Parallel evolution in *Senecio* sect. *Senecio*:

the genetics of adaptive shifts in flowering time reconsidered

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Futuyma (2010) *Evolutionary constraint and ecological consequences*. Evolution, 64, 1865–1884

„ ....one of the most important shifts in evolutionary biology in the past 50 years is an increased recognition of *sluggish evolution and failures to adapt"*.

VOLUME 64, NUMBER 7, JULY 2010

EVOLUTION

INTERNATIONAL JOURNAL OF ORGANIC EVOLUTION

Non-adaptive evolution of plant genome size

Genetic architecture of wing size divergence

Rate of adaptation in asexuals

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a) the structure or paucity of genetic variation in populations
b) the genetic basis of species trait differences

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- phylogenetic niche conservatism (ecology)

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- **organism-wide stasis (palaeontology)**
- **phylogenetic niche conservatism (ecology)**

Is there evidence that genetic variation can limit evolution in *Senecio* – or is there not a prepoderance for rapid and parallel shifts in adaptive traits with a simple genetic basis?

A) Is there insufficiency of selectable genetic variation in populations/species?

- **There IS detectable *additive genetic variance* for both vegetative and floral traits in, e.g., *S. vulgaris* ssp. *vulgaris*/*<u>denticulatus</u>* (Comes, 1994, 1998) ...**

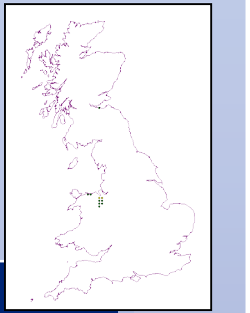
....but the ‚specialist‘ (<u>non-weedy</u>) taxon may lack genetic variation necessary to expand its range.

S. vulgaris ssp. vulgaris var. vulgaris

S. vulgaris ssp. <u>denticulatus</u>

Comes (1994) Ph.D. thesis, Heidelberg University, Germany.
Comes (1998) *Journal of Heredity*, 89, 54–61.

A) Is there insufficiency of selectable genetic variation in populations/species?



- Insufficiency is implied when adaptation is facilitated by infusion of selectable genetic variation (or fitness genes) by hybridization/introgression with other species ...

.....Transfer of ray floret genes (*RAY1/2*) between *S. squalidus* and *S. vulgaris* var. *vulgaris* in the U.K.

S. vulgaris ssp. vulgaris var. vulgaris

S. squalidus

S. vulgaris ssp. v. var. hibernicus

Kim et al. (2008) *Science*, 322, 1116–1119.
Chapman & Abbott (2010) *New Phytologist*, 186, 63–71.

A) Is there insufficiency of selectable genetic variation in populations/species?

In some species it might be that not all traits are genetically variable enough, perhaps especially some ecologically most interesting ones...

....but for which we presently have insufficient knowledge of their underlying genetic basis (few exceptions)

A) Is there insufficiency of selectable genetic variation in populations/species?

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Also, the frequent occurrence of introgression involving selfers (*S. vulgaris*, *S. flavus*) is intriguing ...

....are they particularly prone to introgression (e.g., due to low intra-specific gene flow; Currat et al. 2008) and despite ploidy barriers (2x vs. 4x)?

Currat et al. (2008) *Evolution*, 62, 1908–1920.

A) Is there insufficiency of selectable genetic variation in populations/species?

Also, the frequent occurrence of introgression involving selfers (*S. vulgaris*, *S. flavus*) is intriguing ...

... and what other than floral traits are introgressed, and which genes are linked to the ray floret gene(s)?

B) Genetic correlations, loss of evolutionary potential/reversals, mutational targets

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Selection on the ray floret locus might be constrained by genetic correlation with other ecologically relevant traits...

....but such linkage might also facilitate adaptation and extension of ecological range via introgression (*S. flavus*!?)

B) Genetic correlations, loss of evolutionary potential/reversals, mutational targets

There is some evidence in *Senecio* that developmental pathways are controlled by major genes...

....the loss of one or few such genes may spell the irreversible loss of a character (unless re-introduced by introgression)

B) Genetic correlations, loss of evolutionary potential/reversals, mutational targets

Some authors (e.g., Houle, 1998) pointed out that the mutational traget of such traits is small; mutation rate limits variance available for selection...

....but is has long been recognized that genes of major effect have a higher probability to go to fixation (Kimura, 1983)

Houle (1998) Genetica, 102/103, 241–253
Kimura (1983) *The Neutral Theory of Molecular Evolution.* Cambridge University Press, Cambridge.

B) Genetic correlations, loss of evolutionary potential/reversals, mutational targets

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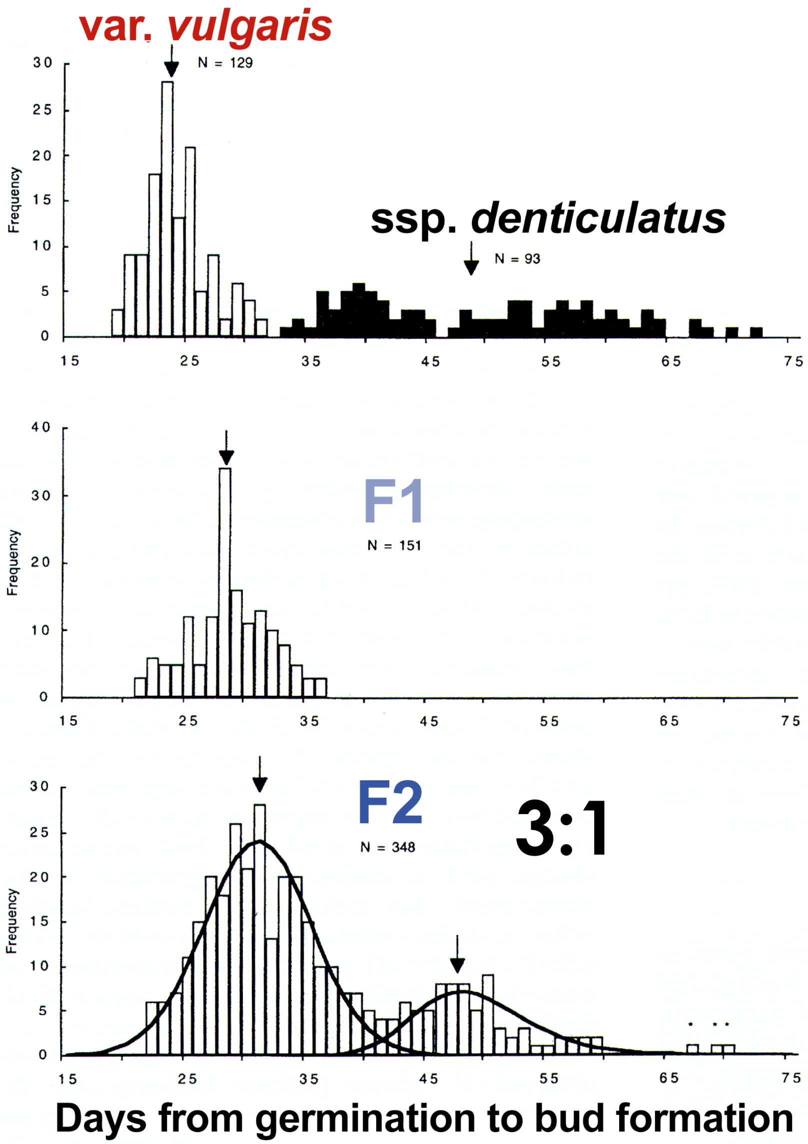
OLIGOGENIC ECOLOGICAL TRAITS MAY SPEED UP – RATHER THAN CONSTRAIN – ADAPTIVE DIVERGENCE, SPECIATION, AND PARALLEL EVOLUTION

Phenological and ecological isolation between two subspecies of *Senecio vulgaris* (Asteraceae)

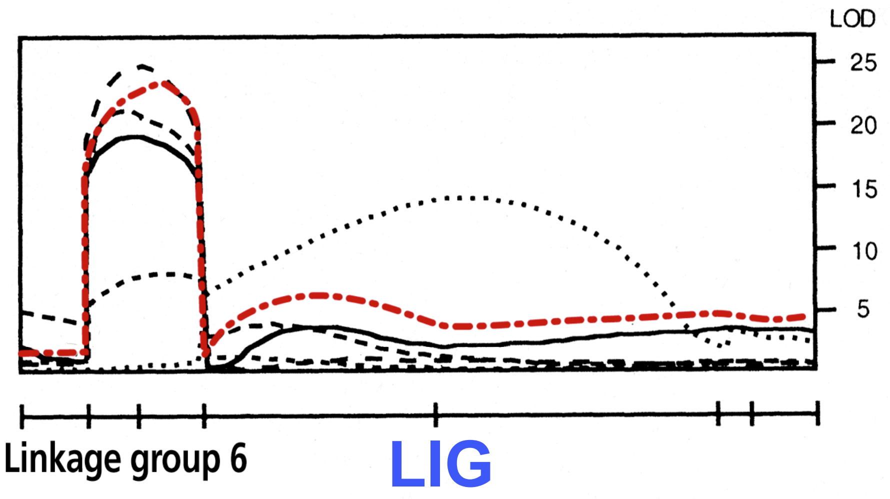


var. *vulgaris* (Derivative) – cosmopolitan weed

ssp. *denticulatus* (Progenitor) – Atlantic/Med Europe

Comes & Kadereit (1996) Heredity 77: 544-554

Phenological isolation governed by a major „speed of development“ gene closely linked to LIG (ray florets)



-·-·-·-	**Days from germination to first bud formation**
———	Plant height at first bud formation (LG6)
- - - -	Leaf no. on main axis at first anthesis
--------	Lateral branch no. along main axis at first fruiting
------·	Outer involucral bracts
··········	Disc florets per capitulum

var. *vulgaris*

ssp. *denticulatus*

Source: Moritz & Kadereit (2001) Pl. Biol. 3: 544-552

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The QTL gene region identified by Moritz & Kadereit (2001), and especially the ‘*SOD*’ locus, might offer promising perspectives for studying variation in an ecologically important trait (flowering time) that considerably varies in a natural system, such as *Senecio* sect. *Senecio*.

Comes & Abbott (2002) Evolution, 55, 1943–1962.

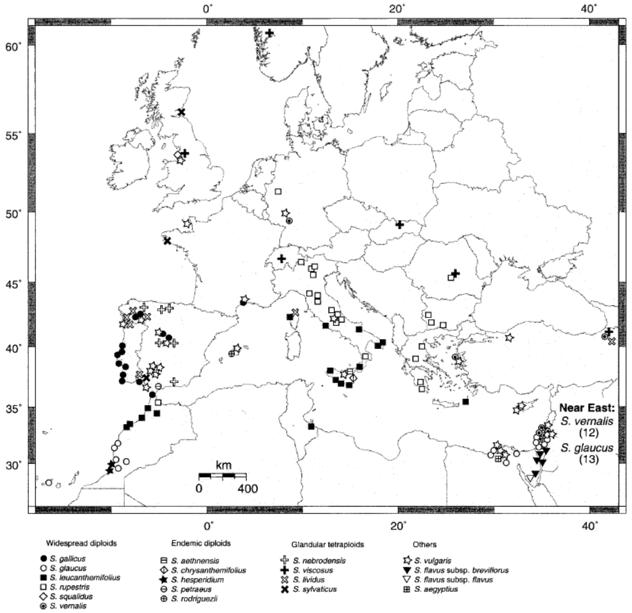


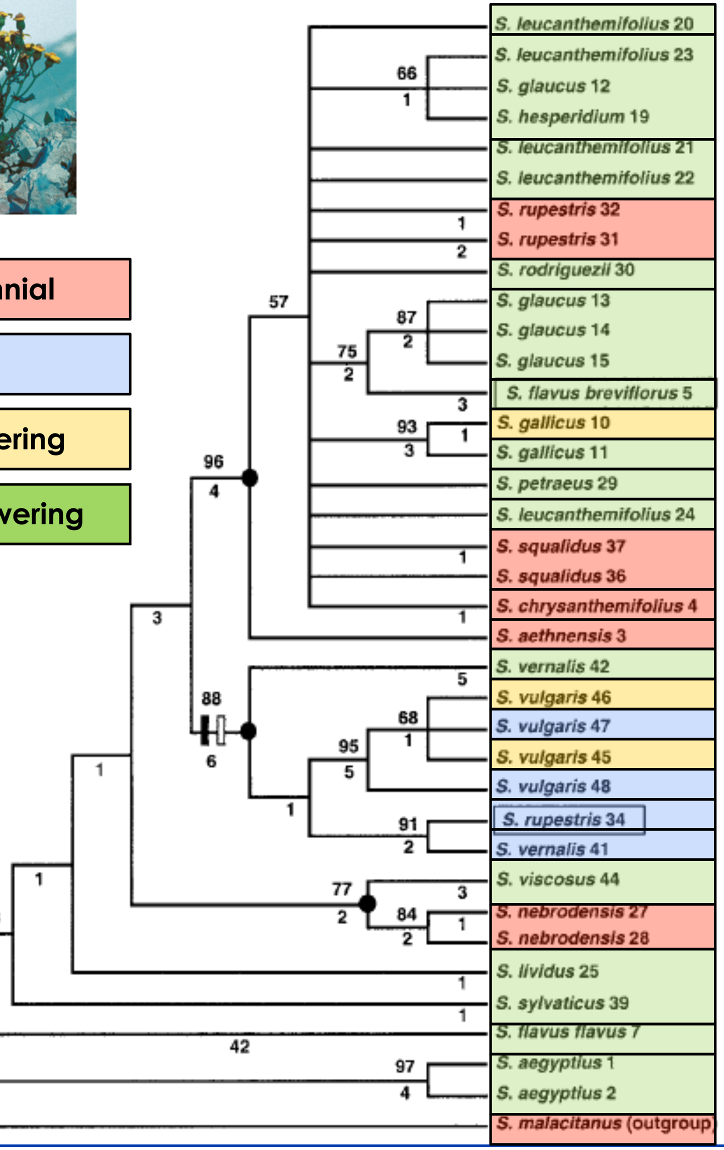
FIG. 1. Map of the sampled populations for 18 species of the Mediterranean *Senecio* complex used in this study for cpDNA haplotype analysis. For two densely collected species from the Near East (*S. vernalis* and *S. glaucus*), numbers in parentheses refer to the actual numbers of populations sampled. See Table 1 for approximate distribution ranges of the species and subspecies analysed.

Short-lived perennial

Winter-annual

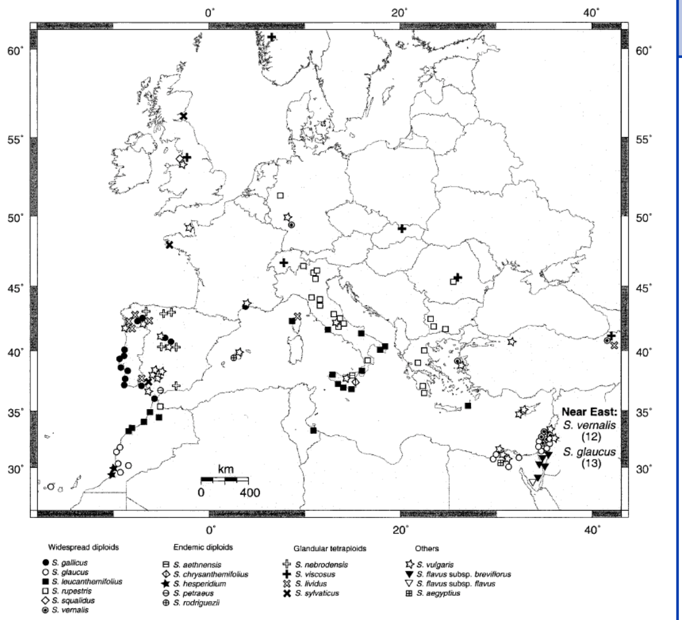
Annual late-flowering

Annual-early flowering



ITS phylogeny *Senecio* sect. *Senecio*

Comes & Abbott (2002) Evolution, 55, 1943–1962.



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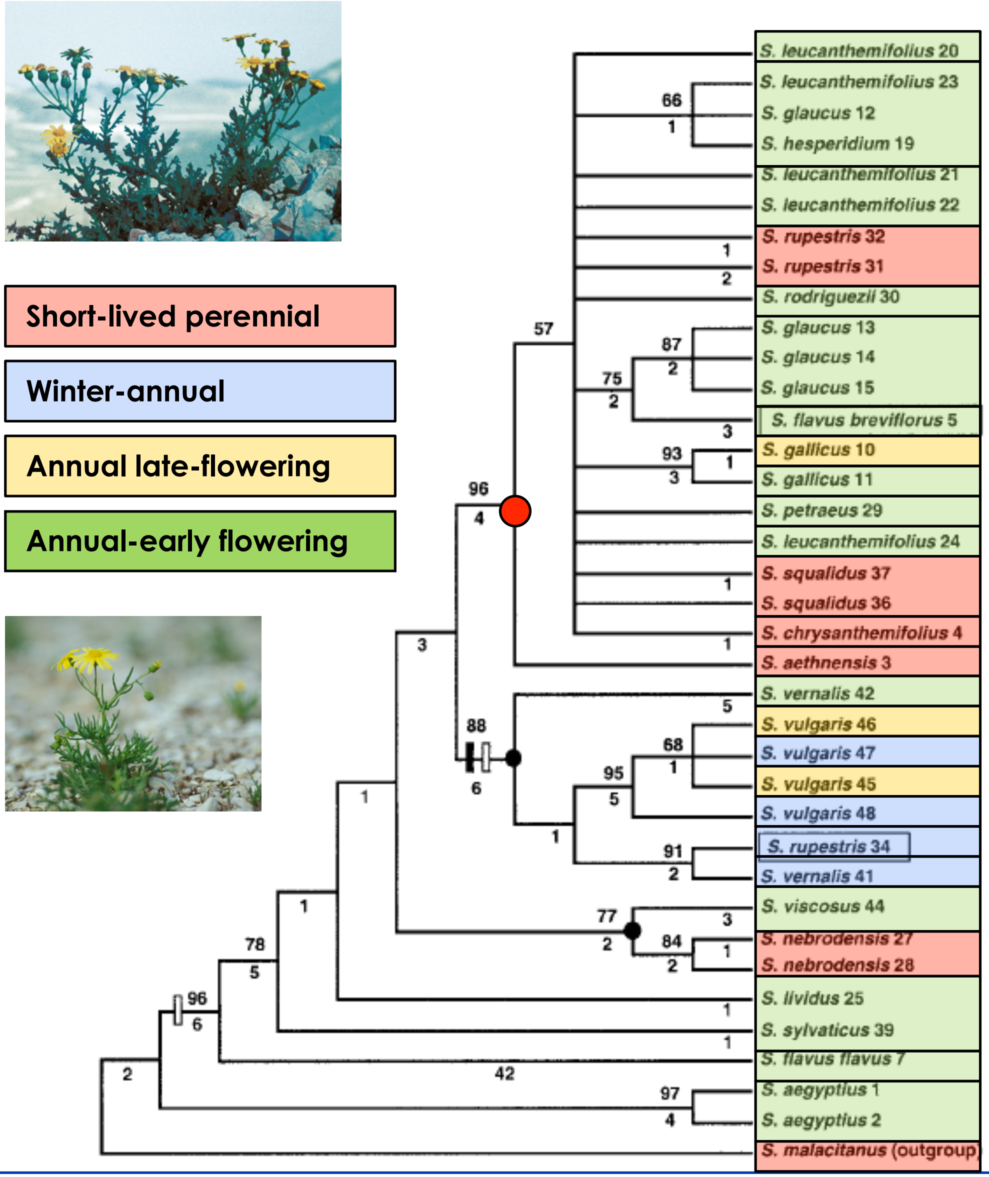


🟥 Short-lived perennial

🟦 Winter-annual

🟨 Annual late-flowering

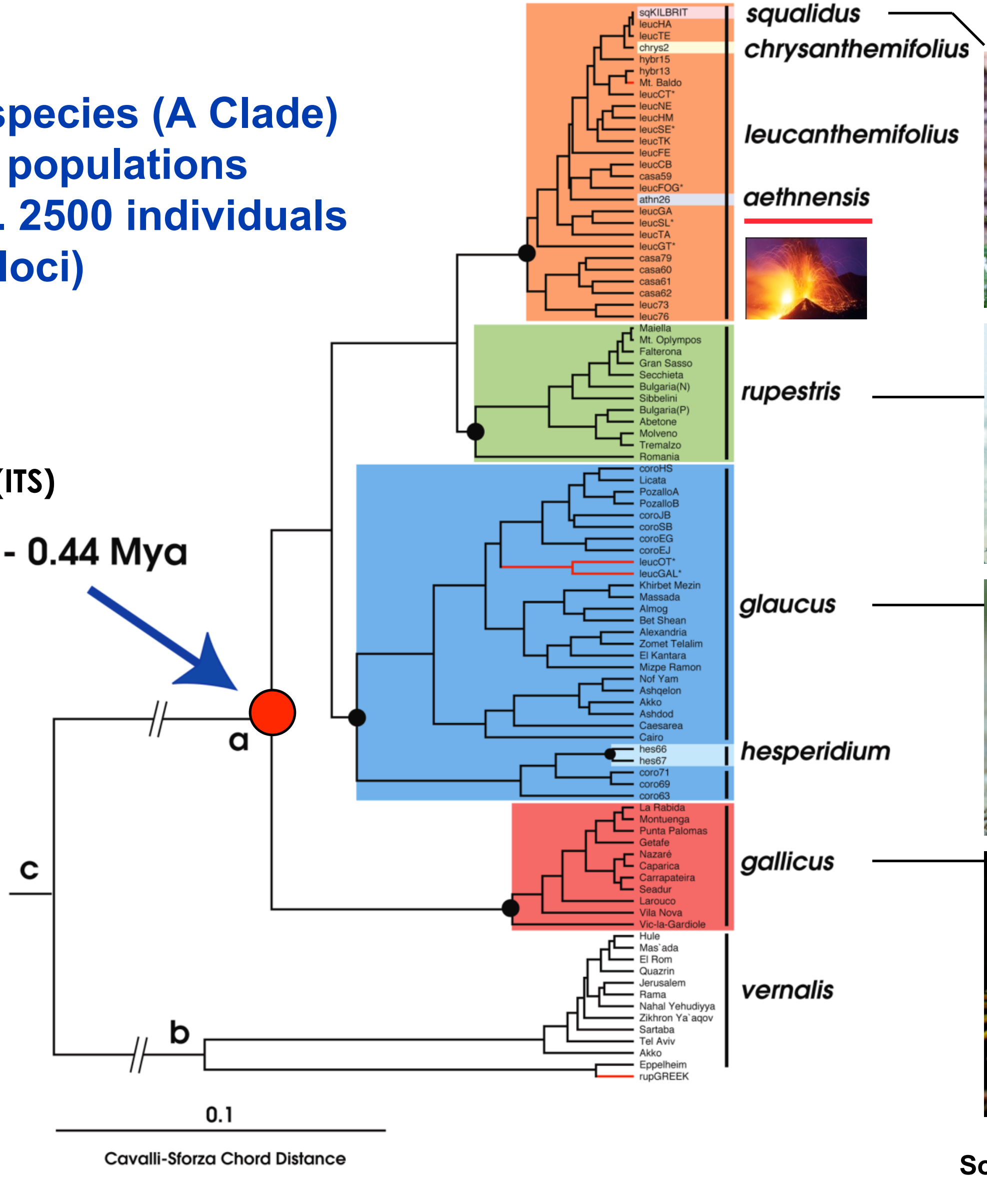
🟩 Annual-early flowering

ITS phylogeny *Senecio* sect. *Senecio*

● Major radiation (1.0–0.44 Ma)

Allozymes (UPGMA)

8 species (A Clade)
80 populations
ca. 2500 individuals
(5 loci)



(ITS)

1.0 - 0.44 Mya

S. squalidus

S. rupestris

S. glaucus

S. gallicus

Source: Comes, Abbott & Forbes (unpubl. data)

There are promising signs of the times which may allow identifying and characterizing the flowering time gene(s) in *Senecio*…

Kim et al. (2008) *Regulatory genes control a key morphological and ecological trait transferred between species*. Science 322: 1116–1119

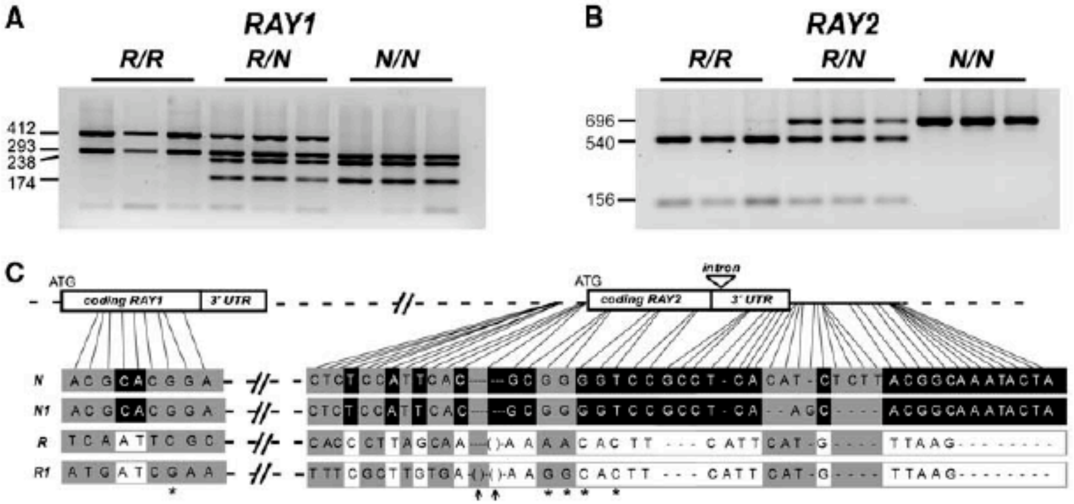


Fig. 3. (**A**) *RAY1*: a 412-bp band that cosegregates with *R* and a 238-bp and a 174-bp band with *N* in an F2 population. (**B**) *RAY2*: a 540-bp and a 156-bp band that cosegregate with *R* and a 696-bp band with *N*. PCR products of *RAY1* and *RAY2* coding regions were digested with *TaqI* and *EcoRI*, respectively. (**C**) Variable sites at *RAY1* and *RAY2* in and around the coding regions for the four haplotypes *N*, *N1*, *R*, and *R1*. Polymorphisms that are diagnostic for *N/N1* versus *R/R1* haplotypes are shown surrounded by black and white, respectively. All other polymorphisms are highlighted in gray. Nucleotide polymorphisms that cause amino acid changes are indicated with asterisks. Positions of deletions of TAAGGAAATCCAAACCCCA and ATAGAAA in the *RAY2-R1* haplotype are marked with arrows.

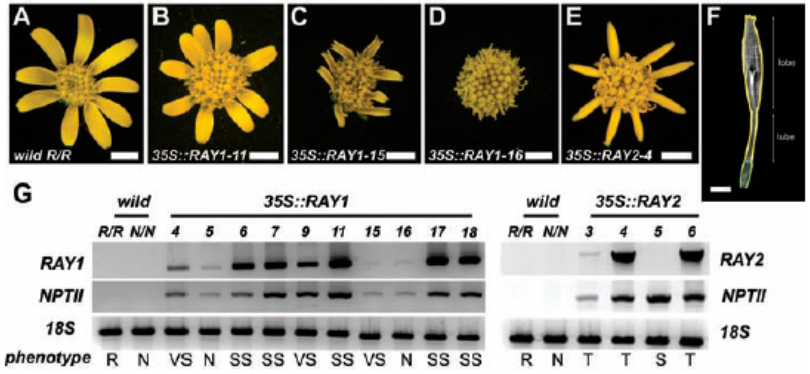


Fig. 4. (**A**) Flower-head phenotypes of *RR* nontransgenic control plant. (**B**) Flower head with slightly short rays from a transgenic plant overexpressing an internal fragment of *RAY1 N* allele coding sequences in a radiate (*R/R*) *S. vulgaris* background. (**C**) Flower head from a *RAY1* transgenic, as in (B), with very short ray florets. (**D**) Flower head from a *RAY1* transgenic, as in (B), giving no ray florets. (**E**) Flower head from a transgenic overexpressing the *RAY2 N* allele coding sequences in a radiate (*R/R*) *S. vulgaris* background, giving tubular ray florets. (**F**) Section through a ventralized ray floret, color-coded as in Fig. 1D. All transgenics are T1 generation, obtained by self-pollinating the primary transformants. (**G**) Semiquantitative RT-PCR showing expression levels of *RAY1* and *RAY2* in the transgenics, together with controls for *18S* RNA and the kanamycin resistance gene (*NPTII*). R, normal radiate head; N, nonradiate or discoid head; SS, slightly short rays; S, short rays; VS, very short rays; T, tubular rays with ventralized petals. Scale bars, 2 mm [(A) to (E)] and 1 mm (F).

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Wang et al. (2010) *PEP1 regulates perennial flowring in Arabis alpina*. Nature 459: 423–427

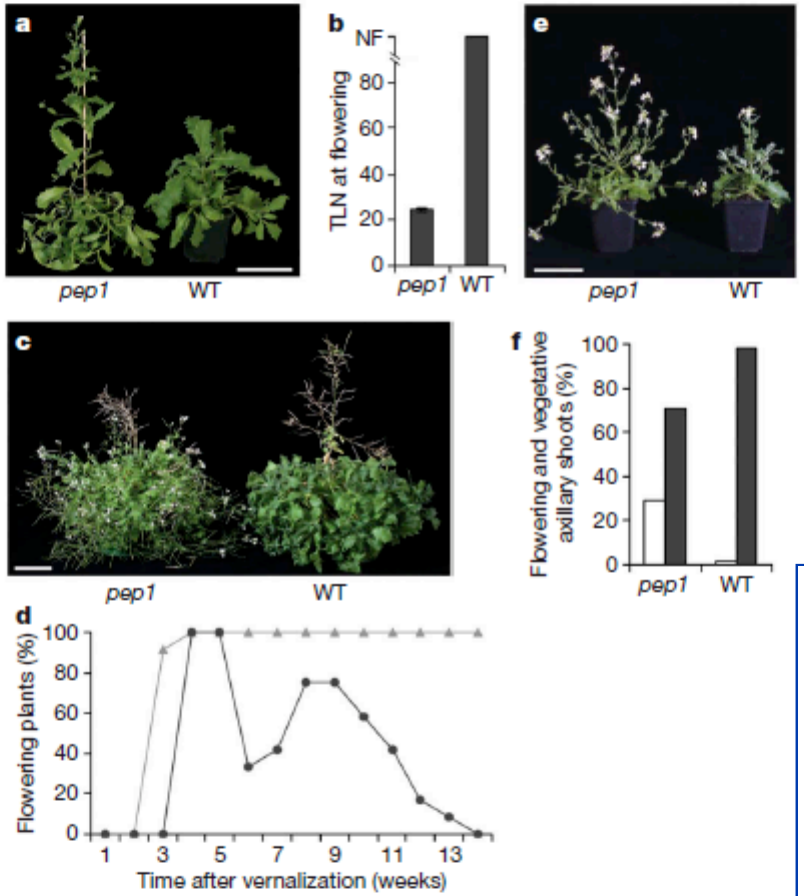

Figure 2 | *PEP1* restricts the flowering phase and enhances polycarpy of *A. alpina*. **a**, The *pep1* mutant flowers without vernalization. **b**, Flowering time of *pep1* plants compared to wild type (WT) without vernalization. NF, no flowering; TLN, total leaf number. **c**, *pep1* mutants show longer periods of flowering than wild-type plants. Plants were grown for 14 weeks under long days after 12 weeks vernalization. **d**, Duration of the flowering phase of *pep1* (grey) and wild-type plants (dark grey) after vernalization. **e**, More axillary branches undergo flowering in *pep1* plants than in wild-type as shown by plants grown for 3 weeks under long days after 12 weeks vernalization. **f**, Percentage of vegetative and flowering axillary shoots produced by *pep1* and wild-type plants. Plants were scored when the first open flower was present. Flowering shoots (white) and vegetative shoots (dark grey) are shown. In **c**–**f**, plants were grown under long days for 5 weeks before being vernalized for 12 weeks. $n = 12$ (**b**, **d**, **f**); scale bars, 10 cm.

Prospects I

Next generation sequencing technologies, recent developments of *Senecio* genomic resources, plus information from model organisms, should make it possible to:

- Map and sequence the genes that control ecologically important traits in natural systems, such as *Senecio* sect. *Senecio*.
- Bring new insights into our understanding of the genetics of parallel plant adaptation and (incipient) speciation.

Prospects I

- ....particularly, the specific gene(s) associated within the QTL region shown to affect flowering (generation) time, a genomic region (*SOD*) (Comes & Kadereit, 1996; Comes, 1998; Moritz & Kadereit, 2001)
- Probably located in the same linkage group of the previously isolated and characterized ray floret genes [*RAY*1/*RAY*2] (Kim et al., 2008).

Comes & Kadereit (1996) *Heredity*, 77, 544–554
Comes (1998) *Journal of Heredity*, 89, 54–61
Moritz & Kadereit (2001) *Plant Biology*, 3, 544–552

Kim et al. (2008) *Science*, 322, 1116–1119

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NOTE: The genetic architecture of flowering time has been extensively studied in model species and crops – but rarely in natural systems at different spatial/temporal scales.

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Prospects II (once identified...)

- Analyse *SOD* gene sequence variation across various intra-specific variants and species of sect. *Senecio*.

- Provide a clear picture of the general phylogenetic trends of flowering time evolution during the Quaternary radiation of this group.

Prospects II (once identified...)

- Compare the genetic basis of similar flowering time traits that have evolved in different locations and species (i.e. test for evolutionary parallelism at the molecular level), caused by, e.g.,

- the same nucleotide substitutions at the same gene(s),
- different changes in the same gene(s),
- entirely different genetic changes at different loci;
- Etc.

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Prospects II (once identified)

- Study phylogeographic patterns of *SOD* allelic sequence variation, including signatures of signatures of recurrent positive selection...
- ... at different spatial scales, e.g., across latitudinal gradients (*S. vernalis, S. gallicus*), across hybrid zones (e.g., *denticulatus* vs. *vulgaris* on Jersey Island), or even in reciprocal transplants.

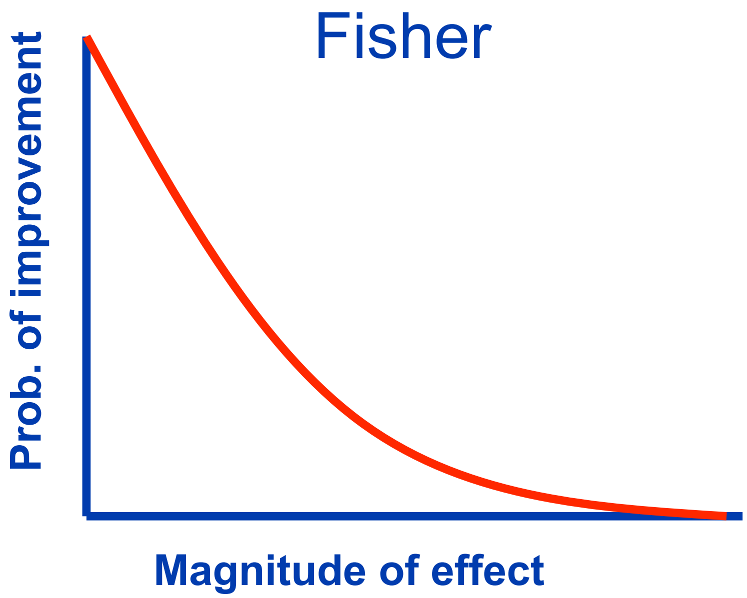
Specific note

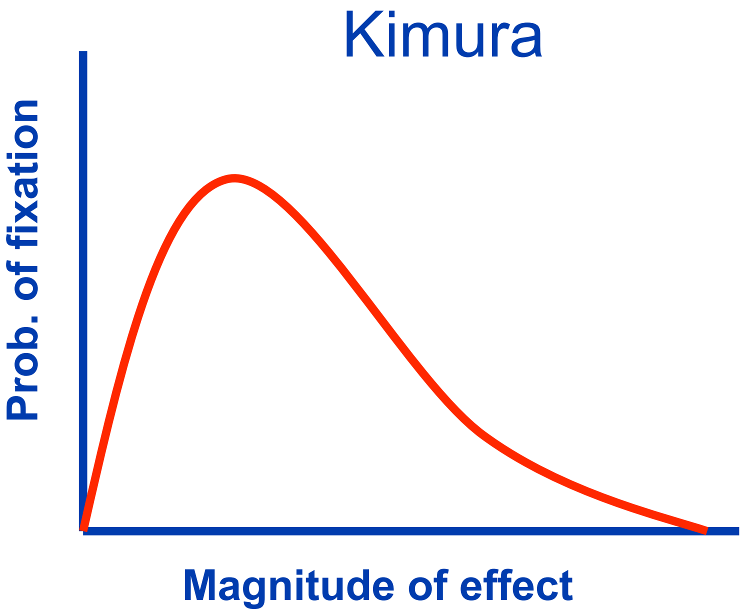
- Recover homeologous copies of the *SOD* gene in tetraploid *S. vulgaris* (similar to the ray floret gene(s); see Chapman & Abbott, 2010).
- Tracing such copies in the close diploid relatives might eventually clarify the origin of this species (allo-/autoploid)...

Chapman & Abbott (2010) ***New Phytologist*****, 186, 63–71**

Kimura (1983) *The Neutral Theory of Molecular Evolution.* Cambridge University Press, Cambridge.

- Fisher ignored probability of fixation
- Probability of fixation of mutation is roughly proportional to its effect
- Mutations of larger effect have higher probability of fixation


Fisher


Kimura

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