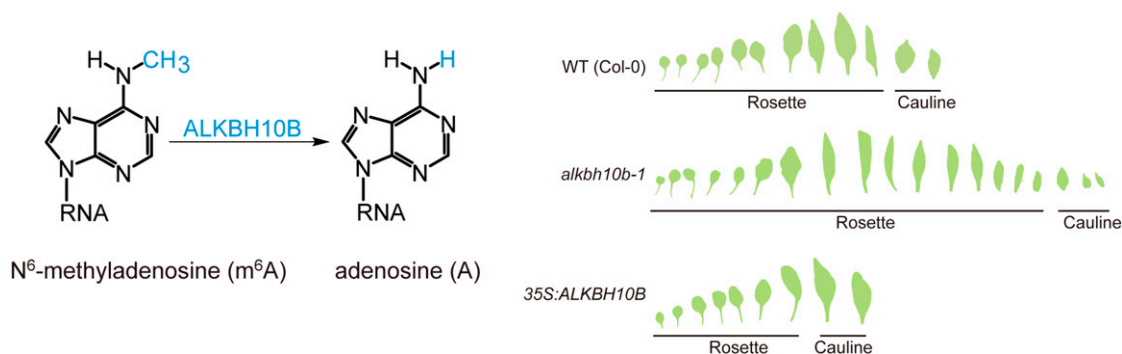
a homolog of the human m^6A demethylase ALKBH5, has m^6A demethylase activity and is involved in defense against viral infection (Martínez-Pérez et al., 2017). Now, **Duan et al. (2017)** report that another m^6A demethylase functions in the regulation of flowering time.

Among five orthologs of human ALKBH5 in *Arabidopsis*, Duan and coworkers found that *ALKBH10B* had the most abundant transcripts in flowers and was expressed in most tissues tested. When ALKBH10B was expressed in *Nicotiana benthamiana*, the purified protein showed m^6A demethylation activity that was dependent on the presence two conserved iron(II) binding residues. Similar to human ALKBH5, ALKBH10B had greater activity with a nonstructured RNA (or single-stranded RNA) substrate than with a stem-loop structured substrate in vitro.

The *alkbh10b-1* mutant had higher levels of m^6A -modified mRNA in both reproductive and vegetative organs, and overexpression of ALKBH10B in wild-type plants decreased m^6A levels, establishing that ALKBH10B indeed functions as an RNA demethylase in vivo. The authors further showed that this activity was specific for m^6A in mRNA.

Notably, the *alkbh10b-1* mutant exhibited late flowering, while the overexpression line flowered early (see figure). Duan et al. found that ALKBH10B activity enhanced the transcript accumulation of the flowering promoter *FT* and that *FT* mRNA had a higher level of m^6A in the mutant than in the wild type. The *FT* regulators *SPL3* and *SPL9* also had lower transcript levels and higher m^6A levels in the *alkbh10b-1* mutant. Furthermore, Duan et al. showed that ALKBH10B directly interacted with *FT*, *SPL3*, and *SPL9* transcripts. Demethylation decreased the degradation of these transcripts, again similar to its effects on human mRNA.

Finally, Duan et al. examined global ALKBH10B-mediated demethylation, identifying 1276 sites in 1190 genes with greater m^6A levels in the mutant compared with the wild type. Their work thus provides a solid foundation for understanding the roles of m^6A -type methylation and demethylation in plants, which appears to share fundamental characteristics with that in mammals. In addition, it is clear that models for the regulation of flowering time must be updated to include posttranscriptional control of multiple transcripts via m^6A marks.



ALKBH10B demethylates m^6A and regulates flowering time. N^6 -methyladenosine and adenosine residues in RNA (left). Number of leaves produced before flowering in the wild type compared with the *alkbh10b-1* mutant and ALKBH10B overexpression line (right). (Adapted from Duan et al. [2017], Figures 2A and 4C.)

Nancy R. Hofmann
Science Editor
nhofmann@aspb.org
ORCID ID: 0000-0001-9504-1152

REFERENCES

- David, R., Burgess, A., Parker, B., Li, J., Pulsford, K., Sibbritt, T., Preiss, T., and Searle, I.R. (2017). Transcriptome-wide mapping of RNA 5-methylcytosine in Arabidopsis mRNAs and noncoding RNAs. *Plant Cell* **29**: 445–460.
- Duan, H.-C., Wei, L.-H., Zhang, C., Wang, Y., Chen, L., Lu, Z., Chen, P.R., He, C., and Jia, G. (2017). ALKBH10B is an RNA *N*⁶-methyladenosine demethylase affecting Arabidopsis floral transition. *Plant Cell* **29**: 2995–3011.
- Martínez-Pérez, M., Aparicio, F., López-Gresa, M.P., Bellés, J.M., Sánchez-Navarro, J.A., and Pallás, V. (2017). *Arabidopsis* m⁶A demethylase activity modulates viral infection of a plant virus and the m⁶A abundance in its genomic RNAs. *Proc. Natl. Acad. Sci. USA* **114**: 10755–10760.
- Zhao, B.S., Roundtree, I.A., and He, C. (2017). Post-transcriptional gene regulation by mRNA modifications. *Nat. Rev. Mol. Cell Biol.* **18**: 31–42.