Regulation of flavonoid biosynthesis involves an unexpected complex transcriptional regulation of TT8 expression, in Arabidopsis

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TT8/bHLH042 is a key regulator of anthocyanins and proanthocyanidins (PAs) biosynthesis in Arabidopsis thaliana. TT8 transcriptional activity has been studied extensively, and relies on its ability to form, with several R2R3-MYB and TTG1 (WD-Repeat protein), different MYB-bHLH-WDR (MBW) protein complexes. By contrast, little is known on how TT8 expression is itself regulated. Transcriptional regulation of TT8 expression was studied using molecular, genetic and biochemical approaches. Functional dissection of the TT8 promoter revealed its modular structure. Two modules were found to specifically drive TT8 promoter activity in PA- and anthocyanin-accumulating cells, by differentially integrating the signals issued from different regulators, in a spatio-temporal manner. Interestingly, this regulation involves at least six different MBW complexes, and an unpredicted positive feedback regulatory loop between TT8 and TTG2. Moreover, the results suggest that some putative new regulators remain to be discovered. Finally, specific cis-regulatory elements through which TT8 expression is regulated were identified and characterized. Together, these results provide a molecular model consistent with the specific and highly regulated expression of TT8. They shed new light into the transcriptional regulation of flavonoid biosynthesis and provide new clues and tools for further investigation in Arabidopsis and other plant species.
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