CHAPTER FOUR

DIFFERENT FATES OF ISLAND BROOMS: CONTRASTING EVOLUTION IN *ADENOCARPUS*, *GENISTA* AND *TELINE* (GENISTEAE, LEGUMINOSAE) IN THE CANARY ISLANDS AND MADEIRA

[This paper has been submitted to the *Botanical Journal of the Linnean Society*]

Abstract

Analysis of sequence data from the internal transcribed spacers (ITS) and 5.8S region of nuclear ribosomal DNA show that Canarian and Madeiran brooms (Genisteae) of the genera *Teline*, *Adenocarpus* and *Genista* are related to Mediterranean species and not to species from adjacent parts of Morocco. Each separate colonization of the islands has resulted in contrasting patterns of adaptation and radiation. The genus *Teline* is diphyletic with both groups (the '*T. monspessulana* group' and the '*T. linifolia* group') separately nested within *Genista*. *Genista benehoavensis* (La Palma) and *G. tenera* (Madeira) form, with *G. tinctoria* of Europe, a single clade characterized by vestigially arillate seeds. The Canarian species of *Adenocarpus* have almost identical sequence to the Mediterranean *A. complicatus*, and are likely to be the result of island speciation after a very recent colonization event. This Canarian/Mediterranean *A. complicatus* group is sister to the afrotropical montane *A. mannii* which is probably derived from an earlier colonization from the Mediterranean, possibly via the Red Sea hills. The independent colonization and subsequent radiation of the two *Teline* groups in the Canary Islands make an interesting comparison: the phylogenies both show geographical structuring, each with a central and western island division of taxa. Within the '*T. monspessulana* group' there is evidence that both continental and Madeiran taxa could be derived from the Canary Islands. The finding of two groups within *Teline* also has implications for patterns of hybridization in those parts of the world where *Teline* species are invasive; in California members of the *T. monspessulana* group hybridize readily, but no hybrids have been recorded with *T. linifolia* which has been introduced in the same areas.

4.1 Introduction

4.1.1 *The tribe Genisteae and its representatives in the Canaries and Madeira*

The Genisteae (brooms, gorse and relatives) form a large (> 450 spp.) and ecologically important group, distributed widely in both Old and New World regions (Polhill, 1976). They are notable for having a particularly confused generic taxonomy. There are several well established genera, such as *Cytisus* and *Genista* but little agreement as to which segregates of these genera are valid (Polhill, 1976; Bisby, 1981). Recent molecular studies have shown the Genisteae to be monophyletic (excluding *Melolobium* and *Argyrolobium uniflorum*, Käss & Wink, 1995, 1997), but further sampling is needed to resolve the generic classification.

The Genisteae are well represented in the Canary Islands and Madeira with 22 native species in six genera. In Madeira there are three endemic species (*Teline maderensis*, *T. paivae* and *Genista tenera*) and one possibly native, non-endemic species (*Adenocarpus complicatus*). The situation is more complicated in the central and western Canary Islands (see map, Fig. 1) where some groups appear to have undergone adaptive radiations. There are no native representatives on the dry, eastern islands (Fuerteventura and Lanzarote). The genus *Teline* is represented by 10 endemic species and *Adenocarpus* by three endemic species (one of which, *A. ombriosus*, may be close to extinction). The two common *Adenocarpus* species (local name: “codeso”) are ecologically important, in places forming the dominant vegetation. *Teline canariensis* (local name: “retamón”) is a prominent component of the fayal-brezal (*Myrica faya* and *Erica arborea*) vegetation on Tenerife, and *T. stenopetala* ssp. *stenopetala* (local name: “gacia”) is cultivated in the islands for fodder (Pérez de Paz *et al*., 1986). *Genista*, however, is represented by a single endemic species, reported as extremely rare in the 1970s (Santos-Guerra, 1975) but recently part of a successful conservation program in the sub-alpine zone of La Palma (Palomares Martínez, 1997). The remaining genera consist of *Spartocytisus* (an endemic genus with two species), *Chamaecytisus* (one variable endemic species) and *Retama* (a single, non-endemic species). The *Adenocarpus*, *Genista* and *Teline* species have been the subject of much generic revision (see Table 1) and have been revised by Gibbs (1967, 1974), Gibbs & Dingwall (1972) and Arco Aguilar (1982, 1983, 1993). These treatments, with minor modifications, have been followed here.

4.1.2 *Molecular approaches to plant biogeography in the Macaronesia flora*

Molecular techniques have been used to interpret the historical biogeography of island taxa by sampling putative continental progenitors in different source areas (Böhle, Hilger & Martin, 1996; Kim *et al.*, 1996; Francisco-Ortega *et al.*, 1997a; Vargas *et al.*, 1999; Panero *et al.*, 1999; Helfgott *et al.*, 2000). Biogeographic links for the Macaronesian flora are diverse. There is evidence for Eurosiberian (*Saxifraga*; Vargas *et al.*, 1999), Mediterranean (*Argyranthemum*, *Bencomia*; Francisco-Ortega *et al.*, 1997a; Helfgott *et al.*, 2000), Southern African (*Phyllis*; Sunding, 1979), North African (*Dracaena*; Sunding, 1979) and New World (*Pericallis*; Panero *et al.*, 1999) origins, as well as Asian and Australasian disjunctions (*Apollonias*, *Picconia*; Sunding, 1979). The ITS-5.8S region often fails to provide adequate sequence divergence for phylogenetic resolution of rapid speciation on islands (Böhle *et al.*, 1996, Kim *et al.*, 1996, Helfgott *et al.*, 2000), and in particular the highly conserved 5.8S region is often uninformative in such studies. However, in this analysis the three informative sites in the 5.8S region include a putative synapomorphy for a group otherwise not recovered with ITS alone (these nodes are indicated in Fig. 2).

The objectives of this study were: (1) to examine phylogenetic relationships of the Macaronesian species; (2) to determine whether the Canarian Genisteae share a common biogeographic origin (i.e. European, Mediterranean or African); (3) to investigate the biogeography of *Teline* within the Canary Islands. The Canarian Genisteae are host to 21 species of plant feeding endemic psyllids (Hemiptera). These are the subject of ongoing parallel studies by one of us (D.P.) and it is hoped that the answers to the above questions may shed light on patterns of speciation in the associated insects.

4.2 Materials and methods

4.2.1 *Taxon sampling*

Ingroup selection

Included in the analyses are 18 of the 22 endemic and native species of Genisteae in the Canary Islands and Madeira, which are considered to have ambiguous or unresolved ancestry. Excluded are the only native non-endemic species, *Retama monosperma* (L.) Boiss., a widespread and common species of less biogeographic interest; and three endemic species in *Spartocytisus* and *Chamaecytisus* which have been shown by Käss & Wink (1997) to have originated from within the *Cytisus*-group. To shed further light on the origins of these species would require extensive sampling among the cytisoid legumes which was beyond the scope of this study. Fourteen species of *Teline* were sampled, representing the entirety of the genus as usually circumscibed (section *Teline*) – with the exception of the restricted endemic *T. osmariensis* (Coss.) P. E. Gibbs & Dingwall, from Morocco. This species, known only from two collections, is described as showing close affinity to *T. monspessulana* (Gibbs & Dingwall, 1972; Maire, 1987). Two Spanish species recently transferred (Talavera & Gibbs, 1999) from *Cytisus* to a separate section of *Teline* (section *Chronanthus*: *T. patens* (DC.) Talavera & P.E. Gibbs and *T. tribracteolata* (Webb) Talavera & P.E. Gibbs) are not considered to be closely related to the Macaronesian species and have not been included here. The majority of intraspecific taxa in the Canary Islands including all five subspecies of *Teline stenopetala* (the most variable species) were also analysed, the only exceptions being the very rare *T. rosmarinifolia* ssp. *eurifolia* del Arco and a recently described subspecies, *T. pallida* ssp. *silensis* del Arco (Arco Aguilar, 2000). Eleven of the 15 *Adenocarpus* species, representing each of the three to five species groups delimited by Lems (1958), Gibbs (1967) and Maire (1987), were used to determine the affiliation of the Canarian taxa. The distinctive basal cytisoid plant, *Argyrocytisus battandieri*, which Talavera & Salgueiro (1999) have suggested should be placed in *Adenocarpus* is also included. Finally, a selection of *Genista* spp. from Africa and Europe were chosen to investigate the biogeographic origins and monophyly of the island species of *Genista* and *Teline*, and to ascertain the specific affiliation of *Genista benehoavensis* from the Canary Islands. Details of all the taxa sampled are given in Table 2.

Outgroup selection

In order to provide rooting of the tree I used a member of a related leguminous tribe (Thermopsideae) as the outgroup (*Anagyris foetida*). As *Anagyris* is highly divergent from the ingroup, an unrooted analysis without *Anagyris* was used as a check that this outgroup did not affect the topology.

4.2.2 *Molecular procedures*

DNA sampling and extraction

A total of 50 individuals, representing 36 species, were sampled, mostly from field-collected leaf material (vouchers in GL). However, herbarium material was used for *Adenocarpus mannii* (E), *A. ombriosus* (TFC) and *Teline stenopetala* spp *microphylla* (from La Gomera) (E). In addition, leaf material was obtained from a cultivated, wild-origin specimen of *G. tinctoria* (RBGE). DNA was extracted from silica-dried leaf material using a QIAGEN DNeasy Plant Mini Kit. Extracts were eluted in 50µl of Buffer AE (included in the kit) and stored at -20°C. For PCR amplification 1µl of this solution was used for each reaction.

PCR and sequencing

Amplification of the ITS1-5.8S rDNA-ITS2 region was accomplished using forward primer ITS 5p: 5'–GGAAGGAGAAGTCGTAACAAG–3' (Möller & Cronk, 1997), and reverse primer ITS 4: 5'–TCCTCCGCTTATTGATATGC–3' (White *et al*., 1990). However, the amplification of *A. mannii* from herbarium sheets dated 1968 (Tanzania) and 1981 (Malawi), and *A. ombriosus* (specimen dated 1985) required the use of internal primers to sequence ITS1 and ITS2 independently. In this instance ITS1 was amplified using primers ITS 5p and ITS 2p: 5'–GCTACGTTCTTCATCGATGC–3' (Möller & Cronk, 1997), and ITS2 was amplified with a forward primer I designed using sequences already obtained (ITS 2-LEG: 5'–GCAGAATCCCGTGAACCAT–3') and primer ITS 4. The primer ITS 2-LEG is likely to give better amplification of the ITS2 region than conventional primers for most papilionoid legumes. Each PCR cycle comprised denaturation at 94°C for four minutes, followed by 30 cycles of 94°C for 30 seconds, annealing at 58°C for one minute and an extension of 72°C for one minute, with a final extension of 72°C for 10 minutes. Amplified PCR products were purified with a QIAGEN QIAquick PCR Purification Kit, and resuspended in 30µl of H_2O for direct sequencing using an automated ABI 377. All the sequences referred to here have been deposited in GenBank database under the accession numbers AF330637 – AF330686.

4.2.3 *Sequence analysis*

Alignment

Sequence boundaries for ITS1-5.8S-ITS2 regions were determined with reference to data for *Vicia faba* (Yokota et al., 1989) and previous sequences for the Genisteae (Käss & Wink, 1997). Alignment of sequences was straightforward and was done manually using the program Se-Al (version 1.0a1; Rambaut, 1998). A single ambiguous region of 3 bp in ITS1 (position 219-221 of the aligned matrix) was excluded in a test analysis, but with no change of topology, so the final analysis was run on the complete matrix. The remaining insertion/deletion events (indels) were unambiguous and small (1-3 bp), with the exception of one notable 17 bp deletion in the ITS1 region of *Genista cinerea* and *G. ramosissima* (position 103-119 of the aligned matrix). The length range of ITS1 was 219-241 bp, and ITS2 217-221 bp, while 5.8S was uniformly 163 bp without gaps. Features of the aligned matrix are given in Table 3. The aligned matrix is given in Appendix 1 and is also available from GenBank; both the aligned matrix and trees (Figs 2 & 3) are available from TreeBASE (http://www.herbaria.harvard.edu/treebase/).

Phylogenetic analyses

Phylogenetic analyses were performed with PAUP* (version 4.0b3; Swofford, 1999). The following heuristic search parameters were employed: 1000 random stepwise addition replicates with TBR branch swapping, other options included MULTREES and COLLAPSE. Bootstrap analyses were performed using 2000 simple addition replicates with TBR branch swapping, and decay indices (Bremer, 1988) were calculated using AutoDecay (version 4.0; Eriksson, 1998) and PAUP*. The analyses were run either with gaps coded as missing, or treated as present/absent in a separate binary matrix using contiguous and simple indel coding (Simmons & Ochoterena, 2000). A maximum likelihood (ML) analysis was performed for a comparison to the MP results. Parameters and assumptions used in the ML search were selected using program Modeltest (Posada & Crandall, 1998). The model selected was Tamura-Nei with gamma distribution. An alternative model, general time-reversible with invariable sites and gamma distribution, selected on the basis of the Akaike Information Criterion (AIC), produced the same tree with slightly improved (lower) log-likelihood score. ML heuristic search parameters included simple addition sequence of taxa with TBR branch swapping, MULTREES and COLLAPSE.

To examine the possible diphyletic origin of *Teline*, three analyses were run with the monophyly of *Teline* constrained for: a) a monophyletic Macaronesian clade; b) a monophyletic genus; c) a paraphyletic clade with the inclusion of *Genista segonnei*. The monophyly of an apparently paraphyletic *Teline stenopetala* (with and without ssp. *spachiana*) was also examined. The Kishino-Hasegawa and nonparametric (Templeton's Wilcoxon signed-rank) tests, as implemented in PAUP*, were used to assess these assumptions by testing significant length differences between constrained and unconstrained trees (Table 4).

4.3 Results

4.3.1 *Molecular results*

Molecular evolution in island and continental Genisteae

A total of 637 sites with an average of 617bp per species (excluding gaps) were used in the analyses; 453 sites were constant, 68 variable sites were parsimony-uninformative and 116 were parsimony-informative, further details of sequence characteristics are provided in Table 3. Sequences obtained using internal primers lacked between 38-107bp of an invariable region of 5.8S. Sequence divergence for the ingroup was greater in ITS1 than in ITS2. The 5.8S region was generally conserved, however, there were seven variable sites (one at the 5' end and six toward the 3' end), four of these were uninformative (three varied only between the ingroup and the outgroup, *Anagyris foetida*) but three provided informative synamomophies in *Adenocarpus* and *Teline*. In particular, position 384 of the aligned matrix in the 5.8S region provided the single nucleotide change supporting the grouping of *T. maderensis* with *T. stenopetala* (subspecies from La Palma and El Hierro). A relatively high proportion of species pairs/groups had identical sequences, these were: (1) *A. foliolosus*, *A. ombriosus* and *A. viscosus*; (2) *Genista cinerea* and *G. ramosissima*; (3) *Teline nervosa* and *T. pallida* ssp. *pallida*; (4) *Teline canariensis* and *T. osyroides*; and (5) *T. salsoloides* and *T. stenopetala* spp. *spachiana*. Uncorrected pairwise distances within the *Teline linifolia* group were 0–2.1% including both continental and island taxa, but only 0–0.8% between island taxa alone. Within the *Teline monspessulana* group, distances were 0–2.1%, with greater distances between taxa within the Canary Islands (e.g. 1.6% divergence between subspecies of *T. stenopetala*) than between the Canary Island and continental or Madeiran taxa (with more divergence, 1.8%, between Madeiran and Canary Island species than between continental and island species).

Results of phylogenetic analyses

A maximum parsimony analysis produced a single tree (length 318, CI 0.643 excluding uninformative characters, RI 0.882). Coding gaps as a separate binary matrix provided a single synapomorphy grouping *Teline salsoloides* with *T. stenopetala* ssp. *spachiana*, within an otherwise totally invariable *T. canariensis* clade. Another single indel grouped the three *Adenocarpus complicatus* samples in an otherwise invariable *complicatus*/Canary Island group. Two trees were recovered in a maximum parsimony search without *Anagyris*. One of these differed from the tree presented, but only by a single change (the reverse placement of the *Genista florida*-*G. tinctoria* and *G. segonnei*-*Teline linifolia* groups) and mid-point rooting gave the same root as that provided by *Anagyris*.

The ITS region provides sufficient resolution of the majority of continental species/group relationships but divergence is insufficient to resolve with confidence the island species groups (Figs 2 & 3). Nevertheless, the phylogeny provides 15 nodes (excluding intraspecific nodes) with bootstrap support ≥ 75% and 6 nodes with decay indices ≥ 5, from which a number of conclusions can be drawn on the relationships between island and continental taxa, and relationships between island groups.

4.3.2 *Taxonomic results*

Genista

The molecular results confirm the transfer of *Cytisus benehoavensis* to *Genista* by Arco Aguilar (1982), and moreover places it together with the Madeiran species, *G. tenera*, in section *Genista* with the type species, *G. tinctoria* (bootstrap 96%). It is interesting that the members of this clade (*G. benehoavensis*, *G. tinctoria*, *G. tenera*) all have a small, or vestigial aril (Gibbs, 1974, Arco Aguilar, 1982). The data also support the transfer to *Genista* of *Adenocarpus segonnei* (Gibbs, 1974), and strongly suggests (bootstrap 93%) affinity with the *Teline linifolia* group.

Adenocarpus

The data support the finding of Käss & Wink (1997) of the very close relationship between *A. foliolosus* and *A. viscosus* of the Canary Islands with the European *A. complicatus* (bootstrap 93%). Our data further show that a third Canarian species, *A. ombriosus*, has identical sequence to *A. foliolosus* and *A. viscosus*. Within *Adenocarpus* as a whole there appear to be three subgeneric groups (although the *anagyrifolius* group is not strongly supported, bootstrap 55%). There is support (bootstrap 78%) for a bracteate clade (possessing distinct floral bracts that are persistent) including *A. nainii*, *A. boudyi* and *A. telonensis*. *A. nainii* has previously been treated as a subspecies of the weakly bracteate *A. complicatus* (with narrow, simple floral bracts, present when the inflorescence is in bud but rapidly caducous on maturation and extension of the inflorescence). However, *A. nainii* is clearly distinct from European *A. complicatus*, and I therefore revert to former taxonomic treatments in recognizing this taxon at specific rank (Maire, 1922). *Adenocarpus mannii* which occurs in montane regions of tropical Africa is of particular biogeographic interest, and is shown to be a sister group of the Mediterranean/Canarian *A. complicatus-viscosus-foliolosus* group (bootstrap 82%). *Adenocarpus* as delimited here is clearly a monophyletic genus (bootstrap 87%). The proximity of *Argyrocytisus battandieri* in the tree topology suggests a more cytisoid than genistoid affinity for *Adenocarpus* (Badr, Martin & Jensen, 1994). A reclassification of *Argyrocytisus battandieri* in *Adenocarpus* has recently been proposed due to the occasional presence of glandular papillae on the fruit surface (Talavera & Salgueiro, 1999). The limited sampling here does not allow us to contradict this classification, and the phylogenies presented by Käss & Wink (1997) do not provide an unambiguous placement of *A. battandieri*.

Teline

Our data show that the genus *Teline* is diphyletic, contrary to cpDNA (rbcL) results presented by Käss & Wink (1995). Forcing *Teline* to be monophyletic results in an extra tree length of 10 steps, which is significant at $P = 0.05$ (Table 4). *Teline* has been considered of genistoid affinity (Polhill, 1976; Bisby, 1981; Norverto, González-Andrés & Ortiz, 1994; González-Andrés & Ortiz, 1995) and I confirm that *Teline* is nested within *Genista*, as shown by Käss & Wink (1997). There therefore seems little reason, other than traditional taxonomic practice, to maintain the genus separate from *Genista*. Each of the two clades of *Teline* ('*linifolia* clade' and '*monspessulana* clade') are monophyletic, with independent origins within the genus *Genista*, and represent two colonizations of the Canary Islands. The continental species, *T. linifolia*, distributed in the western Mediterranean including northwest Africa, is the sister taxon to the island *linifolia* group; and the more widespread continental species, *T. monspessulana*, which has a circum-Mediterranean distribution and also occurs in the Azores, is possibly sister to the island *monspessulana* group (but see further discussion below on the biogeography of *Teline*). The Madeiran *Teline* species are in the '*monspessulana* clade'. *T. maderensis* has more recently been circumscribed to include *T. paivae* (Arco Aguilar, 1983; Cannon & Turland, 1994), but as these two species appear to be distinct in the molecular analysis, I have reverted to recognizing two separate *Teline* species on Madeira.

Within the island *linifolia* group, *Teline pallida* (from Tenerife) is more closely related to *T. nervosa* and *T. rosmarinifolia* (from Gran Canaria, bootstrap 78%) than to the conspecific *T. pallida* ssp. *gomerae* (P. E. Gibbs & Dingwall) del Arco (from La Gomera). It accordingly seems appropriate to revive the name *Teline gomerae* (Kunkel, 1975). Within the island *monspessulana* group, one of the five subspecies of *Teline stenopetala* – ssp. *spachiana* – is identical in sequence to *T. salsoloides* and differs from *T. canariensis* and *T. osyroides* by a single indel. This taxon, originally described as '*Genista* (*Teline*) *spachiana*' by Webb (1845), was later synonomized with *T. canariensis* by Gibbs (1972), who cited the intergradation of the two taxa. Furthermore, when Arco Aguilar (1983) placed this taxon in *T. stenopetala*, he reported it as hybridizing with *T. canariensis*. The four Tenerifean taxa (*T. canariensis*, *T. osyroides*, *T. salsoloides*, *T. stenopetala* spp. *spachiana*) form the homogeneous *canariensis* group (bootstrap 99%), which are distinct from the remaining four subspecies of *T. stenopetala*. Because of the difficulty of separating some *T. stenopetala* ssp. *spachiana* from *T. canariensis*, this plant is probably best treated as *T. canariensis* (*spachiana*-type). Interestingly, ssp. *microphylla* from La Gomera and from El Hierro may not have the same origin (Fig. 3), which would support their independent treatment as advocated by Pitard & Proust (see Gibbs & Dingwall, 1972). Even excluding ssp. *spachiana*, *T. stenopetala* is paraphyletic in this analysis. However, constraining *T. stenopetala* to be monophyletic (excluding ssp. *spachiana*) requires an additional tree length of only two steps which is not significant (Table 4). One should therefore interpret these results cautiously as the possibility that gene flow in the '*monspessulana* clade' is confounding phylogenetic signal is increased by the human introduction and movement of species (Francisco-Ortega *et al*., 1997b; Clarke, Johnson & Murray, 1998).

4.4 Discussion

4.4.1 *The Mediterranean element in the Canarian and Madeiran flora*

The Canarian and Madeiran Genisteae have Mediterranean affinities in common with a large proportion of the Macaronesian flora (Bramwell, 1976; Sunding, 1979; Francisco-Ortega *et al*., 1997a; Helfgott *et al*., 2000). The most northern part of Morocco shares many floristic links with southern Europe (Valdés, 1991). However, this eu-mediterranean element is currently almost completely absent from the northwestern part of Africa (SW Morocco) closest to the Canary Islands.

As the Canary Islands are oceanic in origin (Carracedo *et al*. 1998), long distance dispersal is required. The Canarian Genisteae – with seven colonization events accounting for 18 species in six genera – have been relatively successful at distance dispersal. Quézal (1978) and Hooghiemstra *et al*. (1992) provide evidence for a southward extension of Pleistocene Mediterranean woodland along the northwest edge of Africa, which would have considerably lessened the present day disjunction (Fig. 4, arrow B). Even so, seeds would still have needed to travel more than 200 km to reach suitable habitat on Gran Canaria. Wind, bird and sea dispersal may all have been promoted by prevailing northeasterly trade winds and sea currents that were accentuated during the Pleistocene (Rognon & Coudé-Gaussen, 1996) (Fig. 4, arrow A). The Genisteae have an 'explosive dehiscence' – seeds are ejected from the pod, which remains attached to the parent plant, and have no obvious adaptations to dispersal by air or water. Mature seeds of all genera of Canarian brooms typically sink immediately in sea water (pers. obs.). Endozoochorous bird dispersal is a more likely means by which brooms of Mediterranean origin may have reached the Canary Islands. The endemic avifauna of the Canary Islands is predominantly Palaearctic, with central European and Mediterranean affiliations (Bacallado, 1976), and European migrants occur as vagrants.

4.4.2 *Evolution and biogeography of* Adenocarpus

The three *Adenocarpus* groups recovered in the molecular phylogeny are also circumscribed geographically. First, there is a predominantly montane group with *A. anagyrifolius* (High Atlas), *A. bacquei* (High and Middle Atlas) and *A. decorticans* (montane regions of southern Spain and north Morocco). I also predict that this group includes *A. artemisiifolius* (High Atlas) and *A. hispanicus* (montane regions of central Iberia and north Morocco). It has been suggested by Gibbs (1967) that the latter four species form a group of vicariant species. Secondly, there is the bracteate group which is mainly confined to northwest Africa (Morocco and Algeria). The exception is *A. telonensis* which has a range extending from northern Morocco to southern Iberia and France. The bracteate group is predicted to include three species additional to those sampled here: *A. cincinnatus*, *A. umbellatus* and *A. faurei*, which share similar morphological traits (Gibbs, 1967).

The Canarian species belong to the third group which is otherwise comprised of a single polymorphic species, *A. complicatus* (ssp. *complicatus sensu* Gibbs, 1967), and *A. mannii* which occurs only in tropical and subtropical Africa. *A. complicatus* is widespread across southern Europe to Turkey and Syria, localized in Algeria, and also present on Madeira. This species has a controversial taxonomic history with a number of separate species recognized by different authors (Castroviejo, 1999) as well as numerous intraspecific varieties (Rivas-Martínez & Belmonte, 1989). The lack of molecular divergence between the Canarian and European taxa, implies a rapid and widespread expansion of this group, which contrasts with the relatively localized distributions and greater divergence in the other two *Adenocarpus* groups. The Canarian species are therefore considered part of a recent expansion of *A. complicatus* in southern Europe.

It is interesting that *A. mannii* also exhibits considerable morphological variation throughout its range but no sequence divergence was found between samples from Malawi and Tanzania. Two migration routes (Fig. 5, arrows A & B) have been suggested to explain the presence of European and Mediterranean elements in the montane tropical African flora (Quézel & Martinez, 1958-59; Wickens, 1976; Quézel, 1978). The absence of *A. mannii* or *A. complicatus* in the Moroccan Atlas suggests that the Morocco-Sahara track (shown by Quézel & Martinez (1958-59) to have had a Pleistocene extension of

Mediterranean woodland, Fig. 5, arrow A) is not the route by which *A. mannii* reached central Africa. Instead, the close relationship with *A. complicatus* and the prescence of the latter species around the eastern Mediterranean makes the Red Sea hills migration route (Fig. 5, arrow B) put forward by Wickens (1976) a reasonable alternative hypothesis. Subsequent migration of *A. mannii* to west Africa may have followed the southern migration route (Fig. 5, arrow C) proposed by White (1993).

4.4.3 *Biogeography of* Teline

The origin of the '*linifolia* clade' in the Canary Islands is unambiguous – there is a distinct monophyletic island group derived from a continental progenitor. In contrast, the ITS phylogeny for the '*monspessulana* clade' places both the continental and Madeiran species nested within the Canary Island taxa. However, the possible derivation of the continental and Madeiran species from the Canary Islands is only weakly supported (bootstrap 58%) and the additional tree length required to constrain a monophyletic Macaronesian group, excluding the continental *T. monspessulana*, is not significant (Table 4).

A striking geographical division is evident in the phylogeny between the central and western island groups. This is particularly evident in the *T. linifolia* clade, which has a subclade of three species endemic to the central islands (Gran Canaria and Tenerife) while the remaining two species are restricted to the western islands (La Gomera and La Palma). In the *T. monspessulana* clade the situation is more complex, but the *T. canariensis* group and *T. microphylla* occur only on the central islands, while all the subspecies of *T. stenopetala* are native only on the western islands. One possible explanation for this east-west vicariance, that is consistent with the phylogeny presented here, is that (in both cases) an original colonization of the relatively old island of La Gomera (10-12 Myr) was followed by two independent further colonizations from La Gomera of 1) the central islands and 2) the younger western islands (La Palma and El Hierro, 1-2 Myr). This scenario would accord with the basal position of La Gomera species in both *Teline* clades.

4.4.4 *Island evolution and different patterns of radiation*

Each of the main groups of Canarian Genisteae shows a different pattern of species diversification. These may be summarized as follows:

(1) the *Teline monspessulana* clade: geographical segregation within this clade indicates that inter-island dispersal has been less frequent than diversification within islands. Diversification within islands is, in turn, linked to island size and habitat diversity. Thus, the most morphological diversity in *Teline* can be found on Tenerife (the most

ecologically diverse island) between the five members of the *T. canariensis* group, and this is coupled with the least genetic divergence. The *T. canariensis* group may therefore represent a recent radiation on Tenerife after the main central volcanic and island building period, 1-2 Myr ago (Ancochea *et al*., 1990). In contrast, *T. stenopetala* (excluding ssp. *spachiana*), which has four to five subspecies on three islands, exhibits greater genetic divergence but less morphological diversity.

(2) the *Teline linifolia* clade: radiation of this clade in the Canary Islands is less extensive than the '*monspessulana* clade'. Inter-island dispersal has apparently been more frequent than diversification within islands, with all species endemic to a single island (typically localized or rare in distribution).

(3) *Adenocarpus*: the two widespread taxa are distributed in similar altitudinal and ecological niches on different islands. *A. viscosus* occurs only on the high islands (Tenerife and La Palma) in the subalpine zone and generally more xeric habitats; while *A. foliolosus* occupies a lower altitude, more mesic niche, on four of the five islands with this habitat. However, there are extensive hybrid zones that have developed in locations where habitats intergrade, and often in recently disturbed 'hybrid habitats' (Lems, 1958).

(4) *Genista*: the monotypic island endemism of *Genista* contrasts with the *Teline* and *Adenocarpus* radiations. Neither *Genista* species in Madeira nor the Canary Islands has undergone any diversification or radiation, and *G. benehoavensis* and *G. tenera* may represent relict species.

These taxa thus display a contrast between 'single island lineages' (*Teline*) and inter-island colonization or 'horizontal lineages' (*Adenocarpus*) suggested for other groups (Sanchez-Yelamo *et al*., 1995; Francisco-Ortega *et al*., 1996; Kim *et al*., 1996).

4.4.5 *Convergent morphologies in the Genisteae*

Morphological convergence between unrelated members of the island Genisteae has led to some taxonomic confusion, such as the mistaken synonymization of *Genista tenera* with *Teline osyroides* (noted in Gibbs, 1974). It is also probable that morphological convergence within the '*monspessulana* clade' resulted in the misclassification of *T. stenopetala* ssp. *spachiana*. It is a member of the *T. canariensis* group but shares a number of characteristics (longer racemose inflorescences, larger leaves and flowers) with the *T. stenopetala* group. At a higher level, the diphyletic origin of *Teline* suggests that a number of the characters used to delimit the genus are convergent (e.g. strophiolate seeds and trifoliolate leaves). In the predominantly unifoliolate *Genista sensu stricto* reversals to trifoliolate leaves may have occurred several times (e.g. the trifoliolate *G. clavata* in this analysis). Development of the seed aril appears to have independent origins in *Cytisus* and *Genista*, and may also have independent origins in *Teline*. However, *Genista segonnei*, which is both trifoliolate and possesses strophiolate (rim-aril) seeds, is morphologically similar to *Teline* (Maire, 1987), and constraining *Teline* to be monophyletic with the inclusion of *Genista segonnei* does not require a significant increase in tree length (Table 4).

4.4.6 *Hybridization and species boundaries*

The facility with which island species hybridize when ecological barriers are removed has been addressed for a number of different plant groups (Francisco-Ortega *et al*., 2000). The introduction of several *Teline* species to North America provides a useful comparison to ecological behaviour in the native range. It is notable that the one species reported as ‘a noxious weed’ in California (McClintock, 1993), *T. monspessulana*, is the only widespread species within the genus' native distribution. Particularly revealing is the information on interspecific hybridization in California (McClintock, 1993). Five species of *Teline* have been introduced into cultivation in California and have become naturalized, four of these are members of the *monspessulana* clade and hybrids between *T. canariensis*, *T. monspessulana* and *T. stenopetala* are recorded as ‘common’. Hybrids are also known from Tenerife (and possibly Gran Canaria; Suárez Rodgríguez, 1991) between *T. canariensis* and *T. stenopetala*, where the latter species is not native but cultivated. In contrast, *T. linifolia*, also naturalized in California, has not been reported forming hybrids with any of the *monspessulana* clade; and hybrids do not occur between the two *Teline* clades where these are sympatric in their native distributions. Also unrecorded are hybrids within the *linifolia* clade. However, contact between native species, which is frequent in the *monspessulana* clade, does not occur in the ‘*linifolia* clade’ and no breeding experiments have been conducted to provide further data. These patterns of hybridization reflect the phylogenetic division in *Teline*. Further work is required to determine if hybridization in the *monspessulana* clade in the Canary Islands is contributing to genetic erosion via introgression and assimilation as has been suggested for other groups (Francisco-Ortega *et al*., 2000).

4.5 References

Ancochea E, Fuster JM, Ibarrola E, Cendrero A, Coello J, Hernan F, Cantagrel JM, Jamond C. 1990. Volcanic evolution of the island of Tenerife (Canary Islands) in the light of new K-Ar data. *Journal of Volcanic and Geothermal Research* **44:** 231-249.

Arco Aguilar M del. 1982. Sobre la presencia de *Genista* L. en la Isla de la Palma (I. Canarias): *Genista benehoavensis* (Bolle ex Svent.) M. del Arco, comb. nov. (Fabaceae-Genisteae). *Vieraea* **11 (1981):** 267-282.

Arco Aguilar M del. 1983. Catálogo actualizado de los taxones del género *Teline* Medicus en la región Macaronésica adiciones y nuevas combinaciones. *Vieraea* **12 (1982):** 193-232.

Arco Aguilar M del. 1993. New combinations in the genus *Teline* Medicus. Typification of *Cytisus pallidus* Poiret. *Itinera Geobotanica* **7:** 519-523.

Arco Aguilar M del. 2000. Notes on the taxonomy and nomenclature of *Teline pallida* (Poir.) G. Kunkel. *Taxon* **49:** 17-25.

Arco Aguilar M del, Acebes Ginovés JR. 1982. *Teline salsoloides* M. del Arco & J.R. Acebes. sp. nov. (Fabaceae-Genisteae), una nueva especie para la isla de Tenerife (I. Canarias). *Vieraea* **11 (1981):** 251-266.

Arco Aguilar M del, Acebes Ginovés JR, Wildpret de la Torre W. 1978. *Teline osyroides* (Svent.) Gibbs et Dingw. ssp. *sericea* (O. Kuntze) comb. et stat. nov., una nueva subespecie de la isla de Tenerife. *Vieraea* **7 (1977):** 207-230.

Badr A, Martin W, Jensen U. 1994. Chloroplast DNA restriction site polymorphism in Genisteae (Leguminosae) suggests a common origin for European and American lupines. *Plant Systematics and Evolution* **193:** 95-106.

Bacallado JJ. 1976. Notas sobre la distribución y evolución de la avifauna Canaria. In: Kunkel G, ed. *Biogeography and Ecology in the Canary Islands*. The Hague: Dr. W. Junk Publishers, 413-431.

Bisby FA. 1981. Genisteae (Adans.) Benth. (1865). In: Polhill RM, Raven PH, eds. *Advances in Legume Sytematics.* Kew, Royal Botanic Gardens, 409-425.

Böhle U-R, Hilger HH, Martin WF. 1996. Island colonization and evolution of insular woody habit in *Echium* L. (Boraginaceae). *Proceedings of the National Academy of Science, USA* **93:** 11740-11745.

Bramwell D. 1976. The endemic flora of the Canary Islands. In: Kunkel G, ed. *Biogeography and Ecology in the Canary Islands*. The Hague: Dr. W. Junk Publishers, 207-240.

Bremer K. 1988. The limits of amino-acid sequence data in angiosperm phylogenetic reconstruction. *Evolution* **42:** 795-803.

Cannon MJ, Turland NJ. 1994. *Teline* Medik. In: Press JR, Short MJ, eds. *Flora of Madeira*. London: HMSO, 156-157.

Carracedo JC, Day S, Guillou H, Rodríguez Badiola E, Canas JA, Pérez Torrado FJ. 1998. Hotspot volcanism close to a passive continental margin: the Canary Islands. *Geological Magazine* **135:** 591-604.

Castroviejo S. 1999. *Adenocarpus*. In: Talavera S, Aedo C, Castroviejo S, Romero Zarco C, Sáez L, Salgueiro FJ, Velayos M, eds. *Flora Iberica*. Madrid: Real Jardín Botánico, CSIC, 189-205.

Clarke B, Jonhson MS, Murray J. 1998. How 'molecular leakage' can mislead us about island speciation. In: Grant PR, ed. *Evolution on Islands*. Oxford: Oxford University Press, 181-195.

Eriksson T. 1998. AutoDecay. Version 4.0. (program distributed by the author). Stockholm: Department of Botany, University of Stockholm.

Francisco-Ortega J, Jansen RK, Santos-Guerra A. 1996. Chloroplast DNA evidence of colonization, adaptive radiation, and hybridization in the evolution of the Macaronesian flora. *Proceedings of the National Academy of Science, USA.* **93:** 4085-4090.

Francisco-Ortega J, Santos-Guerra A, Hines A, Jansen RK. 1997a. Molecular evidence for a Mediterranean origin of the Macaronesian endemic genus *Argyranthemum* (Asteraceae). *American Journal of Botany* **84:** 1595-1613.

Francisco-Ortega J, Crawford DJ, Santos-Guerra A, Jansen RK. 1997b. Origin and evolution of *Argyranthemum* (Asteraceae: Anthemideae) in Macaronesia. In: Givnish TJ, Sytsma KJ, eds. *Molecular Evolution and Adaptive Radiation*. Cambridge: Cambridge University Press, 407-431.

Francisco-Ortega J, Santos-Guerra A, Seung-Chul K, Crawford DJ. 2000. Plant genetic diversity in the Canary Islands: a conservation perspective. *American Journal of Botany* **87:** 909-919.

Gibbs PE. 1967. A revision of the genus *Adenocarpus*. *Boletim da Sociedade Broteriana* **41 (series 2):** 67-121.

Gibbs PE. 1974. Taxonomic notes on some Canary Island and North African species of *Cytisus* and *Genista*. *Lagascalia* **4:** 33-41.

Gibbs PE, Dingwall I. 1972 (1971). A revision of the genus *Teline*. *Boletim da Sociedade Broteriana* **45 (series 2):** 269-316.

González-Andrés F, Ortiz J-M. 1995. Isoenzymatic characterization of potentially useful forage shrubs belonging to the genus *Cytisus* and allies (Genistea[e]: Leguminosea). *Biochemical Systematics and Ecology* **23:** 813-824.

Helfgott DM, Francisco-Ortega J, Santos-Guerra A, Jansen RK, Simpson BB. 2000. Biogeography and breeding system evolution of the woody *Bencomia* alliance (Rosaceae) in Macaronesia based on ITS sequence data. *Systematic Botany* **25:** 82-97.

Hooghiemstra H, Stalling H, Agwu COC, Dupont LM. 1992. Vegetational and climatic changes at the northern fringe of the Sahara 250,000-5000 years BP: evidence from four marine pollen records located between Portugal and the Canary Islands. *Review of Palaeobotany and Palynology* **74:** 1-53.

Käss E, Wink M. 1995. Molecular phylogeny of the Papilionoideae (family Leguminosae): rbcL gene sequences versus chemical taxonomy. *Botanica Acta* **108:** 149-162.

Käss E, Wink M. 1997. Phylogenetic relationships in the Papilionoideae (family Leguminosae) based on nucleotide sequences of cpDNA (rbcL) and ncDNA (ITS 1 and 2). *Molecular Phylogenetics and Evolution* **8:** 65-88.

Kim S-C, Crawford DJ, Francisco-Ortega J, Santos-Guerra A. 1996. A common origin for woody *Sonchus* and five related genera in the Macaronesian Islands: molecular evidence for extensive radiation. *Proceedings of the National Academy of Science, USA* **93:** 7743-7748.

Kunkel G. 1975. Novedades y taxones críticos en la flora de La Gomera. *Cuadernos de Botanica Canaria* **25:** 17-49.

Lems K. 1958. Botanical notes on the Canary Islands I. Introgression among the species of *Adenocarpus*, and their role in the vegetation of the Islands. *Boletín del Instituto Nacional de Investigaciones Agronómicas, Madrid* **39:** 351-370.

Maire R. 1922. Contributions a l'étude de la flore de l'Afrique du Nord. *Bulletin de la Société de l'Histoire Naturelle de l'Afrique du Nord* **13:** 209-220.

Maire R. 1987. *Leguminosae. Flore de l'Afrique du Nord.* Volume 16. Paris: Lechevalier.

Marrero A, Suarez C. 1988. Aportaciones corológicas de varias especies arbustivos de interés en Gran Canaria (Islas Canarias). *Botanica Macaronésica* **16:** 3-14.

McClintock E. 1993. *Genista* broom. In: Hickman JC, ed. *The Jepson Manual: Higher Plants of California*. Berkeley and Los Angeles: University of California Press, 609.

Möller M, Cronk QCB. 1997. Origin and relationships of *Saintpaulia* (Gesneriaceae) based on ribosomal DNA internal transcribed spacer (ITS) sequences. *American Journal of Botany* **84:** 956-965.

Norverto C A, González-Andrés F, Ortiz JM. 1994. Leaf and stem anatomy of species of *Cytisophyllum, Cytisus, Chamaecytisus, Genista,* and *Genista* sect. *Teline* (Fabaceae: Genisteae) as an aid for taxonomy. *Israel Journal of Plant Science* **42:** 213-225.

Palomares Martínez A. 1997. Memoria anual. Unpublished report. Parque Nacional de La Caldera de Taburiente.

Panero JL, Francisco-Ortega J, Jansen RK, Santos-Guerra A. 1999. Molecular evidence for multiple origins of woodiness and a New World biogeographic connection of the Macaronesian Island endemic *Pericallis*. *Proceedings of the National Academy of Science, USA* **96:** 13886-13891.

Pérez de Paz PL, Arco Aguilar M del, Acebes Ginovés JR, Wildpret de la Torre W. 1986. *Leguminosas Forrajeras de Canarias*. Santa Cruz de Tenerife: Cabildo Insular de Tenerife. Museo Insular de Ciencias Naturales.

Polhill RM. 1976. Genisteae (Adans.) Benth. and related tribes (Leguminosae). *Botanical Systematics. An Occasional Series of Monographs* **1:** 143-368.

Posada D, Crandall KA. 1998. Modeltest: testing the model of DNA substitution. *Bioinformatics* **14:** 817-818.

Quézel P. 1978. Analysis of the flora of the Mediterranean and Saharan Africa. *Annals of the Missouri Botanical Garden* **65:** 479-534.

Quézel P, Martinez C. 1958-59. Le dernier interpluvial au Sahara central. *Libyca* **6-7:** 211-229.

Rambaut A. 1998. Sequence Alignment Editor. Version 1.0a1. (program distributed by the author). Oxford: Oxford University.

Rivas-Martínez S, Belmonte D. 1989. Sinopsis de *Adenocarpus* DC. (Leguminosae). *Opuscula Botanica Pharmaciae Complutensis* **5:** 69-78.

Rognon P, Coudé-Gaussen G. 1996. Paleoclimates off Northwest Africa (28°-35°N) about 18,000 yr B.P. based on continental eolian deposits. *Quaternary Research* **46:** 118-126.

Sanchez-Yelamo MD, Espejo-Ibañez MC, Francisco-Ortega J, Santos-Guerra A. 1995. Electrophoretical evidence of variation in populations of the fodder legume *Chamaecytisus proliferus* from the Canary Islands. *Biochemical Systematics and Ecology* **23:** 53-63.

Santos-Guerra A. 1975. Notas corológicas. I. *Vieraea* **5:** 89-98.

Simmons MP & Ochoterena H. 2000. Gaps as characters in sequence-based phylogenetic analyses. *Systematic Biology* **49:** 369-381.

Suárez Rodgríguez C. 1991. *Estudio de los relictos actuales del monte verde en Gran Canaria*. Las Palmas de Gran Canaria: Cabildo Insular de Gran Canaria.

Sunding P. 1979. Origins of the Macaronesian flora. In: Bramwell D, ed. *Plants and Islands*. London: Academic Press, 13-40.

Swofford DL. 1999. PAUP*. Phylogenetic Analysis Using Parsimony (*and Other Methods). Version 4. Sunderland, Massachusetts: Sinauer Associates.

Talavera S, Gibbs PE. 1999. *Teline*. In: Talavera S, Aedo C, Castroviejo S, Romero Zarco C, Sáez L, Salgueiro FJ, Velayos M, eds. *Flora Iberica*. Madrid: Real Jardín Botánico, CSIC, 141-147.

Talavera S, Salgueiro FJ. 1999. Sobre el tratamiento de la familia Leguminosae en Flora Ibérica. *Lagascalia* **21:** 155-222.

Valdés B. 1991. Andalucia and the Rif. Floristic links and common flora. *Botanica Chronika* **10:** 117-124.

Vargas P, Morton CM, Jury SL. 1999. Biogeographic patterns in Mediterranean and Macaronesian species of *Saxifraga* (Saxifragaceae) inferred from phylogenetic analyses of ITS sequences. *American Journal of Botany* **86:** 724-734.

Webb PB. 1845. *Genista (Teline) spachiana.* Mr. Spach's Genista. *Curtis' Botanical Magazine* **71:** tab. 4195.

White F. 1993. Refuge theory, ice-age aridity and the history of tropical biotas: an essay in plant geography. *Fragmenta Floristica Geobotanica Supplementum* **2:** 385-409.

White TJ, Bruns T, Lee S, Taylor J. 1990. Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In: Innis MA, Gelfand DH, Sninsky JJ, White TJ, eds. *PCR protocols: a giude to methods and applications*. London: Academic Press, 315-322.

Wickens GE. 1976. *The flora of Jebel Marra (Sudan Republic) and its geographical affinities* (Kew Bulletin Additional Series 5). London: HMSO and Royal Botanic Gardens, Kew.

Yokota Y, Kawata T, Iida Y, Kato A, Tanifuji S. 1989. Nucleotide sequence of the 5.8S rRNA gene and internal transcribed spacer regions in carrot and broad bean ribosomal DNA. *Journal of Molecular Evolution* **29:** 294-301.