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Structure function of MADS complexes in Arabidopsis: In vivo evidence for SEPALLATA-driven MADS tetramerization requirement for flower development.

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MADS transcription factors are present in all eukaryotes, but in flowering plants have undergone a significant expansion over the course of evolution. MADS TFs fulfill many roles in the plant, most notably acting as key regulators of flowering and floral organ development. MADS TFs bind DNA as obligate dimers in animals and fungi. However, in plants, MADS TFs have acquired a tetramerization domain, which allows, in addition to the formation of dimeric complexes, the formation of tetrameric complexes that can bind to two cognate sites simultaneously. In angiosperms, the SEPALLATA (SEP) subfamily of MADS transcription factors act as organizers of MADS complexes, forming both heterodimers and heterotetramers *in vitro*. Whether DNA-binding by SEPALLATA-containing dimeric MADS complexes are sufficient for launching floral organ identity programs or whether tetramerization is mandatory was not yet established *in vivo*. In order to address the fundamental question of the physiological role and requirement of tetramerization in flower development, we performed structural, genome wide, biochemical, and *in vivo* experiments to correlate oligomerization state with DNA-binding and physiological function. Using structure-based design, we generated two MADS (SEP3 and AGAMOUS) mutants with strongly abrogated tetramerization capability, but that retain the capacity to dimerize and bind DNA. In this way, we could decouple tetramerization from DNA binding and clearly show, that tetramerization is essential for the activity of MADS TF complexes *in vivo* in petal, stamen and carpel development in *Arabidopsis thaliana*.