IN BRIEF

Mapping the Barley Chloroplast Transcriptome

Chloroplast genomes encode only 60 to 200 of the several thousand proteins required for photosynthesis and plastid function, the majority having been transferred to the host cell nucleus (or lost) during the course of evolution (Kleine et al., 2009). Genes retained in the chloroplast genome represent some of those required for photosynthesis and gene expression. Many details of the regulation of chloroplast gene expression are unknown, including, for example, regulation of the responses to light quality and quantity, stress, and developmental cues (Barkan, 2011).

Zhelyazkova et al. (pages 123–136) present a genome-wide map of transcription start sites (TSSs) in the chloroplast genome of barley (*Hordeum vulgare*), providing a significant step in our understanding of chloroplast transcription and a valuable resource for further inquiry. The authors used differential RNA sequencing, which distinguishes between primary and processed transcripts, to map transcription start sites and identify noncoding RNAs (ncRNAs) in green and white leaves of the mutant barley line *albostrians* (see figure). There are two major RNA polymerases in the chloroplast of angiosperms, a plastid-encoded (PEP) and a nuclear-encoded (NEP) plastid RNA polymerase. Plastids in white *albostrians* leaves are ribosome deficient and lack plastid-encoded proteins, including the core subunits of PEP. Thus, a comparison of results from white and green sectors allowed the authors to map both PEP- and NEP-active start sites, providing insight into the contribution of each polymerase to chloroplast transcription.

Previously, it was thought that many photosynthesis genes were exclusively transcribed by PEP. As expected, PEP was found to be the major RNA polymerase in the chloroplast, accounting for 88% of TSS in green leaves. However, plastids from white leaves (lacking PEP) yielded bona fide TSS and mRNA for 70 plastid genes, many of which encode photosynthetic proteins. The results showed that a large number of NEP promoters become activated in the absence of PEP, as white plastids yielded a larger number of TSS than green plastids, of which the majority were not found in green plastids. The authors suggest that activation of transcription by NEP in the absence of PEP might serve as a general rescue mechanism in higher plants. Moreover, NEP promoters active in white leaves might also show a weak activity in green leaves or play a relatively more important role in developing leaves and nongreen tissues.

TSSs were categorized based on their genomic location. In both green and white plastids, ~50% of TSS were found just upstream of an annotated gene, 10 to 20% lay within an annotated gene and gave rise to a sense transcript, 20 to 30% were located on the opposite strand of an annotated gene and gave rise to an antisense transcript, and ~10% fell outside of these categories. Intriguingly, these results provide evidence for extensive ncRNA synthesis in barley plastids. Numerous ncRNA transcripts were detected from intergenic regions as well as antisense transcripts to ~35% of genes in green plastids. This represents a rich resource for investigating potential regulatory ncRNA activity in plastids.

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REFERENCES


Operon and TSS map of the barley genome. The outer circle shows gene organization of the barley chloroplast genome. Arrows show the direction of transcription. The inner circle shows genomic position of all mapped TSSs. (Reprinted from Supplemental Figure 5 of Zhelyazkova et al. [2012].)

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