

673 Comparative Genomics of Angiosperm MADS Box Genes

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MADS box genes encode key transcriptional regulators that have been implicated in the control of various aspects of floral development, primarily through work carried out in *Arabidopsis*. We have embarked on a series of analyses to examine the extent to which gene duplication, regulatory diversification and differences in protein interactions have been important in modifying the roles of MADS box genes during the evolution of flowering plant species. As a first step, we have been developing strategies to rapidly identify all MADS box genes from a given species in order to carry out comprehensive phylogenetic and functional analyses. Using these data, we have initiated analyses of the roles of tomato MADS box genes in flower and fruit development. In addition, we have been developing methods to carry out functional analyses in non-model basal eudicot species. By combining in depth sampling of the repertoire of MADS box genes in phylogenetically informative species with functional analyses in a variety of systems, we can trace the evolutionary history of these genes and define their ancestral and derived roles. In addition, we can assess the extent to which developmental modules controlled by MADS box genes have been redeployed to give rise to new morphologies. Together, these approaches are paving the way for a greater understanding of how these genes have diversified to specify different aspects of floral architecture.

674 The evolution of plant architecture in Brassicaceae

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Variation in the disposition of flowers in time and space can greatly affect plant fitness in different environments. Previous work in genetic model systems has suggested that the interplay of shoot and floral meristem identity genes might play a key role in regulating plant architecture and the placement of flowers within shoot systems. In particular, homologs of the shoot meristem identity gene *TERMINAL FLOWER1* (*TFL1*) and the floral meristem identity gene *LEAFY* (*LFY*) have been hypothesized to play a central role in architectural evolution. Brassicaceae have a rather conservative architecture: almost all species produce flowers on inflorescences (elongated portions of stems with reduced or absent leaves). However, at least three lineages have switched to rosette-flowering, a condition in which solitary flowers on long pedicels emerge from the axils of rosette leaves. We used an interspecific transgenic strategy to study the role of meristem identity genes in the switch to rosette-flowering in three lineages that independently acquired rosette-flowering: *Ionopsidium*, *Leavenworthia*, *Idahoia*. The *LFY* homologs from one rosette-flowering species of each genus were introduced with their native promoters into *lfy* mutant *Arabidopsis* plants. The resulting phenotypes suggest that significant changes have occurred at the *LFY* locus during the evolution of *Leavenworthia* and *Idahoia* and implicate changes in the interaction with *TFL1*. Specifically, it appears that the *LFY* transgenes are not properly repressed by *TFL1* due to changes in their *cis*-regulatory regions. These data suggest that changes in the identity of axillary meristems associated with the evolution of rosette flowering were driven by alterations in the regulatory network linking *LFY* and *TFL1*. However, the full evolutionary transition almost certainly entailed changes at more than these two genetic loci.

675 The role of KNOX genes in shoot morphogenesis and compound leaf development

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The evolution of the KNOTTED-like HOMEODOMAIN (KNO1) genes suggests a history of gene loss, duplication and sub-functionalization. Of particular interest to us is the divergence into the Class 1 and Class 2 genes. The KNOX1 genes function in shoot morphogenesis. Analysis of the atypical leaf of *Kalanchoe daigremontiana* shows that KNOX1 genes (together with LEC1) play a role in ectopic shoot production. KNOX1 genes have also been shown by us to be utilized in the developmental cascade leading to compound leaves in most flowering plants. We extended this study to analyze the role of PHANTASTICA, a MYB domain protein that regulates KNOX1 genes. In tomato leaf development PHAN and KNOX1 share a dosage sensitive regulatory relationship and PHAN regulates leaflet placement in numerous compound leaves. We will present data on analysis of leaf development in the tomato and bean families, and in related species in the same genus (e.g. the genus *Lepidium*) or within the same species (e.g. the heteroblastic species *Neobeckia aquatica*) in the mustard family. These data have begun to give us tools to discriminate between the two conflicting hypotheses (the partial shoot and the leaf subdivision hypotheses) on compound leaf origins.

676 Molecular Evolution of LEAFY transcription factor in land plants

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Understanding how molecular mechanisms can be linked to evolution of organism form is the founding theme of “evo-devo” studies. Transcription factors, which regulate and coordinate the expression of thousands of genes during development, represent candidate of choice to address the question of how molecular evolution of a developmental regulator can result in changes in the repertoire of gene it controls and ultimately translate into morphological evolution.

The plant-specific transcription factor LEAFY controls general aspects of the life cycle in a basal plant, the moss *Physcomitrella patens*. In contrast, LEAFY has more specialized functions in angiosperms, where it specifically induces floral fate during the reproductive phase. This raises the question of a concomitant change in the biochemical function of LEAFY during the evolution of land plants. We have identified that the DNA binding domain of LEAFY, although largely conserved, has diverged in activity. On the contrary, other, more rapidly evolving portions of the protein have few effects on LEAFY activity¹. In a broader perspective of functional evolution of LFY among land plants, two testable hypotheses can be proposed. LFY might control similar networks of genes in non-flowering and flowering plants, with co-evolution of target sequences and LFY DNA binding specificity. Alternatively, there may have been a complete change of LFY function between basal taxa and flowering plants, in which an initial, albeit gradual change in biochemical activity was the prerequisite for recruitment and/or intercalation of new targets, such as AP1.

Our results establish a framework for the study of the functional/morphological evolution of plant form and flower invention and tentative unifying scenario will be discussed.

1. Maizel, A. et al. *Science* 308, 260-3 (2005)

677 Evidence of genetic conservation of diverse nectaries within the eudicots

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Nectaries are secretory organs widely present in flowering plants that function to lure floral pollinators. Due to diversity in nectary positions and structures, they are thought to have originated multiple times during angiosperm evolution, with their potential contribution to the diversification of flowering plants and pollinating animals being considerable. Using CRABS CLAW (CRC), a gene required for nectaries in Arabidopsis, the genetic bases of diverse nectary forms in eudicot angiosperm species were investigated. CRC expression is conserved in morphologically different nectaries from several core eudicot species and CRC is required for nectary development in both rosids and asterids, major phylogenetic branches of eudicots. However, in a basal eudicot species, no evidence of CRC expression in nectaries was found. Combining a mapping of nectary positions onto an angiosperm phylogeny and CRC expression analyses in eudicots, we propose that diverse nectaries in core eudicots share conserved CRC gene regulation, and that derived nectary positions in eudicots have altered regulation of CRC. Since the ancestral function of CRC lies in the regulation of carpel development, it was likely co-opted as a regulator of nectary development in basal eudicots, which may have led to the association of nectaries with reproductive organs in derived lineages.

678 Abstract not submitted

679 The roles of genetic integration and constraint in adaptive evolution: a floral case study

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The evolution of phenotypic traits depends on the form and strength of natural selection as well as the genetic variances and correlations among the traits. Genetic correlations may constrain phenotypic evolution, or they may themselves evolve to increase functional integration among traits. Using floral traits as a model system, I will describe a combination of lab, greenhouse, and field studies designed to increase our understanding of the roles of integration and constraint in evolution. A comparative phylogenetic study suggests that a high correlation between the lengths of the stamens and corolla tube has evolved from a lower correlation at least twice in the Brassicaceae, and that this correlation is phylogenetically stable in the clade that includes radish and *Brassica*. This high correlation could be due to selection to maintain proper anther placement for effective pollination. Studies of this high correlation ($r = 0.85$) in wild radish indicate that it is stable across populations and environments and that it is caused by pleiotropy rather than linkage disequilibrium. Thus, this correlation is an extraordinarily strong candidate to cause a constraint. However, when we applied artificial selection to anther position, rapid independent evolution of the stamens and corolla tube occurred. This work should improve our understanding of how rapid adaptive evolution can occur, even in the face of possible constraints caused by genetic correlations.

680 The Nature of Intrinsic Postzygotic Isolation

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Crosses are often made within and between species to infer evolutionary relationships and to determine the nature of intrinsic postzygotic isolation. Here I analyze crossing data from more than 300 genera of plants and animals and present new empirical results from sunflower on the basis of reduced hybrid fertility. I show that contrary to conventional wisdom, plant species are more strongly isolated than animal species and more likely to correspond to reproductively independent lineages. Also, comparisons of F_1 and F_2 hybrid fitnesses indicate that postzygotic isolation in animals and perennial plants is caused mainly by gene incompatibilities, whereas chromosomal rearrangements are the predominant cause of isolation in annuals. This observation is supported by genetic studies of annual sunflowers, where reductions in pollen viability map mostly to chromosomal rearrangements. However, in some cases, pollen viability QTLs behave epistatically, indicating that gene incompatibilities cluster near rearrangement breakpoints as predicted by theory. Finally, genetic-mapped based analyses of hybrid zones demonstrate that rearrangements have a large effect on interspecific gene flow near the center of hybrid zones, but that this effect does not extend to more distant populations as has been reported for some animal species.

