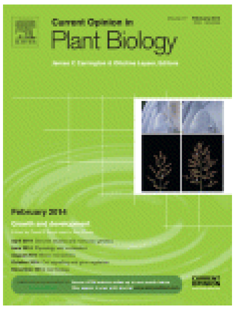

Developmental specialisations in the legume family

Julie MI Hofer[1], TH Noel Ellis[1]
Institute of Biological, Environmental & Rural Sciences, Aberystwyth University, Gogerddan, Aberystwyth, Ceredigion SY23 3EE, UK

Volume 17, February 2014, Pages 153–158

Current Opinion in Plant Biology

DOI: http://dx.doi.org/10.1016/j.pbi.2013.11.014

Developmental specializations in the legume family

Julie M. I. Hofer[1]* and T. H. Noel Ellis[1]

Institute of Biological, Environmental & Rural Sciences, Aberystwyth University, Gogerddan,
Aberystwyth, Ceredigion SY23 3EE, UK.
jmh18@aber.ac.uk; noe2@aber.ac.uk

*corresponding author

[1] Present address: International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), Patancheru 502 324, Andhra Pradesh, India (j.hofer@cgiar.org; n.ellis@cgiar.org)

Abstract

The legume family is astonishingly diverse; inventiveness in the form of novel organs, modified organs and additional meristems, is rife. Evolutionary changes can be inferred from the phylogenetic pattern of this diversity, but a full understanding of the origin of these 'hopeful monsters' of meristematic potential requires clear phylogenetic reconstructions and extensive, species-rich, sequence data. The task is large, but rapid progress is being made in both these areas. Here we review specializations that have been characterized in a subset of intensively studied papilionoid legume taxa at the vanguard of developmental genetic studies.

Introduction

The legume family (Fabaceae, also known as Leguminosae) evolved about 60 million years ago and diversified into three subfamilies; the caesalpinioids, mimosoids, and papilionoids [1]. They make up the third largest family of flowering plants, comprising more than 700 genera and 18,000 species, with an exceptionally wide range of habits, ranging from giant forest trees to aquatic herbs [2-4]. Protein and/or oil-rich papilionoid legumes, such as soybean, peanut and chickpea account for more than one quarter of the world's primary crop production [5••] and are an essential component of low meat consumption diets. Representative genome sequences of some of these important food species have been published recently (www.comparative-legumes.org/) and many more are in the pipeline thanks to cheaper, faster sequencing technologies. This will lead to clusters of intensively sequenced species that are taxonomically closely related, so that, within these clear phylogenetic contexts, comparative genomic studies can be used to investigate genes associated with synapomorphies, characters that are shared due to common ancestry. In this review, the characters we focus on are developmental specialisations (Figure 1). Identification of the sequence variation associated with losses and secondary gains of synapomorphic specialisations provides a deep understanding of the evolution of developmental novelties.

Specialization of inflorescence architecture

The most common type of inflorescence in legumes is a type of shoot axis known as a raceme, which includes flowering spikes, umbels and heads [6, 7]. Even in mimosoid legumes, with their bottle brush or pompom-like floral clusters [8], flowers initiate acropetally on the raceme axis, after which they undergo a remarkable developmental pause until all are initiated, then, together they develop floral organs and open synchronously [6]. There is general agreement that papilionoid legume inflorescences are usually racemes, however there is disagreement on whether panicles are also common, due to differences in the definition of a panicle [6, 9]. Classically defined panicles [10] are not found in legumes, while compound racemes, for example double racemes with two orders of branching, are common [7, 9].

Taking the view that complex inflorescences are a developmental specialization, we can ask if this complexity is regulated by iteration of the same molecular mechanism at sequential developmental time points, or by different mechanisms and whether these mechanisms are shared in other plant species. Berbel et al (2012) [11••] set out to answer these questions in their study of the double raceme in *Pisum sativum*. The *Veg1* gene, encoding a MADS-box transcription factor in the euAGL79-clade, was identified as a novel genetic function that inserts a new meristem (called the I2, or secondary inflorescence, Figure 1) between the primary inflorescence axis and the flowers [11••]. This I2, a short stem terminating in a rudimentary stub, represents an additional meristem upon which novelty can arise [12•]. In *P. sativum* the identity of the I1 primary inflorescence (Figure 1) is determined by a different gene, *Determinate* [13]; its expression is restricted to the I1 by *Veg1* [11••]. Mutations of *Determinate* result in a determinate growth habit much sought after by breeders [14•]. A *Veg1*-like gene is not found in the grasses, where complex inflorescences are generated using a different set of genes [15]. Other papilionoid legumes with double racemes have a *Veg1* gene [11••], but it remains to be shown whether legumes with simple racemes have lost *Veg1* function and whether further subfunctionalization of the *euAGL79* clade occurs in legume species with higher levels of inflorescence complexity.

Novelty in flowers

Most legume flowers are pentamerous with five sepals, five petals, two whorls of five stamens each and a single carpel, but species occur with variations on this plan, featuring floral organ reduction or multiplication [2, 6]. Many members of the papilionoid taxon Swartzieae, for example, are characterised by a single petal [2], while the profuse production of stamens from an innovative developmental feature, the ring meristem, is manifest in all three legume subfamilies [6]. The typical papilionoid flowers of *P. sativum*, *Medicago truncatula* and *Lotus japonicus* have ephemeral meristems known as common primordia (Figure 1) from which both petals and stamens arise [16-

18]. Although the common primordia are entirely consumed by the organs arising from them, they represent an additional step in floral meristem ontogeny, echoing the structural reiteration of compound inflorescences; both remodellings of meristematic potential are ‘hopeful monsters’. In *M. truncatula*, duplicated *APETALA3*-lineage genes are expressed in complementary patterns in the common primordia, petals and stamens and they interact differentially with duplicate *PISTILLATA*-lineage genes to confer petal and stamen identity [19]. Despite the absence of a motif common to other eudicots, *PISTILLATA* proteins in *P. sativum* and *M. truncatula* are fully functional in this role [20, 21].

Mimosoid legumes are characterised by radially symmetrical flowers, whereas papilionoid and caesalpinioid legume flowers are usually zygomorphic (Figure 1), that is they are bilaterally symmetrical; their showy adaxial petal often stands upright and two lateral petals flank two fused keel petals when the flower opens. Like the distantly-related zygomorphic species *Antirrhinum majus*, papilionoid legumes have recruited CYCLOIDEA transcription factor family genes for the regulation of dorsal and lateral petal identity [22-25]. Functional analysis of triplicated *CYCLOIDEA* -like genes in *L. japonicus* showed that *LjCYC1* and *LjCYC2* have roles in dorsal petal patterning, while *LjCYC3* confers lateral petal identity [24, 26•]. Independent genetic loci determine the asymmetry of the lateral and ventral petals [24, 27], but these remain to be identified. Transcriptional profiling of individual floral organs was undertaken in *Vicia sativa* [28] and compared with gene expression in distantly-related non-zygomorphic flowers. In order to capture the particular features of this synapomorphy in papilionoid legumes, a similar approach will need to be taken on species within the same clade, e.g. zygomorphic and non-zygomorphic genistoid legumes [25]. While much attention has focused on floral morphology with respect to pollinators, other factors, for example nectar composition, are also important in pollination systems [29]. Specialisation occurs in other floral organs too, such as the carpel. Legume carpels can be covered in hairs, or hairy on one side only. The former has been deduced as the ancestral state in papilionoid legumes of the tribe Fabeae, while the latter, as either abaxial or adaxial hairiness, characterises different subclades [5••].

Once fertilised, the carpel normally grows out to form the elongated seed-containing legume (or pod) that is characteristic of the entire family. A striking novelty is that some species have coiled pods (Figure 1) and this character has arisen several times independently within papilionoid legumes [2], suggesting it may have adaptive value. A study of species with coiled or uncoiled pods within the genus *Medicago* revealed a clear correlation between pod morphology and a change in the *SHATTERPROOF* gene coding sequence which affected the ability of this MADS-box transcription factor to interact with a partner [30••]. Pod coiling in *Medicago* occurs when cells at the carpel margin fail to elongate and become heavily lignified. As there is only one carpel and only one margin, inhibition of its extension results in coiling of the carpel wall tissues, which extend as usual. Coiled pods can have elaborate patterns of lignin deposition, sometimes resulting in the formation of spines, which allows seed dispersal when the pod becomes attached to the fur of passing animals. The *Medicago* clade, which contains spined and smooth-podded species, is ideally suited to further investigation of this character.

Complex and specialised leaves

Simple leaves have been inferred to be ancestral in Angiosperms and there have been multiple gains and losses of leaf complexity. This means not all compound leaves are homologous, and neither are simple leaves, but terms have not yet been defined to reflect this. It is not clear whether the ancestral leaf form of eudicots, or within these the rosids, from which legumes arose, was simple or more complex [31•]. Maximum likelihood estimates of ancestral states suggest that among Angiosperms there is a strong bias for evolution towards simple leaves, however, within the legumes, a family characterised by compound leaves, there is no such bias, indeed evidence for the opposite trend is found [31•]. Thus the legume compound leaf (Figure 1), once acquired, tended to be retained, and provided further opportunities for developmental specialisation.

The LEAFY protein, known as Unifoliata (Uni) in *P. sativum* [32], SINGLE LEAFLET1 (SGL1) in *M. truncatula* [33, 34] and PROLIFERATING FLORAL MERISTEM (PFM) in *L. japonicus* [35], is known to target many genes involved in floral patterning [36, 37], but it is also proposed to have an earlier role in enabling meristem outgrowth, by inducing the transcription of genes such as *REGULATOR OF AXILLARY MERSTEMS1* [38•]. It may be through this pathway that *Uni*/*SGL1*/*PFM* generates a transient phase of indeterminacy [32] in legume compound leaves. The downstream homeodomainless KNOX transcriptional regulator FUSED COMPOUND LEAF1 has a similar role in promoting leaf complexity, as well as defining leaflet boundaries [39], while *PALMATE-LIKE PENTAFOLIATA1* (*PALM1*) repression of *Uni/SGL1/PFM* is important in ensuring that complexity is eventually curtailed [33, 34]. The *PALM1* gene also regulates leaflet wax deposition and this brings a potentially serendipitous benefit to breeders: reduced wax deposition on the abaxial surface of *palm1* mutant leaves confers resistance to fungal pathogens [40••].

In the mimosoid subfamily bipinnate-leaves (Figure 2A) are common while once-pinnate-leaves (Figure 2B) are more usual in the papilionoid subfamily [2]; whether this additional complexity in mimosoids can be explained by variations in *Uni/SGL1/PFM* and *PALM1* interactions remains an open question. The compound leaves of both these taxa feature astounding leaf specialisations, such as the touch-responsiveness of leaves and of tendrils (Figure 2C). One of the genes required for tendril formation in *Pisum* and *Lathyrus* species, is *Tendril-less* (*Tl*), which is transcriptionally activated by *Uni* [41] and *Lathyroides* (*Lath*) [42•], though direct interactions have not yet been demonstrated. Sequence similarity and genetic map position suggest *Lath* is the ortholog of the *WUSCHEL-related homeobox1* (*WOX1*) transcription factor *STENOFOLIA* (*STF*)*,* which was shown to be critical for leaflet blade development in *M. truncatula* [43]. Genes such as *JAGGED1*, recently identified in soybean [44•], are also involved in this process. In the tendrilled leaf of *P.sativum*, *Lath* appears to have acquired an additional role; not only promoting blade outgrowth in leaflets and the dorsal petal (as in *M. truncatula*) but also suppressing blade outgrowth in tendrils [42•], presumably via activation of *Tl* in the distal domain of the leaf.

Application of the hormone gibberellic acid suppresses terminal leaflet blade development and this activity is proposed to occur through increased *Uni* transcription [45]: again, upregulation of *Tl* would lead to blade suppression. Other hormones are also implicated in blade position and outgrowth, for example, reduced levels of free auxin and changes in the levels of transcription of auxin-regulated genes are seen in narrow-leafleted *stf* mutants [43] while *smooth leaf margin1* mutants have altered auxin distribution and excessive production of terminal leaflets [46]. At this point at least some of the regulators of legume leaflet versus tendril formation have been identified, but their relationships with each other are yet to be fully elucidated. Whether the leaf tendrils of climbing mimosoid legume species represent convergent evolution and whether the same physiological processes are involved in contact-induced coiling remain to be investigated. The terminal leaf tendrils of *Entada scandens* are said to be sensitive over their entire surface [47], as are *Lathyrus aphaca* tendrils, while *P. sativum* tendrils are sensitive on their abaxial surface only [48].

A novel organ, the pulvinus (Figure 2D), mediates leaf movements in some legumes and is driven by changes in turgor pressure [49]. As such responses can be rapid and reversible, whereas leaf repositioning in plants without pulvini occurs more slowly via differential growth of cells in the leaf petiole [50]. Both reversible and non-reversible mechanisms of movement may be important for the tolerance of light, temperature and water stresses. Sensitive leaves, described as seismonastic, due to the suddenness of their response to touch, have a pulvinus enabling rapid flexion from the base of the leaf or leaflet. This intriguingly radialized organ represents an intercalation of non-polarity between the flattened leaflet and the polarised leaf axis. A preliminary study has found evidence of seismonasty evolving multiple times in distinct lineages within the genus *Mimosa* [8]. One hypothesis is that the presence of a pulvinus, acting as a hinge in regular movements, such as solar tracking and day/night leaflet opening and closure, permits exaptation to seismonasty. Analysis of mutants without a pulvinus in *P. sativum*, *M. truncatula* and *L. japonicus* led to the identification of a LOB-domain transcription factor as a key component in the development of a leaflet pulvinus [51•, 52•]. These legumes are all members of the 50 Ma Hologalegina clade of papilionoid legumes [1], so this discovery naturally leads to the question of whether pulvinus formation in phylogenetically remote and distinct clades, such as the 42 Ma Mimosoid clade and the 45 Ma Millettioid clade, with pulvini at the base of their leaves as well as their leaflets, is regulated by the same process.

A novel root organ: the nodule

No discussion of developmental specialisations in legumes can ignore the novelty of nodules (Figure 1), organs that accommodate symbiotic nitrogen-fixing bacteria on underground or adventitious stem roots [3, 53, 54]. Nodules are not unique to legumes as indicated by their multiple independent origins in Angiosperm lineages [55], however, recent attention has focused on their evolution in legumes thanks to a new class of mutant defective in nodule identity [56••]. Maintenance of nodule identity requires suppression of root identity by the *NODULE ROOT*/*COCHLEATA* (*NOOT*/*COCH*)gene, which also has roles in leaf and floral development [56••]. In the pea *coch* mutant the most extreme leaf phenotype is the replacement of the stipules with compound structures resembling those on the rachis; reminiscent of the stipule form in *Delonix regia* [57]. The enlarged meristematic regions and supernumerary organs observed on *noot*/*coch* nodules, leaves and flowers suggests that long and/or short-distance stem cell pool maintenance signals are not correctly interpreted in the mutant. These broad phenotypic effects probably reflect original functions of the ancestral gene in shoot development prior to its co-option in nodule maintenance [56••]. As with many other specialised organs, in the nodule we see a clear example of evolutionary tinkering through gene recruitment.

Conclusions

It happens to be the case that most agricultural legume species are in the papilionoid subfamily, so this group is where genome sequence and genetic information is richest at present. In contrast, fascinating and potentially useful developmental specialisations are distributed throughout the legume family; the examples discussed here are but a sparse sample of the evolutionary inventiveness of this angiosperm family. Among the specializations we have described a common feature is the intercalation of a new meristem, involving gene subfunctionalization, which adds structural complexity. Organ sculpture into specialized shapes is another commonality, whether of leaflets, petals, or pods, members of large gene families have been co-opted to this function. New genes are driving phenotypic evolution, so it is with more extensive sequence information throughout the legume family that we can identify the newcomers and understand how they have been incorporated into existing developmental circuitries to bring about novelty.

References

[1] Lavin M, Herendeen PS, Wojciechowski MF. **Evolutionary rates analysis of Leguminosae implicates a rapid diversification of lineages during the tertiary**. *Systematic Biology* 2005, **54**:575-594.

[2] Lewis GP, Schrire B, Mackinder B, Lock M (ed). In: **Legumes of the World**. Richmond: Kew Publishing; 2005. pp. 1-577.

[3] Capoen W, Oldroyd G, Goormachtig S, Holsters M. ***Sesbania rostrata*: a case study of natural variation in legume nodulation**. *New Phytologist* 2010, **186**:340-345.

[4] Tabosa AB, Matias LQ, Martins FR. **Live fast and die young: The aquatic macrophyte dynamics in a temporary pool in the Brazilian semiarid region**. *Aquatic Botany* 2012, 102:71-78.

[5]●● Schaefer H, Hechenleitner P, Santos-Guerra A et al. **Systematics, biogeography, and character evolution of the legume tribe Fabeae with special focus on the middle-Atlantic island lineages**. *BMC Evolutionary Biology* 2012, **12**:e250.
This exemplary paper uses plastid and nuclear DNA sequences to reconstruct a phylogeny of the Fabeae. The authors use this to study character evolution and to trace the likely origins of different taxa via dispersals from the Eastern Mediterranean. The authors clarify affinities of important crop species, finding that the *Lathyrus* clade includes *Pisum* and a *Vicia* clade includes *Lens*.

[6] Tucker SC. **Floral development in legumes**. *Plant Physiology* 2003**, 131**:911-926.

[7] Benlloch R, Berbel A, Serrano-Mislata A, Madueno F. **Floral initiation and inflorescence architecture: a comparative view**. *Ann Bot* 2007, **100**:659-676.

[8] Simon MF, Grether R, de Queiroz LP et al. **The Evolutionary History of *Mimosa* (Leguminosae): Toward a Phylogeny of the Sensitive Plants**. *American Journal of Botany* 2011, **98**:1201-1221.

[9] Prenner G. **Papilionoid inflorescences revisited (Leguminosae-Papilionoideae)**. *Ann Bot* 2012, 10.1093/aob/mcs258.

[10] Endress PK. **Disentangling confusions in inflorescence morphology: Patterns and diversity of reproductive shoot ramification in angiosperms**. *Journal of Systematics and Evolution* 2010, **48**:225-239.

[11]●● Berbel A, Ferrandiz C, Hecht V et al. ***VEGETATIVE1* is essential for development of the compound inflorescence in pea**. *Nature Communications* 2012, **3**: e797.
This lucid paper identifies the pea *Veg1* gene using *M. truncatula* and *P. sativum* comparative mapping and uses mutants and expression studies to explain the genetic basis of inflorescence development in *P. sativum*.

[12] • Walbot V. **Open questions: Reflections on plant development and genetics**. BMC Biology 2013, **11**: e25.
This is an interesting and thought provoking commentary.

[13] Foucher F, Morin J, Courtiade J et al. ***DETERMINATE* and *LATE FLOWERING* are two *TERMINAL FLOWER1/CENTRORADIALIS* homologs that control two distinct phases of flowering initiation and development in pea.** *Plant Cell* 2003, **15**:2742-2754.

[14] ● Kwak M, Toro O, Debouck DG, Gepts P. **Multiple origins of the determinate growth habit in domesticated common bean** (*Phaseolus vulgaris*). *Annals of Botany* 2012, **110**:1573-1580.
This paper studies *Determinate* gene sequence variation in wild and domesticated accessions of bean and characterises a variety of lesions associated with the determinate habit. A larger study found that a retroelement-associated determinate type is widespread.

[15] Tanaka W, Pautler M, Jackson D, Hirano HY. **Grass Meristems II: Inflorescence Architecture, Flower Development and Meristem Fate**. *Plant and Cell Physiology* 2013, **54**:313-324.

[16] Ferrandiz C, Navarro C, Gomez MD et al. **Flower development in *Pisum sativum*: From the war of the whorls to the battle of the common primordia.** *Developmental Genetics* 1999, **25**:280-290.

[17] Weng L, Tian ZX, Feng XZ et al. **Petal Development in *Lotus japonicus***. *Journal of Integrative Plant Biology* 2011, **53**:770-782.

[18] Benlloch R, Navarro C, Beltran JP, Canas LA. **Floral development of the model legume *Medicago truncatula*: ontogeny studies as a tool to better characterize homeotic mutations**. *Plant Reproduction* 2003, **15**:231-241.

[19] Roque E, Serwatowska J, Rochina MC et al. **Functional specialization of duplicated AP3-like genes in *Medicago truncatula***. *Plant Journal* 2013, **73**:663-675.

[20] Benlloch R, Roque E, Ferrandiz C et al. **Analysis of B function in legumes: PISTILLATA proteins do not require the PI motif for floral organ development in *Medicago truncatula***. *Plant Journal* 2009, **60**:102-111.

[21] Berbel A, Navarro C, Ferrandiz C et al. **Functional conservation of PISTILLATA activity in a pea homolog lacking the PI motif**. *Plant Physiology* 2005, **139**:174-185.

[22] Feng XZ, Zhao Z, Tian ZX et al. **Control of petal shape and floral zygomorphy in *Lotus japonicus***. *Proc Natl Acad Sci USA* 2006, **103**:4970-4975.

[23] Wang JC, Wang YM, Luo D. **LjCYC Genes Constitute Floral Dorsoventral Asymmetry in *Lotus japonicus***. *Journal of Integrative Plant Biology* 2010, **52**:959-970.

[24] Wang Z, Luo YH, Li X et al. **Genetic control of floral zygomorphy in pea** (*Pisum sativum* L.). *Proc Natl Acad Sci USA* 2008; **105**:10414-10419.

[25] Citerne HL, Pennington RT, Cronk QCB. **An apparent reversal in floral symmetry in the legume *Cadia* is a homeotic transformation**. *Proc Natl Acad Sci USA* 2006, **103**:12017-12020.

[26]● Xu SL, Luo YH, Cai ZG et al. **Functional Diversity of *CYCLOIDEA*-like TCP Genes in the Control of Zygomorphic Flower Development in *Lotus japonicus***. *Journal of Integrative Plant Biology* 2013; **55**:221-231.
This paper characterises the petal morphology mutation *kew* in *L. japonicus* and uses transgenic assays to show the roles of each of three *CYCLOIDEA* genes in conferring petal identities. Further studies reveal functional regions of the encoded proteins and their interactions with each other.

[27] Li X, Zhuang LL, Phil MAM et al. **Genetic Analysis of *ele* Mutants and Comparative Mapping of *ele1* Locus in the Control of Organ Internal Asymmetry in Garden Pea**. *Journal of Integrative Plant Biology* 2010, **52**:528-535.

[28] Liu ZP, Ma LC, Nan ZB, Wang YR. **Comparative Transcriptional Profiling Provides Insights into the Evolution and Development of the Zygomorphic Flower of *Vicia sativa* (Papilionoideae)**. *PLoS ONE* 2013, **8**: e57338.

[29] Amorim FW, Galetto L, Sazima M. **Beyond the pollination syndrome: nectar ecology and the role of diurnal and nocturnal pollinators in the reproductive success of *Inga sessilis* (Fabaceae)**. *Plant Biology* 2013; 15:317-327.

[30]●● Fourquin C, del Cerro C, Victoria FC et al. **A Change in SHATTERPROOF Protein Lies at the Origin of a Fruit Morphological Novelty and a New Strategy for Seed Dispersal in *Medicago* Genus**. *Plant Physiology* 2013, **162**:907-917.
This pioneer paper of evolutionary developmental studies in legumes reveals the amino acid residues and the mechanism that determine pod coiling in a representative sample of the genus *Medicago*.

[31] ● Geeta R, Davalos LM, Levy A et al. **Keeping it simple: flowering plants tend to retain, and revert to, simple leaves**. *New Phytologist* 2012, **193**:481-493.
This paper revisits earlier work, but takes a more stringent approach to investigate rates of change from simple leaves to complex and vice-versa.

[32] Hofer J, Turner L, Hellens R et al. ***UNIFOLIATA* regulates leaf and flower morphogenesis in pea**. *Current Biology* 1997, **7**:581-587.

[33] Chen JH, Yu JB, Ge LF et al. **Control of dissected leaf morphology by a Cys(2)His(2) zinc finger transcription factor in the model legume *Medicago truncatula***. *Proc Natl Acad Sci USA* 2010, **107**:10754-10759.

[34] Ge L, Chen J, Chen R. ***Palmate-like pentafoliata1* encodes a novel Cys(2)His(2) zinc finger transcription factor essential for compound leaf morphogenesis in *Medicago truncatula***. *Plant Signal Behav* 2010; 5:1134-7.

[35] Wang ZH, Chen JH, Weng L et al. **Multiple Components are Integrated to Determine Leaf Complexity in *Lotus japonicus***. *Journal of Integrative Plant Biology* 2013, **55**:419-433.

[36] Winter CM, Austin RS, Blanvillain-Baufume S et al. **LEAFY Target Genes Reveal Floral Regulatory Logic, *cis* Motifs, and a Link to Biotic Stimulus Response**. *Developmental Cell* 2011, **20**:430-443.

[37] Pastore JJ, Limpuangthip A, Yamaguchi N et al. **LATE MERISTEM IDENTITY2 acts together with LEAFY to activate *APETALA1***. *Development* 2011, **138**:3189-3198.

[38] ● Chahtane H, Vachon G, Le Masson M et al. **A variant of LEAFY reveals its capacity to stimulate meristem development by inducing *RAX1***. *Plant Journal* 2013, **74**:678-689.
This paper breaks new ground by revealing a separate role for *LEAFY* in meristem emergence.

[39] Peng JL, Yu JB, Wang HL et al. **Regulation of Compound Leaf Development in *Medicago truncatula* by *Fused Compound Leaf1*, a Class M *KNOX* Gene**. Plant Cell 2011, **23**:3929-3943.

[40] ●● Uppalapati SR, Ishiga Y, Doraiswamy V et al. **Loss of Abaxial Leaf Epicuticular Wax in Medicago truncatula *irg1/palm1* Mutants Results in Reduced Spore Differentiation of Anthracnose and Nonhost Rust Pathogens**. Plant Cell 2012, **24**:353-370.
A large scale screen was used to identify mutants resistant to fungal pathogens. By coincidence, this turned out to be the same zinc finger gene that regulates leaf shape complexity identified in [33]. The authors used transcriptome analysis and numerous careful assays to show resistance was mediated by changes in wax biosynthesis.

[41] Hofer J, Turner L, Moreau C et al. ***Tendril-less* Regulates Tendril Formation in Pea Leaves**. *Plant Cell* 2009, **21**:420-428.

[42]● Zhuang LL, Ambrose M, Rameau C et al. ***LATHYROIDES*, Encoding a WUSCHEL-Related Homeobox1 Transcription Factor, Controls Organ Lateral Growth, and Regulates Tendril and Dorsal Petal Identities in Garden Pea (*Pisum sativum* L.)**. *Molecular Plant* 2012, **5**:1333-1345.
A positional cloning tour-de-force in *L. japonicus* enabled the authors to identify the *Lath* gene in *P. sativum*, which reveals a specific connection between adaxial petal and leaflet blade development.

[43] Tadege M, Lin H, Bedair M et al. **STENOFOLIA Regulates Blade Outgrowth and Leaf Vascular Patterning in *Medicago truncatula* and *Nicotiana sylvestris***. *Plant Cell* 2011, **23**:2125-2142.

[44]● Jeong N, Suh SJ, Kim MH et al. ***Ln* Is a Key Regulator of Leaflet Shape and Number of Seeds per Pod in Soybean**. *Plant Cell* 2012, **24**:4807-4818.
The authors used positional cloning and complementation in Arabidopsis to define a long sought after classical locus as a zinc finger gene mutation that shapes narrow leaflets and increases seed number in soybean.

[45] DeMason DA, Chetty VJ. **Interactions between Ga, Auxin, and *Uni* Expression Controlling Shoot Ontogeny, Leaf Morphogenesis, and Auxin Response in *Pisum Sativum* (Fabaceae): Or How the *Uni-Tac* Mutant Is Rescued**. *American Journal of Botany* 2011, **98**:775-791.

[46] Zhou CN, Han L, Hou CY et al. **Developmental Analysis of a *Medicago truncatula smooth leaf margin1* Mutant Reveals Context-Dependent Effects on Compound Leaf Development**. Plant Cell 2011, **23**:2106-2124.

[47] MacDougal DT. **Duplication of Contributions on Physiology of Tendrils**. *Torreya* 1901, **1**:125-127.

[48] Jaffe MJ, Galston AW. **Physiology of Tendrils**. *Annual Review of Plant Physiology* 1968, **19**:417-434.

[49] Moran N. **Osmoregulation of leaf motor cells**. *FEBS Letters* 2007, **581**:2337-2347.

[50] van Zanten M, Pons TL, Janssen JAM et al. **On the Relevance and Control of Leaf Angle**. *Critical Reviews in Plant Sciences* 2010, **29**:300-316.

[51]● Zhou CN, Han L, Fu CX et al. **Identification and characterization of *petiolule- like pulvinus* mutants with abolished nyctinastic leaf movement in the model legume *Medicago truncatula***. *New Phytologist* 2012; 196:92-100.
The authors characterised the same *M. truncatula* gene as in 52, but gave it a different name, which may lead to confusion. Nonetheless, genetic interaction and expression studies provide excellent data that complement 52.

[52]● Chen JH, Moreau C, Liu Y et al. **Conserved genetic determinant of motor organ identity in *Medicago truncatula* and related legumes**. *Proc Natl Acad Sci USA* 2012, **109**:11723-11728.
This paper gives the first genetic insight into the development of the pulvinus, nicely combining independent avenues of research in three different species into one comparative view.

[53] Markmann K, Parniske M. **Evolution of root endosymbiosis with bacteria: how novel are nodules?** *Trends in Plant Science* 2009, **14**:77-86.

[54] Oldroyd GED. **Speak, friend, and enter: signalling systems that promote beneficial symbiotic associations in plants**. *Nature Reviews Microbiology* 2013, **11**:252-263.

[55] Doyle JJ. **Phylogenetic Perspectives on the Origins of Nodulation**. *Molecular Plant-Microbe Interactions* 2011, **24**:1289-1295.

[56] ●● Couzigou JM, Zhukov V, Mondy S et al. ***NODULE ROOT* and *COCHLEATA* Maintain Nodule Development and Are Legume Orthologs of Arabidopsis *BLADE-ON-PETIOLE* Genes**. *Plant Cell* 2012, **24**:4498-4510.
In identifying the first known gene controlling nodule identity, this paper brings a fresh perspective to questions asked decades earlier: are there nodule identity genes and what are the origins of the nodule? The authors have opened a new avenue of research in symbiosis.

[57] Sharma V, Tripathi BN, Kumar S. **Organ-wise homologies of stipule, leaf and inflorescence between *Pisum sativum* genetic variants, *Delonix regia* and *Caesalpinia bonduc* indicate parallel evolution of morphogenetic regulation.** *Plant Syst Evol* 2012, **298**:1167–1175.

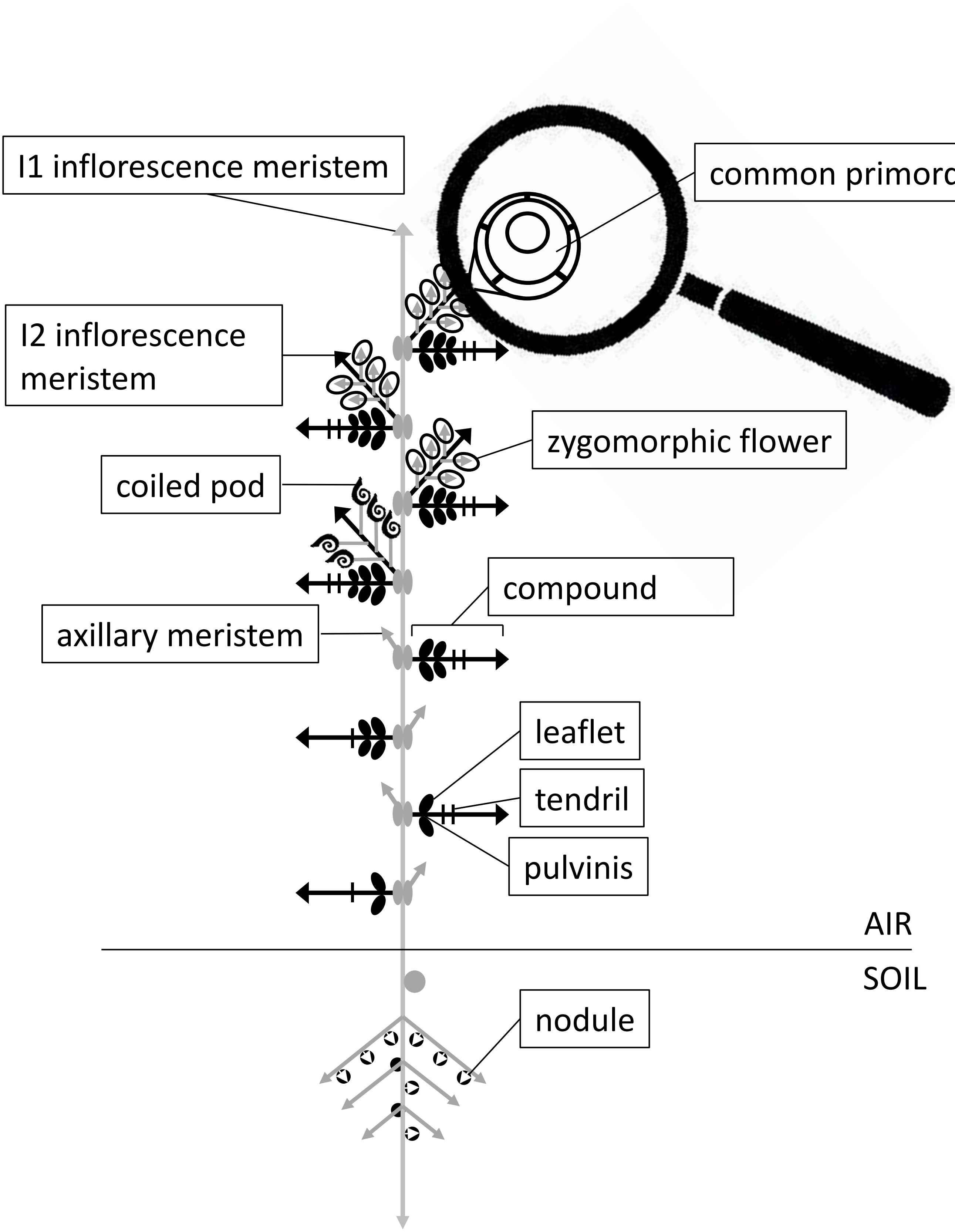

Figure 1. Representation of a hypothetical papilionoid legume highlighting (in black) specialized meristems (arrowheads) and organs discussed in the text.

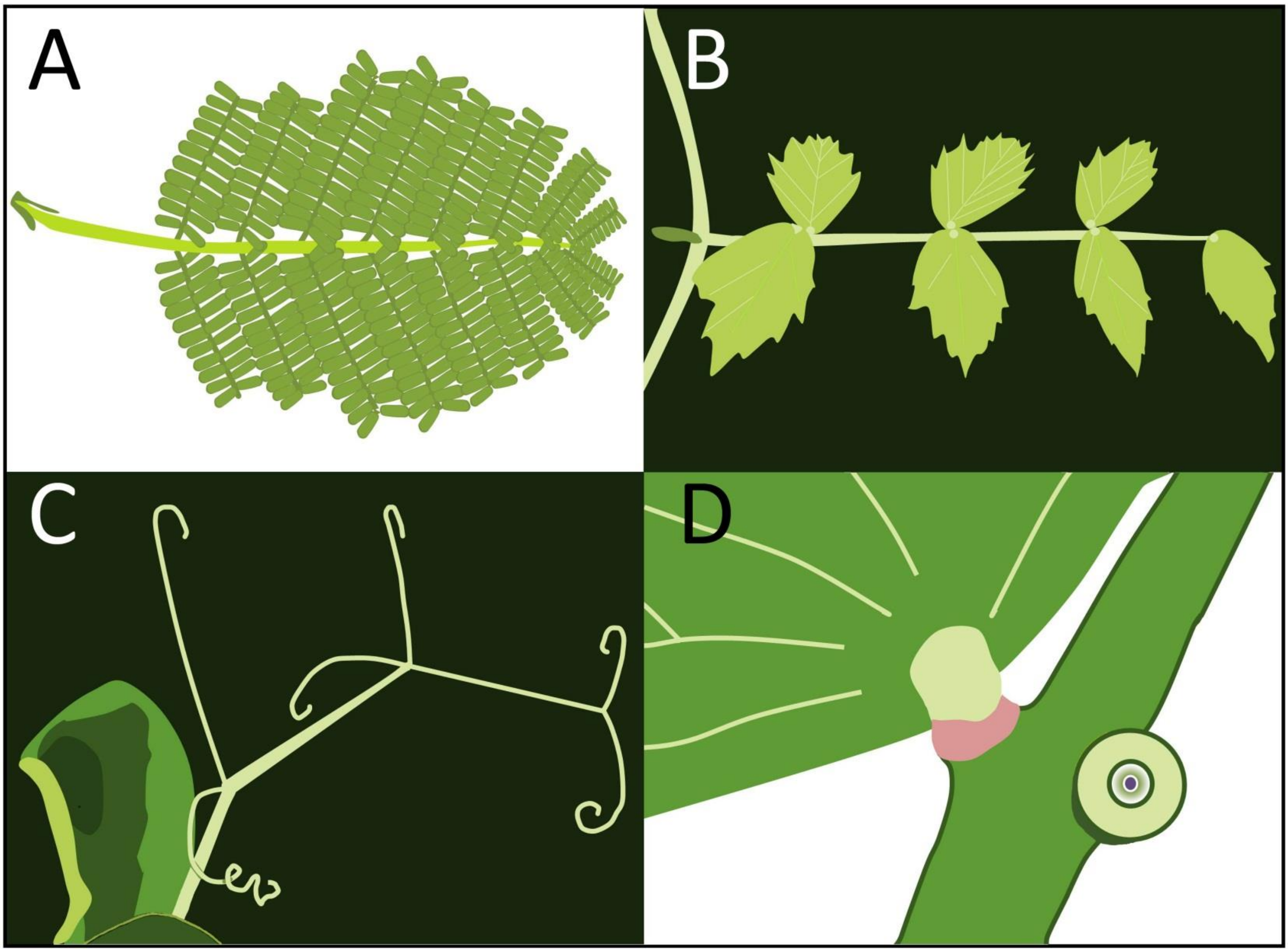

Figure 2. Examples of legume compound leaves and their novel leaf organs.
A. Bipinnate leaf, *Delonix regia.* B. Once-pinnate leaf*, Cicer arietinum.* C. Leaflet tendrils, *Pisum sativum*. D. Pulvini, *P. sativum*. An intact leaflet pulvinis, delimited at its base by red coloured epidermal cells, is shown on the left, a section through a pulvinis on the opposite leaflet of the same leaf, as stained with toluidine blue, is shown on the right.