Zygomorphy and Heteranthery in *Solanum* in a Phylogenetic Context

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Keywords: Solanaceae, heterandry, floral morphology, phylogeny, enantiostyly

Abstract

The majority of species in the large genus *Solanum* (ca. 1500 species) have five-merous, radially symmetrical flowers with equal stamens. However, some *Solanum* species and groups are characterized by four-merous and/or zygomorphic flowers, unequal stamens and enantiostyly (styles deflected to one side of the flower). Previous workers have used flower and seed coat morphology in these unusual *Solanum* species as a guide to interpreting their evolutionary relationships. However, the phylogenetic position of the zygomorphic and heterantherous solanums is only beginning to be examined using molecular data and cladistic methodology. DNA sequence data from both the chloroplast and nuclear genomes are used to infer the phylogenetic position of *Solanum* taxa with variously modified flowers. Zygomorphy and heteranthery have evolved multiple times within *Solanum*, and most frequently within the spiny solanums (*Solanum* subg. *Leptostemonum*). The phylogenies shed light on the disparate morphologies and geographical distributions encountered in the zygomorphic and heterantherous species and pinpoint the likely relatives of these taxa among the actinomorphic *Solanum* species with equal stamens.

INTRODUCTION

Solanum, with approximately 1500 species, is the largest genus in the Solanaceae and one of the largest genera of flowering plants (Frodin, 2004). In general, *Solanum* flowers are easily recognized by their five-merous, radially symmetrical flowers with equal (isantherous) stamens that dehisce by terminal pores. The flowers are adapted for buzz pollination by bees, which vibrate the anthers using their indirect flight muscles and in turn cause pollen to discharge in a stream from the pores (Buchmann, 1983). Within the Solanaceae, only the genus *Lycianthes* also exhibits poricidal anther dehiscence, and in most cases, it can be readily distinguished from *Solanum* by differences in calyx structure (D'Arcy, 1986).

A number of *Solanum* taxa have modified the stereotypical *Solanum* flower ground plan to exhibit such unusual floral features as four-merous and/or zygomorphic flowers, unequal stamens, and enantiostyly (styles deflected to one side of the flower in a left- or right-handed arrangement). Heteranthery (the presence of highly unequal stamens in a single flower; also called heterandry in previous *Solanum* literature) is a particularly notable feature that occurs in many *Solanum* lineages. In *Solanum*, differences in overall stamen size can be due to differences in filament length, anther length, or both. In some cases, these morphological modifications are so striking that they have led previous taxonomists to exclude these species from *Solanum*. For example, species with highly heterantherous and often zygomorphic flowers were segregated into the genera *Normania* Lowe, *Nycterium* Venten., and *Androcera* Nutt. (Whalen, 1984; Lester et al., 1999; Francisco-Ortega et al., 1993), although these genera are now subsumed within *Solanum* (Whalen, 1979, 1984; Bohs and Olmstead, 2001). The occurrence of unusual floral features within various *Solanum* lineages and related genera is mapped onto a summary cladogram derived from Weese and Bohs (2007) in Fig. 1.

The evolution of derived floral morphologies in *Solanum* was most comprehensively examined by Lester et al. (1999), who used SEM studies of pollen grains and seed surface features to determine whether the heterantherous *Solanum* species form a natural group, examined biogeographic patterns among the heterantherous solanums, and attempted to identify the sister groups to the heterantherous taxa. The work reported here focuses on the same key species groups identified in Lester et al. (1999) and builds on this foundation by analyzing most of these species in a phylogenetic framework using DNA sequence data. Our results are compared to those of Lester et al. (1999) and in some cases extend beyond their paper to suggest well supported sister relationships between heterantherous and isantherous *Solanum* species. We also evaluate the biogeographic patterns of the heterantherous species in a phylogenetic context and suggest further morphological and molecular studies that will elucidate evolutionary and developmental patterns in these taxa.

TAXA STUDIED

Within *Solanum*, several groups exhibit striking degrees of heteranthery, often accompanied by other floral modifications such as zygomorphy and enantiostyly. Lester et al. (1999) focused on four *Solanum* groups that show strong heteranthery: 1) the *Normania* group, 2) the *Androceras* group, 3) the *Anisantherum* and *Monodolichopus* group, and 4) the *Nycterium* group. In our study, we included representatives of as many of these groups as possible, along with isantherous species hypothesized to be their closest relatives. We also included *S. thelopodium*, a member of the *S. thelopodium* species group of Knapp (2000). Flowers in this group are highly heterantherous but these species were not examined by Lester et al. (1999). Floral morphologies, breeding systems (here defined as the presence of self-compatibility vs. self-incompatibility), and previous ideas of relationships are summarized below and in Table 1 for the five focal groups. Illustrations of representative flowers of heterantherous *Solanum* species and their relatives are given in Figs. 2 and 3.

Focal Groups in the Non-Spiny Solanums

1. *Solanum thelopodium* Species Group. This group includes three non-spiny species of South American primary rainforest shrubs. These species were neglected until Knapp's recent treatment, in which two of the three species were newly described (Knapp, 2000). Although these taxa are obviously closely related, they have not been properly placed in any of the published *Solanum* sections (see Knapp, 2000 for a summary of the taxonomic and nomenclatural history of the group). The flowers are five-merous with actinomorphic corollas. The androecium is highly heterantherous with a unique combination of stamen morphologies (Fig. 2A). The lowermost stamen is longest due to its very long anther and filament. The uppermost stamens are shortest and the middle two stamens are intermediate in length, with the length difference due mainly to differences in filament length (Knapp, 2000). The style is straight and extends through the groove between the anther thecae of the longest and lowermost stamen. There is no evidence of andromonoecy in this group, and breeding systems of the three species are unknown.

2. *Normania* Clade. This monophyletic group encompasses three species formerly placed in the genera *Normania* and *Triguera*. All were reinstated in or transferred to *Solanum* by Bohs and Olmstead (2001) when they were found to form a clade nested within the genus. *Solanum trisectum* and *S. nava* Webb & Berthel. (former members of genus *Normania*) are native to Macaronesia, whereas *S. herculeum* (former genus *Triguera*) is found in nearby areas of Spain and northwestern Africa. All are five-merous, with actinomorphic to slightly zygomorphic corollas and unusual floral morphologies, but the flowers of *S. herculeum* are quite different from those of the other two species. In this species, the anthers are equal or subequal and each is tipped by two small apical horns (Fig. 2E). They dehisce initially by two subapical pores that enlarge into longitudinal slits with age. Breeding systems and pollination biology have not been investigated in this species.

In contrast, the androecium of *Solanum trisectum* and *S. nava* is highly heterantherous, with two long curved anthers, two medium-sized curved anthers, and one short relatively straight anther (Fig. 2D). The shortest stamen is lowermost in the flower, and the four longer anthers have a projection or horn at the middle or near the base. Dehiscence is ultimately longitudinal, as in *S. herculeum*. The curved style is exserted from and extends between the two long stamens. The breeding system of *S. trisectum* was found to be self-compatible (Bohs and Olmstead, 2001), but that of *S. nava*, one of the rarest species in the Canary Islands, is unknown. Pollination remains to be investigated in these species.

Focal Groups in the Spiny Solanums (*Solanum* subg. *Leptostemonum*)

1. *Solanum* sect. *Androceras*. This section includes 12 species of annual or rhizomatous perennial herbs, often with highly dissected leaves. They range from the central and southwestern USA to Mexico and are found predominantly in warm, semiarid to arid regions. The section was revised by Whalen (1979) and summarized as the *S. rostratum* group in Whalen (1984). The corollas in this section are 5-lobed, zygomorphic, and yellow, white, or purple. Of the five attenuate anthers, the upper four are small, yellow, and straight, whereas the lowermost is much larger, curved, deflected to one side, and often suffused with purple or reddish pigment (Fig. 3A, B). Filament lengths are nearly equal in all five stamens. All species of section *Androceras* exhibit monomorphic enatiostyly, where the curved style is deflected to the side of the flower opposite the large anther, and right- and left-handed flowers alternate along the inflorescence axis. The plants are weakly andromonoecious, with female-sterile flowers on distal parts of the inflorescence and perfect flowers in the proximal half to two-thirds of the inflorescence (Whalen, 1979). Nine species of section *Androceras* grown by Whalen in an experimental garden were all self-compatible (Whalen, 1979; Whalen and Anderson, 1981).

Whalen (1979, 1984) posited that section *Androceras* is most closely related to other heterantherous *Solanum* species such as *S. tridynamum* and *S. azureum* Fern., placed in *Solanum* sect. *Nycterium* and also known as the *S. vespertilio* species group. *Solanum tridynamum* and *S. azureum* are native to Mexico, like most species in section *Androceras*. Jaeger (1985) also considered section *Androceras* to be most closely related to section *Nycterium*. Other authors, however, such as Dunal (1813, 1852), Walpers (1844), Danert (1970) and Lester et al. (1999) believed section *Androceras* to be more closely allied with section *Cryptocarpum*, which includes taxa with actinomorphic, isantherous flowers. Similarities in seed surface morphology between *S. sisymbriifolium* of section *Cryptocarpum* and species of section *Androceras* especially supported this view (Lester et al., 1999).

2. Sections *Anisantherum* and *Monodolichopus*. These two Old World sections include just two species each. Section *Anisantherum* is comprised of *Solanum pubescens* Willd. from India and *S. somalense* Franchet from east Africa, whereas section *Monodolichopus* includes the east African species *S. coagulans* Forssk. and *S. melastomoides* C.H. Wright. *Solanum coagulans* was placed by Dunal (1852) in section *Nycterium* with the other heterantherous spiny solanums, and Jaeger (1985) hypothesized a close relationship among all these groups. Flower structure in sections *Anisantherum* and *Monodolichopus* differs greatly from that of sections *Androceras* and *Nycterium*, however. In both of the former sections flowers are actinomorphic, five-merous, and lack enantiostyly. The five anthers are equal or subequal, but the lowermost stamen in the flower is longer due to an elongated filament (Fig. 3E). These species are poorly known and their breeding systems and pollination biology has not been reported.

Lester et al. (1999) postulated a close relationship among all the heterantherous members of these two sections based on seed surface characters. They also noted similarities between seed characters of the isantherous species *Solanum arundo* and section *Monodolichopus* and between those of the isantherous *S. barbisetum* and section *Anisantherum*. However, this was contradicted by the opposite set of morphological resemblances in leaf shape, habit, and infructescence, with section *Monodolichopus* more similar to *S. barbisetum* and section *Anisandrum* more similar to *S. arundo* (Lester et al., 1999).

3. *Solanum* sect. *Nycterium*. The genus *Nycterium* Venten. was originally created to encompass several *Solanum* species with heterantherous flowers. Included here were the type species *N. cordifolium* Venten., now known as *S. vespertilio* Ait., and several species now removed to *Solanum* sect. *Androceras*. Demoted to sectional rank by Walpers (1844), the traditional circumscription of *Solanum* sect. *Nycterium* (Venten.) Walp. includes heterantherous taxa from both Mexico (*S. tridynamum* and *S. azureum*) and the Canary Islands (*S. lidii* and *S. vespertilio*; Whalen, 1984). Dunal (1852) also included the heterantherous African species *S. coagulans* (as *S. dubium*) in section *Nycterium* until it was removed to *Solanum* sect. *Monodolichopus* by Bitter (1923). Nee (1999), dealing mainly with the New World *Solanum* species, considered the heterantherous Mexican and Canary Island species to be closely related, but he included the American *S. elaeagnifolium* and *S. hindsianum*, both isantherous species or nearly so, in the same group. Some authors have included *S. pubescens* Willd. of India and *S. somalense* Franch. of East Africa in this section, although these species were excluded from it in Whalen's treatment (Whalen, 1984). The seed surface SEM studies of Lester et al. (1999) support a close relationship between *S. tridynamum*, *S. lidii* and *S. vespertilio*, but the seed surface characters of *S. pubescens* and *S. somalense* were more similar to species outside section *Nycterium* (see below). Whalen (1984) furthermore cast doubt on the monophyly of section *Nycterium* as circumscribed to include only the Mexican and Canary Islands species, pointing out differences in floral morphology between the two groups in spite of their shared heteranthery.

Indeed, floral morphology differs considerably between the Mexican and Canary Island heterantherous taxa. In the Macaronesian species *Solanum lidii* and *S. vespertilio*, the flowers are four- to five-merous with strongly zygomorphic corollas. Of the four or five anthers, three to four are relatively short and straight and one long and curved; all the filaments are nearly of equal lengths (Fig. 3G, H). The style is also long and curved and roughly the same length as the longest anther. Enantiostyly has been reported for *S. vespertilio* (G. Anderson, pers. commun.), but this character may be variable among individuals and populations. The plants are weakly andromonoecious and self-compatible. The fruits of the two species are fleshy berries not completely covered by accrescent fruiting calyces.

In contrast, plants of the Mexican species *Solanum tridynamum* bear two distinctly different types of flowers. Both types are five-merous and corollas are actinomorphic or weakly zygomorphic. In staminate flowers, the gynoecia are reduced and nonfunctional, with rudimentary styles. The two uppermost anthers are short and the three lowermost anthers are very long and curved; all filaments are roughly equal in length (Fig. 3K). In this strongly andromonoecious species, the basalmost is the only perfect flower on each inflorescence. The five anthers are equal and relatively short in these flowers and the curved style is long and exserted from the ring of stamens (Fig. 3J). The fruiting calyx is strongly accrescent and completely covers the fruit. The fruit type is unusual in *Solanum*: it is erect, dry, and circumsessile or irregularly dehiscent, forming a splash-cup or censer-type of fruit in which seeds are dispersed by wind or rain, as opposed to the fleshy animal-dispersed berries common in most solanums (Lester and Symon, 1989). Greenhouse crosses showed *S. tridynamum* to be self-compatible (L. Bohs, unpubl. data).

Other Heterantherous Solanums

In addition to the taxa described above, there are other examples of *Solanum* species with unequal stamens, but lacking the highly modified flowers seen in the focal groups. In most of these cases one stamen is longer than the other four, either due to an enlarged filament, an enlarged anther, or both. This morphology is seen in *Solanum* species in several unrelated sections and clades, such as sections *Aculeigerum* Seithe (*S. alternatopinnatum* Steud., *S. glaucescens* Zucc., *S. bicorne*, *S. wendlandii*; Fig. 2B), *Gonatotrichum* Bitter (*S. turneroides*), and the Dulcamaroid clade sensu Bohs (2005) (*S. ipomoeoides*, *S. pensile* Sendtn.; Knapp, 2001, 2002a). This morphology seems to be constant within a species. Two species of section *Geminata* (G. Don) Walp.

(*S. pseudoquina* A. St.-Hil., *S. reitzii* L. B. Smith & Downs) have flowers with two stamens longer than the other three due to both longer anthers and filaments (Knapp, 2002b). Many species of the genus *Lycianthes* also have one stamen longer than the rest (Dean, 2001, 2004).

One other curious floral modification occurs in at least one spiny *Solanum* species from New Caledonia, *S. vaccinioides*, placed by Whalen (1984) in the *S. dunalianum* group. The flowers are five-merous and actinomorphic, with five equal stamens. However, the style is deflected to one side and sticks out horizontally through the stamen cone (Fig. 2C). It is not known if the style is deflected to the same direction in all individuals. This seems to be a case of herkogamy rather than enatiostyly, which implies reciprocal positioning of the style and long anther. The situation in *S. vaccinioides* is referred to as “deflected style” here and in Table 1 and Fig. 4. Pollination or flower visitors have never been described for this rare species, so the function of the deflected style is a mystery.

Representatives of several of the species listed above were included in our analyses to examine the phylogenetic distribution of these modified floral features within *Solanum*.

MATERIALS AND METHODS

Molecular Methods

Sequences of the *trnT-F* region, including the *trnT-L* and *trnL-F* intergeneric spacer regions (Taberlet et al., 1991), and the granule-bound starch synthase (GBSSI or *waxy*) gene (van der Leij et al., 1991; Mason-Gamer et al., 1998) were obtained for 60 *Solanum* and three outgroup species (Table 2) using standard DNA extraction, PCR, and sequencing methods described elsewhere (Bohs, 2004; Levin et al., 2005, 2006). Species known to be bilaterally symmetric, enantiostylous, or heterantherous were sampled, with an emphasis on members of the focal groups identified in Lester et al. (1999). When available, species thought to be closely related to the focal groups based on previous analyses of DNA sequence data and morphology were also sampled. *Capsicum baccatum*, *Lycianthes heteroclita*, and *Jaltomata procumbens* served as outgroups.

Sequence Alignment and Analysis

Sequence alignment for *trnT-F* and *waxy* exons was straightforward and was performed manually using Se-Al (Rambaut, 1996). Although *waxy* intron sequence alignment was more challenging, clearly recognizable sequence motifs that facilitated alignment were identified across all taxa. Similarly, most *trnT-L* spacer and *trnL* intron regions could be aligned with confidence. However, numerous sequence duplications have occurred in the *trnL-F* spacer between the 3' *trnL* and *trnF* exons within the species surveyed, and alignment in this region was highly ambiguous. The *trnT-L* spacer, the *trnL* 5' exon, the *trnL* intron, the *trnL* 3' exon and approximately 320 nucleotides of sequence data following in the *trnL-F* spacer were included in analyses; downstream sequences were excluded because they could not be aligned reliably.

Parsimony analyses were performed on the combined data set using PAUP*4.0b10 (Swofford, 2002). All characters were weighted equally in analyses implementing TBR branch swapping with 1000 heuristic random addition replicates. Bootstrapping (Felsenstein, 1985) was used to evaluate branch support with 1000 random addition replicates and TBR branch swapping.

RESULTS

The aligned length of the combined data after excluding the 3' unalignable sequence region of *trnT-F* was 4016 bases, of which 567 were parsimony informative. The data sets were complete for all taxa except that the *S. mammosum* sequence for *trnT-F* was missing the first 16 bases. The 648 most parsimonious trees had a length of 2011 steps, CI = 0.750, RI = 0.808.

The strict consensus tree was highly resolved, with a number of well-supported nodes (Fig. 4). The overall topology and specific relationships were very similar to those obtained in previous molecular analyses of *Solanum* such as Bohs (2005), Levin et al. (2006), and Weese and Bohs (2007). *Solanum* is well supported as a monophyletic group, including species from the former segregate genera *Normania*, *Triguera*, *Cyphomandra*, and *Lycopersicon*. *Solanum thelopodium* emerges as sister to the remainder of *Solanum* species. The non-spiny and spiny *Solanum* species each do not form monophyletic groups, but most of the spiny *Solanum* species (subgenus *Leptostemonum* sensu stricto of Levin et al., 2006) form a strongly supported clade (97% bootstrap). *Solanum wendlandii* and *S. bicorne*, taxa with spines but without stellate hairs, are separate from the *Leptostemonum* sensu stricto clade and are nested within a group of non-spiny solanums. The Old World spiny members of *Solanum* sampled here form a strongly supported clade (100% bootstrap), in agreement with the Old World clade of Levin et al. (2006). The same major clades named in Bohs (2005), Levin et al. (2006), and Weese and Bohs (2007) are seen in the tree from the present data.

The phylogenetic positions of the heterantherous taxa described above are identified in Fig. 4. Heteranthery, zygomorphy, and enantiostyly can be seen to have evolved multiple times in various *Solanum* lineages. The relationships of *Solanum* taxa with modified flowers are discussed in detail below.

DISCUSSION

Focal Groups in the Non-Spiny Solanums

1. *Solanum thelopodium* Species Group. All molecular datasets that have sampled *S. thelopodium*, including the one presented here, segregate this species from the rest of *Solanum* (Bohs, 2005; Weese and Bohs, 2007). Although Bitter thought that the morphological distinctions of *S. thelopodium* were sufficient to recognize it as a distinct genus (Knapp, 2000), it is well-supported as either sister to the rest of *Solanum* (Bohs, 2005 and this study) or is one branch of a basal polytomy in *Solanum*, with the remainder of the species divided into two large clades (Weese and Bohs, 2007). In any case, the *S. thelopodium* species group represents an independent evolution of heteranthery in the genus. It is highly desirable to know more about pollination and reproductive biology in this group to understand the function of its unique stamen arrangement.

2. Normania Clade. Morphological and molecular data establish that *Solanum herculeum*, *S. trisectum* and *S. nava* are closely related. Francisco-Ortega et al. (1993) and Lester et al. (1999) argued for their close relationship on the basis of seed surface structure and pollen grain sculpturing, as well as similarities in overall flower and fruit characters. However, they considered these taxa to belong to the segregate genera *Normania* and *Triguera* and believed them to be distinct from *Solanum*. The molecular data of Bohs and Olmstead (2001), Bohs (2005), Weese and Bohs (2007) and those presented here establish that *S. trisectum* (former genus *Normania*) and *S. herculeum* (former genus *Triguera*) are nested within *Solanum*. Although the position of this clade (the *Normania* clade of Bohs, 2005) was unresolved in the studies of Bohs and Olmstead (2001) and Bohs (2005), the data of Weese and Bohs (2007) and those presented here suggest a surprising sister group relationship with members of section *Archaesolanum* (represented here by *S. aviculare* and *S. laciniatum*), a group of Australian species with actinomorphic, isantherous flowers and an aneuploid chromosome number based on n=23 (Fig. 2F).

Studies of pollination biology are sorely needed in species of the *Normania* clade to investigate the functional role of their highly specialized flowers. It is clear that the appearance of heteranthery in *S. nava* and *S. trisectum* represents an independent evolution of this trait within *Solanum*. Furthermore, it is curious that the other pair of endemic Macaronesian *Solanum* species, *S. lidii* and *S. vespertilio*, are also heterantherous. However, these latter species belong to *Solanum* subg. *Leptostemonum*, are completely unrelated to *S. nava* and *S. trisectum*, and exhibit a different combination of long and short stamens (Table 1 and text below). Why heteranthery appears to have evolved twice independently in the endemic Macaronesian solanums is an intriguing question.

Focal Groups in the Spiny Solanums (*Solanum* subg. *Leptostemonum*)

1. *Solanum* sect. *Androceras*. The molecular phylogenetic studies reported here and in Levin et al. (2006) refute a close relationship between sections *Androceras* and *Nycterium*, regardless of how the latter section is circumscribed. Instead, they suggest that section *Androceras* is most closely related to section *Crinitum* Child, also known as the *S. crinitum* species group (Whalen, 1984), represented here by *S. crinitum* and *S. lycocarpum*. *Solanum mitlense*, an unplaced Mexican species in Whalen (1984), is also part of the Crinitum clade. Members of the *S. crinitum* group produce some of the largest flowers and fruits in the genus, and the flowers are remarkable in that they change color with age, opening dark purple and fading to almost white over a several-day period. They are actinomorphic, isantherous, and lack enantiostyly (Fig. 3C). The plants are strongly andromonoecious. Baksh and Iqbal (1978) report that *S. macranthum* Dun. of section *Crinitum* is self-compatible, but greenhouse crossing studies on two other species in the section indicate that they may be self-incompatible (L. Bohs, unpubl. data). Members of section *Crinitum* are robust shrubs or large forest trees native to mesic forests or seasonally dry cerrado vegetation of South America. Thus, sections *Androceras* and *Crinitum* are highly divergent in terms of morphology, ecology, and biogeography, and the apparent close relationship among them is quite surprising.

Various authors, most recently Lester et al. (1999), postulated a close relationship between sections *Androceras* and *Cryptocarpum*. *Solanum sisymbriifolium*, a member of section *Cryptocarpum*, emerges as sister to the Androceras/Crinitum clade in the analysis reported here, but this relationship is weakly supported (63% bootstrap). This result was also obtained in the Bayesian but not the parsimony analysis of Levin et al. (2006); in the latter, *S. sisymbriifolium* occupied an unresolved position within the spiny *Solanum* clade. *Solanum sisymbriifolium* is native to South America and has actinomorphic isantherous flowers without enantiostyly (Fig. 3D). The plants are weakly andromonoecious and self-compatible (Whalen and Anderson, 1981). However, despite these differences between *S. sisymbriifolium* and section *Androceras*, the two groups are similar in leaf lobing, calyx morphology, and seed surface sculpturing (Whalen, 1979; Lester et al., 1999). *Solanum sisymbriifolium* should be investigated further as a distant relative of section *Androceras* and its phylogenetic position should be resolved with further sequence data and more extensive sampling of members of section *Cryptocarpum*.

2. Sections *Anisantherum* and *Monodolichopus*. Unfortunately, only one heterantherous species of these two sections (*Solanum coagulans* from section *Monodolichopus*) could be included in the molecular analyses due to lack of DNA material. Likewise, the possible isantherous relatives of these species, *S. barbisetum* and *S. arundo*, were not available. (The "*S. arundo*" sample analyzed in Levin et al. (2006) was misidentified and its determination is still uncertain.) *Solanum coagulans* emerged with moderate support (69% bootstrap) on a clade including African and Macaronesian species with both isantherous and heterantherous flowers (see Fig. 3I for a representative isantherous species). *Solanum aculeastrum*, an actinomorphic isantherous species (Fig. 3F), was sister to *S. coagulans* in the analysis of Levin et al. (2006; *S. coagulans* was identified as *S. thruppii* in their paper), but that position was not supported here. However, relationships are poorly resolved in this part of the tree that includes the Old World spiny solanums. Also, more extensive taxonomic sampling is needed to confirm sister group relationships. It is highly likely, however, that *S. coagulans* illustrates yet another instance of the independent evolution of heteranthery in *Solanum*. Whether heteranthery is also independently derived in section *Anisantherum* remains to be tested. The molecular phylogenies also refute a connection between sections *Monodolichopus*, *Androceras*, and *Nycterium*, as suggested by Jaeger (1985). The wide biogeographic disjunctions seen in these groups also argue against this view.

3. *Solanum* sect. *Nycterium*. All molecular phylogenetic studies to date that have sampled these species (Olmstead and Palmer, 1997; Levin et al., 2006; Weese and Bohs, 2007), including this one, conclusively show that *S. tridynamum* is not closely related to *S. vespertilio* and *S. lidii*, in concordance with their morphological and biogeographical differences. *Solanum vespertilio* and *S. lidii* are sister taxa and occur on a clade with a number of African spiny *Solanum* species from subgenus *Leptostemonum*. Although the Canary Island taxa are part of a well-supported clade including *S. aethiopicum*, *S. anguivi*, *S. capense*, *S. cyaneopurpureum*, *S. hastifolium* and *S. tomentosum*, in Fig. 4 and in the analyses of Levin et al. (2006), sampling is not sufficiently dense among the African spiny solanums to confidently establish the sister taxa to *S. vespertilio* and *S. lidii*. However, the African spiny solanums sampled in this clade all have actinomorphic, isantherous, five-merous flowers and lack enantiostyly (Fig. 3I). The breeding systems and degree of andromonoecy are not well known in the African species of *Solanum* and should be investigated in the future for comparison with the Canary Islands taxa.

Solanum tridynamum is strongly supported as sister to the Mexican *S. hindsianum* and in turn these two are sister to *S. elaeagnifolium*, a species with a disjunct distribution in North and South America. *Solanum hindsianum* and *S. elaeagnifolium* are andromonoecious, with actinomorphic, five-merous, isantherous flowers (or sometimes with subequal anthers in *S. hindsianum*; Fig. 3L). *Solanum hindsianum* has the splash-cup or censer fruit type found in *S. tridynamum*, but *S. elaeagnifolium* has a dryish yellow berry. The breeding system of *S. elaeagnifolium* has not been reported, but greenhouse crossing studies indicate that *S. hindsianum* is self-incompatible (L. Bohs, unpubl. data).

If the sister relationship between *S. tridynamum* and *S. hindsianum* is confirmed upon further study, this species pair may be a useful system for the evolution of derived floral morphologies in *Solanum*. The flowers of the two species are radically different in appearance and present opportunities to examine the genetic control of heteranthery and stylar heteromorphism in sister taxa. Furthermore, since *S. hindsianum* appears to be self-incompatible, the question is raised of whether the extreme floral forms seen in *S. tridynamum* evolved in response to the loss of self-incompatibility in ancestral populations of this species.

Other Heterantherous Solanums

Aside from the focal groups, several *Solanum* species were sampled that have a single long stamen in the flower; these are indicated on Fig. 4. This morphology has evolved multiple times in *Solanum*: at least three times in the sampled species and at least once more in the Geminata clade, represented here by *S. pseudocapsicum*, *S. arboreum*, and *S. argentinum* but without sampling any taxa in the Geminata group with unequal stamens.

Functional Aspects of Heteranthery in *Solanum* Pollination

The biological significance of heteranthery, floral zygomorphy and enantiostyly in *Solanum* pollination is still imperfectly known. The most thoroughly studied species from a functional point of view is *S. rostratum* (section *Androceras*), which is zygomorphic, markedly heterantherous, and has monomorphic enantiostyly, where the style is deflected to the side of the flower opposite the long anther. Successive flowers in an inflorescence are alternatingly right- and left-handed. Bowers (1975) studied the pollination biology of this species and found that while bees grasped and buzzed the four short anthers in the flowers, pollen from the single long anther was deposited under the bee’s wing on one side where it could not easily be groomed off. The deflected style picks up pollen deposited on the bee by the long anther. Depending on the bee visitation patterns and the number of open flowers on a plant at any one time, pollen can be moved between flowers of a single plant (geitnogamy) or between separate plants (xenogamy), but self-pollination is greatly reduced in this syndrome.

Jesson and Barrett (2002, 2005) performed the most thorough experimental analysis of the functional aspects of enantiostyly, again utilizing *S. rostratum* as the model solanaceous species. Using arrays of experimentally manipulated flowers to represent different combinations of stylar deflection, they convincingly demonstrated that enantiostyly functions to reduce geitnogamy and promote outcrossing, as compared to populations that are heterantherous but not enantiostylous (Jesson and Barrett, 2005). They propose that the steps involved in the evolution of enantiostyly begin with a floral morphology where the anthers and stigma are in close proximity, followed by a deflection of the style, and finally culminating in a reciprocal deflection of the style and stamen (Jesson and Barrett, 2005). This prediction might be tested in *Solanum* using *S. lidii* and *S. vespertilio*. Flowers of these two species are highly heterandrous, with the style aligned with the long anther in *S. lidii* and in at least some individuals of *S. vespertilio* (L. Bohs, pers. obs.). However, enantiostyly has been reported for some populations of *S. vespertilio* (Anderson et al., 2006; G. Anderson, pers. commun.). Detailed population-level studies of these two species may allow the testing of the hypotheses of Jesson and Barrett (2005) regarding the evolution of enantiostyly.

Functional explanations for the evolution of heteranthery are less clear. Examining floral features in the monocots, Graham and Barrett (1995) found repeated associations between heteranthery, zygomorphy, outward floral orientation, and enantiostyly. They tested the correlation of these traits in the context of a monocot phylogeny and found significant associations between enantiostyly, heteranthery, and the loss of floral nectaries (Jesson and Barrett, 2003). Their results suggest that the loss of nectaries generally precedes the evolution of heteranthery. However, the sequence of evolution of heteranthery and enantiostyly was less clear. Furthermore, they found only a weak association of enantiostyly with zygomorphy. The latter condition is hypothesized to function in consistent positioning of the pollinator (Faegri and van der Pijl, 1979), which may or may not promote outcrossing. Heteranthery also may be responsible for precision in pollinator positioning (Jesson and Barrett, 2003). Alternatively, heteranthery may promote a division of labor between stamens within a single flower (i.e., between “fertilizing” and “feeding” anthers; Lloyd, 2000). Bowers (1975) examined the relative contributions of pollen from the long and short anthers of *S. rostratum* using fluorescent powders and found that dye placed on the long (“fertilizing”) anther was more likely to be deposited on the stigma than dye placed on the short (“feeding”) anthers. Other heterantherous species of *Solanum* should be tested to determine if pollen grains from all anthers in a flower are equally viable and whether the pollen from various-sized anthers makes differential contributions to plant fitness. In this context, it would be interesting to compare the various morphological versions of heteranthery seen in *Solanum* to deduce their functional significance and possible evolutionary antecedents.

The functional significance of the flower form with a single long stamen in pollination has not been investigated, but presumably it deposits pollen on a part of the bee where it is not easily groomed off. How this pollination system functions without enantiostyly and whether it promotes outcrossing, however, is unknown. Another possibility is that the long stamen merely functions as a landing platform for visiting insects, which then work the remaining anthers for pollen. The long stamen could also be responsible for more precise pollinator positioning, as has been suggested above for other heterantherous *Solanum* species. This does not explain why most *Solanum* species lack the long stamen, however.

Dean (2001) investigated unequal stamens in *Lycianthes* and found several patterns depending on the species investigated. In some species, one filament was much longer than the rest when anthesis occurred, and this length difference was maintained over the life of the flower. In other species, the filaments were nearly equal at anthesis, but one elongated more than the others by the third day of opening. Dean suggests that post-anthesis filament elongation is associated with asynchronous anther dehiscence and correlated with dichogamy (temporal separation of male and female functions in the flower). The longer, lowermost stamen was often (but not always) the last to dehisce, and this occurred when the longest stamen had roughly reached the level of the stigma. Dean hypothesizes that the different-sized stamens perform different functions in pollination, much as Bowers (1975) found in *S. rostratum*. The shorter stamens function as "feeding anthers" in attracting floral visitors, whereas the long stamen primarily functions in pollination. Delaying dehiscence of the long stamen may allow the chance for cross-pollination to occur. Thus, temporal separation of reproductive events in the flower may be important in species with heteranthery but without enantiostyly.

The experimental studies of Jesson and Barrett (2005) suggest that significant inbreeding depression occurs in the self-compatible *S. rostratum*, despite its highly modified floral morphology. Factors such as self-incompatibility (SI) and derived sexual systems such as andromonoecy and dioecy affect the degree of outcrossing in a particular species or population, in addition to floral modifications such as enantiostyly. Outcrossing insurance by mechanical means such as heteranthery and enantiostyly may be much less of an issue if a species is self-incompatible. Thus, a species breeding and sexual system should also be known when evaluating the efficacy of heteranthery and enantiostyly as outcrossing mechanisms.

Future Directions

This study raises as many questions as it answers and suggests many avenues for future investigation of floral evolution in *Solanum*. Even the most basic information on breeding systems, sex expression and floral morphology is lacking for many *Solanum* species, so few generalizations can be made about evolutionary patterns at this time. More extensive sampling of *Solanum* species may help pinpoint the sister taxa to species with highly derived flowers; better sampling of species from sections *Anisantherum* and *Monodolichopus* and their putative relatives is especially desirable. Detailed studies of pollination in heterantherous vs. isantherous taxa are badly needed to determine the functional aspects of derived floral morphologies in the genus and to distinguish between spatial and temporal mechanisms promoting outcrossing, as described above.

The genetic and developmental basis of floral differences between *Solanum* species is virtually unknown, although *Solanum* includes several model organisms and genetic data is accumulating rapidly as a result of the tomato genome sequencing effort. Genes controlling floral traits such as zygomorphy, heteranthery, and enantiostyly have not been studied in *Solanum*, although genes controlling floral symmetry have been investigated in other plant families. Across angiosperms, bilaterally symmetric taxa evolved from radially symmetric ancestors independently numerous times (Stebbins, 1950, 1974; Crepet et al., 1991; Donoghue et al., 1998). In the model of organism *Antirrhinum majus* (Plantaginaceae) bilateral symmetry is established by the adaxial expression of the floral development gene *CYCLOIDEA* (*CYC*) and its paralog *DICHOTOMA* (*DICH*), and homologues of *CYC* and *DICH* also are involved in establishing bilateral symmetry in taxonomically diverse families. Changes in symmetry in other studied angiosperm groups are associated with adaptive evolution at the amino acid sequence level in Fabaceae (Ree et al., 2004), with changes in the timing or location of expression in Veronicaceae (Hileman et al., 2003), and with duplication and subfunctionalization in the Dipsicales (Howarth and Donoghue, 2005) of the regulatory genes *CYC* and *DICH*. Although neither CYC nor DICH have been isolated in the Solanaceae to date (Reeves and Olmstead, 2003), these genes may be identified through the tomato genome sequencing project (http://www.sgn.cornell.edu/solanaceae-project/), which would allow molecular evolutionary genetic analysis of the independent evolution of bilateral symmetry in *Solanum*. Although the specific genes responsible for the development of heteranthery and enantiostyly are not yet known in *Solanum* or in other flowering plants, the Solanaceae, and *Solanum* in particular, could be a model system for the identification and characterization of the genetic basis of derived floral morphologies as well as for the investigation of the evolutionary role of these derived floral forms.

ACKNOWLEDGEMENTS

The authors thank K. Gineva, R. Levin, A. Moore, K. Sebastian and K. Watson for laboratory assistance, and R.G. Olmstead, S. Knapp, M. Nee, D. Spooner, W.G. D'Arcy,

T. Mione, A. Child, J. Francisco-Ortega, R.N. Lester, M. Welman, the Botanic Garden at the University of Nijmegen, The Netherlands and the Missouri Botanical Garden for help in obtaining Solanaceae materials. The authors also appreciate the helpful suggestions of two anonymous reviewers for improvement of the manuscript. This work was supported by NSF grants DEB-9207359, DEB-9996199, DEB-0235339, and DEB-0316614 to LB.

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Tables

Table 1. Summary of floral and reproductive characters among the *Solanum* species mentioned in the text and investigated using molecular data. Species are arranged by clades following Bohs (2005) and Weese and Bohs (2007); within the clades, sectional affiliations are given. Key to symbols: Y = yes, N = no, - = unknown; herm = hermaphroditic flowers, stam = staminate flowers. Range: N Am = North America, C Am = Central America, S Am = South America, Eu = Europe, Mac = Macaronesia, Af = Africa, S Pac = South Pacific. Symmetry: A= actinomorphic, Z = zygomorphic. Enantiostyly: DS = deflected style. Sexual system: H = all flowers hermaphroditic, Aw = weakly andromonoecious, As = strongly andromonoecious, D = dioecious. Breeding system: SC = self-compatible, SI = self-incompatible, N.A. = not applicable.

Species	Range	Merosity	Corolla symmetry	Stamen morphology	Enantiostyly	Sexual system	Breeding system
Thelopodium Clade							
Solanum thelopodium species group							
S. thelopodium Sendtn.	S Am	5	A	1 long, 2 medium, 2 short	N	H	-
Archaesolanum Clade							
Solanum sect. *Archaesolanum* Marzell							
S. aviculare G. Forst.	S Pac	5	A	5 equal	N	H	SC
S. laciniatum Aiton	S Pac	5	A	5 equal	N	H	SC
Normania Clade							
Solanum sect. *Normania* (Lowe) Bitter							
S. herculeum Bohs	Eu, Af	5	± Z	5 ± equal	N	H?	-
S. trisectum Dunal	Mac	5	± Z	2 long, 2 medium, 1 short	N	H	SC
Dulcamaroid Clade							
Solanum sect. *Dulcamara* (Moench) Dumort							
S. calileguae Cabr.	S Am	5	A	5 equal	N	H	-
S. ipomoeoides Sendtn.	S Am	5	A	5 ± equal anthers, 1 long filament	N	H	-
Wendlandii/Allophyllum Clade							
Solanum sect. *Aculeigerum* Seithe							
S. bicorne Dunal	C Am	5	A	5 ± equal anthers, 1 long filament	N	Aw	-

S. wendlandii Hook. f.	C Am	5	A	5 ± equal anthers, 1 long filament	N	D?	N.A.
Brevantherum Clade							
Solanum sect. *Gonatotrichum* Bitter							
S. adscendens Sendtn.	S Am	5	A	5 equal	N	H	SC
S. deflexum Greenm.	N & C Am	5	A	5 equal	N	H	SC
S. turneroides Chodat	S Am	5	A	5 ± equal anthers, 1 long filament	N	H	-
Leptostemonum Clade							
Solanum sect. *Androceras* (Nutt.) Marzell							
S. citrullifolium A. Braun	N Am	5	Z	1 long, 4 short	Y	Aw	SC
S. rostratum Dunal	N Am	5	Z	1 long, 4 short	Y	Aw	SC
Solanum sect. *Crinitum* Child							
S. crinitum Lam.	S Am	5	A	5 equal	N	H to As	-
S. lycocarpum A. St.-Hil.	S Am	5	A	5 equal	N	As	-
Solanum section *Cryptocarpum* Dunal							
S. sisymbriifolium Lam.	S Am	5	A	5 equal	N	H or Aw	SC
Solanum sect. *Leprophora* Dunal							
S. elaeagnifolium Cav.	N Am, S Am	5	A	5 equal	N	H	-
Solanum sect. *Melongena* (Mill.) Dunal							
S. aculeastrum Dunal	Af	5	A	5 equal	N	As	SC
Solanum sect. *Monodolichopus* Bitter							
S. coagulans Forssk.	Af	5	A	4 ± equal anthers, 1 longer anther on 1 long filament	N	H?	-

Solanum sect. *Nycterium* (Venten.) Walp.							
S. lidii Sunding	Mac	4	Z	1 long, 3-4 short	N?	Aw	SC
S. vespertilio Aiton	Mac	4	Z	1 long, 3 short	Y?	Aw	SC
S. tridynamum Dunal	N Am	5	± A	stam: 3 long, 2 short herm: 5 ± equal	N	As	SC
Solanum sect. *Oliganthes* (Dunal) Bitter							
S. aethiopicum L.	Af	5	A	5 equal	N	H	SC
S. anguivi Lam.	Af	5	A	5 equal	N	H	SC
S. capense L.	Af	5	A	5 equal	N	H	-
S. cyaneopurpureum De Wild.	Af	5	A	5 equal	N	H	SC
S. hastifolium Dunal	Af	5	A	5 equal	N	H	-
S. tomentosum L.	Af	5	A	5 equal	N	H	SC
Solanum dunalianum species group							
S. vaccinioides Schltr.	S Pac	5	A	5 equal	DS	H	-
Section not assigned							
Solanum hindsianum Benth.	N Am	5	A	5 ± equal	N	Aw	SI
S. mitlense Dunal	N Am	5	A	5 equal	N	-	-

Table 2. Locality, voucher, and GenBank information for species sequenced. BIRM – seed accession number of the Solanaceae collection at the University of Birmingham, UK. NIJ – seed accession number of the Solanaceae collection at Radboud University, Nijmegen, The Netherlands. Herbarium acronyms where specimens are deposited follow voucher collection numbers.

			GenBank Accession Numbers	
Taxon	Locality	Voucher	trnT-F	waxy
Solanum abutiloides (Griseb.) Bitter & Lillo	BIRM S.0655	*Olmstead S-73* (WTU)	AY266236	AY562948
S. aculeastrum Dunal	NIJ 924750119	*Bohs 3251* (UT)	DQ812102	AY996376
S. adscendens Sendtn.	Bolivia	*Bohs & Nee 2738* (UT)	DQ180421	DQ169013
S. aethiopicum L.	BIRM S.0344	*Olmstead S-74* (WTU)	DQ180394	AY996378
S. allophyllum (Miers) Standl.	Panama	*Bohs 2339* (UT)	DQ180422	AY996379
S. anguivi Lam.	NIJ 974750005	*Cipollini 164* (UT)	DQ812103	AY996380
S. appendiculatum Dunal	Mexico	*Anderson 1401* (CONN)	DQ180461	DQ169018
S. arboreum Dunal	Costa Rica	*Bohs 2521* (UT)	DQ180424	AY996381
S. argentinum Bitter & Lillo	Argentina	*Bohs 2539* (UT)	DQ180425	AY996382
S. aviculare Forst. f.	BIRM S.0809	no voucher	AY562952	AY559238
S. betaceum Cav.	Bolivia	*Bohs 2468* (UT)	DQ180426	AY996387
S. bicorne Dunal	Mexico	*Iltis et al. 29694* (WIS)	DQ812104	AY996460
S. calileguae Cabrera	Argentina	*Nee & Bohs 50809* (NY)	EF068252	DQ169022
S. campanulatum R. Br.	BIRM S.0387	*Olmstead S-78* (WTU)	DQ180395	AY996388
S. candidum Lindl.	Costa Rica	*Bohs 2898* (UT)	AY266237	AY562953
S. capense L.	NIJ 904750116	*Bohs 2905* (UT)	DQ392958	AY996391
S. carolinense L.	BIRM S.1816	*Olmstead S-77* (WTU)	DQ180476	AY996392
S. chenopodinum F. Muell.	BIRM S.0813	no voucher	DQ180396	AY996393
S. citrullifolium A. Braun	BIRM S.0127	*Olmstead S-79* (WTU)	DQ180477	AY996395
S. coagulans Forssk.	NIJ A34750435	*Bohs 3274* (UT)	DQ812105	AY996398
S. cordovense Sessé & Moc	Costa Rica	*Bohs 2693* (UT)	DQ180480	AY996401
S. crinitipes Dunal	Colombia	*Olmstead S-81* (WTU)	DQ180481	AY996402
S. crinitum Lam.	NIJ 924750049	*Bohs 2850* (UT)	DQ180482	AY996403

S. cyaneopurpureum De Wild.	NIJ 874750010	*Bohs 3164* (UT)	DQ392959	AY996405
S. deflexum Greenm.	Costa Rica	*Bohs 2715* (UT)	DQ180427	DQ169025
S. dulcamara L.	Cult. Michigan	no voucher	AY266231	AY996410
S. elaeagnifolium Cav.	Paraguay	*Bohs 3204* (UT; trnT-F) *Bohs 3199* (UT; waxy)	DQ180399	AY996412
S. hastifolium Dunal	NIJ 944750142	*Bohs 2906* (UT)	DQ812106	AY996420
S. herculeum Bohs	Morocco	*Jury 13742* (RNG)	DQ180466	DQ169031
S. hindsianum Benth.	Mexico	*Bohs 2975* (UT)	DQ180402	AY996424
S. ipomoeoides Chodat & Hassl.	Bolivia	*Bohs & Nee 2766* (UT)	DQ180448	DQ169033
S. jamaicense Mill.	BIRM S.1209	*Olmstead S-85* (WTU)	DQ180485	AY562956
S. laciniatum Ait.	New Zealand	*Bohs 2528* (UT)	DQ180467	AY996431
S. lidii Sunding	NIJ 934750022	*Bohs 2903* (UT)	DQ180403	AY996434
S. luteoalbum Pers.	BIRM S.0042	*Bohs 2337* (UT)	DQ180433	AY562957
S. lycocarpum A. St.-Hil.	Paraguay	*Bohs 3212* (UT)	DQ812107	AY996435
S. lycopersicum L.	USA (cultivated)	no voucher	DQ180450	DQ169036
S. macrocarpon L.	BIRM S.0133	*Olmstead S-88* (WTU)	DQ180404	AY996436
S. mammosum L.	BIRM S.0983	*Olmstead S-89* (WTU)	AY266232	AY562958
S. melongena L.	BIRM S.0657	*Olmstead S-91* (WTU)	DQ180406	AY562959
S. *mitlense* Dunal	Mexico	*Whalen & Velasco 825* (BH)	DQ812108	AY996442
S. myoxotrichum Baker	Madagascar	*Bohs 2981* (UT)	DQ392960	AY996445
S. nitidum Ruiz & Pav.	Bolivia	*Nee 31944* (NY)	DQ180451	DQ169039
S. pancheri Guillaumin	New Caledonia	*McKee 41366* (AD)	DQ812109	AY996450
S. physalifolium Rusby var. *nitidibaccatum* (Bitter) Edmonds	USA	*Bohs 2467* (UT)	EF068253	DQ169041
S. pseudocapsicum L.	BIRM S.0870	no voucher	DQ180436	AY562963
S. ptychanthum Dunal	USA	*Olmstead S-94* (WTU)	DQ180454	AY996457
S. pyracanthos Lam.	USA (cultivated)	*Olmstead S-95* (WTU)	DQ180408	AY996459
S. rostratum Dunal	USA	no voucher	DQ180489	AY996463
S. sandwicense Hook. & Arn.	Hawaii	*Bohs 2992* (UT)	DQ180409	AY996464
S. sisymbriifolium Lam.	Argentina	*Bohs 2533* (UT)	AY266235	AY562967

S. thelopodium Sendtn.	Bolivia	*Nee & Bohs 50858* (NY)	DQ180470	AY996471
S. tomentosum L.	NIJ 894750127	*Bohs 3107* (UT)	DQ392961	AY996473
S. torvum Sw.	BIRM S.0839	*Olmstead S-101* (WTU)	AY266246	AY562972
S. tridynamum Dunal	BIRM S.1831	*Olmstead S-102* (WTU)	DQ180412	AY996474
S. trisectum Dunal	France	*Bohs 2718* (UT)	DQ180471	AY996475
S. turneroides Chodat	Bolivia	*Nee et al. 51716* (NY)	DQ180439	DQ169051
S. vaccinioides Schltr.	New Caledonia	*Bohs 3608* (UT)	DQ812110	DQ812111
S. vespertilio Aiton	BIRM S.2091	*Olmstead S-103* (WTU)	DQ180413	AY996476
S. wendlandii Hook. f.	BIRM S.0488	no voucher	DQ180440	AY562974
Capsicum baccatum L. var. *pendulum* (Willd.) Eshbaugh	USA (cultivated)	*Eshbaugh 1584* (MU)	DQ180415	DQ169007
Jaltomata procumbens (Cav.) J. L. Gentry	Mexico	Davis 1189A	DQ180419	AY996374
Lycianthes heteroclita (Sendtn.) Bitter	Costa Rica	*Bohs 2376* (UT)	DQ180414	DQ169010

Figures

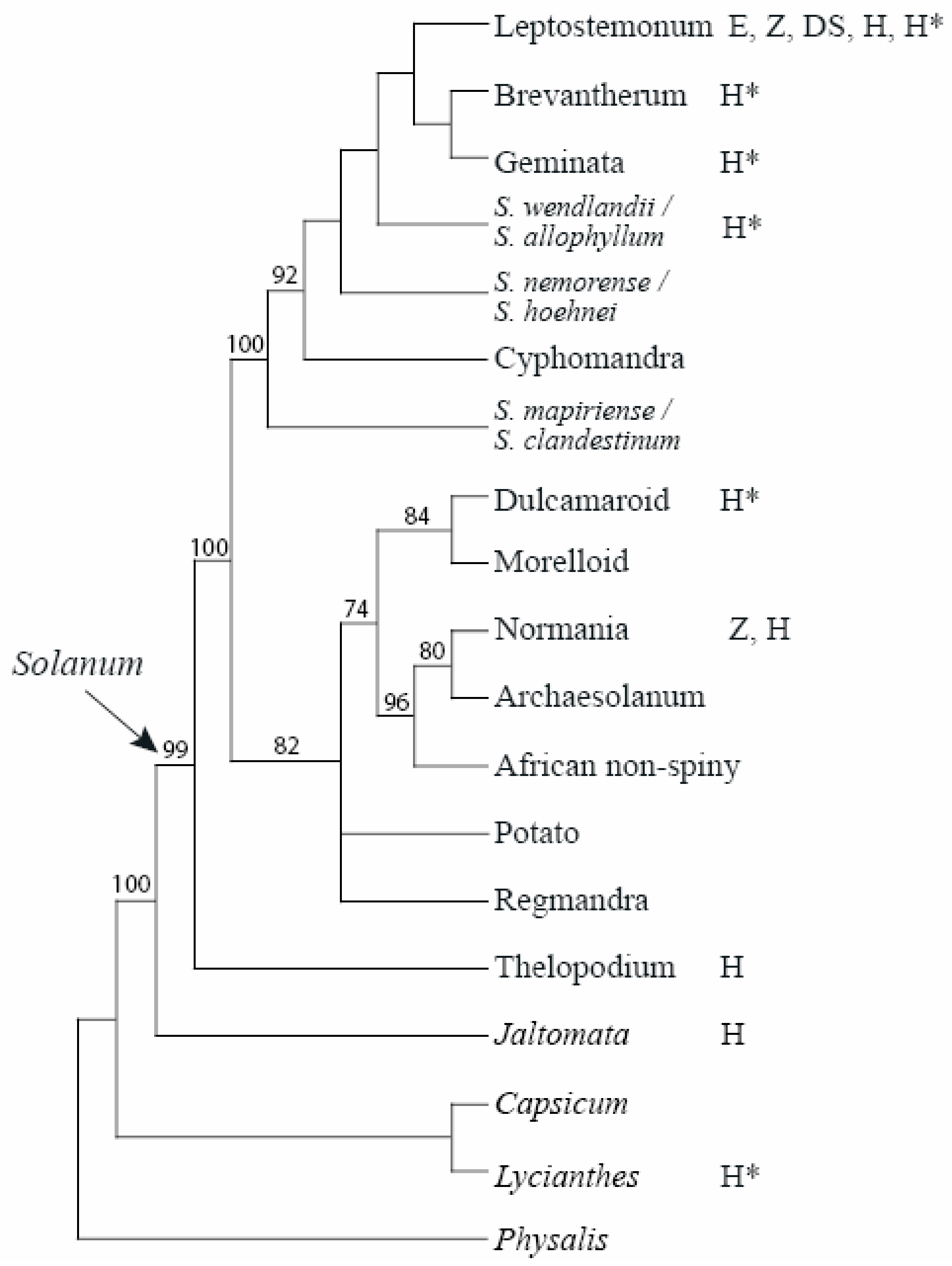

Fig. 1. Summary of relationships among major clades of *Solanum* and selected outgroup genera based on Weese and Bohs (2007) with occurrence of derived floral morphologies mapped onto the relevant clades. Mapped floral features occur in some, but not necessarily all, members of a clade. Clade names follow Bohs (2005) and Weese and Bohs (2007). Key to symbols: E = enantiostyly, Z = zygomorphy, DS = deflected style, H = heteranthery based on highly unequal stamens (corresponds to focal groups mentioned in the text), H* = flowers with one stamen longer than the rest, largely due to one longer filament in the flower. Bootstrap values over 50% are given above branches.

Fig. 2. Representative flowers of *Solanum* species mentioned in the text. A. *S. thelopodium*. B. *B. wendlandii*. C. *S. vaccinioides*. D. *S. trisectum*. E. *S. herculeum* (photo by S. Knapp). F. *S. aviculare*.

Fig. 3. Representative flowers of *Solanum* species mentioned in the text. A. *S. rostratum*. B. *S. citrullifolium*. C. *S. crinitum*. D. *S. sisymbriifolium*. E. *S. coagulans*. F. *S. aculeastrum*. G. *S. lidii*. H. *S. vespertilio*. I. *S. tomentosum*. J. *S. tridynamum*, perfect flower. K. *S. tridynamum*; staminate flower. L. *S. hindsianum*.

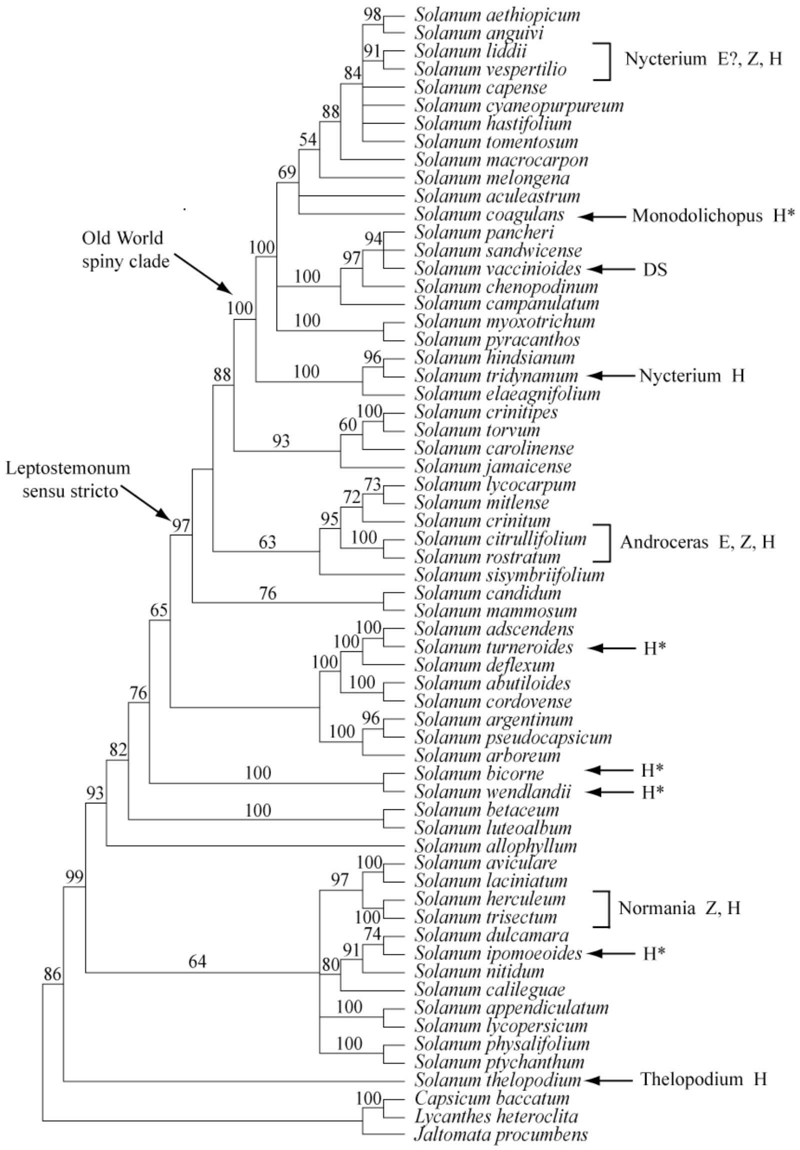

Fig. 4. Strict consensus tree from the phylogenetic analysis. Bootstrap values given on the branches. Symbols for floral features as in Fig. 1.

Fig. 4. Strict consensus tree from the phylogenetic analysis. Bootstrap values given on the branches. Symbols for floral features as in Fig. 1.