CLARA INÉS OROZCO PARDO

EVOLUTIONARY BIOLOGY OF *BRUNELLIA* RUÍZ & PAVÓN (Brunelliaceae, Oxalidales)

EVOLUTIONARY BIOLOGY OF
BRUNELLIA RUIZ & PAVÓN
(Brunelliaceae, Oxalidales)

CLARA INÉS OROZCO PARDO
Profesora Asociada-Instituto de Ciencias Naturales,
Universidad Nacional de Colombia

Evolutionary biology of *Brunellia*
(Brunelliaceae, Oxalidales)

ACADEMISCH PROEFSCHRIFT

ter verkrijging van de graad van doctor aan de Universiteit van Amsterdam
op gezag van de Rector Magnificus

Prof. dr. J.J.M. Franse
ten overstaan van een door het college voor promoties ingestelde
commissie,
in het openbaar te verdedigen in de Aula der Universiteit
op dinsdag 4 december 2001 te 12.00 uur

door

CLARA INÉS OROZCO PARDO

Geboren te Bogotá (Colombia)

To Dr. Thomas Van der Hammen

and my family

To the memory of Dr. José Cuatrecasas

CONTENTS

Pág.

CHAPTER 3 – LEAF ANATOMY IN *BRUNELLIA* RUIZ & PAVÓN

CHAPTER 4 – A COMPARATIVE STUDY OF INFLORESCENSES IN *BRUNELLIA* RUIZ & PAVÓN AND RELATED TAXA

CHAPTER 5 – A STUDY OF FLORAL MORPHOLOGY IN *BRUNELLIA* RUIZ & PAVÓN

CHAPTER 6 – POLLEN MORPHOLOGY OF *BRUNELLIA* RUIZ & PAVÓN RELATED TAXA

CHAPTER 7 – A CLADISTIC ANALYSIS OF *BRUNELLIA* RUIZ & PAVÓN BASED ON MORPHOLOGICAL CHARACTERS

SAMENVATTING

B *runellia* is een geslacht van boomsoorten met een wijde neotropische verbreiding, in de tropische Andes vooral in Colombia. Ook zijn er soorten in Centraal Amerika en de Caraïbische eilanden. Vierenvijftig soorten worden in deze studie formeel erkend. De algemene doelstelling van deze studie is het in termen van evolutie verklaren van de waargenomen kenmerken in *Brunellia*, alsmede de patronen van variatie (Brunelliaceae). Om dit doel te bereiken zijn verschillende disciplines van de biologie gebruikt die niet eerder werden toegepast of tot de nu bereikte resolutie in de kennis van de groep leidden. Twee hoofdstukken betreffende de fylogenie zijn ontwikkeld in deze studie: een met het presenteren van een hypothese betreffende de systematische positie van *Brunellia*, gebaseerd op morfologische kenmerken (hoofdstuk 2), en een betreffende de evolutionaire relaties tussen de soorten van *Brunellia* (hoofdstuk 7). Patronen van geografische verbreiding (hoofdstuk 8), gebieden van speciatie en diversificatie zijn vastgesteld op basis van de fylogenetische studie van *Brunellia* in relatie met andere taxa en geologische gegevens van de Andes, Centraal Amerika en het Caraïbische gebied. Hernieuwd onderzoek van kenmerkende systemen omvatte de anatomie (hoofdstuk 3), patronen van bloeiwijzen (hoofdstuk 4), bloemmorfologie (hoofdstuk 5) en pollenmorfologie (hoofdstuk 6) om te worden gebruikt voor de studie van de evolutionaire relaties van de soorten van het genus (hoofdstuk 7). De dieptestudie van deze systemen van kenmerken leverden zeer belangrijke gegevens op vanuit het gezichtspunt van ontwikkeling en 'gedrag' in relatie met andere taxa gerelateerd aan *Brunellia*.

In **Hoofdstuk 2**, uitgaand van de hypothese dat *Brunellia* een genus van de Cunoniaceae is en niet het enige geslacht in de familie van de Brunelliaceae, is de systematische positie van *Brunellia* onderzocht waarbij de fylogenetische relaties van dit taxon, alsmede 20 morfologische kenmerken vergelijkenderwijs geanalyseerd zijn met de elf geslachten van de Cunoniaceae. Twaalf taxa maken deel uit van de ingroup. Het programma Hennig86 gebaseerd op de methode van parsimonie werd gebruikt voor de selectie van de hypothesen van relaties.

De gegevens van de ingroup werden eerst geanalyseerd met vier outgroups: *Connarus, Fothergilla, Davidsonia* en *Quercus,* vervolgens met drie met uitsluiting van *Quercus,* en tenslotte met elk van deze genera apart; in het laatste geval werden de kenmerken gepolariseerd. De resultaten laten zien dat *Brunellia* een taxon is dat verschillend is van de Cunoniaceae en dat de Cunoniaceae een niet-monophyletische groep is. In deze studie wordt voorgesteld *Brunellia, Spiraeanthemum* en *Acsmithia* te plaatsen in een familie verschillend van de Cunoniaceae. Echter, Bradford (pers. med.), op basis van de resultaten van moleculair onderzoek, signaleert *Brunellia* ook als een natuurlijke eenheid onafhankelijk van *Spiraeanthemum* en *Acsmithia.* Meer onderzoek is nodig om de hypothese betreffende de relaties van deze taxa te ondersteunen. Ook wordt de monofyletische oorsprong van de Cunoniaceae ter discussie gesteld door de aanwezigheid van *Davidsonia* binnen de interne groep. De nauwe verwantschap tussen de basale Rosidae met de lagere Hamamelidae wordt bevestigd. De apomorfie en plesiomorfie van enkele kenmerken wordt bediscussieerd. Van geen enkel tribus van Engler kon de monofylie worden bevestigd.

Een groot deel van de informatie die gebruikt is in de analyse van de evolutionaire verwantschappen van *Brunellia* (**Hoofdstuk 7**) is gebaseerd op de resultaten van de anatomische studie (**Hoofdstuk 3**). In dit hoofdstuk wordt de bladanatomische studie van 24 soorten van *Brunellia* gepresenteerd, inclusief van stomata en nodale anatomie. De nodale anatomie van *Brunellia* is uniloculair. Anatomische kenmerken van de bladsteel en de bladschijf hebben taxonomisch belang. De vaatcilinder van de bladsteel is gewoonlijk continue in het midden en distale deel van de bladsteel, terwijl deze meestal discontinue is in het deel dicht bij de knoop.

Het adaxiale deel van de vaatcilinder van de bladsteel is samengedrukt en vormen in weinig soorten als het ware *B. foreroi* aangetroffen worden. Onder de meest bruikbare kenmerken van de bladschijf zijn de aanwezigheid van holtes of golvingen en de aanwezigheid van een hypodermis. De reductie blaadjes om opvallende of weinig opvallende stipulae op de bladsteel te maken is in veel soorten met de verschijningsvorm van een enkel blad waargenomen. Het onderzoeksresultaat geeft aan dat veel soorten ten onrechte met enkelvoudig blad werden beschouwd; in feite hebben alle soorten van *Brunellia* samengesteld bladeren. De stomata zijn actinocytisch, behalve in *B. cutervensis* met anemocytische stomata.

Zoals anatomische kenmerken zijn cok kenmerken van de archtectuur van bloeiwijzen (**Hoofdstuk 4**) gebruikt voor de analyse van de evolutionaire verwantschappen van *Brunellia.* Dit hoofdstuk omvat het onderzoek aan 35 soorten, die tweederde van de soorten van deze monogenerische familie vertegenwoordigen en representatief zijn voor de totale variatie van de bloeiwijzen in dit genus. Het onderzoek werd gerealiseerd om de bloeiwijzemorfologie te

begrijpen en deze informatie te gebruiken voor taxonomische doeleinden, als ook om de verwantschapsrelaties tussen de soorten te leren kennen. Voor de interpretatie van de bloeiwijzen zijn de concepten van de school van Troll (1964) gevolgd en voor een deel gecombineerd met die van Mora-Osejo (1987). De resultaten laten zien dar *Brunellia* een prolifere monotelische synflorescentie afeen anthocaul-monotelische patroon in de zin van Mora-Osejo (1987) vertoont. Met andere woorden een open synflorescentie met okselstandige bloeiwijzen met einstandige bloemen. Drie algemene patronen werden waargenomen afhankelijk van afwezigheid (1) of aanwezigheid (2, 3) van subthyrsoiden: (1) homocladische thyrsoiden (haplo-thyrsoiden) of (2, 3) heterocladische thyrsoiden, of als (2) diplothyrsoid of (3) pleiothyrsoid, Andere variaties hebben als resultaat verschillen in de grootte van de bloemdelen in relatie tot de totale bloeiwijze, de lengte van de hypopodia en epipodia of de symmetrische of asymmetrische verdeling van de takken. Dit alles beinvloedt de vorm en omtrek van de bloeiwijze. Concaulescentie is altijd aanwezig. In enkele soorten is vroeger proliferatie van paracladia waargenomen.

Gegevens van het onderzoek van de bloemmorfologie (**Hoofdstuk 5**) werden ook gebruikt bij de analyse van de evolutionaire relaties. Bij 46 soorten van *Brunellia* heeft bloemonderzoek plaatsgevonden. Ondanks de intraspecifische variatie van het aantal kelkdelen en carpellen, taxonomisch zijn deze als diagnostische kenmerken bij eerdere studies gebruikt. Bloemen van verschillende collecties werden hierbij (vóór de anthesis) opengesneden en bestudeerd onder de microscoop. De studie omvatte een intraspecifische analyse van de bloem symmetrie, het aantal sepalen, carpellen en dispositie en aantal meeldraden. Veel van de soorten laten een patroon zien van twee kransen van meeldraden, terwijl weinig soorten frequent meer dan twee kransen hebben. Extra kransen van meeldraden worden beschouwd als overgangsstadia (klaarblijkelijk polyandrie) voor die soorten met veelvuldige aanwezigheid van twee kransen, Reductie van de meeldraden werd ook waargenomen. De asymmetrie houdt verband met verschillen in afmeting van de sepalen, aantal carpellen (is verschillend van het aantal sepalen) en veranderingen in het aantal en de rangschikking van beide kransen van meeldraden. Veranderingen in het aantal sepalen hebben invloed op het aantal meeldraden en zeer waarschijnlijk op dat van de carpellen. Protandrie wordt waargenomen. Tweeslachtige en eenslachtige bloemen zijn in verschillende individuen aanwezig.

Enkele kenmerken van de pollenkorrel (**Hoofdstuk 6**) werden gebruikt bij de studie naar de evolutionaire relaties van *Brunellia.* Pollen van 24 soorten van *Brunellia* werd onderzocht met de lichtmicroscoop en met de electronenmicroscoop (SEM) voor aanvullende kenmerken en ten behoeve van de analyse van de fylogenetische verwantschappen. De pollenkorrels zijn 3-colporaat, tectaat en met een grote variatie in de versiering van de exine, dat een bijna volmaakt continuum vormt. Vijf categorieën van ornamentatie werden waargenomen:

striaat-reticulaat (grote lumina en hoge muri), fijn reticulaat, variabel reticulaat (muri en lumina van onregelmatige vorm en op verschillende niveaus), een vorm van rugulaat (onregelmatig en uitstekende elementen) en punctaat (de lumina zijn kleiner en rond to spleetvormig). De versiering van de exine heeft fylogenetische betekenis voor enkele groepen van soorten en is gecorreleerd met enkele kenmerken van vegetatieve organen van bloeiwijze en vrucht. Het punctate type lijkt de plesiomorfe staat te zijn. De relaties tussen het pollen van *Brunellia* en bepaalde geslachten van de Cunoniaceae worden bediscussieerd.

Hoofdstuk 7 omvat de studie naar de evolutionaire relaties van het genus om de indeling onder genusniveau te evalueren en om de kenmerken te identificeren die de natuurlijke groepen van soorten bepalen. Vierenveertig kenmerken van de conventionele morfologie, bladanatomie, nodale anatomie, bloeiwijzen, bloemmorfologie en versiering van de exine van de pollenkorrel, werden gecodeerd voor 31 soorten van *Brunellia* en van een outgroup van *Spiraeanthemum* (Cunoniaceae), die vier soorten omvat. Toepassing van PAUP en NONA resulteerde in de zelfde drie meer parsimone cladogrammen, waarvan één wordt aanbevolen als werkhypothese voor de fylogenetische relaties van *Brunellia*. De aanbevolen boom werd ook gevonden met HENNIG86, Pee-Wee(K=6), en de 'majority rule' concensus boom werd verkregen met Pee-Wee (K=1, K=3). *Brunellia* is monofyletisch. De infragenerische classificatie wordt bediscussieerd.

Het ontbreken van fylogenetische studies voor groepen met een andiene en Caraïbische verbreiding vormt een beperking voor het genereren van een hypothese betreffende de vicariantiebiogeografie. Andere ideeën van de historische biogeografie zijn af te leiden van de vicariantie biogeografie. Als punt van vertrek voor een biogeografische studie wordt een scenario van het ontstaan van de verbreiding van *Brunellia* gepresenteerd (Hoofdstuk 8). Deze stellingname is gebaseerd op de actuele verbreiding van de soorten van *Brunellia*, op die van aan *Brunellia* verwante taxa, de fylogenie van *Brunellia* en de geologische informatie betreffende de vorming van de Andes, Centraal Amerika en de Caraïbische eilanden. Gebieden van soortsvorming en diversificatie zijn voor *Brunellia* in termen van fylogenie gedefinieerd. Het Gondwana ontstaan van *Brunellia* staat ter discussie. De oorsprong van *Brunellia* in Centraal Amerika en de Caraïbische eilanden wordt ook besproken met als uitgangspunt de landconnecties met het noorden van Zuid Amerika gedurende het Eoceen en Mioceen. De grootste speciatie en diversificatie in *Brunellia* voltrok zich gedurende het midden van het Mioceen, de periode van de grootste opheffing van de Andes. *Brunellia* is goed aangepast aan grote hoogte. Enkele soorten hebben bepaalde anatomische structuren ontwikkeld als respons op grote hoogte. Colombia heeft het grootste aantal soorten en het hoogste endemisme. Tenslotte worden ook de patronen van geografische verbreiding bediscussieerd.

SUMMARY

*B**runellia* is a neotropical tree genus widely distributed in the Andes. Species of this genus are also found in Central America and Caribbean islands. In this study 54 species are recognized. The general objective of this study was to explain the observed data for *Brunellia* as well as its patterns of variation in evolutionary terms. To carry out this objective different disciplines of biology were employed which had not previously been used or studied in depth for this group. Two chapters on phylogeny are included: firstly a hypothesis on the position of *Brunellia* based on morphological characters is presented in Chapter 2, and secondly the relationships among the group of species in *Brunellia* is established in Chapter 7. Patterns of geographic distribution, speciation and diversification areas were proposed based on the phylogeny of *Brunellia*, its relationships with other taxa and geological hypotheses for the formation of the Andes and the Caribbean. The reexamination of some characters and the in-depth study of anatomy (Chapter 3), patterns of inflorescence (Chapter 4), floral morphology (Chapter 5) and pollen morphology (Chapter 6) were carried out to provide a source of characters, here called systems of characters. These were then used to define monophyletic groups in *Brunellia*, to establish the relationships among the groups of species and to study the evolutionary behavior of the characters (Chapter 7). In addition to using systems of characters as a basis for the phylogenetic study, they also provided information on their development and behavior as compared to other taxa.

In the **chapter 2** was examined the systematic position of *Brunellia* by studying the phylogenetic relationships of this taxon with eleven ingroup taxa representing Cunoniaceae and twenty characters. This study was formulated to taking into account the hypothesis that *Brunellia* belongs to Cunoniaceae rather than being separated in Brunelliaceae. A total of twelve taxa represent the ingroup. The Hennig86 program based on parsimony was used to choose the hypotheses about relationships. The data of the ingroup was analyzed first with four outgroups: *Connarus, Davidsonia, Fothergilla,* and *Quercus*, then excluding *Quercus* and lastly with each of the outgroups independently. Data

were polarized for the last analysis. Results showed that *Brunellia* does not belong to Cunoniaceae, and that Cunoniaceae is not a monophyletic group. I propose that *Brunellia, Spiraeanthemun,* and *Acsmithia* belong to a family distinct from Cunoniaceae. However, BRADFORD (pers. comm.) based in molecular data indicates that *Brunellia* is a natural group distinct to *Spiraeanthemum* and *Acsmithia*. More research is necessary in order to corroborate the relationship of these taxa. The monophyly of Cunoniaceae is doubtful because some of its genera are nested with *Davidsonia*. The results also confirmed the relationships between Rosidae and the lower Hamamelidae. A discussion about plesiomorphic and apomorphic conditions are given for the Rosidae. The monophyly of none of Engler's tribes is confirmed.

Most of the anatomical characters from **chapter 3** were used in the relationships of *Brunellia* (Chapter 7). In this chapter is presented the results of the study of the leaf anatomy of 24 species of *Brunellia*, including observations on the stomata and nodal anatomy. The nodal anatomy of *Brunellia* was observed as unilacunar. Anatomical characters of the petiole and lamina were found to have taxonomic value at species level. The vascular cylinder is usually continuous in the middle and distal parts of the petiole, while, with few exceptions, it is discontinuous in the proximal part near the node. The adaxial portion of the vascular cylinder is compressed and in a few species semicircular or lumpy-shaped. Cortical bundles are frequent at the distal and middle part of the petiole and in the middle of the petiole, while medullary bundles are only found in *B. foreroi*. Stomatal crypts or undulations and the presence of a hypodermis were found to be among the taxonomically useful characters of the lamina. Reduction of lateral leaflets to form conspicuous or inconspicuous stipels on the petiole resulting in the appearance of a simple leaf was observed. Stomata are actinocytic except in *B. cutervensis* which has anomocytic stomata.

As similar to the anatomical characters, the characters of inflorescence architecture of *Brunellia* (**Chapter 4**) were used to the study of phylogeny of *Brunellia*. In this chapter is presented the study of 35 species, that is, about 2/3 of the species of the monogeneric family, covering the total inflorescence variation of the genus. This study was carried out to understand the morphology and incorporate this important information into taxonomic proposals and for defining phylogenetic relationships among the species. The results indicate, according to the concept of TROLL (e.g. 1964) that *Brunellia* has a proliferating monotelic synflorescence. In the sense of MORA-OSEJO (1987: 65) it follows an anthocaule-monotelic pattern. In other words, the inflorescence architecture corresponds to an open synflorescence with axillary floriferous systems bearing terminal flowers. The ramification pattern of these floriferous systems (paracladia of first order) can be characterized as thyrso-paniculate. Three subpatterns can be distinguished according to the absence (1) or presence (2-3) of subthyrsoids: 1) homocladic thyrsoids (haplo-thyrsoids) or (2-3)

heterocladic thyrsoids either as 2) diplo-thyrsoids or as 3) pleio-thyrsoids. Variations also result from differences in the size of the floriferous part in relation to the total length of the inflorescence, the length of hypopodia and epipodia and the symmetric or asymmetric division of the branches that influence the shape and contours of the inflorescence. Concaulescence is nearly always present. In some species early proliferation of paracladia was observed.

Data from the floral morphology (**Chapter 5**) were also used for the proposed phylogeny of *Brunellia*. It was practiced the floral morphology study on 46 species of *Brunellia*. In spite of intraspecific variation great value has been assigned to sepal and carpel numbers as taxonomic characters. Flowers before anthesis were dissected and studied by light microscopy. This study included an analysis of intraspecific variation in floral symmetry, number of sepals, carpels, the arrangements of the stamens, and breeding systems. Most of the species have two whorls of stamens, the outer whorl alternates with the sepals and the inner whorl is opposite to the sepals. Two whorls of stamens are frequently present in most species although a few species often present additional whorls of stamens. Additional whorls of stamens are considered as transitional states in species that always have two whorls of stamens. It was found that additional whorls of stamens result from rearrangements of the space among floral parts by fusion of sepals and, consequently, the stamens of a previous implied floral arrangement are placed in an extra whorl. Asymmetry is often present and was seen to be related to differences in the size of the sepals, carpel number (different from the sepal number), and changes in the number of stamens and their arrangement. Changes of sepal merosity was found to affect stamen merosity and very probably that of the carpels. Reduction of stamens was also observed. Bisexual and female flowers were frequently observed in different individuals.

Some characters of the pollen grain (**Chapter 6**) were used in the phylogeny of *Brunellia*. In this study pollen of 24 species of *Brunellia* were examined with LM and SEM in order to find additional characters for phylogenetic analysis. The pollen grains were found to be 3- colporate, tectate and to have a variable ornamentation which forms an almost perfect continuum. Five categories of exine ornamentation were observed: striate reticulate (large lumina and high muri), finely reticulate, modified reticulate (muri and lumina irregular in shape and at various levels), modified rugulate (irregular and protruding tectal elements) and punctate (the lumina are smaller and round to slit-shaped). The exine ornamentation provides phylogenetic information for some groups of species, and in some cases it is correlated with vegetative, inflorescence and fruit characteristics. The punctate type could be the plesiomorphic character state. Relationships in the pollen morphology of *Brunellia* and certain genera of Cunoniaceae are discussed.

In the chapter 7 is presented the study of the phylogeny of *Brunellia*. This study was performed to determine the relationships among the species, to evaluate the infrageneric clasification, to identify characters defining natural groups of species. Forty-four characters from conventional morphology and the exine sculpture of the pollen grain were coded for 31 Brunellia species and one outgroup taxon, *Spiraeanthemum* (Cunoniaceae) represents four species of Cunoniaceae. PAUP and NONA found the same three most parsimonious cladograms, of which one is recommended as the current working hypothesis for *Brunellia* relationships. The preferred tree was also found by HENNIG86, Pee-Wee (k=6), and majority rule consensus of trees obtained with Pee-Wee (K= 3, k=1). *Brunellia* is monophyletic, and the infrageneric clasification is discussed.

The absence of phylogentic studies for groups with Andean and Caribbean distribution is a limiting factor in producing a historical biogeographical hypothesis from of point of view of vicariance biogeography. However, given the importance of a starting point for a biogeographic study, a scenario on the origin of the distribution of *Brunellia* is presented here (Chapter 8), based on its current distribution, the phylogeny of *Brunellia*, the related taxa and the geological formation of the Andes, Central America and Caribbean Islands. A diversification and speciaiton area is defined for *Brunellia* in terms of phylogenetic lineages. A Gondwanan origin by mass migration from south to the north is discussed as well as the origin of *Brunellia* in Central America and Greater Antilles by land connections with the north of South-America, during Eocene- Miocene. The highest speciation and diversification of *Brunellia* took place in the Mid-Miocene with the main upheaval of the Andes. *Brunellia* is well adapted to high altitudes and some species have developed special anatomical leaf characters as a response to these altitudes. Colombia has the highest number of *Brunellia* species and the highest rate of endemism. Patterns of geographic distribution are presented.

RESUMEN

*B**runellia* es un género, arbóreo, neotropical ampliamente distribuido en los Andes especialmente en Colombia. Las especies del género se encuentran también en Centro América y las Islas del Caribe. Cincuenta y cuatro especies son aquí reconocidas. Como objetivo general de este estudio se planteó el explicar en términos evolutivos los caracteres observados en *Brunellia*, como también los patrones de variación (Brunelliaceae). Para lograr este objetivo se desarrollaron diferentes disciplinas de la biológia, no estudiadas con anterioridad o en profundidad en el grupo. Dos capítulos de filogenia se desarrollan en este estudio: para presentar una hipótesis sobre la posición sistemática de *Brunellia*, con base en caracteres morfológicos (Capítulo 2) y para establecer las relaciones evolutivas entre grupos de especies en *Brunellia* (Capítulo 7). Patrones de distribución geográfica (Capítulo 8), áreas de especiación y diversificación son establecidas con base en el estudio filogenético de *Brunellia*, en las relaciones con otros taxones y datos geológicos de los Andes, Centro América y el Caribe. Además del reexamen de caracteres se estudiaron sistemas de caracteres como anatomía (Capítulo 3), patrones de inflorescencias (Capítulo 4), de morfología floral (Capítulo 5) y morfología del grano de polen (Capítulo 6) fueron desarrollados para el encuentro de otra fuente de caracteres en el estudio de las relaciones evolutivas de las especies en el género (Capítulo 7). El estudio en profundidad en estos sistemas de caracteres arrojan datos muy importantes desde el punto de vista de su desarrollo y comportamiento en relación con otros grupos relacionados con *Brunellia*.

En el **capítulo 2**, partiendo de la hipótesis de *Brunellia* como género de Cunoniaceae y no como único género de la familia Brunelliaceae, se reexamina la posición sistemática de *Brunellia* analizando las relaciones filogenéticas de este taxón con once taxones que representan a Cunoniaceae, y 20 caracteres morfológicos. Un total de doce taxones hacen parte del grupo interno. El programa Hennig86 basado en el método de simplicidad (parsimony) fue usado

para la elección de las hipótesis de relaciones. Los datos del grupo interno fueron analizados primero con cuatro grupos externos: *Connarus, Fothergilla, Davidsonia* y *Quercus,* con tres excluyendo a *Quercus* y con cada uno de ellos por separado; para este último caso los caracteres fueron polarizados. Los resultados muestran a *Brunellia* como taxón diferente de Cunoniaceae y a Cunoniaceae como grupo no monofilético. Se propone en este trabajo la inclusión de *Brunellia, Spiraeanthemum* y *Acsmithia* bajo una familia diferente de Cunoniaceae. Sin embargo, BRADFORD (com. pers.) de acuerdo con los resultados moleculares señala a *Brunellia* como grupo natural independiente de *Spiraeanthemum* y *Acsmithia.* Mayor investigación es necesaria para corroborar la hipótesis de relaciones de estos taxones. Se cuestiona también la monofilia de Cunoniaceae por la presencia de *Davidsonia* dentro del grupo interno. Se confirma la relación estrecha entre las Rosidae basales con las Hamamelidae inferiores. La apomorfía y plesiomorfía de algunos caracteres es discutida. La monofilia de ninguna de las tribus de Engler es confirmada.

Gran parte de la información usada en el análisis de relaciones evolutivas de *Brunellia* (Capítulo 7), se basa en los resultados obtenidos en el estudio anatómico (Capítulo 3). En este capítulo se presenta el estudio anatómico de la hoja de 24 especies de *Brunellia,* incluyendo observaciones de estomas y de anatomía nodal. La anatomía nodal de *Brunellia* se observó unilacunar. Se encontró que caracteres anatómicos del pecíolo y la lámina tienen valor taxonómico. El cilindro central del pecíolo es usualmente continuo en la parte distal y media del pecíolo mientras que fue observado con pocas excepciones discontinuo en la parte proximal al nodo. La parte adaxial del cilindro vascular del pecíolo es comprimida y en pocas especies los haces vasculares tienen la apariencia de formar arcos. Haces corticales son frecuentes en la parte distal y media del pecíolo, mientras que haces medulares se encuentran en *B. foreroi.* Entre los caracteres taxonómicos más útiles de la lámina se encuentra la presencia de criptas o undulaciones y la presencia de hipodermis. La reducción foliar para formar conspicuas o inconspicuas estipelas sobre el pecíolo se observó anatómicamente en muchas especies con apariencia de hojas simples. Este resultado indica que muchas especies que fueron erróneamente consideradas como especies de hojas simples, son como todas las especies de *Brunellia* de hoja compuesta. Los estomas son actinocíticos excepto en *B. cutervensis* con estomas anomocíticos.

Al igual que los caracteres anatómicos, caracteres de arquitectura de la inflorescencia (**Capítulo 4**) fueron usados en el análisis de relaciones evolutivas de *Brunellia.* En este capítulo se muestra el estudio de 35 especies que corresponden a 2/3 de las especies de la familia monogenérica, y que representan la variación total de la inflorescencia en el género. El estudio fue realizado para entender la morfología de las inflorescencias más allá de los límites netamente descriptivos y usar esta información para propósitos taxonómicos

como también para el encuentro de relaciones de parentesco entre las especies. Para la interpretación de las inflorescencias se siguió la escuela de TROLL (1964) parcialmente combinada con los conceptos de MORA-OSEJO (1987: 65). Los resultados muestran que *Brunellia* presenta (en el sentido de TROLL 1964) una sinflorescencia proliferante monotélica, o un sistema antocaulo-monotélico en el sentido de MORA-OSEJO (1987: 65). En otras palabras, una sinflorescencia abierta, con sistemas floríferos axilares sosteniendo flores terminales. Tres patrones generales fueron encontrados, de acuerdo con la ausencia (1) o presencia (2-3) de subtirsoides: 1) tirsoides homocládicos (haplo-tirsoides) o 2-3) tirsoides heterocládicos 2) diplo-tirsoides o 3) pleio-tirsoides. Otras variaciones resultan de diferencias en el tamaño de las partes floríferas en relación con la inflorescencia total, de la longitud de los hipodios y epipodios o la división simétrica o asimétrica de las ramas. Todos ellos influyen en la forma y el contorno de la inflorescencia. Concaulescencia es siempre presente. Casos de proliferación de inflorescencias se observaron en algunas especies.

Datos del estudio de la morfología floral (**Capítulo 5**) fueron también usados en el análisis de relaciones evolutivas. El estudio floral se practicó en 46 especies de *Brunellia.* A pesar de la variación intraespecífica del número de piezas del cáliz y de carpelos, taxonómicamente han sido usados como caracteres diagnósticos en trabajos previos. Flores antes de la antesis, procedentes de diferentes colecciones fueron disectadas y estudiadas bajo el microscopio. El estudio incluye un análisis intraespecífico de la simetría floral, número de sépalos, carpelos, disposición y número de estambres. Muchas de las especies presentan un patrón de dos verticilos de estambres, mientras que pocas especies presentan frecuentemente más de dos verticilos. Verticilos adicionales de estambres se consideran como estados transicionales (aparente poliandría) para aquellas especies con frecuente presencia de dos verticilos. Reducción de estambres fue también observado. La zigomorfía está relacionada con diferencias de tamaño de los sépalos, número de carpelos (diferente del número de sépalos) y cambios en la condición de dos verticilos de estambres. Estambres y verticilos adicionales son el resultado de rearreglos de los espacios florales cuando hay fusión de sépalos y en consecuencia, estambres de una previa condición de dos verticilos ocupan un extra verticilo. Se observó protandria. Flores bisexuales y unisexuales están presentes en diferentes individuos.

Algunos caracteres del grano de polen (**Capítulo 6**) fueron usados en el estudio de relaciones evolutivas de las especies de *Brunellia.* El polen de 24 especies de *Brunellia* fue examinado en el microscopio de luz (ML) y el microscopio electrónico (MES) para encontrar caracteres adicionales y usar esta información en el estudio de relaciones filogenéticas. El grano de polen es 3-colporado, tectado y con una alta variabilidad en la ornamentación de la exina la cual forma un casi perfecto continuo. Se observaron cinco categorías en la ornamentación de la exina del grano de pollen: reticulado a finamente

reticulado, reticulado modificado (muros y láminas de forma irregular y en varios niveles), rugulada modificada (irregular y proyectando elementos tectales) y la ornamentación de tipo punteado (las láminas son pequeñas, redondeadas o en forma de líneas). La ornamentación de la exina tiene importancia filogenética para algunos grupos de especies y presenta correlación con algunos caracteres vegetativos de inflorescencias y del fruto. El tipo de ornamentación punteado parece ser el estado plesiomórfico. Se discuten las relaciones del grano de polen de *Brunellia* y ciertos géneros de Cunoniaceae.

En el **Capítulo 7** se desarrolló el estudio de las relaciones evolutivas del género, para evaluar la división infragenérica y para identificar qué caracteres definen grupos naturales de especies. Cuarenta y cuatro caracteres de morfología convencional, anatomía de la hoja, nodal, inflorescencias , morfología floral, ornamentación de la exina del grano de polen, fueron codificados para 32 taxones de *Brunellia* y un grupo externo *Spiraeanthemum* (Cunoniaceae) el cual representa cuatro especies de la familia Cunoniaceae. PAUP y NONA encontró los mismos tres más parsimoniosos cladogramas, de los cuales uno es recomendado como la hipótesis de trabajo para las relaciones filogenéticas de *Brunellia*. La hipótesis recomendada fue también encontrada con HENNIG86, Pee-Wee(*K=6*), y el árbol de consenso de mayoría obtenido con Pee- Wee (*K=1, K=3*). *Brunellia* es monofilético. Se discute la clasificación infragenérica.

La ausencia de estudios filogenéticos, en grupos con distribución andina y distribución en las Islas del Caribe, es una limitante para la generación de hipótesis de biogeografía histórica. Otras ideas de biogeografía histórica se apartan de la biogeografía vicariante. Como un punto de partida para un estudio o planteamiento biogeográfico, se presenta un escenario del origen de la distribución de *Brunellia* (**Capítulo 8**). Este planteamiento es basado en la actual distribución de las especies de *Brunellia*, en los taxones relacionados con *Brunellia*, la filogenia de *Brunellia*, y la información geológica sobre la formación de los Andes, Centro América y las Islas del Caribe. Áreas de especiación y diversificación son definidas para *Brunellia* en términos filogenéticos. Es discutido el origen Gondwanico en *Brunellia* por migración de masas de tierra desde el sur de América. El origen de *Brunellia* en Centro América y las Islas del Caribe es también discutido con base en las conexiones de tierra con el norte de Sur América durante el Eoceno y el Mioceno. La mayor especiación y diversificación de *Brunellia* ocurrió a mediados del Mioceno, época del mayor levantamiento de los Andes. *Brunellia* es bien adaptada a altas altitudes. Algunas especies han desarrollado estructuras anatómicas en respuestas a las altas elevaciones. Colombia tiene el mayor número de especies y el más alto endemismo. Son también discutidos los patrones de distribución geográfica.

Chapter 1

INTRODUCTION

CLARA INÉS OROZCO

1. 1 CURRENT STATE OF RESEARCH ON *BRUNELLIA*

According to the evolutionary thought on angiosperm evolution, *Brunellia* is a group with primitive characters. The apetalous flowers, the apocarpous ovary and the presence of five vascular bundles in the carpel structure are characters shared by *Brunellia* and primitive angiosperms. Taxa from the subclass Rosidae related to *Brunellia* (Davidsoniaceae, Eucryphiáceae, Cunoniaceae, Connaraceae) have been studied with regard to different biological aspects, such as floral morphology (DICKISON 1975), leaf, floral and wood anatomy (DICKISON 1971, 1973, 1975 a,b, 1977, 1978,1980), and pollen morphology (HIDEUX and FERGUSON 1976, DICKISON 1979). In the 70´s DICKISON considered these taxa as the core group of Rosidae with close relationships to the paraphyletic subclass Hamamelidae (MANOS et al. 1993). The systematic position of *Brunellia* was studied by HUFFORD and DICKISON (1992) who included *Brunellia* within Cunoniaceae. Doubts with regard to these relationships are due to the absence of a synapomorphy in the group in which *Brunellia* was included within Cunoniaceae, and the lack of complete information about character variation in *Brunellia*. First, this study asks whether *Brunellia* is a taxon of Cunoniaceae or whether it belongs to a different family. With respect to this question the relationship of *Brunellia* was revised by OROZCO (1997, see chapter 2). BRADFORD (in prep.) has focused on the phylogenetic relationships of Cunoniaceae on basis of molecular data. He found that Brunelliaceae has close relationships with Cunoniaceae and the Australian family Cephalotaceae. The research group APG (1998) placed Brunelliaceae, Cunoniaceae, and Connaraceae in the order Oxalidales, having previously considered it to belong to the order Rosales.

1. 1. 2 Phylogenetic research

CUATRECASAS presented the first taxonomic study of Brunelliaceae in 1970. In his supplement of 1985 he presented a taxonomic arrangement similar to that of the 70´s, but including a different species concept, and sections and subsections as well as eight new taxa. In 1985 he also included a hypothesis of the relationships of the subsections based on the assumption that *B. boliviana* shows primitive characters and therefore considered this taxon the primitive form from which remaining taxa were derived.

Today, the biologist sees life as a single pattern of common descent with modifications, such modifications are due to processes including speciation and extinction. The phylogenetic or cladistic approach, based on the principles of HENNIG (1966), is currently used by most researchers who wish to find the pattern of relationships based on observed data as the result of evolution. They also try to define a natural diversity based on monophyletic groups which are defined by shared evolutionary novelties. Phylogeny as a discipline is the result of multiple efforts by the biologist to try to explain the pattern of variation through hypotheses of relationships which are judged by their ability to explain the observations. As in other scientific theories, the phylogenetic hypothesis is continuously evaluated in its concepts and methods. The technological advances, the development of algorithms included in different computational packages and molecular biology have also influenced the development of its concepts.

An unsuccessful attempt to obtain a pattern of relationships for the species in *Brunellia*, based on HENNIG's (1966) concept was carried out by OROZCO in 1993 (unpublished). This preliminary hypothesis showed that additional information for the outgroups used and morphological data for species of *Brunellia* were necessary. Extensive morphological data for species of *Brunellia* were obtained by studying different systems of characters in depth. In addition to these systems of characters providing a good source of morphological characters, a wider knowledge of the biology of *Brunellia* is now available. The evidence presented in the following chapters also contributes to the understanding of DICKISON's core families of Rosidae and their relationships: leaf anatomy (Chapter 3), inflorescence morphology (Chapter 4), floral morphology (Chapter 5), and pollen morphology (Chapter 6). A phylogenetic analysis of *Brunellia* has been carried out based on the morphological data observed and a hypothesis of the relationships within *Brunellia* is proposed in chapter 7. Additional exploration of the relationships of *Brunellia* with other taxa is necessary, for example, with regard to molecular analysis, pollination, and dispersal mechanisms. In the case of *Brunellia* and other dioecious families pollination reports are unknown (RENNER and FEIL 1993).

1. 1. 3 Species concept and speciation in Brunellia

The species concept has been one of the most discussed topics in the last forty years, particularly in the distinction between species as an evolutionary unit or as a category. Most of these discussions were centered on the reality of the species. Species are broadly recognized as evolutionary entities on the nature of which research in biodiversity and conservation is carried out.

Brunellia was previously recognized by the morphological- geographical concept (CUATRECASAS 1970, 1985) which was also used for many years to recognize different taxa. The geographical distribution of *Brunellia* had more importance than morphological characters for defining the species. Most of the species defined under the geographical concept were found to be synonymous to others (OROZCO in prep.). One of these cases is the continuous geographical distribution of populations of the same species through the western Andes from Ecuador to Costa Rica which were named differently; for example, *B. acostae* was described for Ecuador and it is the same species as *B. diversifolia* described for Colombia and *B. darienensis* described for Panama (see chapter 8). These populations are also similar to *B. costaricensis* and *B. standleyana* from Costa Rica, however, additional evidence is necessary to declare these species as synonyms of *B. acostae* as well (OROZCO in prep.). Another problem found in the species definition of *Brunellia* was related to the variations and mistakes in the definition of the characters previously used for recognizing species. Most of these referred to vegetative characters, such as the arrangement, whorled or opposite, and form of the leaves. The character of the leaf indument was also wrongly defined with regard to interspecific differences and therefore it was necessary to redefine and delimit the variation of this character (OROZCO 1999). Most importance was given to the calyx and carpel merosity at interspecific level which are actually very variable within the species (see chapter 5). Due to the weakness of the characters previously used in defining *Brunellia*, a reexamination of these characters and the use of other features were necessary (Chapters 3-6). The autapomorphic concept of species (NIXON and WHEELER 1990, DAVIS and NIXON 1992) is proposed for *Brunellia*. The species are defined by an autapomorphy or by a unique combination of characters working as a unit (unique evolutionary novelty- see chapter 7).

1. 1. 4 Origin of the current distribution of Brunellia

Regarding the distribution of *Brunellia*, CROIZAT (1952) wrote that *Brunellia* was present in the New World before the Andes began to rise, entered from the south, and migrated from Bolivia and Peru to the northern Andes, Central America and the Greater Antilles. However, doubts concerning this theory were presented by CUATRECASAS (1970) who considered that CROIZAT's evidence of the phytogeographical distribution was not enough to affirm that *Brunellia* came from the south of South America. He considered that *Brunellia* could also have

arrived from North America. A preliminary analysis is presented in chapter 8 where the origin of *Brunellia* is considered with basis on the available data. Despite the absence of a study of the biogeographic history of *Brunellia*, a historical scenario as well as diversification and speciation areas are discussed.

1. 2 HOW CAN WE RECOGNIZE *BRUNELLIA*?

Species of *Brunellia* are trees. In primary forest they can reach 25 to 30 m in height while in disturbed forest smaller trees of up to 10 m are found (Fig. 1.1 a). The species have lateral stipules that vary in number (Fig. 1.1 b). Stipels are present on the rachis of the species with compound leaves or on the petiole of species with unifoliolate leaves (Fig. 1.1 c). *Brunellia* species, especially their leaves, are generally covered with unicellular hairs which fall into different indument groups (OROZCO 1999, Fig. 1.1 d-g). The leaves are opposite or whorled, unifoliolate, or compound (Fig. 1.2 a-f). Some species present unifoliolate and compound leaves on the same individual (Fig. 1.2 a). The flowers lack petals, are apocarpous and are disposed in axillary inflorescences (Fig. 1.2 a-f). The fruit is usually covered with indument and in some cases with hirsute hairs. The follicles vary in form depending on the shape of the endocarp (Fig. 1.3 a-d). Seeds are red and arillate (Fig. 1.3 e).

1. 3 SOME ECOLOGICAL DATA

Brunellia is an important element of the Andean vegetation. RANGELet al. (1997) have reported five associations of *Brunellia* in the Andean forest. These associations are distinguished by an arboreal type of vegetation with elements that reach up to 18 m high, as is the case of the two associations of *Brunellia macrophylla* and *Clethra*. These associations share the presence of *Ocotea calophyla* and *Saurauia brachybotrys*. Other species associated with *B. macrophylla*, include *Miconia stipularis*, *Weinmannia pubescens*, and species of *Hedyosmum*. Three additional associations were defined for *Brunellia occidentalis*, distinguished by forest like vegetation of pendent soils, sometimes clearly epiphytics, and localized in a transition zone between Andean and subandean regions. These associations are found between 1980-2400 m and are defined by the presence of *Acnistus arborescens*, *Hedyosmum racemosum*, *Pilea goudotiana*, *Pteris mucronata*, *Xanthosoma jacquini*, and species of *Blakea*, *Clusia*, *Cyclanthus*, *Geonoma*, *Miconia*, *Palicourea*, *Pilea*, and *Oreopanax*.

1. 4 OBJECTIVES

The objectives of this thesis are as follows:

- To explain the observed data for *Brunellia*, and its natural diversity, in terms of evolutionary patterns of variation found by the application of systematic phylogenetics.
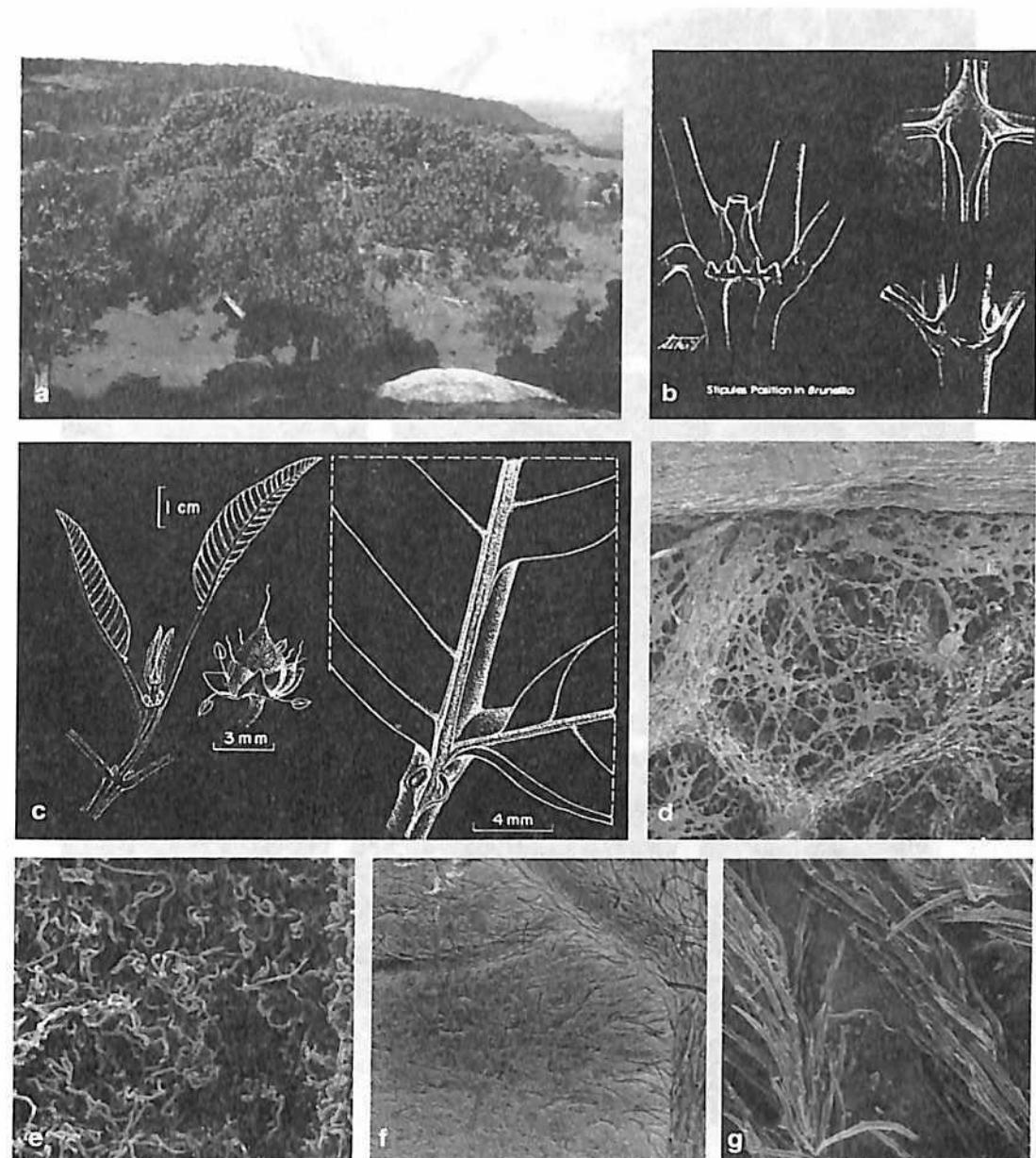
Fig. 1. 1 (a) Habit of *Brunellia goudotii*. (b) Position of stipules in *Brunellia*. (c) Stipel position in *B. amayensis*, unifoliolate and compound leaves. (d) Arachnoid indument in *B. pallida*. (e) Woolly indument in *B. putumayensis*. (f, g) Appressed indument (f) *B. acostae*. (g) *B. boliviana*.

Fig. 1. 2. (a) Unifoliolate and compound leaves in the same node of *B. amayensis*. (b, c) Unifoliolate leaves (b) *B. goudotii*. (c) *B. rufa*. (d-f) Compound leaves, (axillary inflorescence and fruits in f). (d, e) *B. sibundoya*. (f) *B. susaconensis*. (g) Bisexual flowers of *B. comocladifolia* ssp. *dominguensis*. Photos C. I. Orozco (a-f), P. J. M. Maas (g).
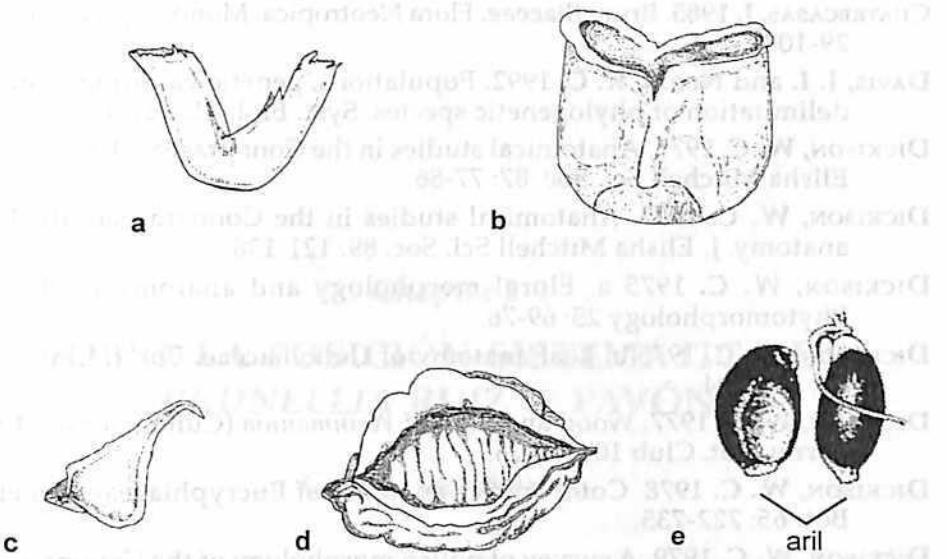
Fig. 1.3 Endocarp shapes in *Brunellia*. (a) U shaped. (b) Urceolate. (c) Navicular modified. (d) Navicular. (e) Arillate seeds.

- To reexamine morphological characters used previously and to identify other sources of characters to contribute to the knowledge of the biology and relationships within *Brunellia* and its relationship with other groups of the subclass Rosidae.

- To find evolutionary novel characters in order to define monophyletic groups in *Brunellia*.

- To propose a species concept for *Brunellia* based on a phylogenetic study.

- To propose a scenario of the current diversification of *Brunellia* using the available information.

1. 5 LITERATURE

APG, 1998. An ordinal classification for the families of flowering plants. Ann. Missouri Bot. Gard. 85 : 531-553.

CROIZAT, L. 1952. Manual of phytogeography, or an account of plant-dispersal throughout the world. W. Junk, The Hague.

CUATRECASAS, J. 1970. Brunelliaceae. Flora Neotropica. Monograph 2: 1-189.

CUATRECASAS, J. 1985. Brunelliaceae. Flora Neotropica. Monograph 2 (suppl.): 29-103.

DAVIS, J. I. and NIXON, K. C. 1992. Populations, genetic variation, and the delimitation of phylogenetic species. Syst. Biol. 41: 421-435.

DICKISON, W. C. 1971. Anatomical studies in the Connaraceae. I. Carpels. J. Elisha Mitchell Sci. Soc. 87: 77-86.

DICKISON, W. C. 1973. Anatomical studies in the Connaraceae. III. Leaf anatomy. J. Elisha Mitchell Sci. Soc. 89: 121-138.

DICKISON, W. C. 1975 a. Floral morphology and anatomy of *Bauera*. Phytomorphology 25: 69-76.

DICKISION, W. C. 1975 b. Leaf anatomy of Cunoniaceae. Bot. J. Linn. Soc. 71: 275-294.

DICKISON, W. C. 1977. Wood anatomy of *Weinmannia* (Cunoniaceae). Bull. Torrey Bot. Club 104: 12-23.

DICKISON, W. C. 1978. Comparative anatomy of Eucryphiaceae. Amer. J. Bot. 65: 722-735.

DICKISON, W. C. 1979. A survey of pollen morphology of the Connaraceae. Pollen et Spores 21: 31-79.

DICKISON, W. C. 1980. Diverse nodal anatomy of the Cunoniaceae. Amer. J. Bot. 67: 975- 981.

HENNIG, W. 1966. Phylogenetic Systematics. Univ. of Illinois Press, Urbana.

HIDEUX, M. J. and FERGUSON, I. K. 1976. The stereostructure of the exine and its evolutionary significance in Saxifragaceae sensu lato. In: The evolutionary significance of the exine (eds. I. FERGUSON and J. MULLER), pp. 327-377. Linn. Soc. Symp. Ser 1. Acad. Press, London.

HUFFORD, L. and. DICKISON, W. C. 1992. A phylogenetic analysis of Cunoniaceae. Syst. Bot. 17: 181-200.

MANOS, P. S., NIXON, K. C., and DOYLE, J. J. 1993. Cladistic analysis restriction site variation within the chloroplast DNA invert repeat region of selected Hamamelidae. Syst. Bot. 18: 551-562.

NIXON, K. C. and WHEELER, Q. D. 1990. An amplification of the phylogenetic species concept. Cladistics 6: 211 - 224.

OROZCO, C. I. 1997. Sobre la posición sistemática de *Brunellia* Ruiz & Pavón. Caldasia 19: 145-164.

OROZCO, C. I. 1999. Indumento del envés foliar en *Brunellia* Ruiz & Pavón y su relativa importancia en la filogenia. Rev. Acad. Colomb. Cienc. 23 (Suplemento especial): 97-102.

RANGEL, O., LOWY, P. D., and AGUILAR, M. 1997. Colombia diversidad biótica II. Tipos de vegetación en Colombia. (ed. O. RANGEL), Santafé de Bogotá.

RENNER, S. and FEIL, J. P. 1993. Pollinators of tropical dioecious angiosperms. Amer. J. Bot. 80: 1100-1107.

Chapter 2

SOBRE LA POSICIÓN SISTEMÁTICA DE *BRUNELLIA* RUIZ & PAVÓN[1]

CLARA INÉS OROZCO

RESUMEN

Bajo el concepto de *Brunellia* como género de Cunoniaceae y no como único género de la familia Brunelliaceae, el presente trabajo examina la posición sistemática de *Brunellia* analizando las relaciones filogenéticas de este taxón con once taxones que representan a Cunoniaceae, y 20 caracteres morfológicos. Un total de doce taxones hacen parte del grupo interno. El programa Hennig86 basado en el método de simplicidad (parsimony) fue usado para la elección de las hipótesis de relaciones. Los datos del grupo interno fueron analizados primero con cuatro grupos externos: *Connarus, Fothergilla, Davidsonia* y *Quercus*, con tres excluyendo a *Quercus* y con cada uno de ellos por separado; para este último caso los caracteres fueron polarizados. Los resultados muestran a *Brunellia* como taxón diferente de Cunoniaceae y a Cunoniaceae como grupo no monofilético. Se propone en este trabajo la inclusión de *Brunellia, Spiraeanthemum* y *Acsmithia* bajo una familia diferente de Cunoniaceae. Mayor investigación es necesaria para corroborar la hipótesis de relaciones de estos taxones. Se cuestiona también la monofilia de Cunoniaceae por la presencia de *Davidsonia* dentro del grupo interno. Se confirma la relación estrecha entre las Rosidae basales con las Hamamelidae inferiores. La apomorfía y plesiomorfía de algunos caracteres es discutida. La monofilia de ninguna de las tribus de Engler es confirmada.

ABSTRACT

Under the concept that *Brunellia* belongs to Cunoniaceae rather than being separated in Brunelliaceae, this paper principally examines the systematic position of *Brunellia* by studying the phylogenetic relationships of this taxon with eleven ingroup taxa representing Cunoniaceae and twenty characters. A total of twelve taxa represent the ingroup. The Hennig86 program based on parsimony was used to choose the hypotheses about relationships. The data of the ingroup was analyzed first with four outgroups: *Connarus*, *Davidsonia*, *Fothergilla*, and *Quercus*, then excluding *Quercus* and lastly with each of the outgroups independently. Data were polarized for the last analysis. Results showed that *Brunellia* does not belong to Cunoniaceae, and that .Cunoniaceae is not a monophyletic group. I propose that *Brunellia*, *Spiraeanthemun*, and *Acsmithia* belong to a family distinct from Cunoniaceae. More research is necessary in order to corroborate the relationship of these taxa. The monophyly of Cunoniaceae is doubtful because some of its genera are nested with *Davidsonia*. The results also confirmed the relationships between Rosidae and the lower Hamamelidae. A discussion about plesiomorphic and apomorphic conditions are given for the Rosidae. The monophyly of none of Engler's tribes is confirmed.

2.1 INTRODUCCIÓN

Engler en 1897 estableció la familia Brunelliaceae y la ubicó dentro del orden Rosales señalando la posición epítropa de los óvulos, como condición principal para considerar a *Brunellia* en una familia separada pero con relaciones muy estrechas a Cunoniaceae. En los más recientes sistemas de clasificación las dos familias son consideradas dentro del orden Rosales (CRONQUIST 1981,THORNE 1983) o dentro del superorden Rosiflorae, orden Cunoniales (DAHLGREN 1980) o Rosanae (TAKHTAJAN 1980). DICKISON (1989) comenta sobre el amplio consenso entre los filogenetistas de que las familias Cunoniaceae, Davidsoniaceae, Brunelliaceae y Eucryphiaceae, esta última considerada como un género de Cunoniaceae (HUFFORD and DICKISON 1992), están muy relacionadas formando una unidad coherente que podría considerarse como primitivas Rosidae, por la posición basal que ocupan dentro del complejo roseliano. Las características de la anatomía del leño de las Cunoniaceae refuerzan su posición basal dentro del complejo.

Diferentes trabajos en los que se discuten la definición de Brunelliaceae y Cunoniaceae como unidades naturales (ENGLER 1897, CUATRECASAS 1970, 1985, DICKISON 1975, 1980, 1989, EHRENDORFER et al. 1984, HUFFORD 1992, HUFFORD and DICKISON 1992). Sin embargo, opiniones originadas en diferentes fuentes cuestionan la monofilia de Cunoniaceae, basadas principalmente en la amplia diversidad morfológica y anatómica y la ausencia de un caracter común para el

grupo (DICKISON 1975, 1989, RUTISHAUSER and DICKISON 1989). Otros como HICKEY and TAYLOR (1991) muestran, con base en caracteres foliares, que Cunoniaceae no es un taxón monofilético, aunque en su investigación la familia no fue suficientemente muestreada.

Los conceptos sobre las relaciones estrechas de *Brunellia* con dos géneros, *Spiraeanthemum* y *Acsmithia* de Cunoniaceae y por ende la no monofilia de Cunoniaceae fue inicialmente insinuada por DICKISON (1975, 1980) y EHRENDORFER et al. (1984) al dar a conocer las diferencias de *Spiraeanthemum* y *Acsmithia* con los restantes taxones de la familia y la estrecha relación de estos dos géneros con *Brunellia*. Estos géneros comparten la condición apétala, la condición apocárpica, la presencia de cinco o cuatro trazas vasculares y la posición epítropa de los óvulos. También tienen en común un xilema no especializado con perforaciones escalariformes (EYDE 1970, DICKISON 1980) condiciones no frecuentes en las Rosales, por lo que se les identifica como géneros basales dentro de Rosidae. Debido a la estrecha relación evolutiva de estos tres taxones, DICKISON (1975, 1980) y EHRENDORFER et al. (1984) coinciden en considerarlos en una familia separada; opinión compartida por CUATRECASAS (1985). Por el contrario, HUFFORD and DICKISON (1992) en su hipótesis de relaciones consideran a *Brunellia* como género de Cunoniaceae y a la familia como grupo monofilético. Sin embargo, aunque *Brunellia*, *Spiraeanthemum* y *Acsmithia* se encuentran dentro de un mismo clado de Cunoniaceae, ninguna sinapomorfía es declarada para estos géneros (Fig. 2.1). En la hipótesis de relaciones muestran también la inclusión de *Eucryphia* dentro de Cunoniaceae considerado dentro de Eucryphiaceae por FOCKE (1895). Por otro lado, los autores confirman las dudas de DICKISON (1975) sobre la monofilia de la tribu *Spiraeanthemeae* tal como fue delimitada por ENGLER (1928).

En este trabajo se examina la posición sistemática de *Brunellia*, reconocido como único género de Brunelliaceae y considerado recientemente como género de Cunoniaceae. El examen se fundamenta en el uso en previos trabajos de interpretación errada de algunos caracteres especialmente de *Brunellia*, taxón del cual he recopilado bastante información. También se pretende reconocer las sinapomorfías de Cunoniaceae, familia reconocida como monofilética pero sin definición de los caracteres por los cuales se considera como grupo natural y aportar mayor conocimiento para Rosidae en general.

2.2 MATERIALES Y MÉTODOS

2.2.1 Caracteres

El estudio se basa principalmente en caracteres morfológicos extraídos para el caso de *Brunellia* de colecciones depositadas en COL y colecciones depositadas en otros herbarios especialmente de US y MO. Todas las especies

actualmente reconocidas ca. de 52 (OROZCO en preparación) fueron examinadas. Colecciones de *Spiraeanthemum* y *Acsmithia* fueron revisadas en US.

Caracteres anatómicos, como también información de los restantes taxones contemplados en el análisis, fueron tomados de trabajos previos (BOGLE 1970, BURGER 1977, CRONQUIST 1981, DICKISON 1971, 1975, 1980, 1989, FORERO 1983, HOOGLAND 1960, 1979, HUFFORD and DICKISON 1992, METCALFE and CHALK 1988, MULLER 1960, RUTHISHAUSER and DICKISON 1989).

Tabla 2. 1 Caracteres y estados de caracteres polarizados con excepción de los caracteres 1, 2 y 3. Los caracteres fueron tratados como no aditivos = desordenados, y aditivos = ordenados.

1. Arreglo de la hoja Alternas = 0 Opuestas = 1 Verticilo = 2	**2. Posición de las estípulas** Incepción lateral = 0 Incepción interpeciolar = 1 Ausentes = 2
3. Hipodermis foliar Presente = 0 Ausente = 1	**4. Forma de los estomas** Anomocítico = 0 Paracítico = 1 Anisocítico = 2
5. Venación Secundaria Brochidódroma = 0 Semicraspedódroma = 1 Craspedódroma = 2	**6. Pétalos** Presentes = 0 Ausentes = 1
7. Número de carpelos 1 ó 2 = 0 Mayor que 2 = 1	**8. Condición del gineceo** Apocárpico = 0 Sincárpico = 1
9. Sutura Ventral de los carpelos Presente = 0 Ausente = 1	**10. Trazas carpelares** Cinco trazas = 0 Tres trazas = 1
11. Estivación de los lóbulos del cáliz Valvada = 0 Imbricada = 1	**12. Número de óvulos** 1 ó 2 = 0 Mayor que 2 = 1
13. Posición de los óvulos Epítropa = 0 Otras formas = 1	**14. Estilo** Acanalado = 0 Sólido = 1
15. Posición del estigma Terminal = 0 Decurrente = 1	**16. Estambres** Diplóstemono = 0 Haplóstemono = 1 Polistemóno = 2
17. Disposición de las flores en Inflorescencias elongadas = 0 En cabezas cortas = 1 Flores solitarias = 2	**18. Posición de la inflorescencia** Axilar = 0 Terminal = 1
19. Superficie estigmática Papilas ausentes = 0 Papilas presentes = 1	**20. Semillas unidas al exocarpo por el funículo** Ausente = 0 Presente = 1

En este análisis se usaron 20 caracteres (Tablas 2.1, 2.2) de los cuales la presencia de sutura ventral de los carpelos (caracter 9), la condición epítropa de los óvulos (caracter 13) y la presencia de papilas en la superficie estigmática (caracter 19) no fueron utilizados en trabajos anteriores. Con base en la información para *Brunellia, Spiraeanthemum* y *Acsmithia* y de acuerdo con los datos de los grupos externos, se modificó la información de los caracteres contemplados por HUFFORD and DICKISON (1992): arreglo de las hojas (caracter 1), número de carpelos (caracter 7), trazas carpelares (caracter 10), disposición de los estambres (caracter 16) y disposición de las flores (caracter 11). Son caracteres multiestados los caracteres 1, 2, 4, 5, 16, 17.

Es de señalar que de los 44 caracteres usados por HUFFORD and DICKISON (1992) solamente son conocidos para el grupo externo 22 estados del caracter (El grupo externo es sintético, basado en Hamamelidaceae, Fagaceae y Rosaceae).

2. 2. 2 *Taxones*

Doce taxones terminales (Tabla 2.2) conforman el grupo interno, uno de ellos corresponde a *Brunellia* (Brunelliaceae) y los once restantes a Cunoniaceae. Los taxones *Spiraeanthemum* y *Acsmithia* están representados en un taxón único, *Spira-Acsmithia*. Los once taxones representan la variación total de Cunoniaceae como también corresponden al muestreo de diferentes clados obtenidos en HUFFORD and DICKISON (1992) Fig. 2.1. El árbol de la figura 2.1 es presentado por los autores como uno de los 47 árboles más simples cuyo índice de consistencia y retención es desconocido.

La elección de once taxones de Cunoniaceae en vez de los 24 utilizados por HUFFORD and DICKISON obedece al muestreo de sólo 20 caracteres (Tablas 2.1, 2.2) por la variación exhibida en los grupos externos o por la ausencia de información confiable para utilizar otras fuentes.

2. 2. 3 *Grupos externos*

Con el propósito de establecer por comparación la polaridad de los caracteres (estados plesiomóficos y apomórficos) se eligieron inicialmente cuatro grupos externos. Estos cuatro grupos externos están representando a Connaraceae (*Connarus*), Davidsoniaceae (*Davidsonia*), Fagaceae (*Quercus*, representando a las Hamamelidae superiores) y Hamamelidaceae (*Fotherghilla*, representando las Hamamelidae inferiores). La elección de los grupos externos se basó principalmente en la revisión de literatura, en donde se destaca la relación existente de Connaraceae y Hamamelidaceae especialmente con los grupos basales de Rosidae (DICKISON 1989, HICKEY and TAYLOR 1991, HUFFORD and DICKISON 1992, MANOS et al. 1993). Rosaceae fue excluída del análisis por presentar gran variación de caracteres.

Los cuatro grupos externos fueron usados todos juntos en un mismo análisis para observar el comportamiento de ellos en relación con el grupo interno, posteriormente con solo tres grupos externos excluyendo a *Quercus* por la ausencia de información para algunos caracteres (Apéndice, Tabla 2.6). Luego se corrieron los datos con cada uno de los grupos externos para conocer cuál de las relaciones es más simple.

Se optó por usar diferentes grupos externos en un mismo análisis para no interpretar erróneamente a un grupo como monofilético cuando se tratan los datos con el programa Hennig86 y un grupo externo (LYNCH com. pers.).

2. 2. 4 Análisis filogenético

Cuando se usaron cuatro y tres grupos externos no se polarizaron los datos por encontrarse en ellos un solo estado del caracter o los dos estados del caracter en el caso de caracteres doble estado (Tabla 2.2). La mayoría de los

Tabla 2. 2 Matriz de datos. El signo interrogante (?) significa polimorfismo para el grupo interno y para el caracter 3 en *Connarus* y *Quercus*. Significa datos confusos o perdidos para los caracteres 3 y 9 en *Fothergilla*, el caracter 3 en *Davidsonia* y los caracteres 10, 11, 14 y 16 en *Quercus*.

Taxones	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
Connarus	0	2	?	1	0	0	0	0	0	0	0	0	0	0	0	0	0	?	0	0
Davidsonia	0	2	?	0	2	1	0	1	0	1	1	1	1	0	0	0	0	1	1	0
Fothergilla	0	0	?	0	2	1	0	1	?	0	0	0	0	1	0	1	?	0	1	0
Quercus	0	0	?	1	2	1	1	1	1	?	?	0	0	?	0	?	0	1	0	0
Brunellia	?	1	1	1	1	1	?	0	0	0	0	0	1	1	1	?	0	1	0	1
Ackama	1	0	0	1	2	0	0	1	0	1	0	1	0	0	0	0	0	0	1	0
Acrophyllum	2	0	0	1	2	0	0	1	0	1	0	1	0	0	0	0	0	1	1	0
Aphanopetalum	1	1	0	1	0	0	1	1	1	1	1	0	0	0	0	0	?	1	1	0
Codia	1	0	1	1	1	?	0	1	0	1	?	?	0	0	0	0	0	1	1	0
Eucryphia	1	0	0	0	?	0	1	1	0	1	1	1	0	1	0	2	2	1	1	0
Gillbeea	1	1	0	1	0	0	1	1	0	1	0	?	0	1	0	0	0	?	1	0
Pancheria	?	0	1	1	1	0	0	1	0	1	1	0	0	0	0	0	0	1	1	0
Pseudoweinmannnia	1	1	0	?	1	1	0	1	0	1	0	1	0	0	0	2	0	1	1	0
Weinmannia	1	0	?	?	1	0	0	1	0	1	1	1	0	0	0	0	0	?	1	0
Spira-Acsmithia	?	0	0	0	0	1	?	0	0	0	0	?	1	?	0	?	0	1	1	0
Spiraeopsis	1	0	0	2	?	0	0	1	0	1	0	1	0.	0	0	0	0	1	1	0

caracteres fueron polarizados, cuando se usó un solo grupo externo (MADDISON et al. 1984) con excepción de los caracteres 1, 2 y 3 (Apéndice, Tablas 2.3 a, 2.3 b, 2.4 - 2.6). Con *Connarus* como grupo externo se practicó un análisis en el que no se polarizaron los caracteres 4 y 5, Tabla 2.1, Apéndice tabla 2.3 b, debido a opiniones encontradas en cuanto al estado plesiomórfico.

Los datos de cada una de las matrices se analizaron con el programa Hennig86 (FARRIS 1988) basado en el método de simplicidad. La condición resuelta de los árboles (libre de politomías) se consideró dentro del concepto de simplicidad.

En el grupo interno el signo interrogante (?) es una condición plesiomórfica del caracter. En los grupos externos el signo interrogante para el caracter 3 significa polimorfismo en *Connarus* y *Quercus* y desconocido para *Fothergilla* y *Davidsonia* (Tabla 2.1, Apéndice, Tablas 2.3-2.6). También son desconocidos para *Quercus* los caracteres 10, 11, 14 y 16. La información para *Fothergilla* de los caracteres 9 y 17 es dudosa.
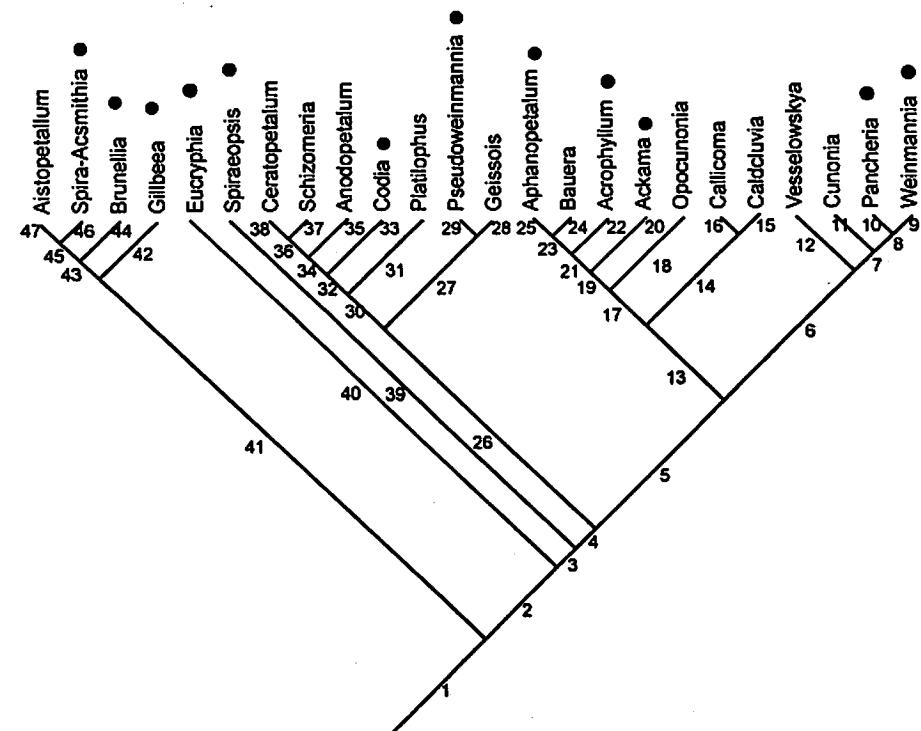
Fig. 2.1 Uno de los 47 cladogramas igualmente parsimoniosos de HUFFORD y DICKISON (1992) sobre las relaciones de Cunoniaceae basado en 44 caracteres. Los puntos negros indican los taxones usados en el análisis.
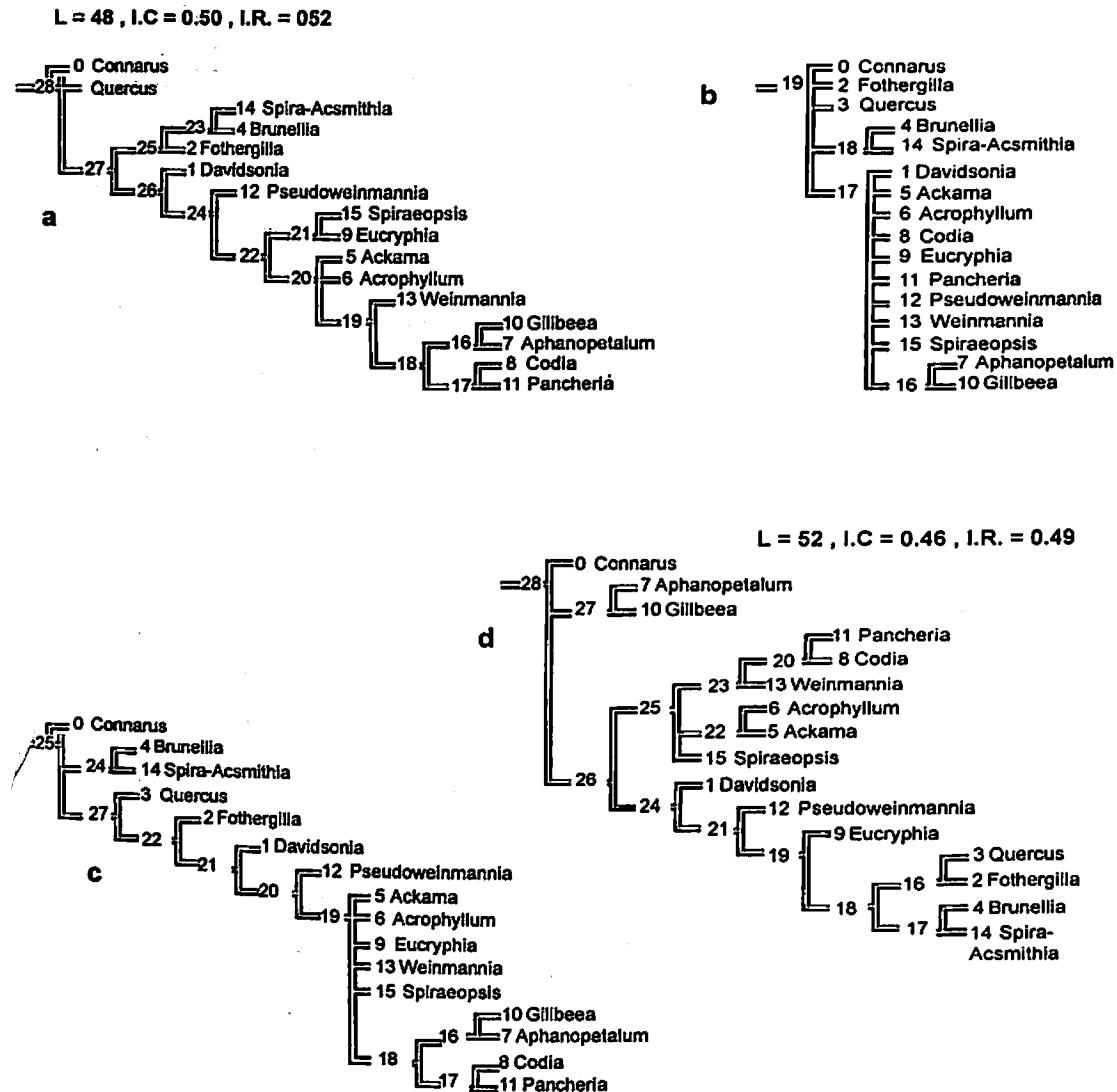
Fig. 2. 2 Cladogramas resultantes de los análisis de las relaciones de *Brunellia* con otros taxones, con cuatro grupos externos. a. Uno de los cladogramas más parsimoniosos obtenido con la opción ie-, caracteres no ditivos. b. Árbol de consenso entre los árboles más parsimoniosos obtenidos con la opción bb*, con caracteres no aditivos. c. Árbol de consenso entre los árboles igualmente parsimoniosos obtenidos al aplicar pesos sucesivos, con caracteres no aditivos. d. 5. Uno de los árboles más parsimoniosos obtenido con la opción ie-, caracteres aditivos.
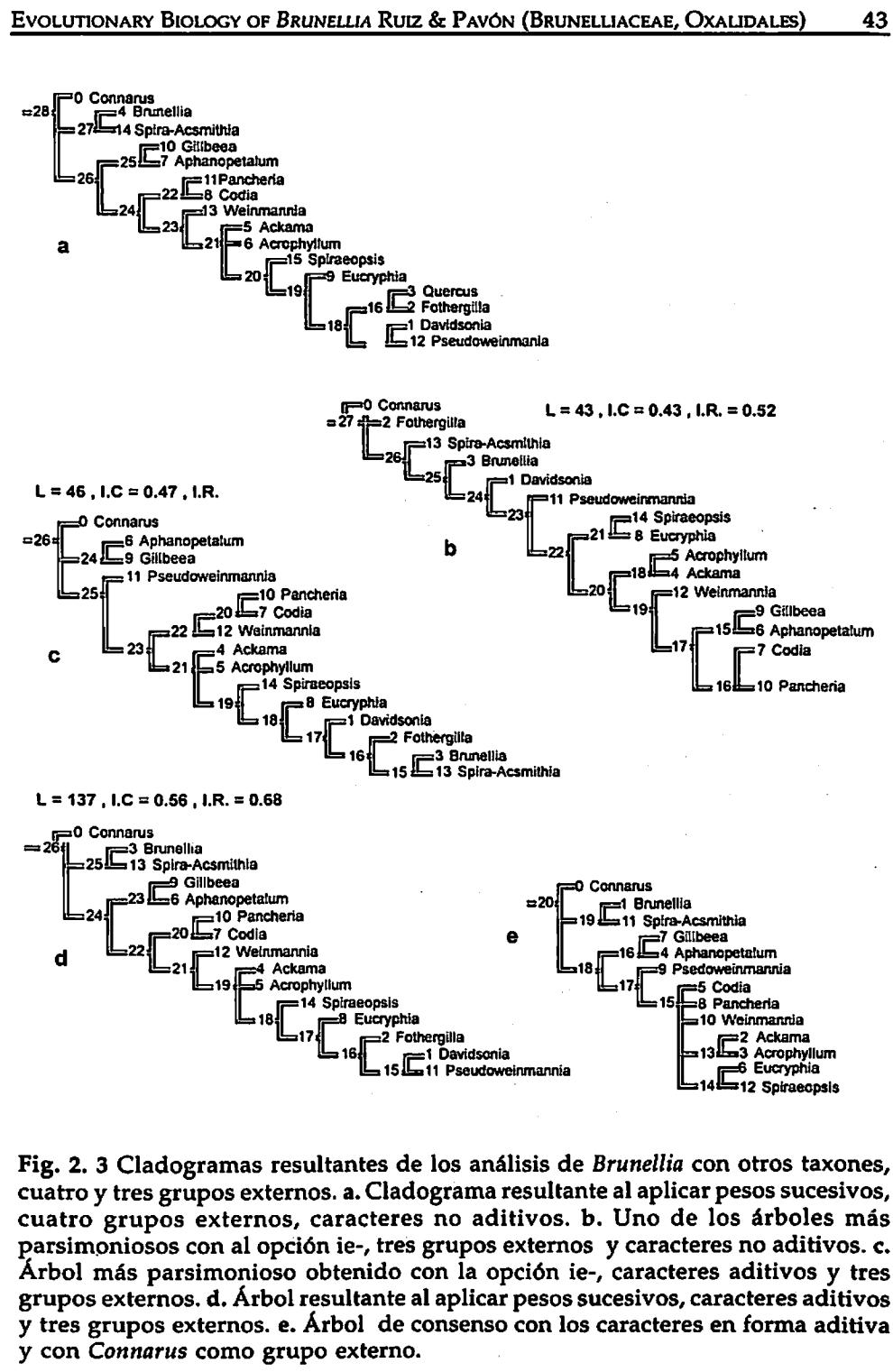
Fig. 2. 3 Cladogramas resultantes de los análisis de *Brunellia* con otros taxones, cuatro y tres grupos externos. a. Cladograma resultante al aplicar pesos sucesivos, cuatro grupos externos, caracteres no aditivos. b. Uno de los árboles más parsimoniosos con al opción ie-, tres grupos externos y caracteres no aditivos. c. Árbol más parsimonioso obtenido con la opción ie-, caracteres aditivos y tres grupos externos. d. Árbol resultante al aplicar pesos sucesivos, caracteres aditivos y tres grupos externos. e. Árbol de consenso con los caracteres en forma aditiva y con *Connarus* como grupo externo.

Cuando se corrieron los datos con cada uno de los grupos externos fueron excluidos del análisis o inactivados por ser autoapomorfías los caracteres 9, 15 y 20. El caracter 20 fue también excluido cuando se corrieron los datos con cuatro y tres grupos externos. El caracter 19, sinapomorfía del grupo interno, se excluyó de los análisis (Tablas 2.1, 2.2, Apéndice, Tablas 2.3-2.6). Aunque estos caracteres fueron excluidos de los análisis, ellos se tuvieron en cuenta en las hipótesis que finalmente se presentan. Los restantes caracteres se trataron en forma ordenada (aditivos), y desordenados (no aditivos).

Se usó la opción ie- con la cual se genera un árbol de longitud mínima, seguida por la opción bb*, que crea un nuevo archivo de árboles por intercambio de ramas y genera todos los árboles más cortos que se puedan encontrar, además permite soluciones exactas. Se optimizaron los datos por pesajes sucesivos, succesive weighting (FARRIS 1989) debido a diferencias del comportamiento de los caracteres en distintas topologías aun cuando los árboles son igualmente simples. Árboles de consenso que reflejan las diferentes topologías originadas de los datos, fueron obtenidos con la opción Nelsen (FARRIS 1988).

2.3 RESULTADOS

Los resultados de los diferentes análisis muestran a *Brunellia*, *Spiraeanthemum* y *Acsmithia* (*Spira-Acsmithia*) como linajes diferentes de Cunoniaceae (Figs. 2.2- 2.5). Los tres taxones están separados de los restantes géneros de Cunoniaceae por ende, la monofilia de Cunoniaceae es cuestionada.

Se enfatizará en los resultados obtenidos con cuatro grupos externos y los caracteres no aditivos debido a que este análisis contiene el mayor número de taxones y caracteres y el menor número de suposiciones (Figs. 2.2 a-d, Tabla 2.7).

2. 3. 1 Cuatro grupos externos

Connarus, Davidsonia, Fothergilla y *Quercus*. Caracteres no aditivos, sólo un paso entre los estados de cada caracter. Longitud de los árboles, número de árboles obtenidos, índices de consistencia y retención para cada una de las opciones tratadas son resumidos en la Tabla 2.7. En la Figura 2.2 a se muestra el cladograma obtenido con la opción ie- y en la figura 2.2 b el árbol resultante de los 120 árboles obtenidos con la opción bb*. En las dos figuras se observan a *Brunellia* y *Spiraeanthemum* y *Acsmithia* (*Spira-Acsmithia*) como géneros hermanos separados de los restantes taxones de Cunoniaceae. *Fothergilla* y *Davidsonia* dentro del grupo interno. El árbol de consenso resultante de optimizar los datos por pesajes sucesivos (Fig. 2.2 c) muestra también a *Brunellia* y *Spira-Acsmithia* en un extremo del árbol y *Davidsonia* dentro del grupo interno como el taxón hermano más cercano de los restantes taxones de Cunoniaceae.

2. 3. 2 Grupos externos por separado (Apéndice, Tablas 2.3-2.6 y 2.8)

Aunque con *Connarus* y *Quercus* se obtienen árboles de igual longitud, Tabla 2.8, para la forma aditiva de los caracteres los índices de consistencia y retención fueron mayores con *Connarus* (I.C. =0.61, I.R.=0.59 vs. I.C.=0.58, I.R.=0.54). Los resultados se discutirán con *Connarus* porque se conoce información de todos sus caracteres (Tabla 2.2, Apéndice, Tablas 3.3 a y 3.3 b), y por mantenerse como género externo en todos los análisis a diferencia de *Davidsonia* o *Fothergilla*.

2. 3. 3 Grupo externo Connarus

Tanto en la forma no aditiva como aditiva, Tabla 2.8, optimizando los datos y cambiando la codificación se obtienen los mismos resultados que en los anteriores análisis indicando que *Brunellia* no es un género de Cunoniaceae y que *Spiraeanthemun* y *Acsmithia* no pertenecen a esta familia. El cladograma de la Figura 2.4 es el único resuelto de los 9 árboles (L= 131, I.C.= 0.83, I.R.=0.81), obtenidos al optimizar los datos cuando los caracteres son tratados en forma no aditiva y polarizando los caracteres con excepción del 1, 2 y 3. La parte resuelta del árbol de consenso, Fig. 2.3 e, resultante de los cinco árboles resueltos (L= 34, I.C. =0.61, I.R=0.59), obtenidos con la opción bb* cuando los datos son tratados en forma aditiva, coincide con el cladograma de la figura 2.4.

2. 3. 4 Cambios en la codificación de los caracteres 4 y 5 (Apéndice Tablas 2.3 b y 2. 8)

Al cambiar la codificación de los caracteres 4 (paracítico =0, anomocítico=1, anisocítico= 2) y la codificación del caracter 5 (brochidódroma = 2, semicraspedódroma= 1, craspedódroma = 0) los árboles conservan la misma longitud e índeces de consistencia y retención que cuando los caracteres 4 y 5 son polarizados. Sin embargo, en la forma aditiva el número de árboles resultantes es menor (L= 130, I.C.=0.84,I.R =0.84) y uno es resuelto de los seis obtenidos al optimizar los datos (Fig. 2.5). Diferencias menores son encontradas en las topologías de los cladogramas de las figuras 2.4 y 2.5. Estas diferencias son debidas al cambio de posición de *Codia* y *Eucryphia*.

Una tercera diferencia, cuando los caracteres 4 y 5 no son polarizados, está relacionada con la longitud de los caracteres todos son igualmente parsimoniosos tanto en la forma aditiva como no aditiva con excepción del caracter 3.

2. 3. 5 Caracteres y Taxones

En las dos hipótesis se registran 11 homoplasias (Figs. 2.4, 2.5) incluyendo una regresión que corresponde a la presencia de no más que dos óvulos en *Pancheria*. Con excepción de los caracteres 8, 10, 13, 17 y 18 (Tabla 2.1) los

caracteres restantes presentan cada uno una homoplasia y cuando los caracteres son aditivos el caracter 3 presenta dos homoplasias (Fig. 2.5).

Los caracteres 2 (2), 9, 15, y 20 son autoapomorfías. *Brunellia* es definida por los caracteres 15 y 20, *Aphanopetalum* por el caracter 9 y *Connarus* por la ausencia de estípulas caracter 2. Otros caracteres aparecen en el análisis como autoapomorfías como son los caracteres 1(2), 4(2), 17(2) y 17(1).

Dos grupos se observan en las figuras 2.4 y 2.5 *Brunellia* y *Spira-Acsmithia* conforman un grupo definido por la sinapomofía 13 (1). Un segundo grupo comprende los restantes géneros del grupo interno y es definido por las sinapomorfías 8 (1) y 10 (1). Todo el grupo interno comparte el caracter 19 (1) que también es una sinapomorfía. Otro grupo es el de *Pseudoweinmannia, Ackama, Acrophyllum, Spiraeopsis, Eucryphia, Weinmannia* y *Codia* definido por la sinapomorfía 12(1) y *Ackama* y *Acrophyllum* definidos por la venación craspedódroma, 5(0) polarizado, 5(2) no polarizado.

2. 4. DISCUSIÓN Y CONCLUSIONES

2. 4. 1 Posición Sistemática de Brunellia

La hipótesis planteada por HUFFORD and DICKISON (1992), Fig. 2.1, es falseada en cuanto a que *Brunellia* no es un género de Cunoniaceae. Tampoco puede considerarse como único género de Brunelliaceae debido a las relaciones encontradas entre *Brunellia* y *Spiraeanthemum* y *Acsmithia* sustentadas por las sinapomorfías de la posición epítropa de los óvulos (Figs. 2.4, 2.5) y la

Tabla 2.7 Comparación de resultados de las relaciones de *Brunellia* con otros taxones. Con cuatro grupos externos *Connarus, Davidsonia, Fothergilla* y *Quercus*; con tres grupos externos *Connarus, Davidsonia* y *Fothergilla*. L= número de pasos, I.C.= índice de consistencia, I.R.= índice de retención, bb*= intercambio de ramas; xs w= pesajes sucesivos, cc- = caracteres no aditivos, cc+= caracteres aditivos.

Grupos externos	Caracteres	Opciones	L	I.C.	I.R.	Número de árboles	Número de árboles resueltos
Con cuatro grupos externos: Connarus, Davidsonia, Fothergilla y Quercus	cc-	bb*	48	0.50	0.52	120	
		xs w	106	0.76	0.83	32	7
	cc+	bb*	52	0.46	0.49	72	
		xs w	136	0.53	0.69	1	
Con tres grupos externos: Connarus, Davidsonia y Fothergilla	cc-	bb*	43	0.51	0.52	110	
		xs w	96	0.76	0.82	32	7
	cc+	bb*	46	0.47	0.50	50	10
		xs w	137	0.56	0.68	1	

presencia de un xilema no especializado con perforaciones escalariformes (EYDE 1970, DICKISON 1980). Se propone en este trabajo, como resultado de los análisis practicados, ubicar a los tres géneros, *Brunellia, Spiraeanthemum* y *Acsmithia*, en una familia diferente de Cunoniaceae. La hipótesis presentada aquí, insta a la investigación de otro tipo de fuentes taxonómicas como por ejemplo profundizar en los caracteres anatómicos especialmente a nivel del leño, desarrollo floral, polinización y caracteres moleculares.

2. 4. 2 Cunoniaceae grupo no monofilético

La hipótesis planteada por HUFFORD and DICKISON (1992) en la que además, de la inclusión de *Brunellia* dentro de Cunoniaceae, propone la monofilia de la familia es también falseada bajo esta última consideración. La no monofilia del grupo fue observada por HICKEY and TAYLOR (1991). En este trabajo no sólo es
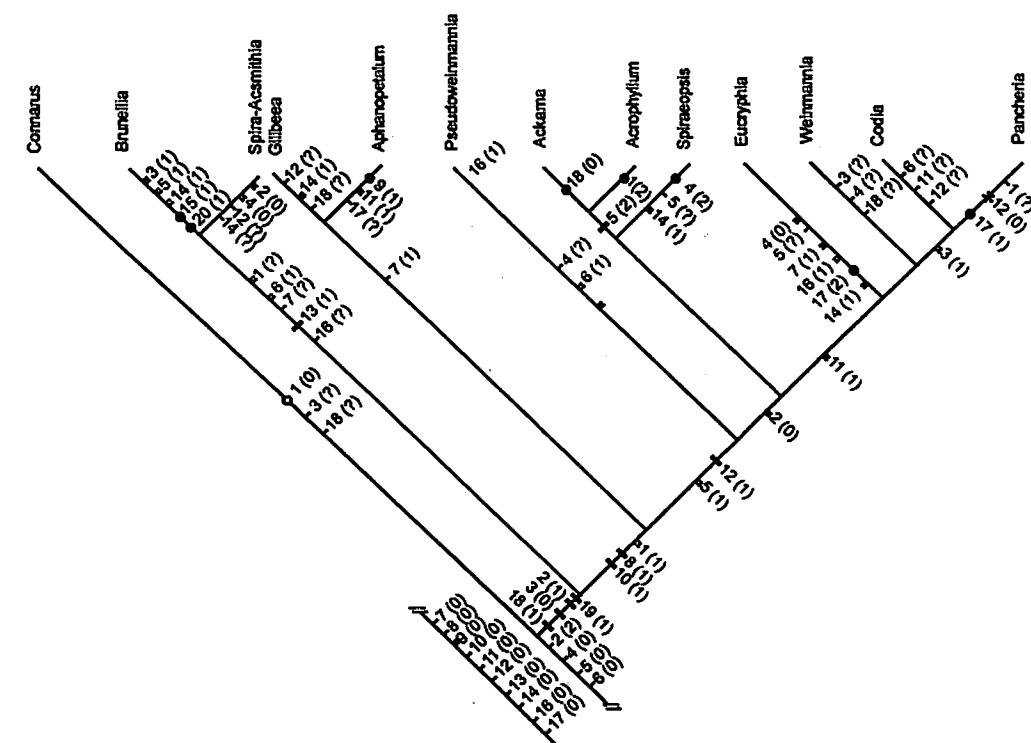
Fig. 2.4 Uno de los cladogramas más parsimoniosos obtenido con *Connarus* como grupo externo, pesos sucesivos, caracteres polarizados con excepción de los caracteres 2 y 3. Este cladograma coincide con uno de los más parsimoniosos obtenido con la opción bb*.= paralelismos, * = regresión, ▆▆= sinapomorfía.

reconocida por el aislamiento de *Brunellia, Spiraeanthemum* y *Acsmithia* de los restantes taxones del grupo interno (Figs. 2.2, 2.3 a-d) sino también, por la presencia de *Davidsonia* dentro del grupo interno cuando se usaron diferentes grupos externos en un mismo análisis; caso contrario, *Davidsonia* siempre se observaría externo al utilizar a este género como el único taxón del grupo externo (Figs. 2.2 d, 2.3 d). El género fue segregado de Cunoniaceae por BANGE (1952) y es reconocido actualmente como único género de Davidsoniaceae por la presencia de hojas alternas, óvulos epítropos y semillas sin albúmina. *Davidsonia* podría ser también el grupo hermano más cercano a Cunoniaceae en sentido estricto (sin considerar a *Brunellia, Spiraeanthemum* y *Acsmithia*), Figs. 2.2 a, 2.2 c, 2.3 b, concepto compartido por HUFFORD (1992). En sentido estricto Cunoniaceae está definida por un gineceo sincárpico y la presencia de tres haces vasculares (Figs. 2.4, 2.5).
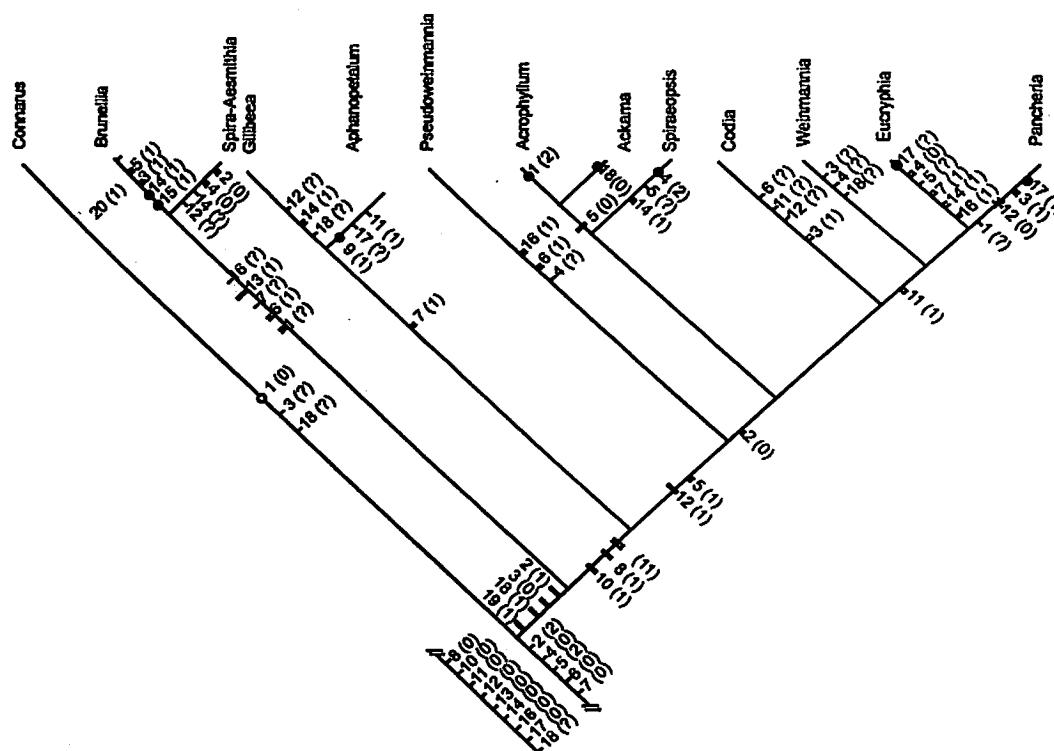
Fig. 2.5 Uno de los cladogramas más parsimoniosos obtenido con *Connarus* al aplicar pesajes sucesivos, caracteres aditivos, obtenido con la opción bb, *Connarus* como grupo externo, caracteres aditivos, polarizados con excepción de los caracteres 4 y 5. = paralelismos, * = regresión, ▬ = sinapomorfía.

2. 4. 3 *Eucryphia* género de Cunoniaceae

En este trabajo se corrobora la inclusión de *Eucryphia* dentro Cunoniaceae según HUFFORD and DICKISON (1992). El género fue considerado por FOCKE (1895) como único género de Eucryphiaceae. Llama, sin embargo, la atención la presencia de un gran número de paralelismos (Figs. 2.4, 2.5) indicando una débil relación ancestro-descendientes con los restantes taxones de Cunoniaceae.

2. 4. 4 *Relaciones de Rosidae y Hamamelidae*

Los resultados del análisis sustentan en parte los conceptos de DICKISON (1989) y de HUFFORD (1992) en cuanto a que las relaciones son más estrechas entre los grupos basales de Rosidae y las Hamamelidae inferiores representadas en el análisis por *Fothergilla* (Figs. 2.2 a, 2.3 b, 2.3 c) relaciones debidas a ancestro común, que aquellas entre Rosidae y Hamamelidae superiores, representadas en el análisis por *Quercus*.

Tabla 2.8 Comparación de resultados de las relaciones de *Brunellia* con otros taxones usando cuatro grupos externos por separado. L= número de pasos, I.C.= índice de consistencia, I.R. = índice de retención. bb * = intercambio de ramas; xs w= pesajes sucesivos, cc- = caracteres no aditivos, cc+ = caracteres aditivos.

Grupos Externos	Caracteres	Opciones	L	I.C.	I.R.	Número de árboles	Número de árboles resueltos
Connarus	Polarizados						
	cc-	bb*	33	0.63	0.58	28	8
		xs w	131	0.83	0.81	9	1
	cc+	bb*	34	0.61	0.59	11	5
		xs w					5
No polarizados							
	cc-	bb*	33	0.63	0.58	28	8
		xs w	183	0.83	0.81	9	1
	cc+	bb*	34	0.61	0.58	31	14
		xs w	130	0.84	0.84	6	1
Davidsonia	Polarizados						
	cc-	bb*	34	0.58	0.53	14	5
		xs w	142	0.83	0.8	1	1
	cc+	bb*	35	0.57	0.53	8	3
		xs w					8
Fothergilla	Polarizados						
	cc-	bb*	34	0.61	0.59	1	1
	cc+	bb*	36	0.58	0.57	2	2
Quercus	Polarizados						
	cc-	bb*	32	0.62	0.58	4	2
		xs w	126	0.85	0.83	3	1
	cc+	bb*	34	0.58	0.54	7	5

2. 4. 5 Tribus de Cunoniaceae

Los géneros muestreados para Cunoniaceae representan la variación de la familia. Sólo tres tribus (DICKISON and RUTISHAUSER 1990) de las cinco consideradas por ENGLER se encuentran en las hipótesis aquí presentadas. Las tres tribus *Spiranthemeae*, *Cunonieae* y *Pancheriae* no son monofiléticas. Sin embargo, se mantienen algunas relaciones que corresponden a los grupos originales de ENGLER. Esas relaciones son dadas entre *Gillbeea* - *Aphanopetalum* de *Spiranthemeae*, *Acrophyllum* - *Ackama* y menos frecuente con *Pseudoweinmannia* de *Cunonieae* y *Codia-Pancheria* de *Pancheriae* (Figs. 2.4, 2.5).

2. 4. 6 Conceptos sobre apomorfía y plesiomorfía

Para Rosidae la apocarpía y la presencia de cinco trazas carpelares son condiciones plesiomórficas. La distribución de estos caracteres en *Connarus*, *Brunellia*, *Spiraeanthemum* y *Acsmithia* y la posición de los taxones en el cladograma (Figs. 2.4, 2.5) podrían indicar la retención de caracteres ancestrales (concepto sustentado por CUATRECASAS 1985 con especial referencia a la apocarpía). La apocarpía en *Brunellia* y *Spiraeanthemum* tiene un origen común. Concepto diferente es dado por HUFFORD and DICKISON (1992), Fig. 2.1.

Los resultados muestran que, a diferencia del concepto de DICKISON (1989) la condición uniovulada (caracter 12) presente en *Connarus* y *Aphanopetalum* es plesiomórfica. Por otro lado, la distribución del caracter dentro de los taxones (Figs. 2.4, 2. 5) coincide con las observaciones de STERLING (1966) y ROBERTSON and ROHRER (1987) en cuanto a que la condición multiovulada es un estado derivado, presente en *Pseudoweinmannia*, *Acrophyllum*, *Ackama*, *Spiraeopsis*, *Weinmannia*, *Eucryphia* y en algunas especies de *Codia*, y no una condición plesiomórfica de Rosidae. Independientemente a partir de un estado uniovular o multiovular se origina la condición biovulada. La condición biovulada está presente en *Brunellia* y *Spiraeanthemum*, en algunas especies de *Acsmithia* (HOOGLAND 1979) de *Gillbeea*, *Aphanopetalum* y *Codia* y en *Pancheria*. Se encuentra la tendencia a la correlación entre el bajo número ovular y la presencia de cinco trazas carpelares con algunas excepciones, por ejemplo en *Aphanopetalum* y en algunas especies de *Gillbeea* y *Codia*.

Los resultados corroboran la suposición de DICKISON (1989) en cuanto a que la pérdida de pétalos en Rosidae es secundaria y adquirida en diferentes linajes independientemente. En las hipótesis planteadas en las Figs. 2.4 y 2.5 la condición apétala 6(1) es un estado apomórfico presente en *Brunellia*, *Spiraeanthemum* y *Acsmithia* y adquirido independientemente en *Pseudoweinmannia*.

La condición epítropa de los óvulos se encuentra también en otros taxones de Rosidae, *Davidsonia* y Spiraeoideae, esta última considerada primitiva dentro de Rosaceae. La presencia de este caracter, 13 (1), en *Spiraeanthemum*, *Brunellia* (Tabla 2.6), las relaciones de estos taxones con *Davidsonia* (Figs. 2.2 a, 2.2 c,

3.3 a, 3.3 d), y la presencia de estos caracteres en Spiraeoideae indican que la condición epítropa de los óvulos tiene diferente origen dentro de Rosales.

Los resultados coinciden con la opinión de DICKISON en cuanto a que una condición intermedia del número de estambres es básica dentro de Rosidae y Hamamelidae con un consecuente incremento o reducción. La condición polistémona es derivada, 16(1), Figs. 2.4, 2.5 presente en diferentes líneas filéticas como *Eucryphia* y *Pseudoweinmannia* mientras, que el estado diplostémono, 16(0), es el patrón básico. En *Brunellia* ocurren los tres estados; la aparente poliandria, un patrón básico diplostémono y reducción del número de estambres a partir del patrón básico (Capítulo 5). Caso similar también ocurre en *Acsmithia* (HOOGLAND 1979). HUFFORD and DICKISON (1992) registran la polistemonía en más de dos líneas filéticas.

También hay coincidencia de los resultados obtenidos con el concepto de DICKISON en cuanto a que la condición valvada es un estado plesiomórfico para las Rosidae. Este patrón básico valvado, 11(0), es observado en la mayoría de los géneros (Figs. 2.4, 2.5). La condición apomórfica imbricada, 11 (1), es una característica de *Weinmannia*, *Eucryphia* y *Pancheria* aunque se encuentra en paralelismo al estar presente en *Aphanopetalum*. Según los resultados de HUFFORD and DICKISON (1992) la condición valvada se observa en diferentes líneas de evolución, presente también en *Callicoma* (Fig. 2.1).

La presencia de un estigma decurrente en *Brunellia*, 15(1), es una condición derivada, mientras que el estigma terminal presente en el resto de taxones es plesiomórfica. DICKISON señala la polinización anemófila para *Brunellia*, imperfecta aún, por la relación de un estigma decurrente con la polinización anemófila encontrada en *Vessellowskya*. Para *Spiraeanthemum* y *Acsmithia*, con estigma terminal, se sugiere una adaptación parcial a la polinización anemófila, debido a que la unisexualidad es una condición que favorecería la anemofilía. No es clara la extrapolación sobre la anemofilía señalada por DICKISON para *Brunellia*. Se cree que el sistema de polinización es entomófilo por la presencia de pequeños escarabajos capturados en las flores masculinas (obsv. pers.).

Los resultados indican que la condición bicarpelar, 7(1), con cualquiera de los grupos externos, es plesiomórfica. Sin embargo, DICKISON (1989) afirma para el ancestro de Hamamelidae- Rosidae un gineceo compuesto de cinco carpelos. En HUFFORD and DICKISON (1992) el estado plesiomórfico o apomórfico es desconocido para el grupo sintético, el cual funciona como grupo externo a pesar de que en Hamamelidaceae y Fagaceae (taxones usados además de Rosaceae para construir el grupo sintético) exhiben dos carpelos. Por otro lado, señalan tanto para *Brunellia* y *Spiraeanthemum* la presencia de tres a cinco carpelos, aunque la condición bicarpelar está presente en *Gillbeea*, *Aphanopetalum* y *Eucryphia* (Figs 2.4, 2.5) y en varias especies de *Brunellia* (ver Capítulo 5).

La condición brochidódroma (cerrada, nervios secundarios se juntan antes de llegar a la margen de la hoja) es plesiomórfica con *Connarus* (Fig. 2.4). Este concepto es opuesto al de DICKISON (1989), quien propone para Rosidae una condición semicraspedódroma, plesiomórfica, nerviación secundaria semiabierta, 5(1). La condición abierta o semiabierta de lo nervios, semicraspedódroma, 5(1), y craspedódroma, 5(2), (terminación de los nervios en la margen de la hoja o al menos una de sus ramas) son condiciones derivadas que comparten los taxones con más de dos óvulos por carpelo con excepción de *Brunellia* con semicraspedódroma nerviación. De estas dos condiciones, la craspedódroma es la más derivada presente en *Ackama* y *Acrophyllum* y en algunas especies de *Eucryphia* y en *Spiraeopsis*. La condición abierta (craspedódroma) es plesiomórfica cuando los datos se tratan con *Fothergilla* y *Davidsonia* como grupos externos por separado.

Al cambiar la codificación el estado plesiomórfico es craspedódromo (condición abierta) y coincide con lo establecido por DICKISON para Rosidae. Al obtener una hipótesis resuelta cuando los datos son aditivos, menos datos en conflicto (Fig. 2.5), se puede inferir que el paso de una condición abierta craspedódroma (estado plesiomórfico) a una cerrada brochidódroma es más simple que el paso de la condición cerrada a la abierta. El caracter debe tratarse con esta codificación y en forma no aditiva al encontrarse, estados iguales del caracter, en líneas filéticas independientes. En HUFFORD and DICKISON (1992) la condición plesiomórfica es desconocida.

El estado anomocítico de los estomas, las células acompañantes no obedecen a un determinado arreglo, (4-0), Fig. 2.4 y es aquí considerado un estado plesiomórfico a partir del cual existe la tendencia evolutiva hacia la diferenciación de las células acompañantes en relación con el tamaño, estado anisocítico (4-2), o a la organización de las mismas a lo largo del eje del poro y las células de guarda, estado paracítico, (4-1). El patrón básico anomocítico (CUATRECASAS, 1970) se observa en *Brunellia, Gillbeea, Acrophyllum, Codia, Pancheria, Aphanopetalum* y en algunas especies de *Pseudoweinmannia* y *Weinmannia*. No hay un acuerdo sobre la condición plesiomórfica para Angiospermas (METCALFE and CHALK, 1988), algunos opinan que el estoma paracítico es plesiomórfico. Dentro del grupo Rosidae los resultados de HUFFORD and DICKISON (1992) coinciden con los presentados en este trabajo, al considerar la condición anomocítica plesiomórfica. Se recomienda tratar el caracter en forma no aditiva por presentarse estados iguales del caracter, en diferentes linajes.

2. 5 AGRADECIMIENTOS

2. 6 LITERATURA

BANGE, G. J. 1952. A new family of dicotyledons: Davidsoniaceae. Blumea 7: 293-296.

BOGLE, A. L. 1970. Floral morphology and vascular anatomy of the Hamamelidaceae: The apetalous genera of Hamamelidoideae. J. Arnold Arbor. 51: 310-366.

BURGER, W. 1977. Fagaceae. Fieldiana Bot. 40: 59-82.

CRONQUIST, A. 1981. An integrated system of classification of flowering plants. Columbia, Univ. Press, New York.

CUATRECASAS, J. 1970. Brunelliaceae. Flora Neotropica Monograph 2. 1-189.

CUATRECASAS, J. 1985. Brunelliaceae. Flora Neotropica Monograph 2 (suppl.): 29-103.

DAHLGREN, R. M. 1980. A revised system of classification of the angiosperms. J. Linn.Soc. Bot. 80: 91-124.

DICKISON, W. C. 1971. Anatomical studies in the Connaraceae. I. Carpels. J. Elisha Mitchell Sci. Soc. 87: 77-86.

DICKISON, W. C. 1975. Studies on floral anatomy of the Cunoniaceae. Amer. J. Bot. 62: 433-477.

DICKISON, W. C. 1980. Comparative wood anatomy and evolution of Cunoniaceae. Allertonia 2: 281-321.

DICKISON, W. C. 1989. Comparisons of primitive Rosidae and Hamamelidae. In: Evolution, systematics, and fossil history of the Hamamelidae, vol 1: Introduction and 'lower' Hamamelidae (eds. P.R. CRANE and S. BLACKMORE), pp. 47-73. Clarendon Press. Oxford.

DICKISON, W. C. and RUTISHAUSER, R. 1990. Developmental morphology of stipules and systematics of the Cunoniaceae and presumed allies. II. Taxa withouth interpetiolar stipules and conclusions. Bot. Helvetica 100: 75-95.

EHRENDORFER, F., MORAWETZ, W., and DAWE, J. 1984. The neotropical angiosperm families Brunelliaceae and Caryocaraceae: First karyosystematical data and affinities. Pl. Syst. Evol. 145: 183-191.

ENGLER, A. 1897. Brunelliaceae. In: Die natürlichen Planzenfamilien, 2 nd (eds. A. ENGLER and K. PRANTL), pp. 182-184 W. Engelmann, Leipzig.

ENGLER, A. 1928. Cunoniaceae. In: Die natürlichen Pflanzenfamilien, 2 nd (ed. A. ENGLER and K. PRANTL), pp. 229-262 W. Engelmann, Leipzig.

EYDE, R. 1970. Anatomy in Brunelliaceae. In: (ed. J. CUATRECASAS). Flora Neotropica Monograph 2. 32-43

FARRIS, J. S. 1988. Hennig86. Version 1.5. Port Jefferson Station, New York.

FARRIS, J. S. 1989. The retention index and the rescaled consistency index. Cladistics 5: 321-424.

FOCKE, W. O. 1895. Eucryphiaceae. In : Die natürlichen Pflanzenfamilien (eds. A. Engler and K. Prantl), pp. 129-131. W. Engelmann, Leipzig.

FORERO, E. 1983. Connaraceae. Flora Neotropica Monograph 36: 1-207.

HICKEY, L. J. and TAYLOR, D. W. 1991. The leaf architecture of *Ticodendron* and the application of foliar characters in discerning its relationships. Ann. Missouri Bot. Gard. 78: 105-130.

HOOGLAND, R. D. 1960. Studies in the Cunoniaceae. I. The genera *Ceratopetalum, Gillbeea, Aistopetalum, and Calycomis*. Austral. J. Bot. 8: 318-341.

HOOGLAND, R. D. 1979. Studies in the Cunoniaceae. II. The genera *Caldcluvia, Pullea, Acsmithia,* and *Spiraeanthemum.* Blumea 25: 481-505.

HUFFORD, L. 1992. Rosidae and their relationships to other nonmagnoliid dicotyledons: a phylogenetic analysis using morphological and chemical data. Ann. Missouri Bot. Gard. 79: 218-248.

HUFFORD, L and DICKISON, W. 1992. A phylogenetic analysis of Cunoniaceae. Syst. Bot. 17: 181-200.

MADDISON, W., DONOGHUE, M., and MADDISSON, D. 1984. Outgroup analysis and parsimony. Syst. Zool. 33: 83-103.

MANOS, P. S., NIXON, K. C., and DOYLE, J. J. 1993. Cladistic analysis restriction site variation within the chloroplast DNA invert repeat region of selected Hamamelidae. Syst. Bot. 18: 551-562.

METCALFE, C. R. and CHALK, L. 1988. Anatomy of the dicotyledons. Vol. 1. Oxford University Press.

MULLER, C. H. 1960. Fagaceae. Ann. Missouri Bot. Gard. 47: 95-104.

ROBERTSON, K. R. and ROHRER, J. R. 1987. Are the apples pears? Illinois Natural History Survey Reports. 267. Champaign, Illinois.

RUTISHAUSER, R. and DICKISON, W. C. 1989. Developmental morphology of stipules and systematics of the Cunoniaceae and presumed allies. I. Taxa with interpetiolar stipules. Bot. Helvetica 99: 47-169.

STERLING, C. 1966. Comparative morphology of the carpel in the Rosaceae IX. *Spiraeoideae: Quillajeae, Sorbarieae.* Amer. J. Bot. 53: 951-960.

TAKHTAJAN, A. L. 1980. Outline of the classification of flowering plants (Magnoliophyta). Bot. Review 46. 225-359.

THORNE, R. F. 1983. Proposed new realignments in the angiosperms. In : New evidence of relationships and modern systems of classification of the angiosperm. Symp. Proc. (eds. F. EHRENDORFER, R. DAHLGREN) Nord. J. Bot. 3: 85- 117.

APÉNDICE

Tabla 2. 3 a Matriz de observaciones de 13 taxones incluido *Brunellia* y 19 caracteres. *Connarus* como grupo externo. Polarizados los caracteres 4 y 5. Inactivados los caracteres 9, 15 y 19.

Taxones	Caracteres																		
	1	2	3	4	5	6	7	8	9	1 0	1 1	1 2	1 3	1 4	1 5	1 6	1 7	1 8	1 9
Connarus	0	2	?	0	0	0	0	0	0	0	0	0	0	0	0	0	0	?	0
Brunellia	?	1	1	0	1	1	?	0	0	0	0	0	1	1	1	?	0	1	1
Ackama	1	0	0	0	2	0	0	1	0	1	0	1	0	0	0	0	0	0	1
Acrophyllum	2	0	0	0	2	0	0	1	0	1	0	1	0	0	0	0	0	1	1
Aphanopetalum	1	1	0	0	0	0	1	1	1	1	1	0	0	0	0	0	?	1	1
Codia	1	0	1	0	1	?	0	1	0	1	?	?	0	0	0	0	0	1	1
Eucryphia	1	0	0	1	?	0	1	1	0	1	1	1	0	1	0	1	2	1	1
Gillbeea	1	1	0	0	0	0	1	1	0	1	0	?	0	1	0	0	0	?	1
Pancheria	?	0	1	0	1	0	0	1	0	1	1	0	0	0	0	0	1	1	1
Pseudoweinmannia	1	1	0	?	1	1	0	1	0	1	0	1	0	0	0	1	0	1	1
Weinmannia	1	0	?	?	1	0	0	1	0	1	1	1	0	0	0	0	0	?	1
Spira-Acsmithia	?	0	0	1	0	1	?	0	0	0	0	?	1	?	0	?	0	1	1
Spiraeopsis	1	0	0	2	?	0	0	1	0	1	0	1	0	1	0	0	0	1	1

Tabla 2. 3 b Matriz de observaciones de 13 taxones incluidos. *Brunellia* y 19 caracteres. *Connarus* como grupo externo. No polarizados los caracteres 1, 2, 3, 4, y 5. Inactivados los caracteres 9, 15 y 19.

Taxones	Caracteres 1 2 3 4 5 6 7 8 9 1 1 1 1 1 1 1 1 1 1 0 1 2 3 4 5 6 7 8 9
Connarus	0 2 ? 1 2 0 0 0 0 0 0 0 0 0 0 0 0 ? 0
Brunellia	? 1 1 1 1 1 ? 0 0 0 0 1 1 1 ? 0 1 1
Ackama	1 0 0 1 0 0 0 1 0 1 0 1 0 0 0 0 0 0 1
Acrophyllum	2 0 0 1 0 0 0 1 0 1 0 1 0 0 0 0 0 1 1
Aphanopetalum	1 1 0 1 2 0 1 1 1 1 0 0 0 0 0 ? 1 1
Codia	1 0 1 1 1 ? 0 1 0 1 ? ? 0 0 0 0 0 1 1
Eucryphia	1 0 0 0 ? 0 1 1 0 1 1 1 0 1 0 1 2 1 1
Gillbeea	1 1 0 1 2 0 1 1 0 1 0 ? 0 1 0 0 0 ? 1
Pancheria	? 0 1 1 1 0 0 1 0 1 1 0 0 0 0 0 1 1 1
Pseudoweinmannia	1 1 0 ? 1 1 0 1 0 1 0 1 0 0 0 1 0 1 1
Weinmannia	1 0 ? ? 1 0 0 1 0 1 1 1 0 0 0 0 0 ? 1
Spira-Acsmithia	? 0 0 0 2 1 ? 0 0 0 0 ? 1 ? 0 ? 0 1 1
Spiraeopsis	1 0·0 2 ? 0 0 1 0 1 0 1 0 1 0 0 0 1 1

Tabla 2. 4 Matriz de observaciones de 13 taxones incluidos *Brunellia* y 19 caracteres. *Davidsonia* como grupo externo. Todos los caracteres polarizados con excepción de los caracteres 1 y 3. Inactivados los caracteres 9, 15 y 19.

Taxones	Caracteres 1 2 3 4 5 6 7 8 9 1 1 1 1 1 1 1 1 1 1 0 1 2 3 4 5 6 7 8 9
Davidsonia	0 0 ? 0 0 0 0 0 0 0 0 0 0 0 0 0 0 1 1
Brunellia	? 0 1 1 1 0 ? 1 0 1 1 1 0 1 1 ? 0 1 1
Ackama	1 1 0 1 0 1 0 0 0 0 1 0 1 0 0 0 0 0 1
Acrophyllum	2 1 0 1 0 1 0 0 0 0 1 0 1 0 0 0 0 1 1
Aphanopetalum	1 0 0 1 2 1 1 0 1 0 0 1 1 0 0 0 ? 1 1
Codia	1 1 1 1 1 ? 0 0 0 0 ? ? 1 0 0 0 0 1 1
Eucryphia	1 1 0 0 ? 1 1 0 0 0 0 0 1 1 0 1 2 1 1
Gillbeea	1 0 0 1 2 1 1 0 0 0 1 ? 1 1 0 0 0 ? 1
Pancheria	? 1 1 1 1 1 0 0 0 0 0 1 1 0 0 0 1 1 1
Pseudoweinmannia	1 0 0 ? 1 0 0 0 0 0 1 0 1 0 0 1 0 1 1
Weinmannia	1 1 ? ? 1 1 0 0 0 0 0 1 0 0 0 0 ? 1
Spira-Acsmithia	? 1 0 0 2 0 ? 1 0 1 1 ? 0 ? 0 ? 0 1 1
Spiraeopsis	1 1 0 2 ? 1 0 0 0 0 1 0 1 1 0 0 0 1 1

Tabla 2. 5 Matriz de observaciones de 13 taxones incluido *Brunellia* y 18 caracteres. *Fothergilla* como grupo externo. Caracteres polarizados con excepción de los caracteres 1, 3 y 17. Inactivados los caracteres 9, 15, y 19.

Taxones	Caracteres 1 2 3 4 5 6 7 8 9 1 1 1 1 1 1 1 1 1 0 1 2 3 4 5 6 7 8 9
Fothergilla	0 0 ? 0 0 0 0 0 ? 0 0 0 0 0 0 1 ? 0 1
Brunellia	? 1 1 1 1 0 ? 1 0 0 0 0 1 0 1 ? 0 1 1
Ackama	1 0 0 1 0 1 0 0 0 1 0 1 0 1 0 0 0 0 1
Acrophyllum	2 0 0 1 0 1 0 0 0 1 0 1 0 1 0 0 0 1 1
Aphanopetalum	1 1 0 1 2 1 1 0 1 1 1 0 0 1 0 0 ? 1 1
Codia	1 0 1 1 1 ? 0 0 0 1 ? ? 0 1 0 0 0 1 1
Eucryphia	1 0 0 0 ? 1 1 0 0 1 1 1 0 0 0 2 2 1 1
Gillbeea	1 1 0 1 2 1 1 0 0 1 0 ? 0 0 0 0 0 ? 1
Pancheria	? 0 1 1 1 1 0 0 0 1 1 0 0 1 0 0 1 1 1
Pseudoweinmannia	1 1 0 ? 1 0 0 0 0 1 0 1 0 1 0 2 0 1 1
Weinmannia	1 0 ? ? 1 1 0 0 0 1 1 1 0 1 0 0 0 ? 1
Spira-Acsmithia	? 0 0 0 2 0 ? 1 0 0 0 ? 1 ? 0 ? 0 1 1
Spiraeopsis	1 0 0 2 ? 1 0 0 0 1 0 1 0 0 0 0 0 1 1

Tabla 2. 6 Matriz de observaciones de 13 taxones incluido *Brunellia* y 19 caracteres. *Quercus* como grupo externo. Caracteres polarizados con excepción de 1, 3, 10, 14 y 16. Inactivados los caracteres 9, 15 y 19.

Taxones	Caracteros 1 2 3 4 5 6 7 8 9 1 1 1 1 1 1 1 1 1 1 0 1 2 3 4 5 6 7 8 9
Quercus	0 0 ? 0 0 0 0 0 ? ? 0 0 ? 0 ? 0 ? 0 0 0
Brunellia	? 1 1 0 1 0 ? 1 1 0 0 0 1 0 1 ? 0 0 1
Ackama	1 0 0 0 0 1 1 0 1 1 0 1 0 1 0 0 0 1 1
Acrophyllum	2 0 0 0 0 1 1 0 1 1 0 1 0 1 0 0 0 0 1
Aphanopetalum	1 1 0 0 2 1 0 0 0 1 1 0 0 1 0 0 ? 0 1
Codia	1 0 1 0 1 ? 1 0 1 1 ? ? 0 1 0 0 0 0 1
Eucryphia	1 0 0 1 ? 1 0 0 1 1 1 1 0 0 0 1 2 0 1
Gillbeea	1 1 0 0 1 1 0 0 1 1 0 ? 0 0 0 0 0 ? 1
Pancheria	? 0 1 0 1 1 1 0 1 1 1 0 0 1 0 0 1 0 1
Pseudoweinmannia	1 1 0 ? 1 0 1 0 1 1 0 1 0 1 0 1 0 0 1
Weinmannia	1 0 ? ? 1 1 1 1 0 1 1 1 0 1 0 0 0 ? 1
Spira-Acsmithia	? 0 0 1 2 0 ? 1 1 0 0 ? 1 ? 0 ? 0 0 1
Spiraeopsis	1 0 0 2 ? 1 1 0 1 1 0 1 0 0 0 0 0 0 1

Chapter 3

LEAF ANATOMY IN *BRUNELLIA* RUIZ & PAVÓN[1]

CLARA INÉS OROZCO AND BERTHA COBA[2]

ABSTRACT

The leaf anatomy of 24 species of *Brunellia* was studied, including observations on the stomata and nodal anatomy. The nodal anatomy of *Brunellia* was observed as unilacunar. Anatomical characters of the petiole and lamina were found to have taxonomic value at species level. The vascular cylinder is usually continuous in the middle and distal parts of the petiole, while, with few exceptions, it is discontinuous in the proximal part near the node. The adaxial portion of the vascular cylinder is compressed and in a few species semicircular or lumpy-shaped. Cortical bundles are frequent at the distal and middle part of the petiole and in the middle of the petiole, while medullary bundles are only found in *B. foreroi*. Stomatal crypts or undulations and the presence of a hypodermis were found to be among the taxonomically useful characters of the lamina. Reduction of lateral leaflets to form conspicuous or inconspicuous stipels on the petiole resulting in the appearance of a simple leaf was observed. Stomata are actinocytic except in *B. cutervensis* which has anomocytic stomata.

RESUMEN

Se estudió la anatomía de la hoja de 24 especies de *Brunellia*, incluyendo observaciones de estomas y de anatomía nodal. La anatomía nodal de *Brunellia*

---

se observó unilacunar. Se encontró que caracteres anatómicos del pecíolo y la lámina tienen valor taxonómico. El cilindro central del pecíolo es usualmente continuo en la parte distal y media del pecíolo, mientras que fue observado con pocas excepciones discontinuo en la parte proximal al nodo. La parte adaxial del cilindro vascular del pecíolo es comprimida y en pocas especies los haces vasculares tienen la apariencia de formar arcos. Haces corticales son frecuentes en la parte distal y media del pecíolo, mientras que haces medulares se encuentran en *B. foreroi*. Entre los carateres taxonómicos más útiles de la lámina se encuentra la presencia de criptas o undulaciones y la presencia de hipodermis. La reducción foliar para formar conspicuas o inconspicuas estipelas sobre el pecíolo se observó anatómicamente en muchas especies con apariencia de hojas simples. Los estomas son actinocíticos excepto *B. cutervensis* con estomas anomocíticos.

3.1 INTRODUCTION

Brunellia has been considered the single genus of Brunelliaceae (Cuatrecasas 1970, 1985) and has about 53 species (Orozco in preparation). The species are distributed in the Neotropics from Panama to Bolivia and also in Central America and the Greater Antilles. Species grow in humid high Andean or subandean woodlands.

Dickison (1989) comments on the wide agreement among phylogeneticists that Cunoniaceae, Davidsoniaceae, Brunelliaceae, and Eucryphiaceae are closely related, forming a coherent unit which can be considered as primitive Rosidae due to the basal position they occupy within the Rosales complex. Currently the family Brunelliaceae is placed in the order Oxalidales (APG 1998) and recently Bradford (pers. comm.) confirms *Brunellia* as the only genus of the family. However, Hufford and Dickison (1992) proposed that *Brunellia* should be part of the Cunoniaceae, and Orozco (1997) suggested that due to the relationships among *Brunellia, Spiraeanthemum* and *Acsmithia* (Cunoniaceae), these three genera could be included in a family separate from Cunoniaceae.

Species of *Brunellia* show wide morphological variation, especially in the shape and consistency of the lamina. Leaves are opposite, or less frequently in whorls and can be composed of various leaflets or of just one foliar lamina. In some species with one foliar lamina, stipels on the petiole are present. Species with one foliar lamina and no stipels on the petiole have been considered to have simple leaves (Cuatrecasas 1970, 1985). The leaves have long or short petioles, where stipels are frequently present, however, stipels are also found on the rachis of the multifoliolate species. Petioles are ventrally sutured and in some cases have longitudinal furrows.

Anatomical studies of *Brunellia* are scarce. There are only the contributions of Metcalfe and Chalk (1965) on the foliar lamina and Eyde

(1970) on the foliar lamina and wood (*B. tomentosa, B. comocladifolia,* and *B. putumayensis*). Due to the wide morphological variations in *Brunellia*, this study was carried out with the objective of using the leaf anatomy to differentiate species of the genus. The information will be used to establish the relationships between the species, and it is hoped that this will provide a clearer understanding of the relationships of *Brunellia* with other taxa of Oxalidales.

3.2 MATERIALS AND METHODS

The anatomy of the foliar lamina of 24 species of *Brunellia*, the petiole of 18, and the nodal anatomy of 9 species, was studied under a light microscope. Stomata were also studied in 10 species (Table 3.1). Data for stomata were studied for 4 species by scanning electron microscope (SEM). The species selected for analysis represent the total morphological variation of the genus.

In order to observe stomata under the light microscope, the epidermis was obtained by disintegrating the mesophyll and then the strips of lamina were treated with 5% sodium hypochlorite and water in equal parts. The 0.5 cm^2 cuts of lamina were left in the solution for 5 days, washed and stained with 5% carmine in water.

Samples of lamina and petiole were fixed in 70% alcohol, hand-cut and bleached in 5% sodium hypochlorite, washed and coloured with methyl green and aluminic carmine (Salomon and Cantino 1983) and sealed with glycerine jelly. Sections of the lamina were taken at the center, and for some species at the proximal and distal levels of the central midrib. In the petiole, sections were made at the basal, middle, distal part and close to the base of the lamina.

The dry lamina material was cut into 2 cm sections and rehydrated by boiling and then hand-sectioned as above. In order to observe the nodal anatomy, cuts were made at the node and three levels above and below it. They were cut into small pieces and rehydrated by boiling in water and glycerine, and sectioned by microtomy. Resulting sections were stained with fast green and safranin. Most of the preparations were photographed whith a Wild Makroskope, M420,125.

3.3 RESULTS

3.3.1 Nodal anatomy

All species observed have a unilacunar nodal structure (Fig. 3.1 a, b,) with the exception of *B. darienensis* which has two additional incipient pairs of lacunae associated with the proximal stipules (Fig. 3.1 d). Additional lacunae were often observed at both levels immediately below and above the node (Fig. 3.1 e, f). These additional lacunae correspond to those left by the bundles

Table 3.1 Species of *Brunellia* used in the anatomical analysis.

Species	Collections	Node	Petiolo	Petiolulo	Stomata	Lamina
B. acostae Cuatrec.	Cuamaçás & Gudiño, E. et al. 243	+				
	Nell, D. et al. 9789	+				
	Palacios, W. et al. 4885	+	+			
B. acutangula H. & B.	Orozco, C.I. et al. 1961		+			+
	Orozco, C.I. et al. 1247		+			+
	Orozco, C.I. et al. 1963					+
	Orozco, C.I. et al. 1083		+			+
B. amayensis Orozco	Orozco, C.I. et al. 1426		+			+
B. boliviana Cuatrec.	Lewis, M. et al. 40829		+	+		
B. boqueronensis Cuatrec.	Orozco, C.I. et al. 1458		+		+	+
	Orozco, C.I. et al. 1639		+			+
B. cayambensis Cuatrec.	Orozco, C.I. et al. 1187	+	+	+		
B. comocladifolia H. & B.	Orozco, C.I. et al. 1163A					
	Orozco, C.I. et al. 1133		+			
	Orozco, C.I. et al. 1265		+			+
B. cutervensis Cuatrec.	Diaz, C. et al. 2270		+		+	+
B. darienensis Cuatrec. & Porter	Orozco, C.I. et al. 1539	+	+			+
B. dulcis Macbride	Orozco, C. I. et al. 1522		+			+
B. ecuadorensis Cuatrec.	Palacios, W. et al. 3705		+	+	+	+
B. elliptica Cuatrec.	Orozco, C.I. et al. 2970		+			+
B. foreroi Orozco	Forero, E. et al. 2260	+	+		+	+
B. glabra Cuatrec.	Lozano, G. et al. 6709		+		+	
B. goudotii Cuatrec.	Orozco, C.I. et al. 1129		+			+
B. integrifolia Szyszyl.	Orozco, C.I. et al. 1640		+			+
	Orozco, C.I. et al. 1589	+				
	Orzoco, C.I. et al. 1596	+	+		+	
B. littlei Cuatrec.	Orozco, C.I. et al. 2894		+			+
B. macrophylla Cuatrec.	Orozco, C.I. et al. 2784		+		+	+
	Orozco, C.I. et al. 2812		+			+
B. morii Cuatrec.	Davidse, G. et al. 25489		+			+
B. ovalifolia H. & B.	Van der Werff, H. et al.9406	+	+		+	
B. pauciflora Orozco & Cuatrec.	Palacios, W. et al. 6973	+				
	Palacios, w. et al. 5273	+	+			
	Tipaz, G. et al. 23	+	+			
B. penderiscana Cuatrec.	Orozco, C.I. et al. 2800		+			+
B. pitayensis Cuatrec.	Orozco, C.I. et al. 2901		+			+
B. propinqua H. & B.	Orozco, C.I. et al. 1767		+			+
B. putumayensis Cuatrec.	Orozco, C.I. et al. 1201		+			+
B. racemifera Tul.	Orozco, C.I. et al. 1100		+	+		+
B. rufa Cuatrec.	Orozco, C.I. et al. 2853		+		+	+
B. sibundoya Cuatrec.	Orozco, C.I. et al. 1163A		+			
	Orozco, C.I. et al. 1099		+			+
B. stuebelii Hieronymus	Orozco, C.I. et al. 1187		+			
	Orzoco, C.I. et al. 1176	+	-			
B. subsessilis Killip & Cuatrec.	Lozano, G. 3968		+			+
B. tomentosa H. & B.	Gudiño, E. et al. 82	+				
	Orozco, C.I. et al. 1175	+	+			
	Orozco, C.I. et al. 1214	+				
	Orozco, C.I. et al. 1217		+			+
	Orozco, C.I. et al. 1218		+			+
	Orozco, C.I. et al. 1176		+			+
B. trigyna Cuatrec.	Lozano, G. 5537					
	Orozco, C.I. et al. 2975		+	+		
	Orozco, C.I. 2916		+	+		+
B. velutina Cuatrec.	Lozano,G. et al. 4860	+				

that vascularise the stipules. In relation to the nodal structure and the vascular bundle traces, DICKISON (pers. comm.) considered the presence of a multilacunar node in *Brunellia*. According to DICKISON (pers. comm.) the vascular bundle observed at the node (Fig. 3.1 a, b) does not correspond to initial traces but they are the product of trace divisions of the initial ones.

3. 3. 2 Petiole

Most of the species have wax on the epidermis of the petiole (Fig. 3.2 b). The epidermal cells are longer than cells of the cortical tissues and they vary in shape (Fig. 3.2 a-c). They can end in a point or can be round, squared, or rectangular.

Two layers of hypodermis are usually present in the petiole, (Table 3.2), with the exception of *B. boliviana*, *B. boqueronensis*, and *B. acostae* having just one layer. In *B. cayambensis* and *B. trigyna* the hypodermis is absent.

Angular collenchyma is present in most species (Fig. 3.2 b), whereas laminar collenchyma was observed only in *B. boliviana*, *B. ecuadorensis*, and *B. elliptica* (Fig. 3.2 c). The cells of the cortical and medullated parenchyma are polyhedral in shape with abundant druses in some species. Sclerenchyma fibres protect the vascular bundles externally, except in *B. pauciflora* (Fig. 3.3 e) whose vascular system is surrounded by angular collenchyma.

The principal vascular system of the petiole is cylindrical and medullated. Cortical vascular bundles are usually present (Fig. 3.3 a-c), with the exception of *B. acostae*, *B. ecuadorensis*, and *B. pauciflora* (Fig. 3.3 d-f) which have no cortical bundles at any level of the petiole. In other species, cortical bundles are not present along the petiole, for example, cortical bundles were not observed at any level of the petioles of *B. boliviana* and *B. foreroi* (Fig. 3.3 g, h). Medullated bundles were only present in *B. foreroi* which were observed at the middle levels but may also be present at the proximal levels (Figs. 3.3 h, 3.4 d).

The petiole is generally bifacial at almost all levels, with some exceptions as in Fig. 3.3 f, g. The adaxial portion is compressed or flat (Fig. 3.3 d, e), semicircular, lumpy-shaped, and discontinuous (Fig. 3.3 a) or convex (Fig. 3.3 b). The presence of cortical bundles is almost always associated with the adaxial band of the petiole. They are separated from the principal vascular system of the petiole and can be incorporated into the vascular system of the mesophyll. In some species such as *B. ovalifolia*, a central bundle is observed in the abaxial part of the main vascular bundle of the petiole (Fig. 3.3 c).

In most of the species with only one foliar lamina, the anatomical sections, usually at the proximal levels of the petiole, show bundles like wings, incorporated or not into the central vascular system (Fig. 3.4 a-d). These bundles correspond to the vascularization of reduced leaflets which were also observed
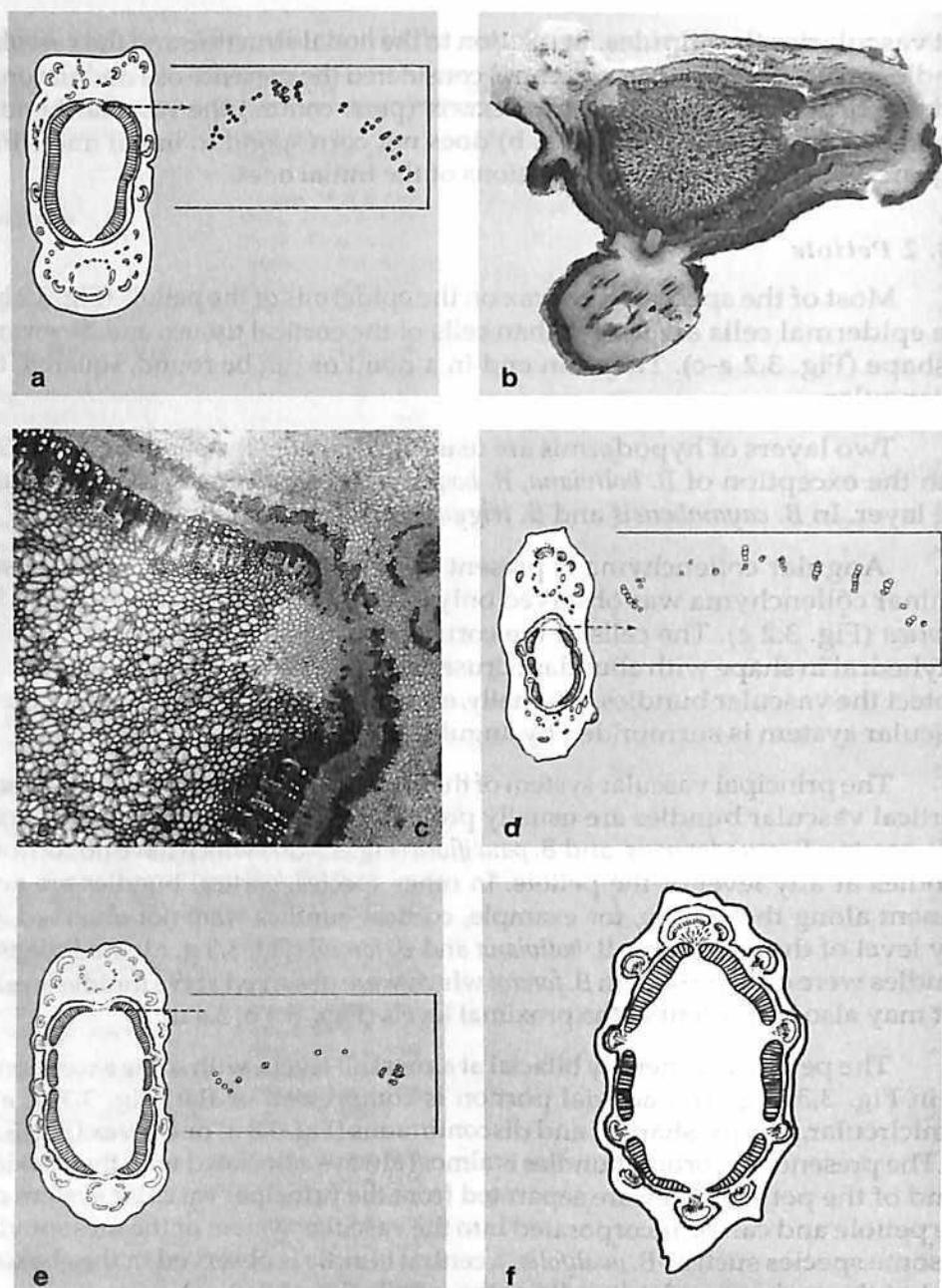
Fig. 3.1 Nodal and internodal anatomy of *Brunellia*, estereoscope observations. (a) Node of *B. acostae*. (b) Node of *B. glabra*. (c) Detail of the node with vascular bundles. (d) Node of *B. darienensis*. (e) Level immediately below the node of *B. acostae*. (f) Level immediately below the node of *B. foreroi*. (a) 36x (b) 146x (c) 230x (d) 36x (e) 83x (f) 93x.

at the distal level (Figs. 3.3 a, b). In some species, specially those with one foliar lamina, reduced folioles are externally seen as conspicuous stipels on the petiole.

3. 3. 3 Petiolule

There are no major differences in the anatomy of the petiolule and the vascularization is similar to that of the petiole (Figs. 3.4 e-h). In *B. boliviana*, the petiolule is monofacial, at the distal level but bifacial at the proximal level (Fig. 3.4 g, h).

3. 3. 4 Foliar lamina

The presence of papillae on the abaxial epidermis, crypts, undulations or straight shape of the abaxial epidermis, presence or absence of hypodermis, and the shape of the central vascular bundle are the differentiating characters among species (Table 3. 3). Hairs are present in all species with few exceptions. A study of the variation of leaf indument was carried out by OROZCO (1999).

The cells of the upper epidermis have thick walls. The cells are triangular, rectangular, square and occasionally polyhedral (Fig. 3.5). In many species, periclinal divisions can be seen in the epidermis, especially close to the vascular bundles, giving the appearance of having various layers (Fig. 3.5 a).

The upper epidermis has a thick cuticle, which varies in thickness, between 2.5 μm in *B. macrophylla* to 200 μm in *B. pauciflora*. The cuticle forms layers of wax which vary in number and shape (Fig. 3.5 a). The cells of the lower epidermis have thin walls and form dense papillae in all species except, *B. cutervensis* (Fig. 3.5 b). The papillae are cylindrical, and look like short or long fingers, with a surface varying from smooth or soft to densely striated (Fig. 3.6 a- d). On the lower side of the epidermis, some species present crypts or undulations (Fig. 3. 5 c, e) or the epidermis is absolutely straight (Fig. 3.5 b).

Stomata are present in the lower epidermis. SEM observations show sunken or raised stomata (Fig. 3.6 c, d). Stomata under light microscope (LM), (Table 3.1) are mostly actinocytic due to the radial arrangement of the accompanying cells, (Fig. 3.6 e), except in *B. cutervensis* which has anomocytic stomata (Fig. 3.6 f).

3. 3. 5 Mesophyll

The mesophyll is dorsiventral, with clear differentiation between the palisade and spongy tissues with the exception of some species having a transition area between the two zones. Variation in the mesophyll consists mainly of the presence or absence of a hypodermis. Hypodermal cells are arranged parallel to the surface, have round edges and are elliptical, square,
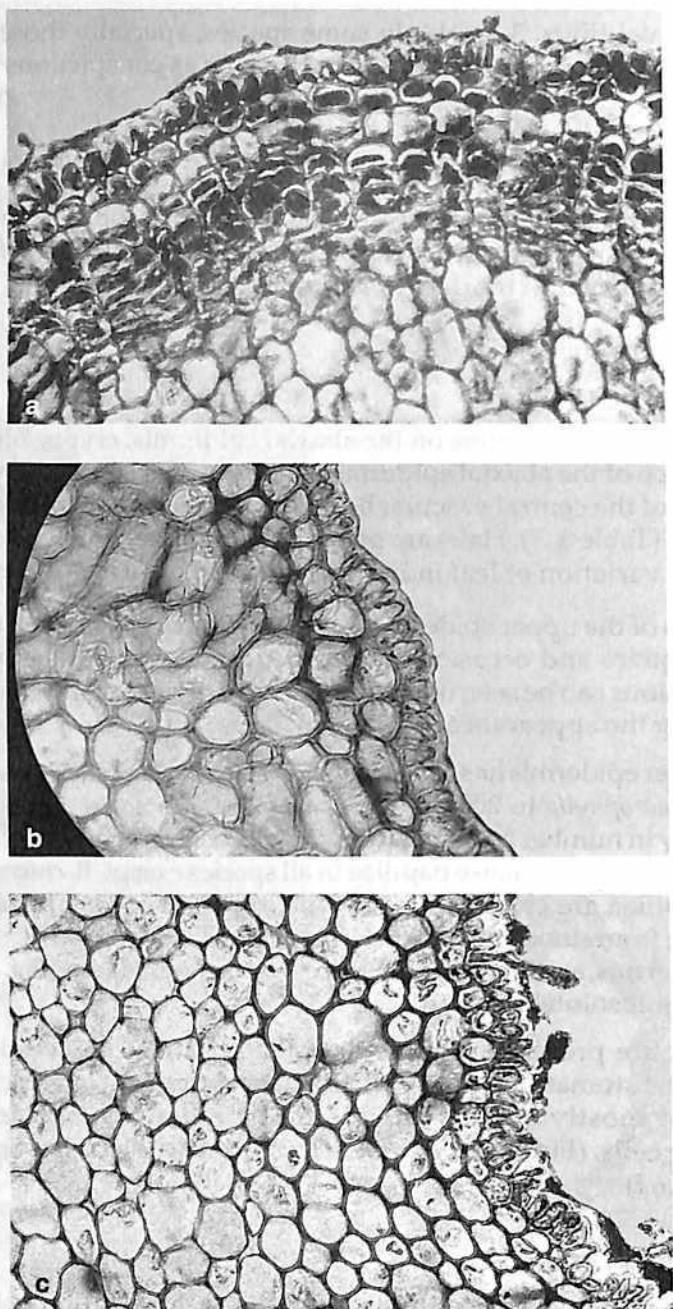
Fig. 3. 2 Cortical section of the petiole of *Brunellia*, light microscope observations. **(a)** Presence of epicuticular wax and cork layer in *B. trigyna*. **(b)**. Epidermis of extended cells, lumen ending in a point, and epicuticular wax in *B. rufa*. **(c)** Laminar collenchyma in *B. boliviana*. **(a)** 313x **(b,c)** 300x.

or rectangular (Fig. 3.5 d). Fourteen of the 24 species examined have no hypodermis (Table 3.3) although they do have periclinal divisions of the epidermis which could in some cases be confused with a hypodermis (Fig. 3.5 a). The number of layers of hypodermis ranges from 1 in *B. foreroi* to 4 in *B. pauciflora* (Table 3.3).

The number of layers of spongy and palisade tissue varies from one species to another. *B. acostae*, *B. darienensis*, *B. integrifolia*, *B. subsessilis*, and *B. velutina* have the least number of layers of palisade (1-2). In other species (Table 3.3) there are 3-4 layers (Fig. 3.5 a, c). The number and size of cells vary, especially in the first and last layers. There were no great differences in the cell arrangement of the spongy tissue, but variations in the number of layers were observed (Table 3.3). The highest number of layers was observed in *B. boliviana*, *B. boqueronensis*, and *B. darienensis*, the lowest with 3-4, is most frequent.

3. 3. 5. 1 Vascular bundles

The vascular bundles of the mesophyll are connected to the lower and upper epidermis by sclerenchym fibers which vary in number within the same sample (Fig. 3.5 a, c, e).

Two bands, adaxial and abaxial, usually V-shaped, can be observed in the central vascular bundle of the midrib. The bands are separated by sclerenchymatous tissue which surrounds them inside and outside (Fig. 3.7). The adaxial and abaxial portions may be discontinuous due to the projection of the sclerenchymal cells towards the center of the bundle. Medullated bundles, apparently formed by evagination of the adaxial band were observed in *B. foreroi* (Fig. 3.7 d, h).

The abaxial band is continuous as in *B. acostae* and *B. racemifera* (Fig. 3.7 a, e) or interrupted as in *B. cayambensis*, *B. foreroi* and *B. stuebelii* (Fig. 3.7 b, d, f) (Table 3.2). In some species such as *B. cayambensis* and *B. racemifera* several packets of bundles are formed (Fig. 3.7 b, e).

There are some differences in the shape of the adaxial band. All species have an internally concave adaxial band with exception of *B. foreroi*, which is slightly convex (Fig. 3.7 d, h). The adaxial band is straight on the outside, or slightly compressed as in *B. acostae* and *B. stuebelii* (Fig. 3.7 a, f). In *B. glabra* there are prolongations at the ends (Fig. 3.7 c) whereas, the adaxial band is V-shaped in *B. racemifera* (Fig. 3.6 e), and convex in *B. foreroi* (Fig. 3.7 d).

3.3.5.2 Druses

Druses forming prisms are found in different parts of the foliar lamina, very often in the proximal cortical parenchyma and in the medullated

parenchyma of the central bundle. They are also found in the phloem of the central bundle, in the palisade parenchyma and close to the vascular bundles.

3.4 DISCUSSION

Some anatomical characters of the foliar lamina and the petiole are important for recognising species of *Brunellia* (Tables 3.2, 3.3). Among the
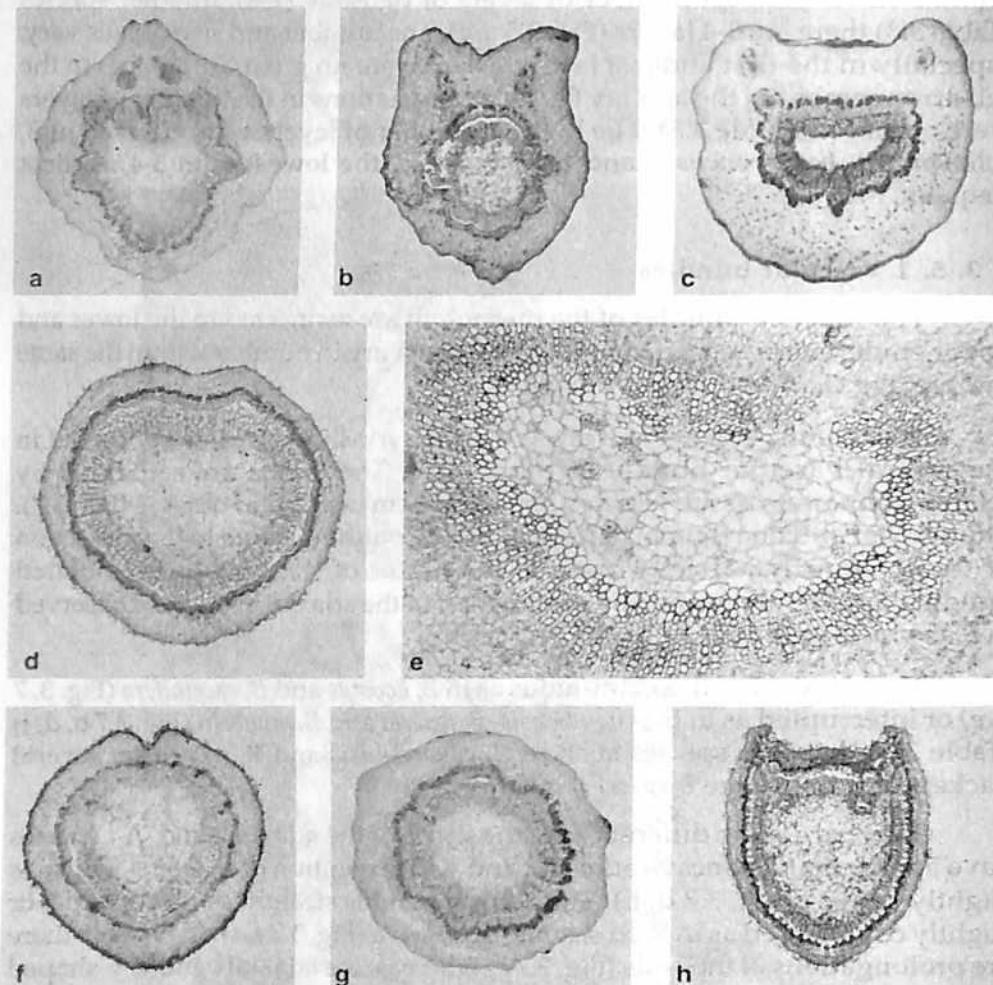
Fig. 3. 3 Anatomy of the petiole of *Brunellia*, stereoscope observations (a- e) Distal level. (a) *B. boliviana* (b) *B. macrophylla* (c) *B. ovalifolia* (d) *B. ecuadorensis* (e) light microscope observation, *B. pauciflora* (f-h) Medial level. (f) *B. acostae* (g) *B. boliviana* (h) *B. foreroi*. (a) 32X (b) 23X (c) 33X (d) 45X (e) 158X (f) 31X (g) 42X (h)15X.

anatomical characters which do not vary greatly between species are the position of the stomata in the lower epidermis, the actinocytic type of stomata and the presence of papillae on the lower epidermis. Exceptions were found in *B. cutervensis*, with anomocytic stomata and imperceptible papillae on the epidermis. In all the species studied, the mesophyll is dorsiventral, has vascular bundles running across it, and a vascular adaxial band slightly separated from
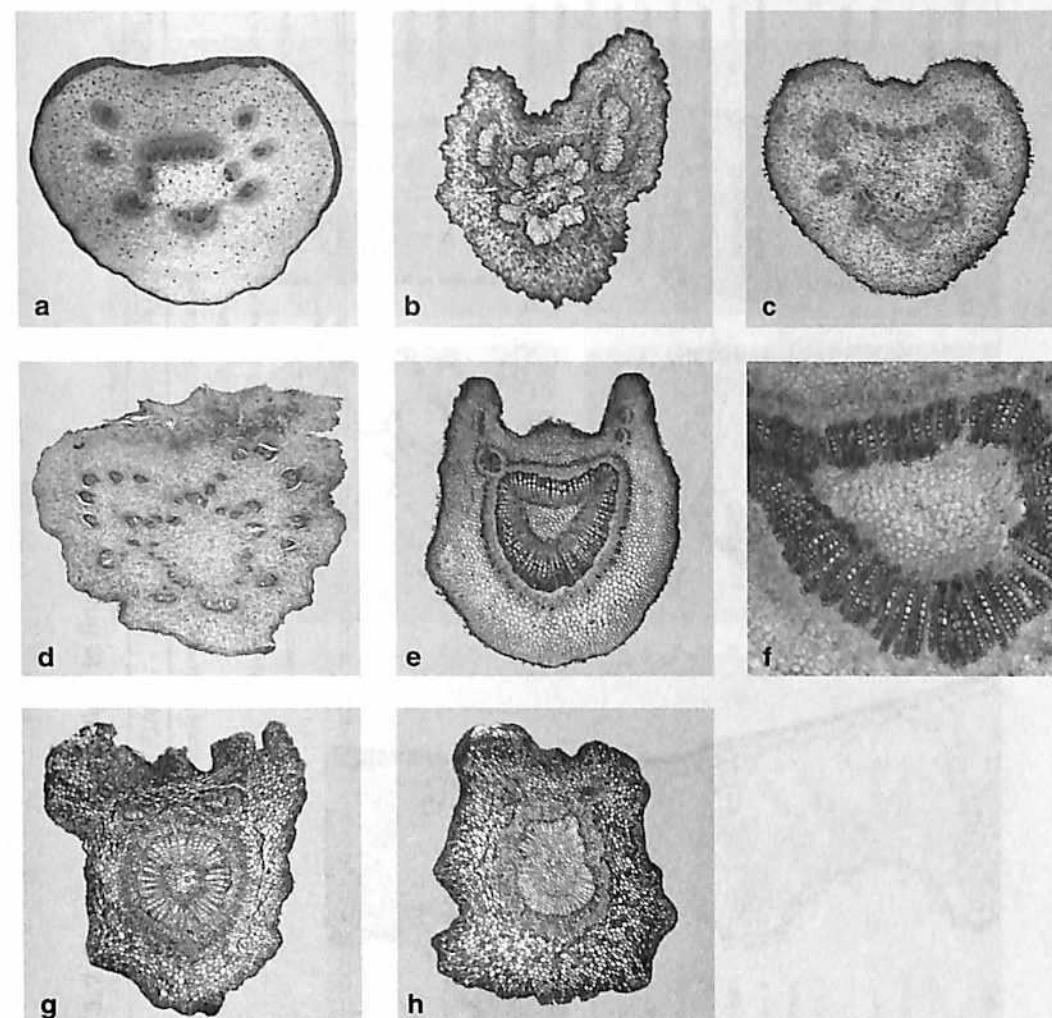
Fig. 3. 4 Anatomy of the petiole and petiolule of *Brunellia*, stereoscope observations. (a-d) Proximal level of the petiole. (a) *B. glabra* (b) *B. tomentosa* (c) *B. ecuadorensis* (d) *B. foreroi* (e-h) Petiolule, microscope observations. (e, g) Distal level (e) *B. acostae* (g) *B. boliviana* (f, h) Proximal level. (f) *B. acostae* (h) *B. boliviana*. (a) 27X (b) 60X (c) 23X (d) 18 X (e.g.h) 26 X (f) 30X .

the ventral band at the midrib. The separation of the bands occurs in some species from the proximal levels of the leaf, in the petiole and in the petiolules.

The presence of crypts and undulations in the lower portion of the foliar lamina has been related to plants growing in dry environments, but it has not been detected in any of the Cunoniaceae species in such environments (DICKISON 1975). For *Brunellia*, there was initially thought to be a correlation between
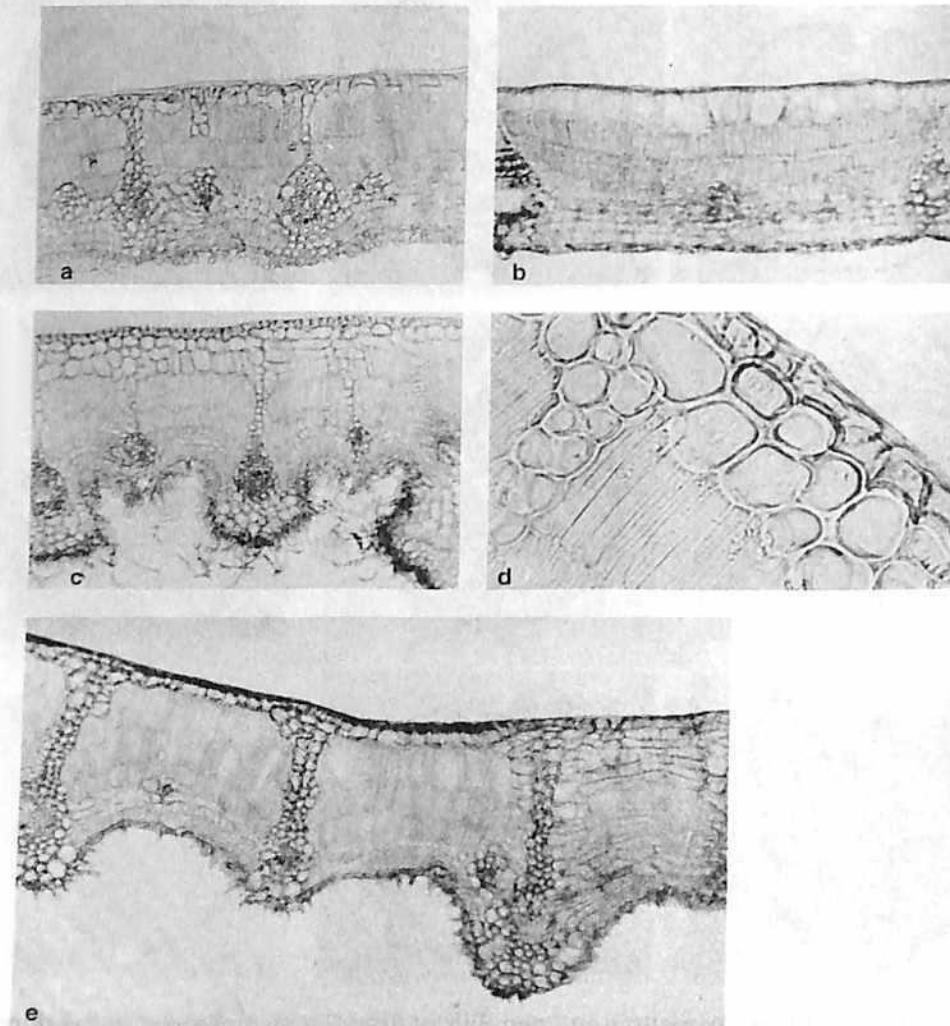
Fig. 3. 5 Anatomy of foliar lamina, light microscope observations. (a) *B. trigyna* (b) *B. cutervensis* (c) *B. rufa.* (d) Detail of the mesophyll of *B. rufa* (e) *B. ecuadorensis.* (a) 79X (b) 60X (c) 85 X (d) 336X (e) 212 X.

Table 3. 2 Characters of the petiole in *Brunellia.*

	CORTICAL				CENTRAL VASCULAR BUNDLE			
Species	Petiole wax	Hypodermis	Collenchyma (A= Angular, L= Laminar)	Accessory Vascular Bundles (Dis= distal level, Mid=Middle level, Pro= Proximal level)	Support Tissue Central Bundle (S= Sclerenchyma, C= Collenchyma)	Adaxial band Distal level	Adaxial band Middle level / Proximal level	Vascular bundles as foliar relicts
B. acostae	+	+	A		S	Ribbon-shaped	Continuous	Pro
B. boliviana	+	+	L	Dis	S	Ribbon-shaped	Continuous	Dis-Pro
B. boqueronensis	+	+	A	Dis-Mid-Pro	S	Compressed	Discontinuous	*?
B. cayambensis	+	Cork	A	Dis-Pro	S	Compressed	Discontinuous	Pro
B. cutervensis	+	+	A		S			Absent
B. darienensis	+	+	A	Dis-Mid-Pro?	C	Compressed	Continuous	Pro
B. ecuadorensis	+	+	L		S	Slight-compressed	Continuous	·
B. elliptica		+	L	Dis-Mid	S	Slight-compressed	Discontinuous	·
B. foreroi	+	+	A	Medullate-Mid	S	Compressed	Continuous	Pro
B. glabra	+	+	A	Dis-Mid-Pro	S	Compressed	Continuous	Pro
B. integrifolia	+	·	A	Dis	S	Convex	Discontinuous	Dis-Pro
B. macrophylla		·	A	Dis-Mid-Pro	S	Concave - compressed	Continuous	Absent
B. ovalifolia	+	+	A	Dis-Mid-Pro	S	Straight	Continuous	Absent
B. pauciflora	+	+	A		C	Compressed	Discontinuous	
B. pitayensis	+	+	A		S			
B. racemifera	+	+	A	Dis-Mid-Pro	S	Compressed	Continuous	Midi-Pro
B. rufa	+	+	A	Dis-Mid-Pro	S	Concave	Continuous	Pro
B. stuebelii		·	A		S			
B. subsessilis	+	+	A	Dis-Mid-Pro	S	Concave	Continuous	Dis
B. tomentosa	+	+	A	Dis	S	Compressed	Discontinuous	Pro
B. trigyna	+	Cork	A		S			
B. velutina	+	+	A		S			

this and the texture of the leaf, but *B. acutangula* and *B. trigyna* (Fig. 3.5 a) have a straight epidermis and leathery laminae, while the presence of crypts and undulations was observed in species with thin and leathery textures (Table 3.3). In *Brunellia* this could be associated with a low level of transpiration and minimum water loss through stomata. Slow transpiration takes place in many plants in humid forest due to the high relative humidity of the surrounding
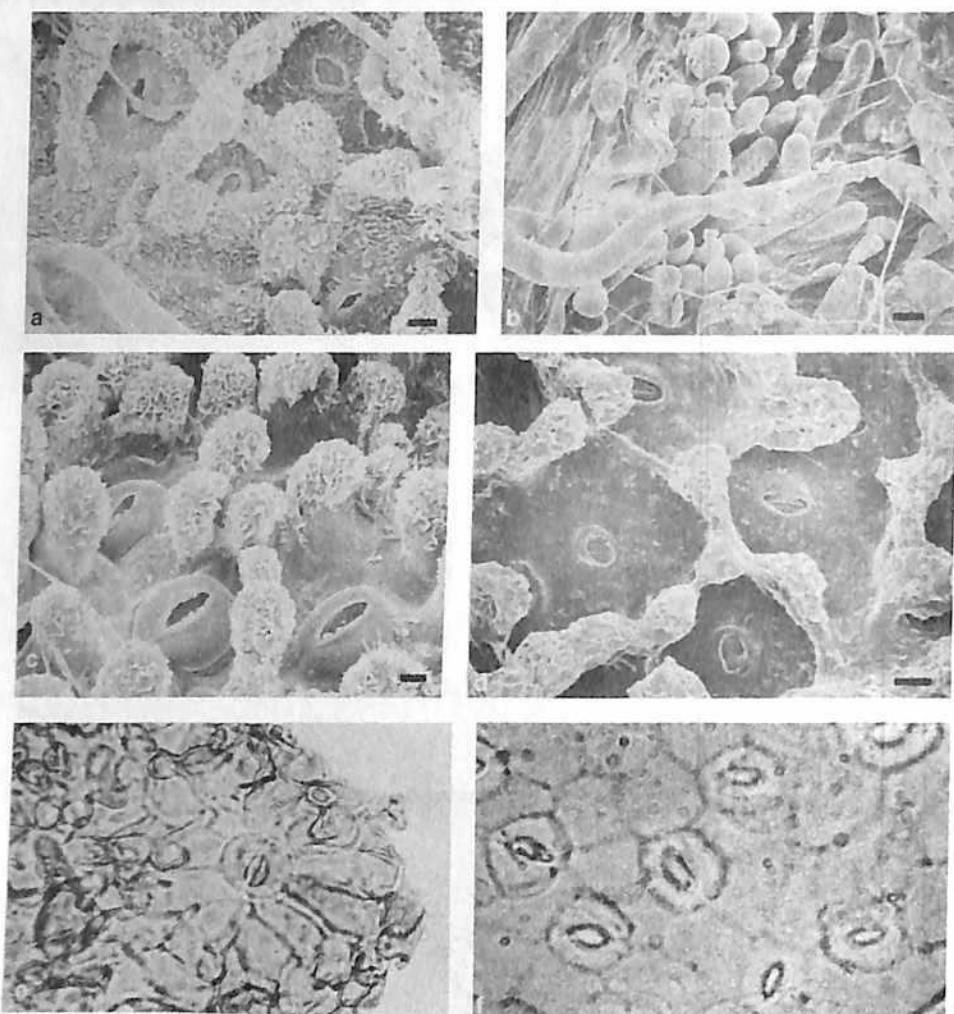
Fig. 3. 6 Abaxial epidermis of *Brunellia* under scanning electron microscope **(a-d)** and light microscope **(e-f)**. **(a)** Stomata sunken, short and striated papillae, epidermis with folds in *B. standleyana*. **(b)** Papillae forming long smooth fingers in *B. acostae*. **(c)** Stomata raised, short and striated papillae. **(d)** Submerged stomata, short and striated papillae in *B. trigyna*. **(e)** Actinocytic stomata in *B. ecuadorensis*. **(f)** Anomocytic stomata in *B. cutervensis*. **(a, b, c, d)** 10µ **(e)** 336 X **(f)** 1000 X.

air. There is a relationship between the presence or absence of crypts in the lower epidermis and the foliar area exposed. Species with several leaflets are more likely to have a straight lamina while crypts and undulations are more frequent in unifoliolate species which are usually found at higher altitudes (see chapter 1).

The presence of a hypodermis is another anatomical feature of some species of *Brunellia*, as observed by EYDE (1970) and the number of layers may be associated with the leatheriness of the lamina (Table 3.3). Thin laminae such as in *B. velutina* and *B. acostae* have no hypodermis, *B. cayambensis* and *B.*
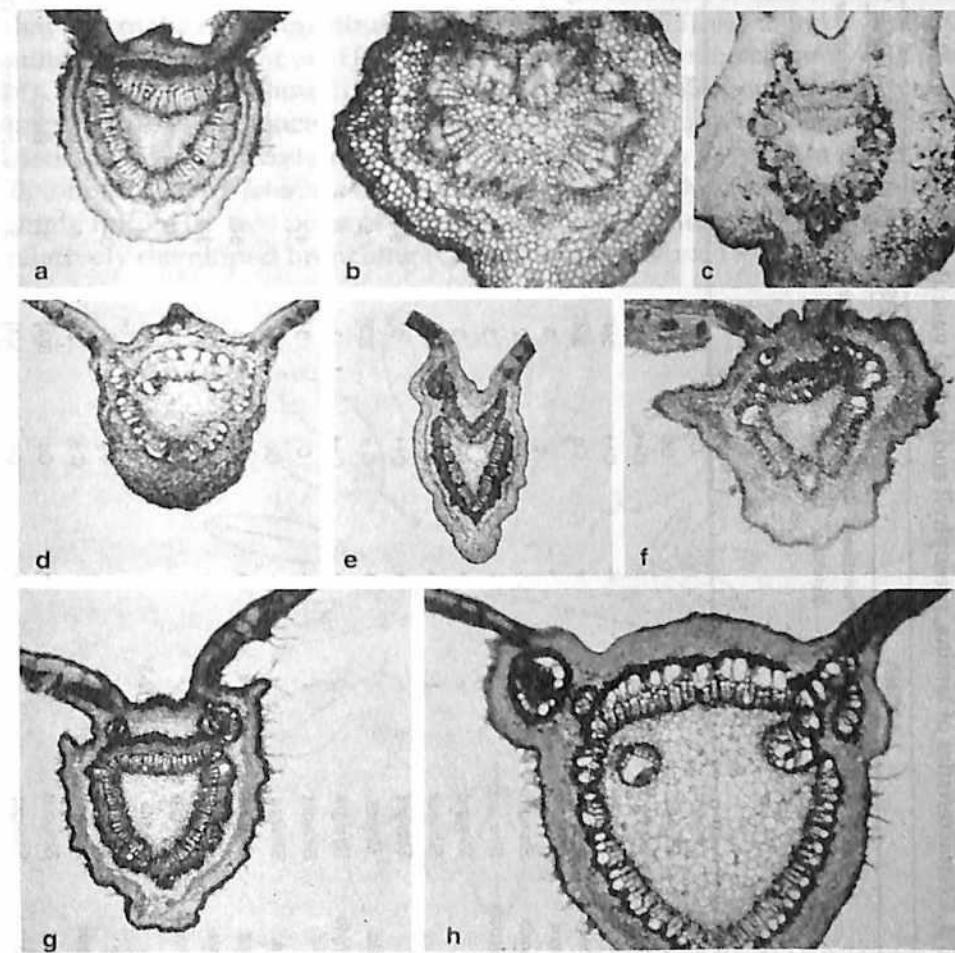
Fig. 3. 7 Central bundle of the midrib of the foliar lamina, ligth microscope observations. **(a)** *B. acostae* **(b)** *B. cayambensis* **(c)** *B. glabra* **(d)** *B. foreroi* **(e)** *B. racemifera* **(f)** *B. stuebelii* **(g)** *B. tomentosa* **(h)** *B. foreroi*, at proximal level of the midrib. **(a, d)** 6X **(b,h)** 16X **(c, e, f)** 7X **(g)** 11X.

Table 3. 3 Characteristics of lamina, mesophyll and central vascular bundle in *Brunellia*.

Species	LAMINA Lower epidermis	Hypodermis	Periclinal divisions	C=cartaceous Co=coriaceous (++ highly)(+ very)(- not very)	MESOPHYLL Palisade layers	Spongy layers	CENTRAL VASCULAR BUNDLE Adaxial band Outer	Inner	Abaxial band C=continuous D=discontinuous
B. acostae	Straight	-	-	C	2	5	Straight	Convex	C
B. acutangula	Straight	2(3)	-	Co	3-4	4-5	Straight	Convex	C
B. boliviana	Straight	-	+	C++	3	7	Concave	Concave	C
B. boqueronensis	Crypts	(1)3(2)	-	C++	3(4)	5-7	Straight	Straight	C
B. cayambensis	Straight	-	+	Co	3(4)	3	Compressed	Concave	D
B. cutervensis	Straight	-	+	C	3	6			C
B. darienensis	Straight	-	-	C	2	8	Straight	Straight	C
B. ecuadorensis	Sinuous	-	+	C	3	3			
B. elliptica	Straight	-	+	C+	3	5			
B. foreroi	Sinuous	1	+	C++	3	4	Concave	Concave	D
B. glabra	Straight	-	+	C+	3	5-6	Compressed	Convex	C
B. integrifolia	Crypts	-	+	C++	2(3)	5	Compressed	Concave	C
B. macrophylla	Straight	2(3)	+	C++	3	4			
B. morii	Straight	-	-	C	3	4-6			
B. ovalifolia	Straight	-	-	Co	3		Compressed	Concave	C
B. pauciflora	Sinuous	2-4	+	Co	3	5-6	Straight	Concave	C
B. pitayensis	Crypts	2	-	Co	3	3-4			C
B. racemifera	Straight	-	-	C	3	4	Compressed	Convex	C
B. rufa	Crypts	(1)2-3	-	C	3	4	Compressed	Convex	
B. stuebelii	Crypts	1	-	Co	3	3-4	Straight	Concave	
B. subsessilis	Straight	-	-	C+	2	5	Compressed	Concave	D
B. tomentosa	Sinuous	1-2	+	Co	3(4)	3	Straight	Concave	C
B. trigyna	Straight	-	+	C+	3(4)	5	Compressed	Concave	D
B. velutina	Sinuous	-	-	C	1	3-5			

*trigyna* are exceptions, with leathery laminae and no hypodermis. *B. pauciflora* has the highest number of layers of hypodermis, its leaves have a leathery texture while *B. foreroi* has only one layer and the foliar lamina is thin, both species have one foliar lamina. The texture of the foliar lamina could also be related to the number of layers of palisade parenchyma, *B. velutina* has only one layer and its foliar lamina is thin (Table 3.3).

There is little interspecific variation in the arrangement of the vascular tissues in the central bundle of the midrib. This variation seems to be constant with few exeptions. In *B. foreroi* the medullary bundles could contribute to the vascularisation of the laminae (Fig. 3.7 h). HOWARD (1974) does not consider that this makes any contribution. DICKISON (1973, 1975) also found medullary bundles in some genera of Connaraceae and in *Spiraeanthemum* and *Acsmithia* of Cunoniaceae. Although the homology of the medullary bundle structure is uncertain, its presence in Connaraceae, *Spiraeanthemum* and *Acsmithia* is associated with the relatively advanced state of these taxa within the group. Taxonomically, *B. foreroi* is easily recognizable from other *Brunellia* taxa with a single foliole by two pairs of stipels on the petiole and an inflorescence with relatively developed branching (OROZCO and WEBERLING 1999).
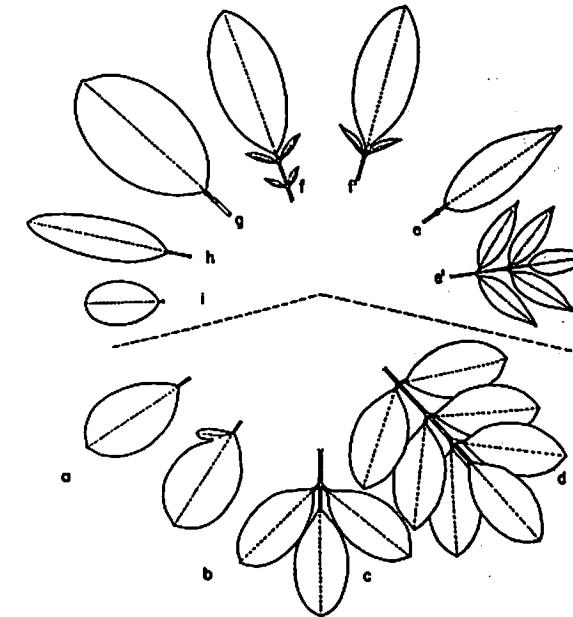

Fig. 3. 8 Morphological variations of the *Brunellia* leaf. (a-d) Developmental stages of the compound leaf in *B. propinqua*. (e, e') Developmental stages of the compound leaf in *B. darienensis*. (f, f') Developmental stages of the compound leaf in *B. amayensis*. (g) Reduced leaf in *B. foreroi*. (h) Reduced leaf in *B. rufa*. (i) Reduced leaf in *B. pauciflora*.

In *Brunellia*, as in *Acsmithia* and *Spiraeanthemum*, with which *Brunellia* shares some characteristics (OROZCO 1997), there is no relation between the texture of the foliar lamina and the presence of medullate bundles, as occurs in other genera of Cunoniaceae (DICKINSON 1975). The presence of phloem, initially in the medullary bundles is highly characteristic of *Acsmithia* and *Spiraeanthemum*, but absent in other species of Cunoniaceae (DICKISON 1975) while it seems to be present in *Brunellia* (Fig. 3.7 h).

The cortical bundles present in the petiole of some genera of Cunoniaceae, *Ceratopetalum*, *Pancheria*, and *Acsmithia* are considered to be relatively advanced in anatomical terms, compared to other anatomical characteristics of the family (DICKISON 1975). Their presence in other groups such as Connaraceae and Dilleniaceae (DICKISON 1969, 1973) is considered to be a special condition of the petiole. In the case of *Brunellia*, it could be said that the petiole is relatively advanced in anatomical terms, due to the presence of cortical bundles in most species examined (except *B. acostae*, *B. ecuadoriensis*, and *B. pauciflora*).

According to HARE's (1943) classification, the species of *Brunellia* with more than one foliar lamina have a principal vascular system with an elongated, cylindrical petiole, except for *B. macrophylla* and *B. foreroi*, with a single and well developed foliar lamina. A short cylindrical form was observed in species with a single foliar lamina such as *B. pauciflora*, *B. glabra*, *B. tomentosa*, and *B. stuebelii* (Figs. 3.3 e; 3.4 a, b). In *B. ovalifolia* (Fig. 3.3 c) the same short cylindrical shape can be seen, but leaves can be both unifoliolate or with additional leaflets. All this indicates, as expected, that the shape of the principal vascular system of petiole is related to the foliar mass supported.

Vascular bundles in the form of wings, are joined to the principal vascular system of the petiole in most species and indicate the presence of stipels homologous to undeveloped leaflets in *Brunellia*. In some species they can be easily recognised by their external morphology, but in others, especially those with a single lamina, they can only be recognised anatomically.

Different stages of development of lobulation from a foliar lamina have been observed in compound leaves, as is illustrated by *B. propinqua* (Figs. 3.8 a-d). Also, in *B. darienensis*, *B. inermis* and *B. amayensis* (OROZCO 1985, 1986) reduction to a single foliar lamina (with stipels on the petiole) has been observed, usually at the distal end of branches, which also bear leaves with several leaflets (OROZCO 1985). There is no anatomical evidence of stipels on the three levels of the petiole of *B. ovalifolia* and *B. pauciflora*, however stipels were seen in specimens of *B. ovalifolia* from the AAU herbarium. In specimens of *B. ovalifolia*, compound leaves were observed on the proximal branches, while leaves with a single lamina are to be found on the distal branches. We can not explain the absence of vascular bundle relicts in the petiole of *B. ovalifolia*

with one foliar lamina, or the minuscule and not easily perceptible anatomical presence of stipels. The same applies to *B. pauciflora*.

The reduction of compound leaves with several foliar lamina to a single leaflet is clearly seen in *B. amayensis*, with a well developed terminal leaflet, with very reduced lateral leaflets; or there is a large single foliar lamina whose petiole has a pair of stipels corresponding to the reduced lateral leaflets (OROZCO 1986, Fig. 3.8 f, f'). This process of reduction may also have occurred in *B. foreroi* (OROZCO, 1981) and *B. latifolia* (CUATRECASAS 1970, 1985) with single foliar laminae, perhaps the largest of the genus and lateral leaflets reduced to two pairs of stipels (Fig. 3.8 g). In species such as *B. macrophylla*, *B. rufa*, *B. tomentosa*, and *B. stuebelii*, reduction of lateral leaflets is seen anatomically. All these species develop one large foliar lamina, and it seems that the reduction of lateral leaflets is related to the increased development of the single lamina or the terminal leaflet, as observed in *B. amayensis* (OROZCO 1986).

The presence of a single foliar lamina might represent a basic state from which leaves with more than one foliar lamina could develop (Fig. 3.8 b-d). During the ontogeny of the leaves, in species of compound and unifoliolate leaves, one single leaflet was always observed as the initial stage of the development (OROZCO in prep.). However, phylogenetic analysis of *Brunellia* indicates that foliar reduction is a reversable character (Chapter 7).

Of the species examined with a single foliar lamina, *B. glabra*, *B. rufa*, *B. macrophylla*, *B. tomentosa*, and *B. stuebelii* should be considered as compound leaved species. We think that the remaining species with one foliar lamina could show the same pattern of vascularization.

According to DICKISON (pers. comm.) the results of nodal anatomy analysis of *Brunellia* are not convincing. Unfortunately DICKISON's death left his research on the *Brunellia* material that I sent him unfinished. Whether or not the node of *Brunellia* is unilacunar is still a matter of research. Unilacunar and trilacunar nodes in Cunoniaceae were reported by DICKISON (1980). Unilacunar for *Aphanopetalum* and *Baurea*, and trilacunar for *Spiraeanthemum*.

3. 5 ACKNOWLEDGMENTS

3. 6 LITERATURE

APG. 1998. An ordinal classification for the families of flowering plants. Ann. Missouri Bot. Gard. 85: 531-553.

CUATRECASAS, J. 1970. Brunelliaceae. Flora Neotropica Monograph 2: 1 - 189.

CUATRECASAS, J. 1985. Brunelliaceae. Flora Neotropica Monograph 2 (suppl.): 29-103.

DICKISON, W. C. 1969. Comparative morphological studies in Dilleniaceae, IV. Anatomy of the node vascularization of the leaf. J. Arnold Arb. 50: 384-410.

DICKISON, W. C. 1973. Anatomical studies in the Connaraceae. III. Leaf Anatomy. J. Elisha Mitchell Sci. Soc. 89: 121-38.

DICKISON, W. C. 1975. Leaf anatomy of Cunoniaceae. Bot. J. Linn. Soc. 71: 275-294.

DICKISON, W. C. 1980. Diverse nodal anatomy of the Cunoniaceae. Amer. J. Bot. 67: 975 - 981.

DICKISON, W. C. 1989. Comparisons of primitive Rosidae and Hamamelidae. In : Evolution, systematics, and fossil history of the Hamamelidae, vol. 1 : Introduction and 'lower' Hamamelidae (eds. P.R. CRANE and S. BLACKMORE), pp. 47-73. Clarendon Press, Oxford.

EYDE, R. 1970. Anatomy in Brunelliaceae. In: (ed. J. CUATRECASAS), Flora Neotropica. Monograph 2: 32 - 43.

HARE, C. L. 1943. The anatomy of the petiole and its taxonomic value. Proc. Linn. Soc., London. 155: 223-229.

HOWARD, R. A. 1974. The stem node leaf continuum of the dicotyledoneae. J. Arnold Arbor. 55: 125-181.

HOWARD, R. A. 1979. The stem node-leaf continuum of the dicotyledoneae. The petiole. In: Anatomy of the dicotyledons 1. Systematic anatomy of the leaf and stem (eds. C. R. METCALFE. and L. CHALK), pp. 76-96. Science Publications, Oxford.

HUFFORD, L . and DICKISON, W. C. 1992. A phylogenetic analysis of Cunoniaceae. Syst. Bot. 17: 181- 200.

METCALFE, C. R. and CHALK, L. 1965. Anatomy of the dicotyledons. Leaves, stem, and wood in relation to taxonomy with notes on economic uses. Vol. 1. Clarendon Press, Oxford.

OROZCO, C.I. 1981. Contribución al estudio de Brunelliaceae I. Mutisia 50 : 4-8.

OROZCO, C.I. 1985. Dos especies nuevas de Brunellia para Colombia. In: (ed. J. CUATRECASAS), Flora Neotropica Monograph 2 (suppl.): 20-24.

OROZCO, C.I. 1986. Contribución al estudio de Brunelliaceae de Colombia III. Caldasia 15: 177-185.

OROZCO, C.I. 1997. Sobre la posición sistemática de Brunellia. Caldasia 19: 145-164.

OROZCO, C.I. 1999. Indumento del envés foliar de Brunellia Ruiz & Pavón y su relativa importancia en la filogenia. Rev. Acad. Colomb. Cienc. Exact. Fis. Nat. 23 (Suplemento Especial): 97- 102.

OROZCO, C. I. and WEBERLING, F. 1999. A comparative study of inflorescences in Brunellia Ruiz & Pav. (Brunelliaceae) and related taxa. Beitr. Biol. Pflanzen 71: 261-279.

SALOMON, F. and CANTINO, M. H. 1983. Curso de prácticas de Biología general Tomo II. Universidad Complutense de Madrid. Herman Blume, Madrid.

A COMPARATIVE STUDY OF INFLORESCENCES IN
BRUNELLIA RUIZ & PAVÓN AND RELATED TAXA[1]

CLARA INÉS OROZCO[2] AND FOCKO WEBERLING[3]

ABSTRACT

The inflorescence architecture of *Brunellia* was studied in 35 species, that is, about 2/3 of the species of the monogeneric family, covering the total inflorescence variation of the genus. This study was carried out to understand the morphology and incorporate this important information into taxonomic proposals and for defining phylogenetic relationships among the species. The results indicate, according to the concept of TROLL (e.g. 1964) that *Brunellia* has a proliferating monotelic synflorescence. In the sense of MORA-OSEJO (1987: 65) it follows an anthocaule-monotelic pattern. In other words, the inflorescence architecture corresponds to an open synflorescence with axillary floriferous systems bearing terminal flowers. The ramification pattern of these floriferous systems (paracladia of first order) can be characterized as thyrso-paniculate. Three subpatterns can be distinguished according to the absence (1) or presence (2-3) of subthyrsoids: 1) homocladic thyrsoids (haplo-thyrsoids) or (2-3) heterocladic thyrsoids either as 2) diplo-thyrsoids or as 3) pleio-thyrsoids. Variations also result from differences in the size of the floriferous part in

relation to the total length of the inflorescence, the length of hypopodia and epipodia and the symmetric or asymmetric division of the branches that influence the shape and contours of the inflorescence. Concaulescence is nearly always present. In some species early proliferation of paracladia was observed.

RESUMEN

La arquitectura de la inflorescencia de *Brunellia* fue estudiada en 35 especies que corresponden a 2/3 de las especies de la familia monogenérica, y que representan la variación total de la inflorescencia en el género. El estudio fue realizado para entender la morfología de las inflorescencias más allá de los límites netamente descriptivos y usar esta información para propósitos taxonómicos como también para el encuentro de relaciones de parentesco entre las especies. Para la interpretación de las inflorescencias se siguió la escuela de TROLL (1964), parcialmente combinada con los conceptos de MORA-OSEJO (1987: 65). Los resultados muestran que *Brunellia* presenta (en el sentido de TROLL 1964) una sinflorescencia proliferante monotélica, o un sistema antocaulo-monotélico en el sentido de MORA-OSEJO (1987: 65). En otras palabras una sinflorescencia abierta, con sistemas floríferos axilares sosteniendo flores terminales. Tres patrones generales fueron encontrados, de acuerdo con la ausencia (1) o presencia (2-3) de subtirsoides: 1) tirsoides homocládicos (haplo-tirsoides) o 2-3) tirsoides heterocládicos 2) diplo-tirsoides o 3) pleiotirsoides. Otras variaciones resultan de diferencias en el tamaño de las partes floríferas en relación con la inflorescencia total, de la longitud de los hipodios y epipodios o la división simétrica o asimétrica de las ramas. Todos ellos influyen en la forma y el contorno de la inflorescencia. Concaulescencia es siempre presente. Casos de proliferación se observaron en algunas especies.

4.1 INTRODUCTION

Brunellia is a neotropical genus widely distributed in the Andes and recognized as the sole genus of the family Brunelliaceae (CUATRECASAS 1970, 1985). HUFFORD and DICKISON (1992), however, considered *Brunellia* as a genus of the family Cunoniaceae. OROZCO (1997) recognized close relationships among *Brunellia* and two genera of the family Cunoniaceae, *Spiraeanthemum* and *Acsmithia*. She suggested that either these two genera should be included in Brunelliaceae or form a family apart from Brunelliaceae and Cunoniaceae, the latter being considered as not monophyletic. The genus *Brunellia* with ca. 53 species, has apetalous flowers disposed in open axillary inflorescences which have peduncles of various lengths.

Comparisons of inflorescences made using external characters only have sometimes made the true relationship between the inflorescences of species of any taxonomic group difficult to interpret. The study of the inflorescence in *Brunellia*

has, until now, not gone beyond simple description (CUATRECASAS 1970, 1985; OROZCO 1985, 1986, 1991). The usual description often is insufficient to detect corresponding characteristics among the parts of the inflorescences of different species. Only these characteristics, however, enable us to understand how the various forms of inflorescence of larger or smaller taxonomic groups, although being very different in their appearance are modifications of the same basic plan (WEBERLING 1989 a: 201). For such proposals, the typological study of inflorescences based on TROLL's work (1954, 1955, 1957, 1964, 1969 see also; TROLL and WEBER 1955, TROLL and WEBERLING 1989, and WEBERLING 1988, 1989 b) is recommended.

According to TROLL there are two basic types of inflorescences: monotelic and polytelic. In monotelic inflorescences the main axis ends in a terminal flower. The same applies to all branches preceding the terminal flower, which in this respect repeat the structure of the main axis and therefore are called paracladia (repeating shoots). In cases of a rather recent loss of the terminal flower (truncate or proliferating monotelic inflorescence) this can still be deduced from the structure of the flowering branches. In polytelic inflorescence the main axis ends in a racemose or thyrsoid florescence instead of a terminal flower and remains indefinite. Often the main florescence is preceded by branches which themselves end in a florescence; thus repeating the structure of the main axis. Again they are called paracladia (of a polytelic system). In order to distinguish their florescences from the main-florescence these are named co-florescences (see WEBERLING 1988, 1989 a).

Within both the monotelic and polytelic types, a corresponding zonation can be observed. The terminal flower or the main florescence is preceded by an enrichment zone (paracladial zone, PZ or BZ, "Bereicherungszone") and the inhibition zone (Inh-Z) in which the development of the paracladia is inhibited. In perennials the inhibition zone is preceded by an innovation zone (INN.Z) which carries the buds of innovation (WEBERLING 1989 a). In herbaceous plants these buds serve to produce a new flowering shoot in the following vegetation period. In ligneous plants they usually originate the shoots which continue the vegetative branching system. Here they are often placed immediately below the paracladial zone, which means that an inhibition zone is absent (Fig. 4.2).

Some general patterns of the inflorescence structure, whether monotelic or polytelic were explained by MORA-OSEJO (1987) and MORA-OSEJO and GONZÁLEZ (1995). Basically three models were established: the anthocaule, holocaule and anthoblast. Anthocaule (Span. antocaulo) means that the synflorescence is proliferating in the sense of TROLL (1959). In this case the synflorescence is an open system, which after the production of floriferous paracladia turns to vegetative growth again. Holocaule means presence of floriferous systems on the principal axes and in the branches of different order. Both principal axes and branches have normal leaves (Span. nomófilos) and buds. Anthoblast is a modification of a polytelic or monotelic holocaule.

The present study tries to find a more profound understanding of *Brunellia* inflorescence than earlier approaches. This may help to reveal the comparable elements of different inflorescences in order to use these differences as taxonomic characters at level of species or species groups. Finally this might lead to a phylogenetic interpretation of the relationships in *Brunellia*. The study includes additional characters as compared with earlier studies.

4. 2 MATERIALS AND METHODS

Inflorescences of 35 species of *Brunellia* (Table 4.1), were studied with a dissecting microscope and were sketched. These species cover the total inflorescence variation within the genus and represent different phylogenetic branches of cladistic analysis of *Brunellia* (Chapter 7). The research was carried out on herbarium material and on some material fixed in FAA.

Specimens of most species studied were deposited in COL, MO, and US. The interpretation of the pattern of the inflorescences was based on TROLL´s typology, as carried out by WEBERLING (1961, 1989 a) SELL (1976), also using the experience of SELL and CREMERS (1987), MORA-OSEJO (1987), MORA-OSEJO and GONZÁLEZ (1995), GONZÁLEZ (1991) and LOIZEAU (1994). The level of ramification of the branches as well as the number of inflorescence branches were registered for every species examined. The figures that accompany this paper represent the total variation of the inflorescences in *Brunellia*. Several collections of *Acsmithia* and *Spiraeanthemum*, (Cunoniaceae) from the Rijksherbarium, Leiden (L) were studied.

The ramification pattern of the 1st order paracladia of the proliferating inflorescences of *Brunellia* is described here as thyrsoid or thyrso-paniculate. A "thyrsoid" by contrast to the panicle, is defined as an inflorescence "with cymose partial inflorescences" where "cymose branching" is branching exclusively from the axils of the prophylls, which are the only leaf organs preceding the individual flowers (WEBERLING 1989 a: 211).

The larger part of our study was based on the analysis of herbarium material which for reasons of its limited size does not always offer sufficient information about the entire branching system and its growth rhythm. In some cases, however, the presence of bud scars at the bases of the branches or any other marks indicating the age of the twigs was found to be helpful.

4. 3 RESULTS

Brunellia exhibits a monotelic proliferating[3] synflorescence with thyrsoid or thyrso-paniculate paracladia (Figs. 4.1-4.7). In *Brunellia* the prophylls are

---

3 Synflorescence is "a system of florescences (in the polytelic type) or a system of the terminal flower and monotelic paraclades (in the monotelic type)" (WEBERLING 1989, p. xx).

often bracteose and small (subulate), but in some species they reach 2.0-4.0 x 1.0-2.0 cm. In some species the prophylls of vigorous paracladia are petiolate. In other species the size of the prophylls is maintained through all the branches of the paracladia.

Within the general pattern of the monotelic-proliferating synflorescence there are several subpatterns in the architecture of the 1st order paracladia, the so called "axillary inflorescence". The differences refer to the presence or absence of subthyrsoids. The simplest paracladia bear three flowered cymes, which in *B. pauciflora* (Fig. 4.3 a) originate in the axils of foliage leaves. In other species mono-and diplo-thyrsoids are found. In most of the species examined the paracladia are heterocladic and often very complex attaining a high order of branching. The presence of complex branching even in the most distal paracladia is of value as a distinguishing character (Table 4.1 f).

Another variation influencing the contour of the paracladia, which is important for species identification, refers to differences in the length of the
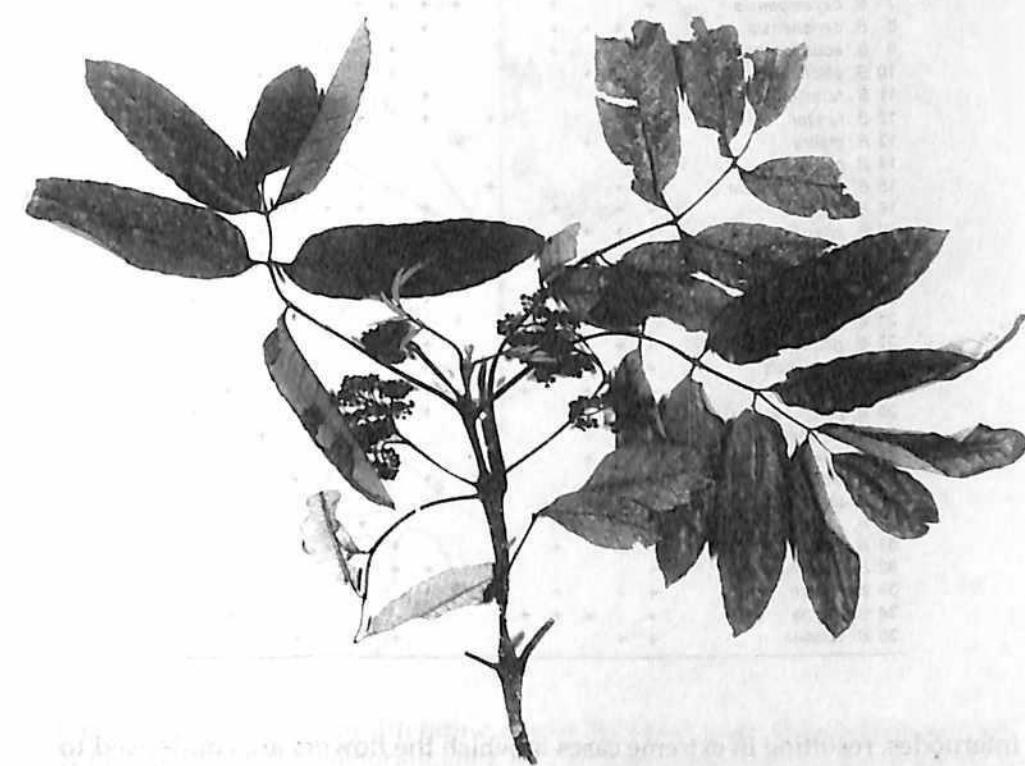

Fig. 4.1 *Brunellia costaricensis*, proliferating inflorescence. Tree near Cerro de la Muerte, Cordillera de Talamanca, Costa Rica (WEBERLING, 8563, ULM).

Table 4.1 Variations of the proliferating monotelic synflorescence in *Brunellia*. 1. Triadic partial inflorescences (paracladia) only (haplo-thyrsoid with triadic to pentadic cymes). 2. Haplo-thyrsoid partial inflorescences. 3. Diplo or Pleio-thyrsoid inflorescence. a. Complexity of proximal paracladia. b. Complexity of distal paracladia. Branching order: c. 2nd to 3rd order, d. 3rd to 4th order, e. 4th to 5th order, f. > 5th order. Ratio between floral zone and length of total inflorescence, g. < 50%, h. 50-70%, i. > 70%., j. Acrotonic promotion of paracladia. k. Formation of glomerules. l. Symetric dichotomy of paracladia. m. Proliferation. n. Presence of a reduced paracladium (Pc'$_3$) in the middle of the main axis. o. Inflorescence represented by one paracladium of more complex synflorescence.

SPECIES	1	2	a	b	c	d	e	f	g	h	i	j	k	l	m	n	o
1 *B. acostae*			+	+		+				+							
2 *B. acutangula*			+	+	+	+				+							
3 *B. amayensis*			+	+		+				+							
4 *B. boliviana*			+	+		+				+	+						
5 *B. boqueronensis*			+		+					+							
6 *B. briquetii*	+								+								
7 *B. cayambensis*			+			+			+	+	+	+					
8 *B. darienensis*			+	+	+	+				+							
9 *B. ecuadorensis*			+	+		+				+	+						
10 *B. elliptica*			+		+					+						+	
11 *B. farallonensis*	+									+							
12 *B. foreroi*			+	+				+		+		+		+			
13 *B. glabra*			+		+				+								
14 *B. goudotii*	+																
15 *B. hygrothermica*			+	+				+				+	+				
16 *B. inermis*			+	+						+		+	+	+	+		+
17 *B. integrifolia*			+	+	+					+			+		+	+	
18 *B. littlei*			+		+					+			+		+		
19 *B. morii*			+	+		+				+	+						
20 *B. ovalifolia*	+				+					+						+	
21 *B. pauciflora*	+							+									
22 *B. penderiscana*			+	+	+					+							
23 *B. pitayensis*			+	+	+	+				+							
24 *B. propinqua*			+	+		+		+		+						+	
25 *B. putumayensis*			+	+	+					+							+
26 *B. racemifera*			+	+						+					+	+	
27 *B. rufa*			+		+					+							
28 *B. sibundoya*			+	+				+		+	+			+	+		+
29 *B. stenoptera*			+	+	+					+		+					
30 *B. stuebelii*	+									+							
31 *B. subsessilis*			+	+	+					+		+					
32 *B. tomentosa*			+						+	+							
33 *B. trianae*			+	+	+					+	+						
34 *B. trigyna*			+		+	+	+			+						+	
35 *B. velutina*			+	+						+					+		

internodes, resulting in extreme cases in which the flowers are condensed to form glomerules. The size of the floral portion in relation to that of the total inflorescence as well as the length of the hypopodium and epipodium (Fig. 4.2) also influence the shape of the inflorescence. Concaulescence and

recaulescence often occur in the inflorescence of *Brunellia*. Anaphysis is rare. The presence of foliage leaves in the floral zone of *B. putumayensis* (Figs. 4.4 a) could be considered as a metatopic displacement.

First order paracladia formed by a three flowered cyme are only observed in *B. pauciflora* (Fig. 4.3 a). Sometimes, however, seven flowered cymes were observed in the same species.

More complex floriferous paracladia of first order were observed in *B. goudotii* (Fig. 4.3 b). However, the ramification of the cymes remained low and the formation of subthyrsoids was not yet completed. In *B. tomentosa* (Fig. 4.3 c) the degree of cymose ramification of second order paracladia is somewhat
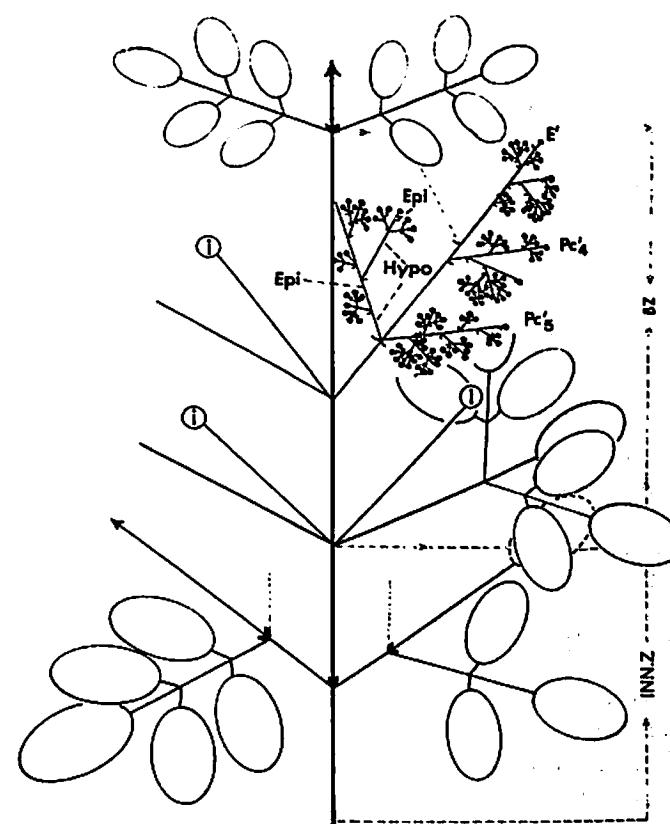
Fig. 4.2 Diagram of a proliferating monotelic (Span. antocaulo) synflorescence of *Brunellia* ('open indeterminate'). BZ , enrichment zone. INN.Z = innovation zone (Trophotagma, MORA-OSEJO and GONZÁLEZ 1995). Hypo = hipopodium. Epi = epipodium. Pc'$_4$, Pc'$_5$, Pc'$_6$... branching order of paracladia. i represents a (partial) inflorescence. E´ = terminal flower.

higher. In *B. putumayensis* the paracladia may be (reduced to) haplo-thyrsoids (Fig. 4.4 a). Mostly they are diplo-thyrsoids, their proximal second order paracladia forming haplo - thyrsoids (Fig. 4.4 a), as does the proximal second order paracladium in Fig. 4.4 b

For the majority of the species examined compound thyrsoids, i.e. heterocladic inflorescences, are characteristic (Table 4.1-4.3, Figs. 4.3 b - d, 4.4 - 4.7). Within this pattern some of the aforementioned variations are characteristics of groups of species (Table 4.1 a-o).

In some species the complexity of inflorescence is due principally to the strong development of the proximal paracladia. This applies to the elongate diplothyrsoids of *B. rufa* (Fig.4.3 d), as to *B. racemifera* and *B. propinqua* (Figs. 4.4 c, 4.5 a). In the remaining species complex thyrsoids are also present in the distal portions of the floriferous paracladia (Figs. 4.5 b, 4.6 a, 4.7 b).

The paracladia of the higher order, from 3rd order upward may appear most complex as a result of repetitive cymose ramification combined with the shortening of the epipodia as is observed in *B. inermis, B. foreroi, B. hygrothermica* and *B. sibundoya* (Figs. 4.5 c, 4.7 b, Table 4.1 k). In other species the ramification appears more paniculate than cymose because the displacement of the flowers from their original positions is caused by the enlargement of hypopodia and epipodia, as can be seen in *B. putumayenis* (Fig. 4.4 a, b)

Repetitive dichasial ramification often occurs in species with diplo-to pleio- thyrsoid paracladia. In these species (Figs. 4.4 c, 4.6 a, 4.7 b) the higher order paracladia tend to surpass the main axis (Table 4.1 j, l). In some species with normally well branched paracladia, specimens with highly reduced paracladia were found, as in *B. putumayensis* with complex and less complex inflorescences (Fig. 4.4 a, b).

In the species group of *B. propinqua* (Fig. 4.5 a), *B. trigyna* and *B. susaconensis* (Table 4.1 n) a remarkable case of branching was observed repeatedly (Pc$_5$). This paracladium is localized in the middle part of the main axis of the inflorescence. It is less ramified than its distal and proximal counterparts.

A further differentiation of the paracladia is found in the species group of *B. elliptica, B. littlei, B. racemifera, B. integrifolia*, and *B. inermis* (Table 4.1 m, Figs. 4.6 d, 7a). The first order paracladia start their growth with the production of floriferous second order paracladia and then turn over to vegetative growth. Thus, they differ from the common paracladia by their indeterminate growth. In a certain way they resemble the long-paracladia of Leguminosae (with the difference that these are of polytelic structure, see Weberling 1989 b, p. 38). We may regard this as a secondary differentiation of paracladia and distinguish the proliferating paracladia as long paracladia from the floriferous short paracladia, which arise from the basal part of the long paracladia.
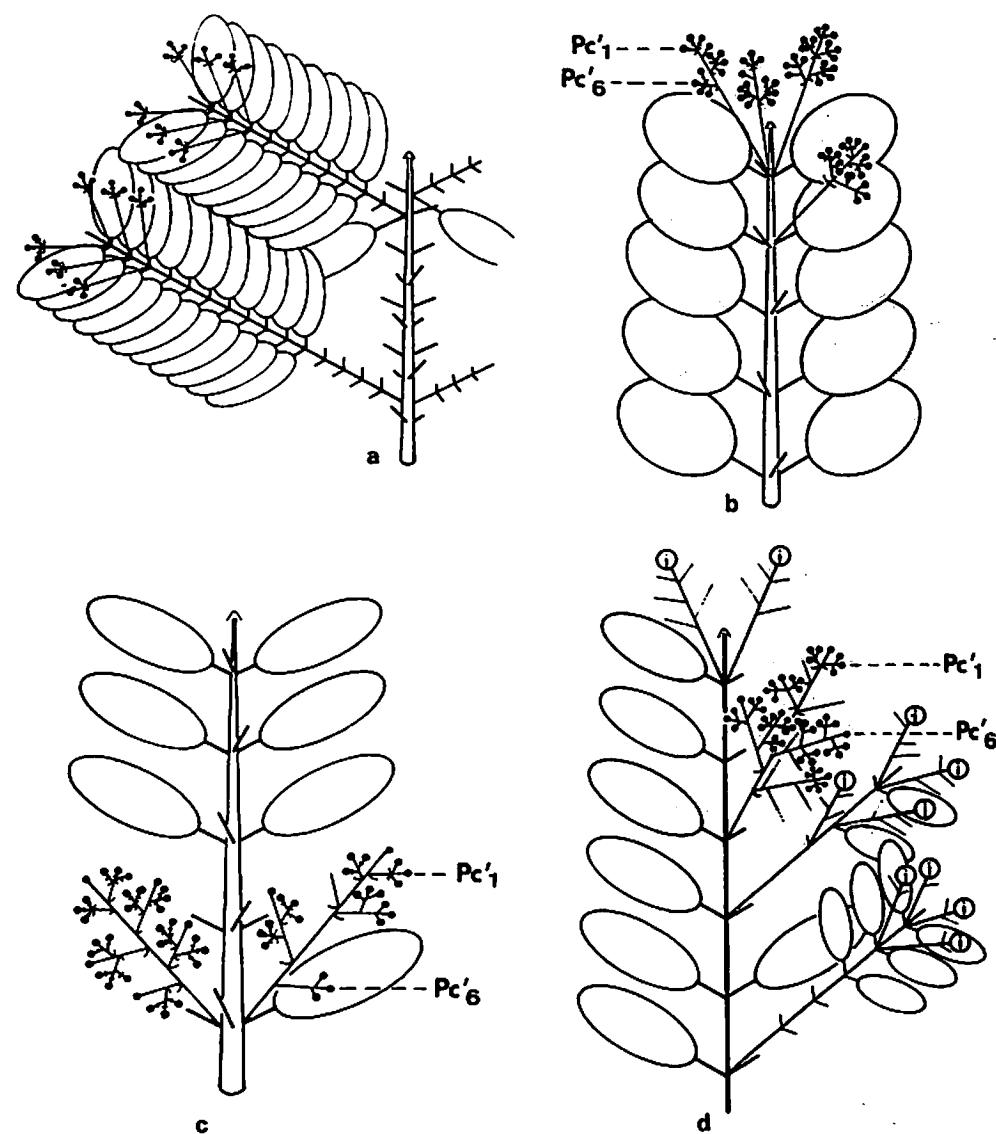
Fig. 4.3 Homocladic or monothyrsoid synflorescences with triadic cymes **(a)** *B. pauciflora*, branching system with flowering twigs with a thyrsoid inflorescence composed of triadic paracladia (Palacios and van der Werff 3897). **(b)** Heterocladic (diplo-thyrsoid) paracladia, *B. goudotii*, (Orozco 1129, 1130). **(c)** Heterocladic inflorescence diplo-thyrsoid, *B. tomentosa* (Orozco 1214, 1175). **(d)** elongate di - (pleio-) thyrsoid in *B. rufa* (Orozco 2850). Pc' branching order of paracladia. i. Thyrsoid paracladia.
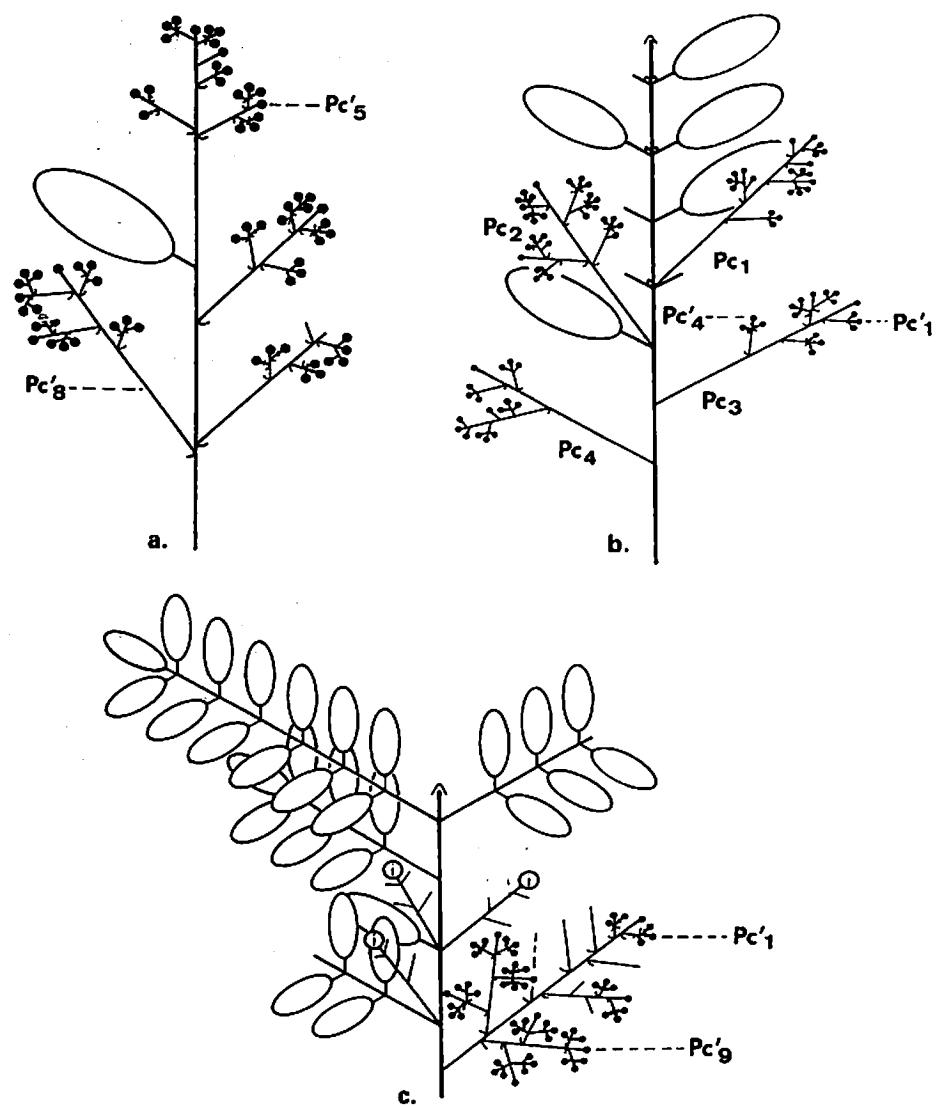
Fig. 4.4 Synflorescences with heterocladic paracladia (diplo thyrsoids). (a) *B. putumayensis*, (OROZCO 1201, 1208, 1213) a richly ramified paracladium; the presence of a foliage leaf in the middle of the paracladium is probably due to a metatopic displacement. (b) *B. putumayensis* proliferating synflorescence with haplo-thyrsoid paracladia and a diplo- thyrsoid proximal paracladium. (c) *B. racemifera* (BARCLAY 3413, KILLIP 380109, OROZCO 1100), complexity of the proximal paracladia. Pc' branching order of paracladia. i. Thyrsoid paracladia.
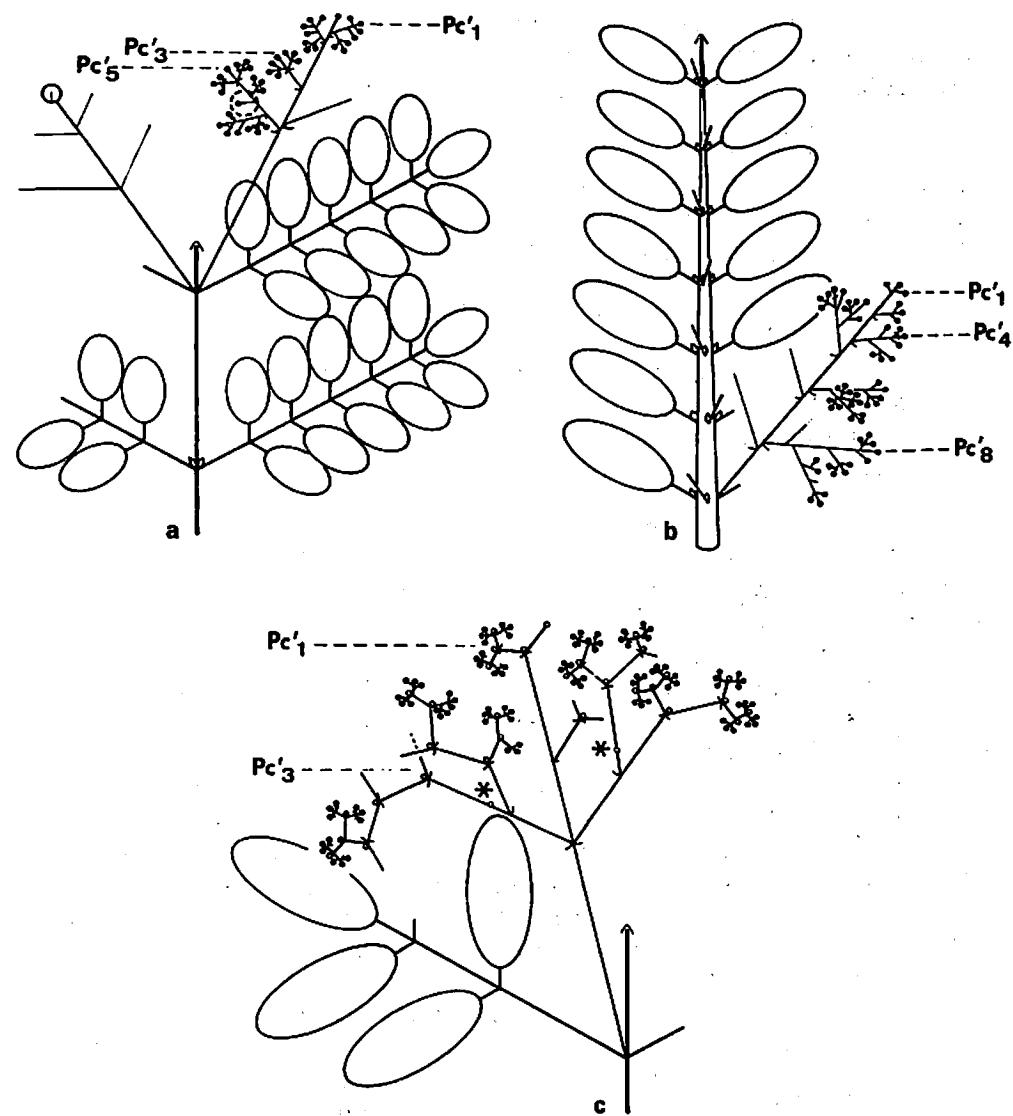
Fig. 4.5 Synflorescences with pleio-thyrsoid (disjunctive heterocladic) partial inflorescences (a) *B. propinqua* (CUATRECASAS 27329, OROZCO 1767). (b) *B. acutangula* (OROZCO 1422) synflorescence with triplo-thyrsoid (quadruplo) paracladia. (c) *B. inermis* (DÍAZ 2625, FOSTER and GENTRY 44878, VAN DER WERFF 6328, 9196) repetitive dichasial ramification of the paracladia, flowers in glomerules. Scars indicate the place of a further ramification. Pc' branching order of paracladia. i. Thyrsoid paracladia.

4. 4 DISCUSSION AND CONCLUSIONS

In *Brunellia* all species exhibit the same proliferating monotelic synflorescence with thyrso- paniculate partial inflorescences (usually as paracladia of first order). Differences among the species correspond to variations of the partial inflorescences due to the presence of triads, their mode of ramification as haplo-thyrsoid or diplo-to pleio-thyrsoids (homocladic or heterocladic thyrsoids), variations in the complexity of branching, the ratio of floriferous part to the total length of the inflorescence and shortening of the internodes especially of the hypopodia and epipodia.

The occurence of triadic (three-flowered) paracladia in *B. pauciflora* (Fig. 4.3 a) will certainly be interpreted as a case of reduction, since 7- flowered cymes have also been found. However, until now we have not found any indication that they may be derived from paracladia of thyrsoid structure, unless the position in their axis of foliage leaves might be compared to the position of the thyrsoid paracladia of the other species.

The proliferation of paracladia and the differentiation of short and long paracladia which were reported here for *B. littlei, B. elliptica* and other taxa (p. 66) have also been reported for inflorescenses of other monotelic and polytelic families. The terms have been coined by TROLL (1965) in order to define elements of polytelic truncate synflorescences which are found in many Leguminosae. A similar differentiation occurs in Poaceae (TROLL 1968, p. 105/106, CAMARA-HERNANDEZ and GAMBINO 1990, CAMARA - HERNANDEZ and RUA 1991, see WEBERLING et al. 1993/97), in Marantaceae (KUNZE 1989) and other polytelic families. Among families with monotelic synflorescences the Datiscaceae (TROLL 1968, p. 97/98), Euphorbiaceae (TROLL 1975, p. 138), Apocynaceae (TROLL and WEBERLING 1989, STECK and WEBERLING 1989) may be mentioned here. It must be emphasized, however, that in all these families the differentiation of long - and short paracladia appears in a certain way specific for each family. This means, that the long - and short paracladia described here for some taxa of *Brunellia* cannot be identified in a strict sense with the long- and short-paracladia of the other families mentioned. The differentiation of long - and short paracladia is rather due to a specific development in each of the families. In case of *Brunellia* this is evident and is made clear by the fact that the differentiation of long - and short-paracladia is connected with the phenomenon of proliferation, which e.g. is not the case in Apocynaceae or Euphorbiaceae.

One could regard the paracladia of *B. elliptica* in Fig. 4.7 a as equivalent with the entire synflorescences as presented by Fig. 4. 2. The example of *B. littlei* in Fig. 4. 6 d, however shows that these paracladia can end their growth with a distal floriferous (thyrsoid) zone and finally a terminal flower, thus forming a transition between the form of paracladia in Fig. 4.7 a and Fig. 4.2. In a similar way the proximal flowering branches of *B. rufa* in Fig. 4.3 d might be

regarded either as long paracladia or as separate proliferating synflorescences equivalent to those in Fig. 4.2. Finally the growth rhythm of the entire branching system has to be taken into consideration, which in some cases is not possible by research on herbarium material only (see p. 62).

It is important to point out that in *Brunellia* there is a clear separation between the three different forms of inflorescences. The homocladic thyrsoids with simple triadic partial inflorescence (Fig. 4.3 c) and homocladic to heterocladic inflorescence with a low degree of ramification are often present in unifoliolate species, with big flowers and fruits (Fig. 4.3 b, c). The most complex inflorescences were often observed in species with small flowers and fruits (Figs. 4.6 a, 4.7 b) with exception of *B. foreroi* with unifoliolate leaves and two pairs of stipels on the petiole which represent reduced leaves (Chapter 3).

The acrotonic promotion of paracladia overtopping the main axis which was observed in some samples of species of different groups (Figs. 4.6 a-c, 4.7 b) could be associated in some way to pollination mechanisms. Research on the biological consequences of inflorescence architecture is rare. These effects, which are related to gene flow, include pollination and seed dispersal. Even though these topics have not been studied in *Brunellia*, the great production of flowers by those species with complex inflorescences versus few flowers produced by other species might be also related to the behavior of pollinators or dispersal agents. Scanty knowledge exists of the pollinators of *Brunellia*. Nevertheless, beetles were seen and captured in the flowers and they vary in size depending on the kind of flower (hermaphrodite or female flower). *Thomasomys*, a small mouse, (LÓPEZ et al. 1993) may be a possible dispersal agent for *Brunellia propinqua*. This mouse eats seeds from the soil and an analysis of its stomach contents showed the presence of *Brunellia* seeds. Research carried out by ROSSELLI (pers.comm.) indicates that many species of birds eat the aril of the seeds of *Brunellia propinqua*, but analysis of fecal samples has not yet been performed.

Any affirmation about the tendency in *Brunellia* towards reduction or increase in complexity of the inflorescence is still premature. However, reduction from more complex inflorescences was observed in different species (Table 4.1), especially in species with leaf and carpel reduction, and with an increase of flower size (Chapter 7).

There is no doubt, that proliferating monotelic synflorescences are a prevailing character in Cunoniaceae as well. This especially applies to the two sister genera of *Brunellia*, *Spiraeanthemum* A. GRAY and *Acsmithia* HOOGL., as can be seen from the figures presented by HOOGLAND (1979: 497, 503 and 1987: 395). This was confirmed by research on several species of both genera. Thus, the so-called axillary inflorescences of these taxa ought to be interpreted as paracladia which arise from the axils of one or more leaf-pairs or tri- or tetramerous whorls of foliage leaves. They usually show a triplo - or quadruplo-

Pc'₉

Pc'₁₀

a

ℸ = (diplo-) thyrsoid   paracladium.

d

ℸ = (diplo-) thyrsoid   paracladium.

Pc'₅

b

c
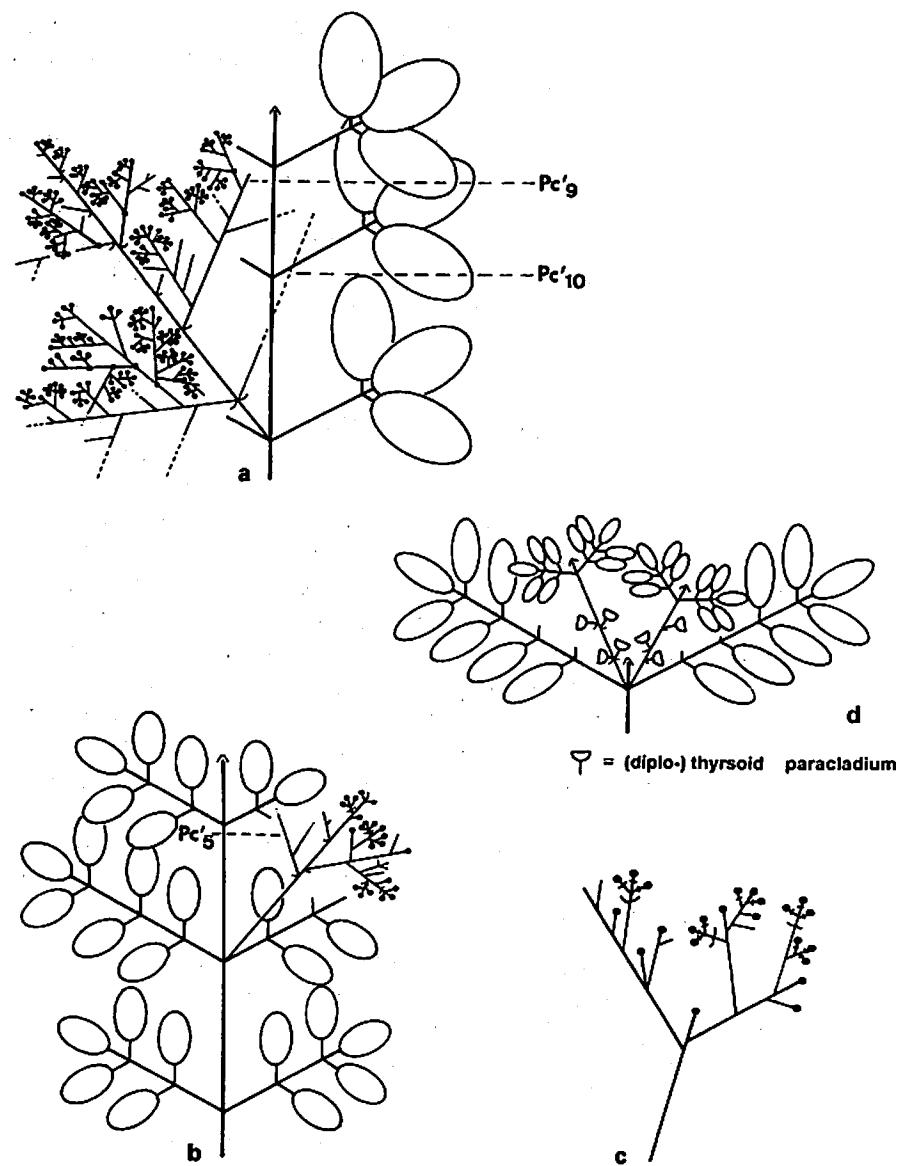
Fig. 4.6 Synflorescences with pleio-thyrsoid (disjunctive heterocladic), partial inflorescences (paracladia). **(a)** *B. velutina* (LOZANO 4860) acrotonic support of paracladia and tendency to repetitive dichasial ramifications in most distal branches. **(b, c)** *B. cayambensis* (CUATRECASAS 11944, ESPINAL 980, IDROBO 3864, OROZCO 1187, PALACIOS 3700) acrotonic promotion of 1ˢᵗ order paracladia. **(d)** *B. littlei* (CUATRECASAS 7556, 8058, HULMAN 6295) proliferation. Pc' branching order of paracladia.

Pc'₃   E'

Pc'₅

i

b
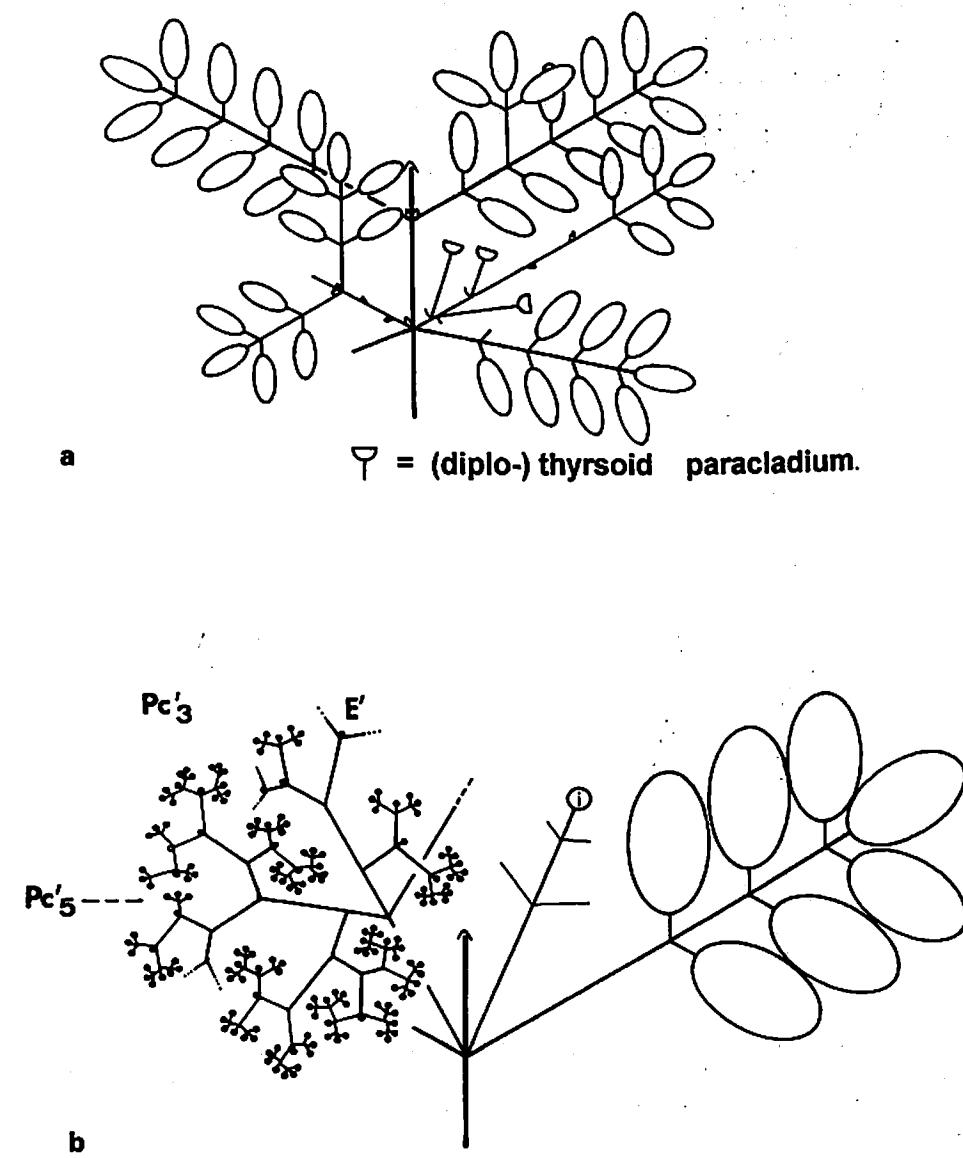
**Fig. 4.7** Synflorescence with pleio-thyrsoid (disjunctive heterocladic), partial inflorescences. **(a)** Proliferation of paracladia in *B. elliptica* (CUATRECASAS 12464, OROZCO et al. 2971). **(b)** *B. sibundoya* (NEILL 4401), one of the most complex branching patterns: repetitive dichasial ramification with acrotonic promotion. E¹ = terminal flower. i. Thyrsoid paracladia.

paniculate-thyrsoid ramification pattern (Fig. 4.8 a)[4]. Their main axis and all its ramifications (Fig.4. 8 b) end in a terminal flower, thus revealing the monotelic character of the synflorescence. After the production of a smaller or greater number of such paracladia the synflorescence axis returns to vegetative growth. Sometimes the formation of a terminal bud indicates a period of rest. Among a great number of specimens we observed only one synflorescence of *Spiraeanthemum katakata* SEEM. (A. C. SMITH 8845, Fiji, Namosi, L) terminating in a panicle. This demonstrates that under exceptional conditions the normally
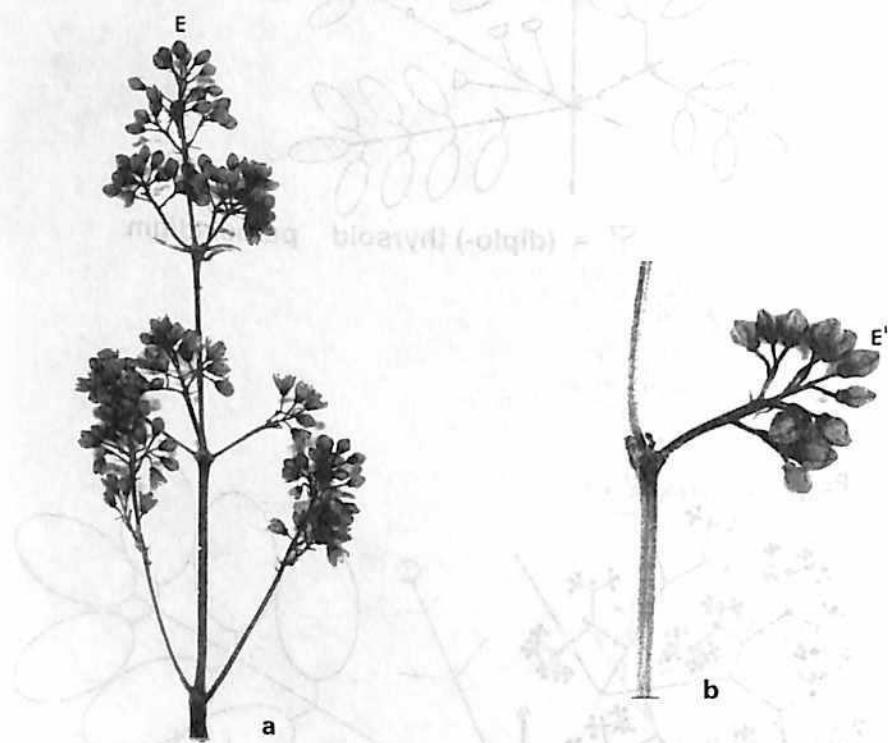
Fig. 4.8 Acsmithia austrocaledonica (Brongn. & Gris.) Hoogl., (Herbarium material, GORDON MCPHERSON 3733, New Caledonia, Mt. Panié ; L). **(a)** paracladium of first order. **(b)** of second order. E. terminal flower of the paracladium of first, E' of second order.

---

4 Thus the two sister genera of *Brunellia*, *Spiraeanthemum* and *Acsmithia*, also present proliferating monotelic synflorescences (with diplo-to pleio-thyrsoid branches). This could indicate that heterocladic condition, still not complex, could be considered as a plesiomorphic state, whereas complex branching could be apomorphic. Simpler inflorescences could be considered as secondary reductions.

proliferating synflorescences might retain the ability to form a complete synflorescence instead of a truncate synflorescence.

In *Cunonia capensis* L. the paniculate ramification pattern of the (usually two) paracladia is obscured because the hypopodia of the branches remain undeveloped, thus causing a more or less spike-like shape of the paracladia. In the distal part of these paracladia, however, the second order paracladia remain 1-, 2-, 3-, 4-, 5-, or 6- flowered, one of the flowers always being in a terminal position. The terminal flower of the entire paracladium is preceded by several sterile bracts (hypsophylls) (unpublished notes of W.TROLL). The structure of the paracladia of the *Weinmannia* -species appears to be similar. Sometimes the flower bearing part of the paracladium-axis remains short, thus causing a globose shape of the partial inflorescence (*Pancheria elegans* BRONGN. & GRIS, *Callicoma serratifolia* ANDR.).

4. 5 ACKNOWLEDGMENTS

4. 6 LITERATURE

CAMARA - HERNÁNDEZ, and GAMBINO, J. S. 1990. Ontogeny and morphology of *Zea diploperennis* inflorescences and the origin of maize (*Zea mays* ssp. *mays*). Maydica 35: 113-124

CAMARA - HERNÁNDEZ, J. and RUA, G. H. 1991. The synflorescence of Poaceae. Beitr. Biol. Pflanzen 66: 297-311.

CUATRECASAS, J. 1970. Brunelliaceae. Flora Neotropica Monograph 2:1-189.

CUATRECASAS, J. 1985. Brunelliaceae. Flora Neotropica Monograph 2 (suppl.): 29-103.

GONZALEZ, F. 1991. Notes on the systematics of *Aristolochia* subsect. *Hexandrae*. Ann. Missouri Bot. Gard. 78: 497-503.

HOOGLAND, R. D. 1960. Studies in the Cunoniaceae. I. The genera *Ceratopetalum*, *Aistopetalum*, and *Calycomis*. Austral. J. Bot. 8: 318-341.

HOOGLAND, R. D. 1979. Studies in the Cunoniaceae. II. The genera *Caldcluvia*, *Pullea*, *Acsmithia*, and *Spiraeanthemum*. Blumea 25: 481-505.

HOOGLAND, R. D. 1987. Studies in the Cunoniaceae. IV. Further notes on New Caledonian *Acsmithia*. Adansonia 4: 393-397.

HUFFORD, L. and DICKISON, W.C. 1992. A phylogenetic analysis of Cunoniaceae. Syst. Bot. 17: 181-200

KUNZE H. 1989. Probleme der infloreszenztypologie von W. TROLL. Pl. Syst. Evol. 163: 128

LOIZEAU, P. A. 1994. Les Aquifoliaceae péruviennes. Boissiera 48: 9-306.

LÓPEZ, H., MONTENEGRO, O., and CADENA, A. 1993. Ecología de los pequeños mamíferos de la Reserva Biológica Carpanta, en la cordillera oriental colombina. Studies on Neotropical Fauna and Evironment 28: 193-210.

MORA-OSEJO, L. 1987. Estudios morfológicos, autoecológicos y sistemáticos en angiospermas. Academia Colombiana de Ciencias Exactas, Físicas y Naturales. Serie JORGE ÁLVAREZ LLERAS 1: 1-195.

MORA-OSEJO, L. and GONZÁLEZ, F. 1995. Tipología de las unidades de crecimiento y floración. (UCF) y consideraciones sobre la evolución del género *Hypericum* en la cordillera oriental de Colombia. In: Biodiversity and conservation of neotropical montane forests, (eds. S. P. CHURCHILL et al.), pp. 377-395. The New York Botanical Garden.

OROZCO, C. I. 1985. Dos especies nuevas de *Brunellia* de Colombia. In: (ed. J. CUATRECASAS) Flora. Neotrópica. Monograph 2. [suppl.]: 20-24.

OROZCO, C. I. 1986. Contribución al estudio de Brunelliaceae de Colombia III. Caldasia 15: 177-185.

OROZCO, C. I. 1991. Contribución al estudio de las Brunelliaceae IV. Dos especies del Ecuador. Caldasia 16: 453-458.

OROZCO, C. I. 1997. Sobre la posición sistemática de *Brunellia*. Caldasia 15: 177-185.

PIRANI, J. R. 1993. Inflorescence morphology and evolution in the genus *Picramnia* (Simaroubaceae). Candollea 48: 119-153.

SELL, Y. 1976. Tendences évolutives parmi les complexes inflorescentiels. Rev. Gen. Bot. 83: 247-267.

SELL, Y. and CREMERS, G. 1987. Les inflorescences des Melastomataceae guyanaises, leur filiation et leur valeur taxonomique. Canad. J. Bot. 65: 999-1010.

STECK, J. and WEBERLING, F. 1989. Infloreszenzuntersuchungen an Apocynaceen. Akad. Wiss. Lit. Mainz, Tropische u. subtrop. Pflanzenwelt 71.

TROLL, W. 1950. Über den Infloreszenzbegriff und seine Anwendung auf die blühender Region Krautiger Pflanzen. Akad. Abh. Math. Naturwiss. Nr. 15.

TROLL, W. 1954. Bericht d. Komm. f. Biolog. Forschg., Akad. Wiss. Lit. Mainz, Jb. 1954: 38-44.

TROLL, W. and WEBER, H. 1955. Morphologie einschliesslich Anatomie. IV. Infloreszenzen. Fortschr. Bot. 17: 36-42.

TROLL, W. 1957. Bericht d. Komm. f. Biolog. Forschg., Akad. Wiss. Lit. Mainz, Jb. 1956: 36-42.

TROLL, W. 1958. Bericht d. Komm. f. Biolog. Forschg., Akad. Wiss. Lit. Mainz, Jb. 1957: 39-50.

TROLL, W. 1959. Bericht d. Komm. f. Biolog. Forschg., Akad. Wiss. Lit. Mainz, Jb. 1959: 127-144

TROLL, W. 1960. Bericht d. Komm. f. Biolog. Forschg., Akad. Wiss. Lit. Mainz, Jb. 1959: 112-130.

TROLL, W. 1964/69. Die Infloreszenzen, Typologie und Stellung im Aufbau des Vegetationskörpers. Bd.I, II/1. G. Fischer, Jena.

TROLL, W. 1965. Bericht d. Komm. f. Biolog. Forschg., Akad. Wiss. Lit. Mainz, Jb. 1964: 93-111.

TROLL, W. 1968. Bericht d. Komm. f. Biolog. Forschg., Akad. Wiss. Lit. Mainz, Jb. 1969: 88-109.

TROLL, W. 1969. Bericht d. Komm. f. Biolog. Forschg. Akad. Wiss. Lit. Mainz, Jb. 1968: 88-109.

TROLL, W. 1974. Bericht d. Komm. f. Biolog. Forschg., Akad. Wiss. Lit. Mainz, Jb. 1975: 128-142.

TROLL, W. 1975. Bericht d. Komm. f. Bot. Forschg., Akad. Wiss. Lit. Mainz, Jb. 1974: 128-142

TROLL, W. and. WEBERLING, F. 1989. Infloreszenzuntersuchungen an monotelen Familien. G. Fischer, Stuttgart.

WEBERLING, F. 1961. Die Infloreszenzen der Valerianaceen und ihre systematische Bedeutung. Akad. Wiss. Mainz, Abh. Math.-Naturwiss. Kl. Nr.15: 1-131.

WEBERLING, F. 1963. Homologien im Infloreszenzbereich und ihr systematischer Wert. Ber. Deutsch. Bot. Ges. 76: 102-112.

WEBERLING, F. 1965. Typology of inflorescences. J. Linn. Soc., Bot. 59: 215-221.

WEBERLING, F. 1981. Morphologie der Blüten un der Blütenstande. Ulmer, Stuttgart.

WEBERLING, F. 1983. Fundamental features of modern inflorescences morphology. Bothalia 14: 917- 922.

WEBERLING, F. 1985. Aspectos modernos de la morfología de las inflorescencias. Boletin Soc. Argent. Botanica 24: 1-8.

WEBERLING, F. 1988. The architecture of inflorescences in the Myrtales. Ann. Missouri Bot. Gard. 76: 226- 310.

WEBERLING, F. 1989 a. Morphology of flowers and inflorescences. Cambridge Univ. Press, Cambridge. 405 pp. English translation of the German edition 1981 Ulmer, Stuttgart.

WEBERLING, F. 1989 b. Structure and evolutionary tendencies of inflorescences in the Leguminosae. In: (eds.C.H. STIRTON & J. L. ZARUCCHI), Advances in Legume Biology. Monogr. Syst. Bot. Missouri Bot. Gard. 29: 35-58.

WEBERLING, F., MULLER-DOBLIES, W., and MULLER-DOBLIES, D. 1993. Zur deskriptiven und vergleichend-morphologischen terminologie komplexer infloreszenzen. Beitr. Biol. Pflanzen 67: 453-473

WEBERLING, F., MULLER-DOBLIES, U., MÜLLER-DOBLIES, D., and RUA, G. H. 1997. Hacia una terminología descriptiva y morfológico-comparativa para inflorescencias complejas. Bol. Soc. Argent. Bot. 32: 171 - 184.

Chapter 5

A STUDY OF FLORAL MORPHOLOGY IN *BRUNELLIA* RUIZ & PAVÓN [1]

CLARA INÉS OROZCO

ABSTRACT

The floral morphology of 46 species of *Brunellia* was studied. In spite of intraspecific variation great value has been assigned to sepal and carpel numbers as taxonomic characters. Flowers before anthesis were dissected and studied by light microscopy. This study included an analysis of intraspecific variation in floral symmetry, number of sepals, carpels, the arrangements of the stamens, and breeding systems. Most of the species have two whorls of stamens, the outer whorl alternates with the sepals and the inner whorl is opposite to the sepals. Two whorls of stamens are frequently present in most species although a few species often present additional whorls of stamens. Additional whorls of stamens are considered as transitional states in species that always have two whorls of stamens. It was found that additional whorls of stamens result from rearrangements of the space among floral parts by fusion of sepals and, consequently, the stamens of a previous implied floral arrangement are placed in an extra whorl. Asymmetry is often present and was seen to be related to differences in the size of the sepals, carpel number (different from the sepal number), and changes in the number of stamens and their arrangement. Changes of sepal merosity was found to affect stamen merosity and very probably that of the carpels. Reduction of stamens was also observed. Bisexual and female flowers were frequently observed in different individuals.

---

[1]   To be submitted to Botanische Jahrbücher.

RESUMEN

Se practicó un estudio floral en 46 especies de *Brunellia*. A pesar de la variación intraespecífica del número de piezas del cáliz y de carpelos, taxonómicamente han sido usados como caracteres diagnósticos en trabajos previos. Flores antes de la antesis, procedentes de diferentes colecciones fueron disectadas y estudiadas bajo el microscopio. El estudio incluye un análisis intraespecífico de la simetría floral, número de sépalos, carpelos, disposición, y número de estambres. Muchas de las especies presentan un patrón de dos verticilos de estambres, mientras que pocas especies presentan frecuentemente más de dos verticilos. Verticilos adicionales de estambres se consideran como estados transicionales (aparente poliandría) para aquellas especies con frecuente presencia de dos verticilos. Reducción de estambres fue también observado. La zigomorfía está relacionada con diferencias de tamaño de los sépalos, número de carpelos (diferente del número de sépalos) y cambios en la condición de dos verticilos de estambres. Estambres y verticilos adicionales son el resultado de rearreglos de los espacios florales cuando hay fusión de sépalos y en consecuencia, estambres de una previa condición de dos verticilos ocupan un extra verticilo. Se observó protandria. Flores bisexuales y unisexuales están presentes en diferentes individuos.

5.1 INTRODUCTION

Brunellia is a neotropical genus widely distributed in the Andes and currently recognized as the only genus in the family Brunelliaceae. Discussions of the systematic position of *Brunellia* are given by HUFFORD and DICKISON (1992), OROZCO (1997), BRADFORD (pers. comm.). The genus is distributed in the subandean and Andean forests and it is an important element of the Andean ecosystem. The genus belongs to the subclass Rosidae (CRONQUIST 1981), order Oxalidales (APG 1998). The species exhibit opposite or verticillate leaves, apetalous flowers and apocarpous fruits. The flowers are disposed in axillary inflorescences. A proliferating synflorescence with floriferous axillary thyrsoid paracladia is present in *Brunellia* (OROZCO and WEBERLING 1999).

The apetalous condition occurs in different lineages of Rosidae (DICKISON 1989) and has been considered as an apomorphic character in *Brunellia* and in some genera of Cunoniaceae (OROZCO 1997).

Merosity is understood as the number of sepals and petals, but in the merosity of the flower all the whorls of the flower must be considered. Pentamery is considered as a basic and universally widespread condition in Rosidae, Dilleniidae, and Asteridae. Among the possible causes for changes in the merosity of the flowers are the reduction of the number of carpels and zygomorphy (RONSE, DECRAENE and SMETS 1994). According to CUATRECASAS (1970), the flowers are actinomorphic or subactinomorphic, isomerous or heteromerous, and have four

whorls. A pentamerous pattern was considered for the genus (CUATRECASAS 1970, HUFFORD and DICKISON 1992), however, tetramerous, hexamerous, and less often, heptamerous flowers also exist in *Brunellia* (CUATRECASAS 1970, OROZCO in prep.). In spite of intraspecific variation in the number of sepals and carpels these features have been used as specific characters.

CUATRECASAS (1970) reported the existence of diplostemonous flowers in *Brunellia* and considered as anomalous, although not rare, the presence of one or more additional stamens. *B. boliviana* has a different floral arrangement, in that it often has more than the two whorls of stamens present in most species of the genus. CUATRECASAS also reported a duplication of the inner whorl in this species but he did not consider this to be additional whorls of stamens. This apparent polyandrous condition is indicated by the presence of an additional whorl with one, two or three stamens.

In *Brunellia* staminodes were reported only in female flowers, thus usually all stamens are fertile or all are sterile. The sexuality of flowers varies. Bisexual and unisexual flowers, and monoecious and dioecious conditions were reported in *Brunellia* (CUATRECASAS 1970). Male flowers were described as having small, presumably sterile, carpels (CUATRECASAS 1970, 1985, OROZCO 1985, 1986, 1991).

In Rosales and in related groups to *Brunellia* such as Cunoniaceae and Eucryphiaceae polyandrous development, associated with a whorled pattern (complex polyandry) was observed (RONSE DECRAENE and SMETS 1992). This kind of development in Rosales usually starts from antesepalous complex primordia, rarely antepetalous (RONSE DECRAENE and SMETS 1992: 635).

The floral organ arrangements known as diplostemonous, obdiplostemonous and their variations are related to the position of the outer or inner whorls of stamens with respect to the sepals and petals. There are several concepts about the origin and kind of the obdiplostemonous condition (RONSE DECRAENE and SMETS 1993, 1995). This condition, where the outer whorl alternates with the sepals and the inner one is opposite to the sepals, tends to be a common character among Rosidae (RONSE DECRAENE and SMETS 1995).

This study seeks an understanding of the intraspecific variations of the floral organ arrangement in *Brunellia* which, for purposes of differentiation of species, would permit a better use concerning the information of carpel, sepal, and stamen numbers and stamen arrangement. Additionally, this study was undertaken to define the floral character states in *Brunellia* for phylogenetic purposes, and to understand the evolutionary changes of floral characters, as well as the tendencies of floral plan variation within every species of *Brunellia*.

5.2 MATERIALS AND METHODS

In order to study the floral organ arrangement and number of floral parts in *Brunellia* species, several closed flowers prior to anthesis from different

collections of 46 species (Table 5.1) were softened by heating in water and subsequently observed under a dissecting stereomicroscope. For every species, the basic floral pattern is printed in a box and every variation of this pattern is illustrated. However, many species often present more than one basic floral pattern, all of which are included in the box.
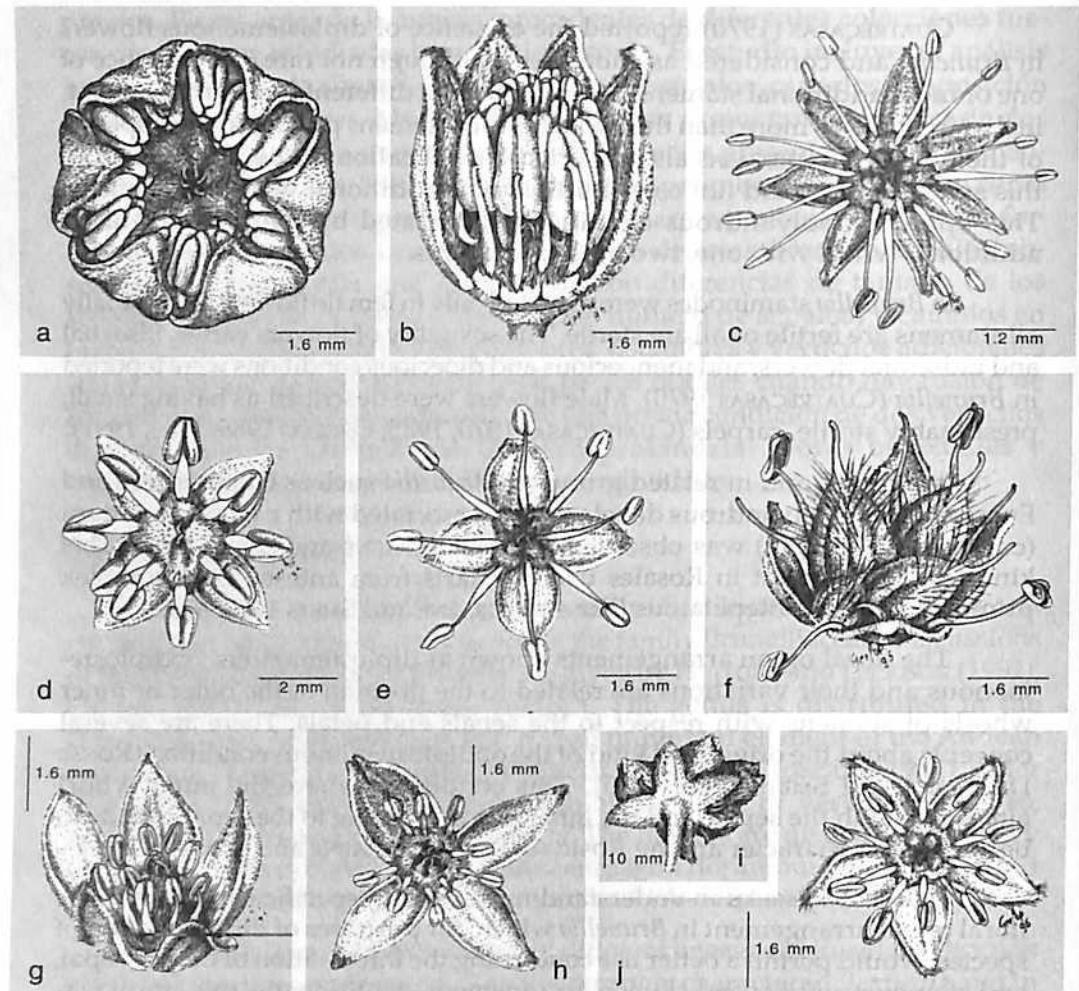
Fig. 5.1 Bisexual flowers with two or additional whorls of stamens. **(a-c)** flowers with additional whorls of stamens, *B. boliviana* (DAVIDSON 4775). **(a, b)** position and insertion of stamens. **(c)** dissected flower. **(d-f)** flowers with two whorls of stamens or additional stamens in a third whorl. **(d)** flower dissected, with an additional stamen in a third whorl of *B. propinqua* (URIBE 6739). **(e-f)** *B. comocladifolia* (FERNÁNDEZ-ALONSO et al. 10236). **(g- h)** altered floral organ arrangement, *B. pauciflora* (PALACIOS et al. 6978). **(i, j)** *B. stuebelii* (OROZCO et al. 1530). **(i)** unisexual flower. **(j)** bisexual.

5.3 RESULTS

Flowers of *Brunellia* are apetalous, bisexual or unisexual, and generally present an asymmetric pattern. The stamens are inserted on a disk and the flowers usually exhibit two stamen whorls. The outer whorl alternates with the sepals and the inner is opposite to the sepals (Fig. 5.1). This condition is called obdiplostemony and tends to be present in Rosidae as pointed out by ROSE DECRAENE and SMETS (1995 : 211). A few species usually exhibit apparent polyandry, and in these species an additional whorl of stamens is observed, and the presence of only two whorls of stamens is rare (Fig. 5.2 a-h). The basic condition of two whorls of stamens, without alterations (increase or reduction of stamens), is reported for 13 of the 41 species of *Brunellia* examined. Alterations indicate reductions from a larger floral organ arrangement and the tendency to the continuous reduction. Additional studies are necessary in those species that appear with one asterisk in Table 5.1. The species for which the least data are available (little floral material) is *B. weberbaueri* where the stamens (10) seem localized in a single whorl (Table 5.1). The anthers are introrse (Fig.5.1), sagittate, versatile, and dorsifixed.

The diameter of the flowers in *Brunellia* usually varies between 4 and 16 mm. Unifoliolate species generally have larger flowers (9-16 mm in diameter) with the exception of *B. foreroi* whose flowers have calyces between 3.5-4.5 mm in diameter. Some species with pinnate leaves also have large flowers as were observed in *B. boliviana, B. cuzcoensis, B. ovalifolia* (the last species also has branches with unifoliolate leaves) *B. propinqua, B. susaconensis,* and *B. trigyna.*

Flowers are bisexual or, if unisexual, they are always female. Female and bisexual flowers generally occur in different individuals. Some species such as *B. pauciflora* and *B. stuebelii* however, have bisexual and female flowers on the same individual (Figs. 5.1 i, j). Pronounced protandry was observed in which the flowers often give the impression of being unisexual male flowers, especially prior to pollination and when the carpels have not begun to ripen (Figs. 5.1a, c-e, h, j). Flowers in which carpels have developed into fruit often show withered stamens (Fig. 5.1 f). Male flowers are uncommon in the genus. However, in *B. comocladifolia* and *B. sibundoya*, male flowers, without small carpels, were observed as terminal flowers of a dichasial division of the branches of the inflorescence. On the other hand, female flowers sometimes have staminodes which can be recognized by their small size and inflexed anthers. Staminodes were also observed in bisexual flowers (Figs. 5.3 e, g; 5.4 c, d, f, l; 5.5 a, b, f - j; 5.6 h-j ; 5.7 c-g, i, j).

Variations in the number of carpels, differences in the size of the sepals, and alterations of stamen number of the two whorls by increase or reduction of stamens, influence the symmetry of the flowers in *Brunellia*. Very few species of *Brunellia* have isomerous flowers. In only 23 species, the number of sepals

Table 5.1 List of species of *Brunellia* with floral organ arrangements. K = sepals, A = stamen number, G = carpel number. * * = polystemonous plan, actino = actinomorphic, asym = asymmetric ( ) = rare, * = more study needed.

Species	Basic floral organ arrangement	Number of staminal flowers examined	Altered obdiplostemonous	Symmetry	Total number of flowers examined
B.acostae	K4A4+4G4(5)	17	-	actino	17
B.acutangula	K4 (3)A4+4G4(3)	32	5	actino (asym)	58
	K5A5+5G5(4)				
B.boliviana**	K6(5)A6+5+3G6(8)	23	16	asym	40
	(5) (9)				
	K7A7+7+1G8				
B. boqueronensis	K5A5+5G4	6	1	asym	15
B.brunnea	K5A5+5G5	11	-	actino	11
* B. cutervensis	K5G5	4	-	actino	4
B. cuzcoensis**	K6A6+6+3G6-7-10	6	6	asym	6
B. cayambensis	K5A5+5G5(6)	19	1	actino-asym	11
B. costancensis	k5(6)G5(6)			?	19
B. comocladifolia	K5A5+5G5	5	1	actino (asym)	12
	K4A4+4G4				
B. darienensis	K5(4-6)A5+5G5(4)	22	3	asym	51
* B. dichapetaloides	K4A4+4G4	5	-	actino	5
B. dulcis	K5A5+5G5	7	3	actino-asym	20
	K4A4+4G4(3)				
B. ecuadorensis	K5A5+5G2	9	-	asym	9
*B. farallonensis	K5G3(2)			asym	6
*B. foreroi	K4(3)G4(3)		-	actino	6
*B. glabra	K5A5+5G5	7	7	asym	7
	(K8A6+6G5)				
B. goudotii	K5A5+5G2-3(4)	6	2	asym	14
B. hygrothermica	K5A5+5G5	16	4	actino (asym)	31
	K4 A4+G4				
B. inermis	K5A5+5G5			actino	27
B. integrifolia	K5A5+5G5(4)	9	3	actino	37
	K6G6				
B. latifolia	K5A5+5G5	9	6	asym	19
B. littlei	K5A5+5G5	20	3	actino	30
B. macrophylla	K5A5+5G4	15	4	asym	15
B. mexicana	K5A5+5G5	9	-	actino	21
	K5G5				
B. morii	K5A5+5G5	5	1	actino (asym)	11
*B. occidentalis	K5A5+5G5	2	-	actino	21
B. oliveri	K4A4+4G4	7	-	actino	7
B. ovalifolia	K5A5+5G5	5	1	actino (asym)	22
B. pauciflora	K5A5+5G5	15	6	actino (asym)	21
	K4A4+4G4				12
*B. penderiscana	K4G4			?	
B. propinqua	K5A5+5G3-2	68	47	asym	129
B. putumayensis	K5A5+5G2	9	2	asym	15
B. racemifera	K5A5+5G5(4)	15	2	actino-asym	47
	K6A6+5 G5-4				
B. rhoides	K5(6)A5+5G5(3(4))	23	-	asym	23
B. rufa	K4A4+4G4	7	-	actino	15
	K5A5+5G3				
B. standleyana	K5A5+5G5	9	-	actino	36
	K(4)G(4)				
B. stenoptera	K5A5+5G5	14	2	actino (asym)	14
	K4 A4+4 G4				
B. stuebelii	K5A5+5G5	7	1	actino (asym)	10
B. subsessilis	K5A5+5G5	7	3	actino (asym)	10
	K6A6+6+6G6				
B. susaconensis	K5A5+5G4-3(2)	12	11	asym	48
B. tomentosa	K5A5+5G2(3)	7	-	asym	14
B. tnanae	K5A5+5G5-3-4	22	15	asym	55
	K4A4+4G4				
	K5G5				
B. tngyna	K5A5+5G3(5)	28	13	asym	62
	K6A6+6 G4				
B. velutina	K5A5+5G5	27	-	actino	27
*B. weberbauen	k5A10?	1	-	-	1
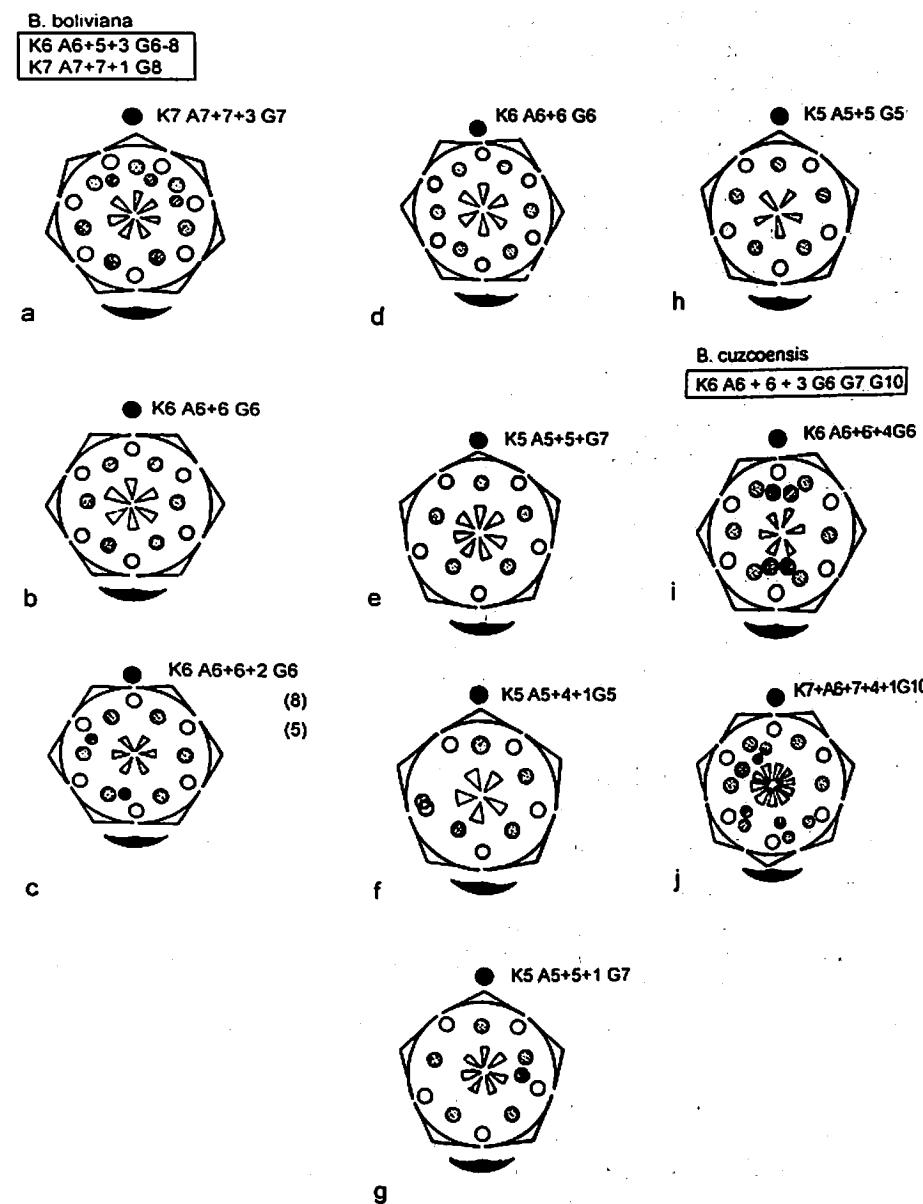
Fig. 5.2 Floral diagrams of *B. boliviana* (a-h) and *B. cuzcoensis* (i, j). a, b, c. (SOLOMON 8028). g, h (SOLOMON 1643). d (DAVIDSON 4775). e (SOLOMON 1643). i, j, (VARGAS 17827). O first whorl of stamens, ⊕ second whorl of stamens, ⦿ third whorl of stamens, ● fourth whorl of stamens.

equals the number of carpels (Table 5.1), as in *B. hygrothermica, B. latifolia, B. morii, B. ovalifolia, B. pauciflora, B. stenoptera, B. stuebelii,* and *B. subsessilis,* but some of these species present alterations of the androecium, due to the presence of additional stamens in additional whorls or fewer stamens in the two whorls. Reduction of stamens was observed as fusion of anthers or filaments or in the presence of staminodes.

Variations in the number of carpels are common, whereas sepal number is relatively stable. Thus, the terms tetramerous, pentamerous etc., refer to sepal number. Twenty two species have fewer or more carpels than sepals (Table 5.1). For example the following species usually have 5 sepals but there are variations in the number of carpels: *B. ecuadorensis* (2 carpels), *B. farallonensis* (2-3 carpels), *B. propinqua* (2-3), *B. rufa* (3-4), *B. susaconensis* (3-4), and *B. tomentosa* (2-3) (Table 5.1). Flowers having three sepals and carpels were found in *B. foreroi* and *B. acutangula.* A larger number of carpels than sepals was seen in *B. subsessilis* (7-8 carpels). In species with additional stamens or apparent polystemonous floral organ arrangement such as *B. cuzcoensis,* flowers with 6 sepals usually have 7 and 10 carpels; in *B. boliviana* flowers with 6 sepals have 8 carpels.

Flower symmetry also varies due to difference in width of the sepals. Sepal fusion is related to the increase or decrease of the number of stamens such as in *B. goudotii* (Fig. 5.4 l), *B. integrifolia* (Fig. 5.6 f), *B. macrophylla* (Fig. 5.6 b), *B. pauciflora* (Figs. 5.1 h ; 5.8 d) and *B. susaconensis* (Fig. 5.4 c). Weak fusion initially joins two sepals by a median line which is completed later. Each one of the two united sepals possesses its own midvein and its two lateral veins. Complete fusion of the sepals is seen in *B. pauciflora* (Fig. 5.1h). Simultaneously with sepal fusion there is a change in the size of the disk where the stamens are positioned, thus some stamens are now localized in an apparent third whorl.

The usual floral pattern has 5 sepals (rarely 4 sepals). However, increase and reduction of stamens affect the symmetry of the flower. In species with a pattern of two whorls of stamens such alterations are considered transitional stages (Figs. 5.1d, g, h; 5.3; 5.4; 5.5 c, d, f-j; 5.6; 5.7; 5.8 b-f). It is rare to find flowers with two whorls of stamens in species which often have additional whorls (Fig. 5.2).

Few cases of additional whorls with one or two or more stamens were seen in *B. darienensis, B. littlei, B. macrophylla, B. morii, B. racemifera, B.ovalifolia,* and *B. goudotii* (Figs. 5.4 g-l; 5.5 c; 5.6 b). These variations of the androecium are probably related to the inter-specific variations of the sepal merosity. In this case, changes occurred from flowers with 6 sepals and two whorls of stamens, due to the current variation of floral arrangement within each species. However, in *B. ovalifolia* and *B. racemifera,* floral arrangements larger than K6 A6+6 were not observed and the presence, of one additional whorl with one stamen in the case of *B. ovalifolia* and five stamens in a third whorl in *B.*
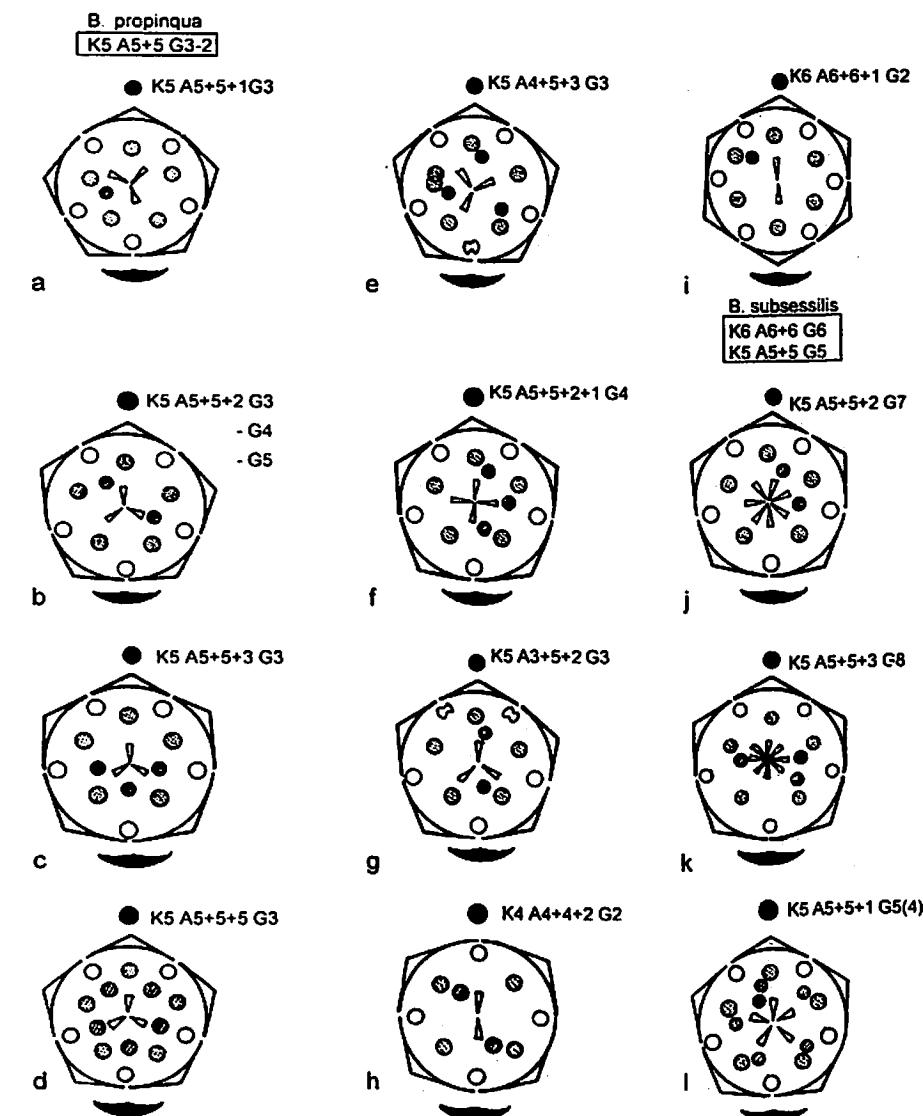
Fig. 5.3 Floral diagrams of *B. propinqua.* (a-i) and *B. subsessilis.* (j, k). a (CUATRECASAS 27399, OROZCO et al. 1750). b (FORERO 3701, HAWKES 145, OROZCO et al.1778). c (FORERO 370, OROZCO et al. 1778). d (FORERO 3701). e, h , g (OROZCO et al. 1778). j (OROZCO et al. 1750). f (OROZCO et al. 1772). j, k, l (ARCHER 1519). O first whorl of stamens, ⊕ second whorl of stamens, ● third whorl of stamens, ⊃ staminode.
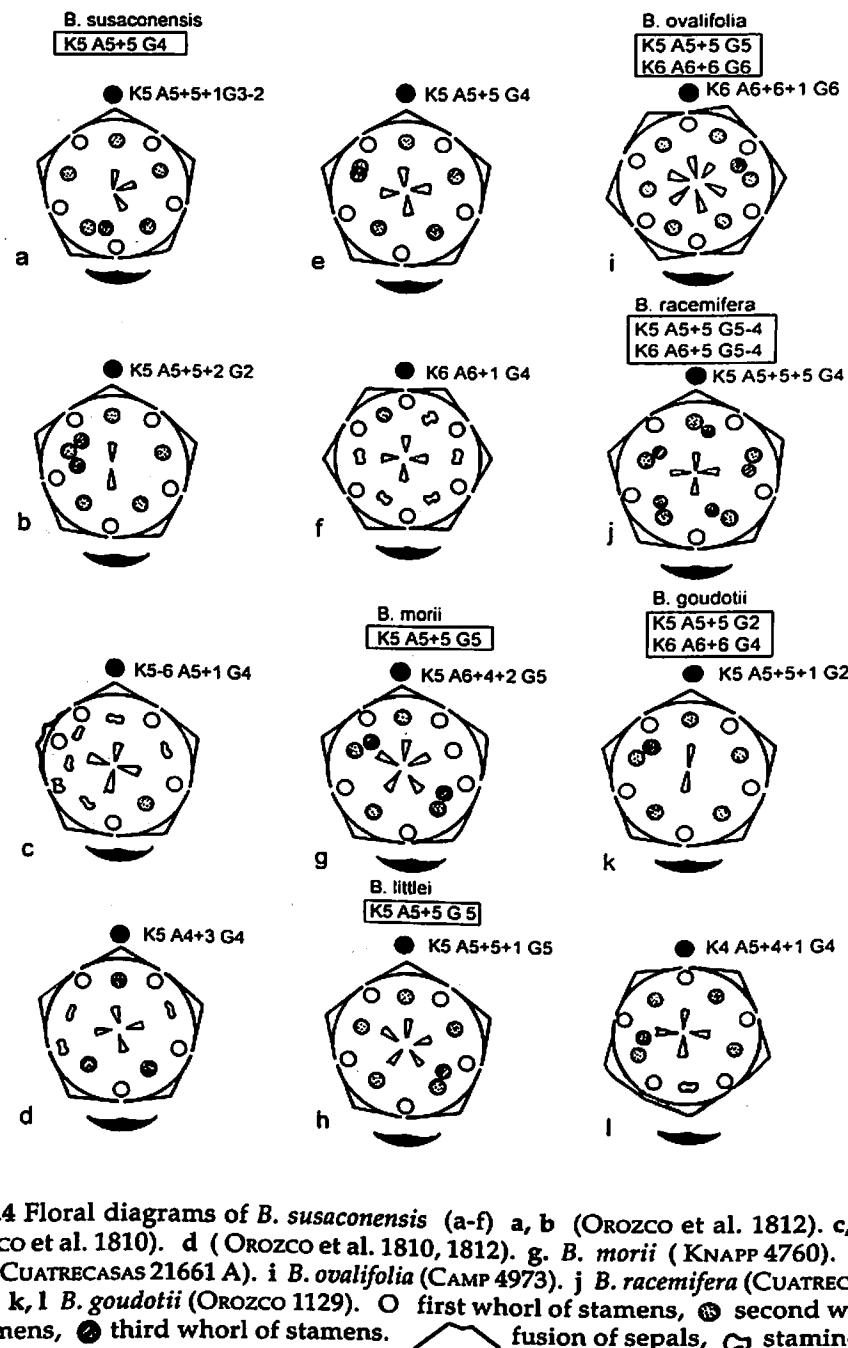
Fig. 5.4 Floral diagrams of *B. susaconensis* (a-f) **a, b** (OROZCO et al. 1812). **c, e, f** (OROZCO et al. 1810). **d** (OROZCO et al. 1810, 1812). **g.** *B. morii* (KNAPP 4760). **h** *B. littlei* (CUATRECASAS 21661 A). **i** *B. ovalifolia* (CAMP 4973). **j** *B. racemifera* (CUATRECASAS 8028). **k, l** *B. goudotii* (OROZCO 1129). O first whorl of stamens, ⊛ second whorl of stamens, ● third whorl of stamens. ⌒ fusion of sepals, ⌀ staminode.

racemifera, indicates reduction or increase in number from a larger number than K6 A6+6 and from an even greater number in *B. racemifera* (Fig. 5.4 i, j). In *B. integrifolia*, the variation of sepal merosity leads to the androecium variation. The frequent presence of the floral organ arrangement K6 A6+6 indicates variation from this plan. The reduction and increase of stamens relating to sepal merosity is due to the transition from K6 A6+6 to probably K4 A4+4 because of the presence of fewer stamens than ten (Fig. 5.6 g)

Reductions or a tendency to reduction to less than K5 A5+5 were observed in *B. acutangula*, *B. glabra*, *B. hygrothermica*, *B. integrifolia*, *B. latifolia*, *B. macrophylla*, *B. pauciflora*, *B. putumayensis*, *B. stenoptera*, and *B. trigyna* (Figs. 5.5 d, e - j; 5.4; 5.6 c, d, g, h-j; 5.7 c-l; 5.8 a-f). An example where the reductions to less than K5 A5+5 is seen clearly in *B. pauciflora* (Figs. 5.8 a-d). The presence of an additional stamenK 4A4+4+1G4 (Fig. 5.8 d), indicates a transitional state from an arrangement of K5A5+5G5 toward the more typical arrangement of flowers with K4A4+4G4. Three transitional states (K5 A5+4G5, K5A5+3 G5, and K4A4+4+1G4) were found between the floral plans K5A5+5G5 and K4A4+4G4 (Figs.5.8b, c). In Fig. 5.8 d, fusion of two sepals is shown, which is correlated with reductions of stamen number in the same whorl. Reductions of stamens by fusion of filaments is illustrated at Figs. 5.2 f, 5.4 e, 5.8 c.

The greatest reduction was observed in *B. trianae* (Figs.5.7 g-l); but two whorls of stamens are usually present. This reduction was observed in collections from Llanos de Cuibá and Cerro del Padre Amaya in Antioquia, Colombia.

In some cases, staminodes were observed in an outer extra-whorl e.g. in *B. dulcis*, *B. darienensis* (Figs. 5.5 a, b). These cases could imply reductions of whorls in the floral organ arrangement of *Brunellia*.

5.4 DISCUSSION AND CONCLUSIONS

Flowers in *Brunellia* have been described previously as bisexual or unisexual (male or female), but flowers described as male have carpels which hold ovules. Protandry occurs in *Brunellia*, the presence of carpels was always confirmed in the flowers of species examined. The protandry facilitates outcrossing.

Dorsifixed, introrse, and more or less sagittate anthers with a slight connective protrusion, not clearly observable in all species of *Brunellia*, are characters that correspond to the 'Lower Rosidae' as was pointed out by ENDRESS and STUMPF (1991).
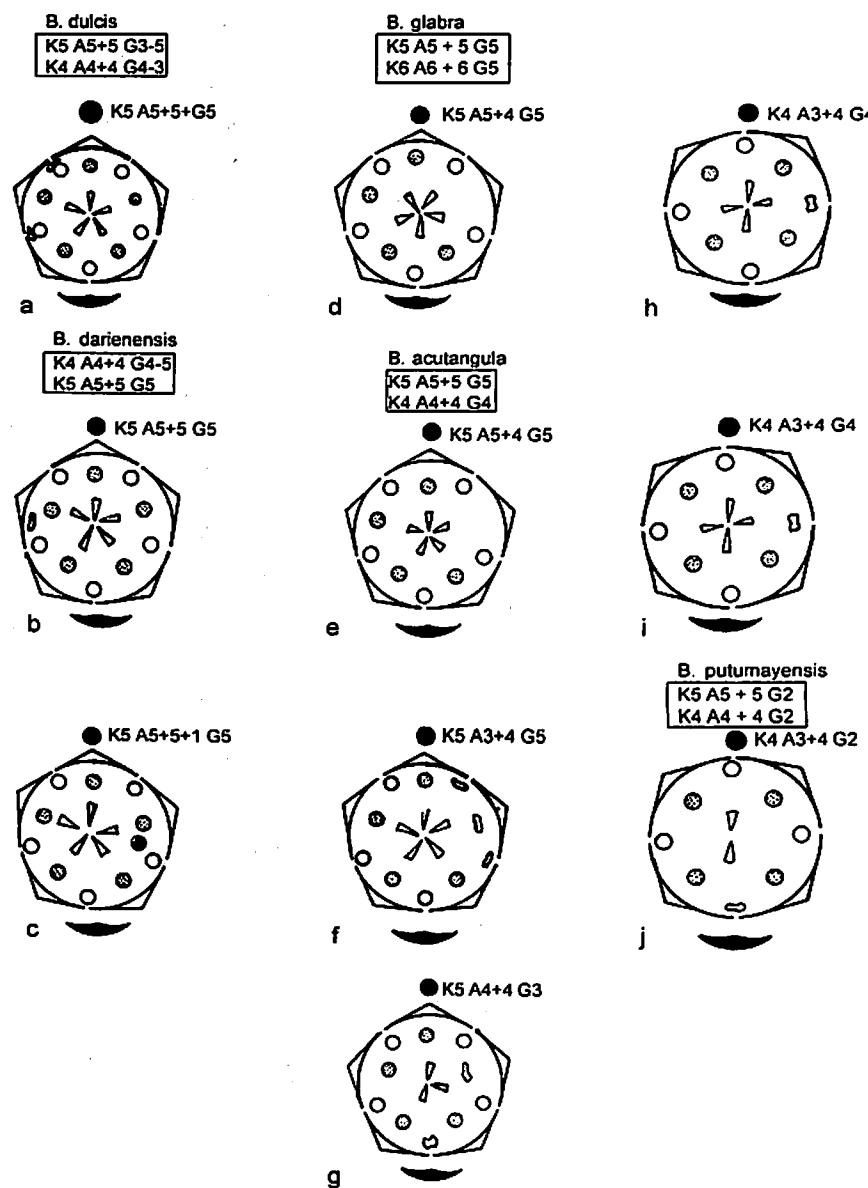
Fig. 5.5 Floral diagrams of *B. dulcis*. a (MACBRIDE 74), *B. darienensis*. b-c BRANT and LUTEYN 12135). d *B. glabra* (LUTEYN 7387), *B. acutangula* (e-i). j *B. putumayensis* (CUATRECASAS 29005). f (CUATRECASAS 29005). g-h (VIDAL 114). i (CUATRECASAS 29005). O first whorl of stamens, ⊛ second whorl of stamens, ⊘ third whorl of stamens, ꙮ staminode.
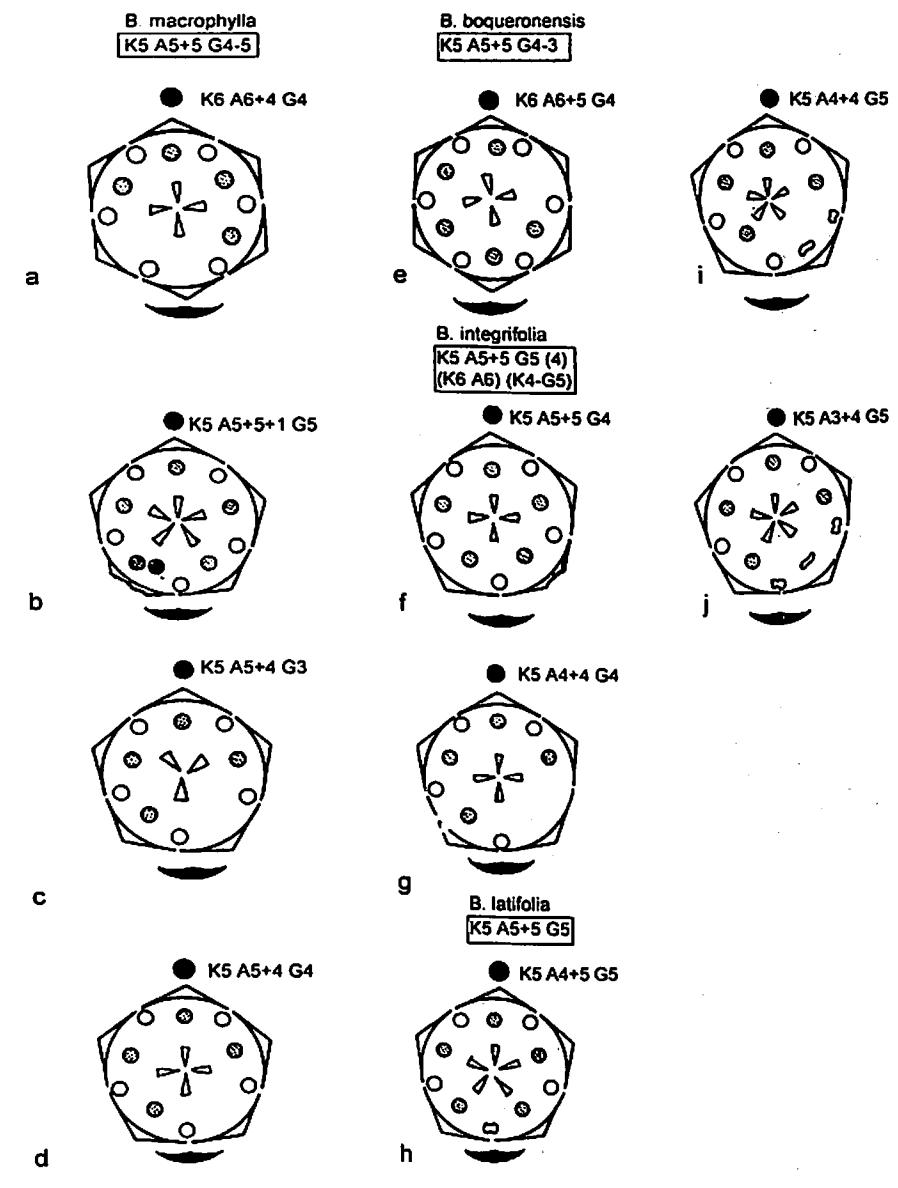
Fig. 5.6 Floral diagrams of *B. macrophylla* a-d (CUATRECASAS 8575 A). e *B. boqueronensis* (OROZCO 1416). f, g *B. integrifolia* (ESPINAL 3189). h-j *B. latifolia* (CUATRECASAS 18109). O first whorl of stamens, ⊛ second whorl of stamens, ⬤ third whorl of stamens. ⌢⌢⌢ fusion of sepals.
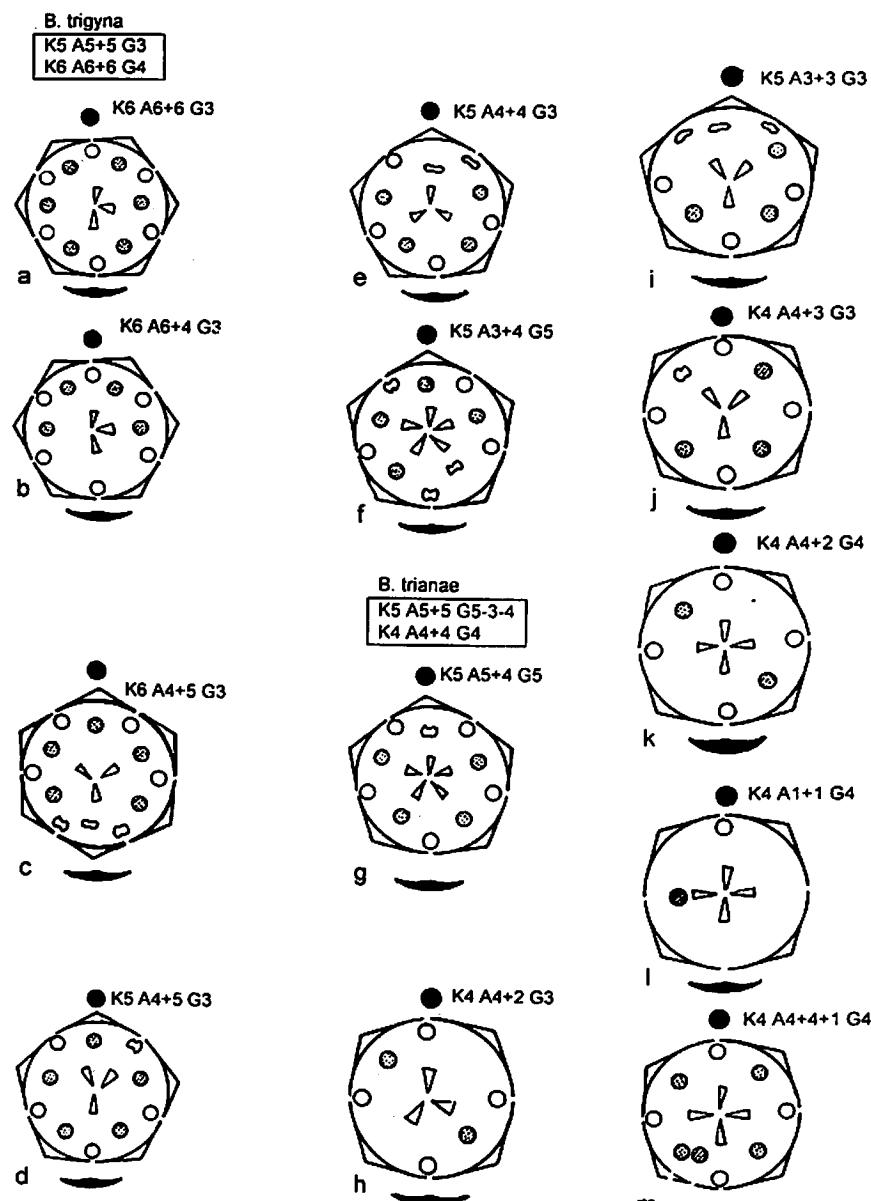
Fig. 5.7 Floral diagrams of *B. trigyna* (a-f) *B. trianae* (g-m). a-d (RUIZ-TERÁN 1203). b (LOZANO 5518). c-f (LUTEYN 5330) g-m (LOZANO 3969). h (LLANOS s.n ; LOZANO 3969). i, k, f (LLANOS s.n.). j (LLANOS s.n ; LOZANO 3969). O first whorl of stamens, ● second whorl of stamens, ◉ third whorl of stamens, ◯ staminode.

Observation of numerous individuals shows that the flower in *Brunellia* tends to an asymmetric condition. Only very few species exhibit an actinomorphic symmetry (Table 5.1). The asymmetric condition in *Brunellia* is expressed in sepal size, the number of carpels being larger or smaller than the number of sepals, and increase and reductions of stamen number. Two whorls of stamens are usually present in *Brunellia*, however, some species, *B. boliviana* and *B. cuzcoensis*, have an apparent polystemonous floral arrangement. They have flowers with additional whorls of stamens (Fig. 5.2).

For most of the species, reduction (sepals and stamens) in number of parts occurs. Fusion of sepals and, as consequence, alterations in the number of stamens of each whorl, owing to alterations in the position of the stamens, was also observed by LEINS and ERBAR (1991) who found a relationship between the changes in merosity of trimerous and pentamerous flowers and the position of the stamens. ENDRESS (1992) also found alterations in the relative size (by increase or reduction) of a particular primordium and of the entire floral area. These observations also imply that the polystemonous condition is an artifact and is not a real condition in *Brunellia* and that changes in the space occupied by the stamens produce an apparent polystemonous condition by fusion of sepals.

The very common presence of additional whorls of stamens in flowers of some species of *Brunellia* seems to be also related to changes in merosity of the sepals. This could be a tendency toward a stable flower with two whorls due to the continuous reductions, of the number of parts, observed in flowers of these species.

From a phylogenetic analysis of *Brunellia* (Chapter7) the increase in stamen whorls is seen as secondary condition from a basic floral pattern of two whorls of stamens as was also pointed out by (HUFFORD and DICKISON 1992; OROZCO 1997).

A high correlation of 49% between the alternation of the outer whorl of stamens with the sepals and absence of petals was recorded by RONSE DECRAENE and SMETS (1995). There is no evidence of a concomitant shift in stamen position as a consequence of the loss of petals in *Brunellia*. This is due to the absence of petals remnants and all species of *Brunellia* lack petals.

Although two whorls of stamens are also reported in Cunoniaceae, the alternate condition of the outer stamens to the sepals, as was found in *Brunellia*, is not present in most of the genera of Cunoniaceae. HOOGLAND (1979) noticed the presence of extra stamens in additional whorls in two genera of Cunoniaceae, *Spiraeanthemum* and *Acsmithia*, closely related to *Brunellia* (HUFFORD and DICKISON 1992, OROZCO 1997, BRADFORD pers. comm.) These genera lack petals and the presence of an alternate position of the outer stamens to the sepals, is possible, as occurs in *Brunellia*.

The relation between changes of androecium merosity with changes in carpel merosity as was suggested by RONSE and DECRAENE (1994) for other groups of angiosperms is not observed in *Brunellia*. However, if the fusion of sepals alters the position of the stamens, this could also affect the merosity of carpels by obstruction of their development. A larger number of carpels was
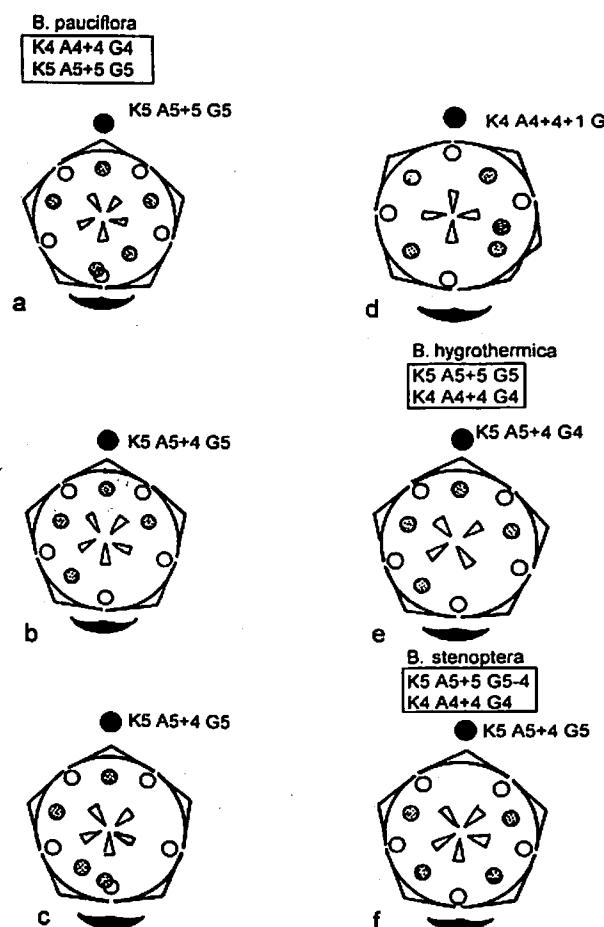
Fig. 5. 8 Floral organ arrangements of *B. pauciflora* (a-d). a (PALACIOS et al. 6987). d (PALACIOS et al. 3897,6978). e *B. hygrothermica* (BERNAL et al. 1105). f *B. stenoptera* (ASPLUND 18698). O first whorl of stamens, ⊗ second whorl of stamen, ● third whorl of stamens. ⌒⌒ fusion of sepals.

observed in those species that usually have additional whorls of stamens, but few carpels, two or three, are also present in species with additional whorls of stamens such as in *B. propinqua* and *B. susaconensis*.

This is the first attempt to understand the biological causes of the merosity variation in *Brunellia* and many ambiguities must be elucidated in future work. The study of early floral states of taxa related to *Brunellia* is necessary to understand the relation between the floral organ arrangement and the lack of petals. These studies could be applied to genera with species having flowers with and without petals. The interdependence of carpel and stamen merosities, among other questions, could be resolved by study of early stages of floral development in *Brunellia* by electron scanning microscopy.

5.5 ACKNOWLEDGMENTS

5.6 LITERATURE

APG, 1998. An ordinal classification for the families of flowering plants. Ann. Missouri Bot. Gard. 85 (4): 531-553.

CHATIN, A. 1855. Sur les types obdiplostémones et diplostémones direct ou de l'existence et des caractères de deux types symmétriques distincts chez les fleurs diplostémones. Bull. Soc. Bot. France 2: 615-622.

CLINCKEMAILLIE D. and SMETS, E. F. 1992. The systematic importance of the floral similarities between Plumbaginaceae and Primulaceae. Belg. J. Bot. 125: 151-153.

CORNER, E. 1946. Centrifugal stamens. J. Arnold Arbor. 27: 423-437.

CRONQUIST, A. 1981. An integrated system of classification of flowering plants. Columbia Univ. Press. New York.

CUATRECASAS, J. 1970. Brunelliaceae. Flora Neotropica Monograph 2: 1-189.

CUATRECASAS, J. 1985. Brunelliaceae. Flora Neotropica Monograph 2 (suppl.): 29-103.

DICKISON, W.C. 1989. Comparisons of primitive Rosidae and Hamamelidae. In: Evolution, systematics, and fossil history of the Hamamelidae, vol. 1:

Introduction and 'Lower' Hamamelidae (eds. P.R. CRANE and S. BLACKMORE), pp. 47-73. Oxford, Clarendon Press.

EAMES, A. J. 1963. Morphology of the angiosperms. McGraw-Hill. New York.

ENDRESS, P. and STUMPF, S. 1991. The diversity of stamen structures in 'Lower Rosidae (Rosales, Fabales, Proteales, Sapindales). Bot. J. Linn. Soc. 107: 217-293.

ENDRESS, P. 1992. Evolution and floral diversity: The phylogenetic surroundings of *Arabidopsis* and *Anthirrhinum*. Int. J. Plant Sci. 153: 106-122.

ERBAR, C. 1988. Early developmental patterns in flowers and their value for systematics. In: Aspects of floral development (eds. LEINS P, TUCKER SC, ENDRESS PK), pp. 7-23. Berlin.

HOOGLAND, R.D. 1979. Studies in the Cunoniaceae II. The genera *Caldcluvia*, *Pullea, Acsmithia*, and *Spiraeanthemum*. Blumea 25: 481-505.

HUFFORD, L. and DICKISON, W 1992. A phylogenetic analysis of Cunoniaceae. Syst. Bot. 17: 181-200.

LEINS, P. and ERBAR, C. 1991. Fascicled androecia in Dilleniidae and some re-marks on the *Garcinia* androecium. Bot. Acta 104: 336-344.

OROZCO, C.I. 1985. Dos especies nuevas de *Brunellia* para Colombia. In: (ed. J. CUATRECASAS). Flora Neotropica Monograph 2 (suppl.): 20-24.

OROZCO, C.I. 1986. Contribución al estudio de Brunelliaceae de Colombia III. Caldasia 15: 177-185.

OROZCO, C.I. 1991. Contribución al estudio de Brunelliaceae IV. Dos especies del Ecuador. Caldasia 16: 453-458.

OROZCO, C.I. 1997. Sobre la posición sistemática de *Brunellia* Ruiz & Pavón. Caldasia 19: 145-164.

OROZCO, C. I. and WEBERLING, F. 1999. A comparative study of inflorescences in *Brunellia* Ruiz & Pav. (Brunelliaceae) and related taxa. Beitr. Biol. Pflanzen 71: 261-279.

RONSE DECRAENE, L. P. and SMETS, E. F. 1992. Complex polyandry in the Magnoliatae: definition, distribution and systematic value. Nord. J. Bot. 12: 621-649.

RONSE DECRAENE, L. P. and SMETS, E. F. 1993. The distribution and systematic rel-evance of the character polymery. Bot. J. Linn. Soc. 113: 285-350.

RONSE DECRAENE, L. P. and SMETS, E. F. 1994. Merosity in flowers: definition, origin, and taxonomic significance. Pl. Syst. Evol. 191: 83-104.

RONSE DECRAENE, L. P. and SMETS, E. F. 1995. The distribution and systematic relevance of the androecial character oligomery. Bot. J. Linn. Soc. 118: 193-247, 285-350.

WEBERLING, F. 1989. Morphology of flowers and inflorescences. Cambridge University Press. Cambridge.

Chapter 6

POLLEN MORPHOLOGY OF *BRUNELLIA* RUIZ & PAVÓN AND RELATED TAXA[1]

CLARA INÉS OROZCO

ABSTRACT

Pollen grains of 24 species of *Brunellia* were examined with LM and SEM in order to find additional characters for phylogenetic analysis. The pollen grains were found to be 3-colporate, tectate and to have a variable ornamentation which forms an almost perfect continuum. Five categories of exine ornamentation were observed: striate reticulate (large lumina and high muri), finely reticulate, modified reticulate (muri and lumina irregular in shape and at various levels), modified rugulate (irregular and protruding tectal elements) and punctate (the lumina are smaller and round to slit-shaped). The exine ornamentation provides phylogenetic information for some groups of species, and in some cases it is correlated with vegetative, inflorescence and fruit characteristics. The punctate type could be the plesiomorphic character state. Relationships in the pollen morphology of *Brunellia* and certain genera of Cunoniaceae are discussed.

RESUMEN

El polen de 24 especies de *Brunellia* fue examinado en el microscopio de luz (ML) y el microscopio electrónico (MES) para encontrar caracteres adicio-nales y usar esta información en el estudio de relaciones filogenéticas. El gra-no de polen es 3-colporado, tectado y con una alta variabilidad en la

---

ornamentación de la exina la cual forma un casi perfecto continuo. Se observaron cinco categorías en la ornamentación de la exina del grano de pollen: reticulado a finamente reticulado, reticulado modificado (muros y lúminas de forma irregular y en varios niveles), rugulada modificada (irregular y proyectando elementos tectales) y la ornamentación de tipo punteado (las lúminas son pequeñas redondeadas o en forma de líneas). La ornamentación de la exina tiene importancia filogenética para algunos grupos de especies y presenta correlación con algunos caracteres vegetativos de inflorescencias y del fruto. El tipo de ornamentación punteado parece ser el estado plesiomórfico. Se discuten las relaciones del grano de polen de *Brunellia* y ciertos géneros de Cunoniaceae.

6.1 INTRODUCTION

The genus *Brunellia* Ruiz & Pavón includes 54 species (Orozco in prep.) and is currently considered the only genus of the family Brunelliaceae. *Brunellia* is neotropical and widely distributed in Andean and subandean forests where high humidity and frequent cloudiness prevail. Species of *Brunellia* also occur from Mexico to Bolivia and in the Greater Antilles. The highest number of species, 34, is present in Colombia (Chapter 1)

Most of the earlier taxonomic classifications indicated relationships between Brunelliaceae and Cunoniaceae and often both families were placed in the order Rosales (CRONQUIST 1981, DAHLGREN 1980, TAKHTAJAN 1981, THORNE 1983). CUATRECASAS (1970) also noted a close relationship between *Brunellia* and Cunoniaceae, especially with the genera *Spiraeanthemum* and *Acsmithia*, based on vegetative and anatomical characters. The relationship of the two families, however, is currently under discussion; both families have been included in the order Oxalidales (APG 1998). Research on the position of *Brunellia* has also been carried out by HUFFORD and DICKISON (1992) who placed *Brunellia* in the family of Cunoniaceae, whereas OROZCO (1997) suggested that *Brunellia, Spiraeanthemum* and *Acsmithia* form a separate family, closely related to the Cunoniaceae. However, BRADFORD and BARNES (2001) from his studies on molecular data, considers that Brunelliaceae must be retained as a monotypic family. He indicates that Brunelliaceae show close relationships with *Cephalotus* (Cephalotaceae) and with Cunoniaceae, but Brunelliaceae probably does not fall within the Cunoniaceae clade. It was indicated by HICKEY and TAYLOR (1991) and also by OROZCO (1997) that Cunoniaceae is not a monophyletic group.

Cunoniaceae, Brunelliaceae, *Eucryphia* (Eucryphiaceae) and *Davidsonia* (Davidsoniaceae) are considered by DICKISON (1989) to comprise the basal group in the rosalean complex. Connaraceae is another closely related family to Brunelliaceae. It used to be included in Rosales but now it is placed together with Brunelliaceae and Cunoniaceae in the order Oxalidales (AGP 1998). Most

of the genera of Connaraceae, with exception of *Agelaea, Cnestidium* and *Cnestis* share the presence of five vascular traces in the carpels with *Brunellia, Spiraeanthemum* and *Acsmithia* (DICKISON 1971). Other characteristics shared among *Brunellia, Spiraeanthemum* and *Acsmithia* and also by *Connarus*, include the absence of petals and the apocarpous gynoecium. The epitropous ovule and the absence of a specialized xylem with scalariform pitting are also seen in *Brunellia, Spiraeanthemum* and *Acsmithia* (DICKISON 1975, 1980, CUATRECASÁS 1970, 1985). From molecular data it appears (BRADFORD and BARNES 2001), that Brunelliaceae is closer to Cunoniaceae and Cephalotaceae than to Connaraceae among the families of the order Oxalidales. HUFFORD and DICKISON (1992), OROZCO (1997) and BRADFORD and BARNES (2001), consider the genera *Eucryphia* and *Davidsonia* within Cunoniaceae.

The first observations of pollen grains of *Brunellia* were made in *B. comocladifolia* by ERDTMAN (1952) who with regard to sexine described the grains as having a rough surface and somewhat irregular sculpture. Basak (1967) described the pollen grain of *B. boliviana* and *B. comocladifolia*. He noticed differences in the surface pattern of the two species, in *B. boliviana* an echinulate reticulate pattern being observed, whereas the pattern in *B. comocladifolia* was seen as irregular to slightly echinulate. Fossil pollen of *Brunellia* was first reported by GRAHAM and JARZEN (1969) from Puerto Rico. They did not find differences between the fossil grains of *Brunellia* and the modern species including *B. comocladifolia*. SALOMONS (1986) described fossil pollen of *B. goudotii* using LM. He observed intectate or semitectate pollen grains with verrucate, fossulate, scabrate or microreticulate sculptures. MARTICORENA (1970) studied several species of *Brunellia* using LM. He found two basic patterns of exine structure, one with a finely reticulate sexine without differentiated columellae and the other intectate with verrucose processes. Results reported by OROZCO (1991) differ from those of Marticorena as the former did not find intectate pollen grains in the SEM observations.

There are few complete studies on the pollen morphology of groups considered close to *Brunellia*. HIDEUX and FERGUSON (1976) studied pollen grains, under light (LM) and scanning electron microscopy (SEM) of Saxifragaceae *sensu lato*, including Saxifragaceae *sensu stricto*, Cunoniaceae, Escalloniaceae and Hydrangeaceae. Pollen morphology in Connaraceae was studied by DICKISON (1979) using LM and SEM.

Observations on pollen grains of groups related to *Brunellia*, such as those of WALKER and DOYLE (1975), indicate that tricolporate grains and finely reticulate (small lumina) sculpture are especially common in many basal groups of Rosidae. It seems that the oblate triangular grains with apertures at the angles (angulaperturate) is a character present in the basal groups of Rosidae as it is present in Cunoniaceae. This character is also present in Connaraceae, however, this family was not considered a basal group of Rosidae (DICKISON

Table 6. 1 Morphological characteristics of pollen grains of *Brunellia* species. All sizes, are given in μm; (mean sizes in parentheses). Polar axis (P), equatorial diameter (E), ratio of polar axis and equatorial diameter P/E, colpus length (L), and width (W), pore length (L) and width (W), distance between the apices of the colpus (A), polar index (A/E). Exine sculpture categories: Finely reticulate (FR), Modified reticulate (MR), Punctate (Pu), Striate reticulate (SR), Rugulate (Ru).

Species	P	E	P/E	Colpus L	Colpus W	Endopore L	Endopore W	A/E	Exine thickness	Exine sculpture
B. boliviana Cuatrec	18.5-20.0 (10.5)	14.5-18.0 (17)	1.05	12.0-16.0 (14)	0.8-2.0 (1.2)	3.0-4.0 (3.2)	3.0-4.0 (3.2)	0.32	1.3-1.5 (1.5)	Ru
B. brunnea Macbride	19.0-25.0 (22.1)	19.0-25.0 (21.7)	1.02	16.0-22.0 (18)	1.0-2.0 (1.6)	2.0-3.0 (2.9)	1.0-2.0 (2.0)	0.24	1.5-2.0 (1.7)	Pu
B. cayambensis	24.5-32.5 (30.1)	20.0-26.0 (23.6)	1.28	10.0-12.0 (11)	0.5-1.5 (0.8)	2.0-3.0 (3.0)	1.0-2.0 (2.0)	0.27	1.0-1.5 (1.4)	Ru
B. comocladifolia H. B. K subsp. *comocladifolia*	15.0-16.0 (15.5)	11	1.48	10.0-12.0 (11)			1.0-2.0 (2.0)		1.0-1.5 (1.4)	SR
B. comocladifolia H. B. K subsp. *cundinamarcensis* Cuatrec	13.0-14.5 (13.6)	11.0-12.0 (11.8)	1.15	20.0-25.0 (22.4)	1.0-3.0 (2.2)			0.3	1.8-2.6 (2.2)	FR
B. costaricensis Standley	22.5-24.0 (23.4)	18.0-22.0 (19.8)	1.18	17.5-20.0 (19)	0.5-1.5 (0.7)			0.32	1.5-1.8 (1.8)	Pu
B. costanensis Standley	15.0-21.0 (19.6)	14.5-18.5 (16.6)	1.11					0.33	1.0-1.5 (1.3)	Pu
B. darienensis Cuatrec. & Porter	16.5-18.0 (18.0)	12.5-18.0 (17.0)	1.05	13.0-14.5 (13.5)	0.5-0.8 (0.5)			0.3	1.6-2.5 (1.8)	FR
B. dulcis Macbride	16.0-19.2 (17.4)	16.0-17.0 (16.5)	1.05	12.0-15.0 (13.5)	0.5-1.5 (1.0)			0.37	1.3-1.8 (1.6)	MR
B. glabra Cuatrec.	22.0-28.0 (25.3)	19.0-24.0 (22.0)	1.15	17.0-20.0 (20)	1.0-1.5 (1.1)	4.0-8.0 (5.2)		0.31	1.0-1.8 (1.5)	MR
B. goudotii Tulasne	23.5-26.0 (24.0)	20.0-23.5 (21.3)	1.13	8.0-9.5 (9)	0.5-1.0 (1.0)			0.29	1.0-1.2 (1.1)	MR
B. hygrothermica Cuatrec.	11.0-12.5 (11.7)	12.0-14.5 (13.0)	0.9	13.0-18.0 (16)	0.5-2.5 (0.8)			0.42	1.3-1.7 (1.6)	FR
B. inermis Ruiz & Pavón	17.0-23.0 (21.3)	17.0-19.0 (17.7)	1.2	12.0-17.0 (13)	1.0-2.5 (1.6)			0.29	1.0-1.8 (1.6)	MR
B. latifolia Cuatrec.	15.5-21.0 (17.3)	17.0-20.0 (18.2)	0.92	11.0-13.5 (12)	1.0-1.5 (1.0)			0.32	1.5-2.0 (1.8)	Ru
B. propinqua H. B. & K.	15.0-18.0 (16.05)	15.5-18.0 (17.1)	0.83	13.5-22.0 (19)	1.8-2.5 (2.0)			0.31	1.2-1.8 (1.6)	Ru
B. propinqua H. B. & K.	18.0-28.0 (24.0)	18.0-23.0 (21.6)	1.11	16.0-20.0 (19)	1.5-2.0 (1.7)			0.22	1.2-1.5 (1.4)	Pu
B. putumayensis Cuatrec.	21.8-25.0 (23.3)	16.5-19.5 (18.1)	1.29	14.0-18.0 (17.2)	0.5-1.5 (0.8)	2.0-3.0 (2.4)	5.0-6.5 (6.4)	0.26	1.6-2.0 (1.6)	MR
B. racemifera Tulasne	18.0-20.8 (21.7)	14.0-18.5 (17.4)	1.24	14.0-19.0 (16.5)	0.5-1.0 (0.9)	2.0-3.5		0.3	1.2-1.5 (1.4)	MR
B. rhodes Rusby	12.5-18.0 (15.8)	13.0-15.0 (13.9)	1.13	9.0-15.0 (12.3)	0.5-1.5 (0.8)	2.5-4.0 (3.1)			1.0-2.0	FR
B. sibundoya subsp. *antioquensis* Cuatrec.	11.5-14.5 (13.3)	10.5-12.5 (11.3)	1.17	9.5-12.0 (10.3)	0.5-1.0 (0.8)			0.27	1.0-2.0 (1.1)	Ru
B. sibundoya Cuatrec. subsp. *sibundoya*	13.0-14.0 (13.5)	9.0-11.5 (10.5)	1.3	9.1-11.5 (9.2)						
B. stuebelii Hieronymus	14.0-18.0 (16.6)	14.5-18.0 (16.3)	1.01	10.0-15.0 (12.6)	0.5-1.5 (0.8)			0.3	1.0-2.0 (1.7)	MR
B. subsessilis Kslip & Cuatrec.	19.0-23.0 (20.6)	17.0-19.0 (17.7)	1.16	14.0-19.0 (16.5)	1.0-2.0 (1.2)			0.21	1.3-2.3 (1.5)	Pu
B. susaconensis Cuatrec.	22.8-29.5 (27.0)	21.0-28.0 (23)	1.33	21.0-25.0 (23)	2.0-4.0 (2.9)			0.27	1.5-2.0 (1.7)	Pu
B. tomentosa H. & B	17.5-19.5 (18.8)	15.5-17.5 (16.5)	1.01	14.0-16.0 (14.5)	0.5-1.0 (0.6)			0.31	1.0-2.0 (1.2)	MR
B. trianae Cuatrec.	17.5-19.5 (18.8)	16.5-18.5 (17.6)	1.07	13.0-15.0 (14.4)	1.0-2.5 (1.8)			0.31	1.3-1.8 (1.6)	MR
B. trigyna Cuatrec.	14.0-16.0 (14.7)	15.0-18.0 (16.7)	0.88	11.5-13.0 (11.8)	0.5-2.0 (1.2)			0.25	1.0-1.6 (1.2)	Pu
B. weberbaueri Loesener	19.0-23.0 (20.4)	16.5-19.0 (18.1)	1.12	16.0-17.0 (16.8)	0.8-1.5 (1.1)	2.0-4.0 (3.2)	4.0-8.0 (5.65)	0.32	1.3-1.5 (1.4)	Ru
Spiraeanthemum katakata Seem.	7.5-9.0 (8.5)	10.0-11.5 (10.6)	0.79	8.0-10.5 (8.7)	0.5-1.0 (0.5)			0.35	0.8-1.0 (0.9)	Pu

---

1989). HIDEUX and FERGUSON (1976) consider that the finely reticulate grains present in Cunoniaceae (*Vesselowskya* excepted) are unspecialized, whereas grains with complete tectum and supratectal processes are specialized conditions present in the grains of Saxifragaceae *sensu stricto*.

The present paper provides detailed information on the pollen grains of 24 species of *Brunellia*, using both LM and SEM. Micrographs of pollen grains of some species of Cunoniaceae, *Aistopetalum viticoides*, *Caldcluvia paniculosa*, *Geissois superba*, *Lamanonia tomentosa* were compared with grains of *Brunellia* as a close relationship was expected.

6.2 MATERIALS AND METHODS

Anthers from open flowers were removed from herbarium specimens (COL, NY, MO, QCA and US) of 22 species of *Brunellia* and of *Spiraeanthemum katakata* belonging to Cunoniaceae (Table 6.1). Data of two additional species of *Brunellia* previously studied were included in the present analysis (OROZCO 1991). Flowers of *Acsmithia* were not available at that time. Grains were acetolysed as outlined by Erdtman (1966). For SEM, pollen was mounted on stubs, coated with palladium gold and examined and photographed with a Hitachi S-570 scanning electron microscope. The species examined represent about half of the total number of *Brunellia* species and represent almost all of the clades from a phylogenetic analysis of *Brunellia* (Chapter 7).

Slides for LM were made with glycerine jelly and sealed with paraffin. Sets of these preparations were deposited in US and COL. Measurements are based on at least 10 pollen grains (Table 6.1). The size range and the mean values are given for each species. Descriptions of pollen grains are based on observations under LM and SEM. Shape classes are in accordance with ERDTMAN (1952). Pollen morphology terminology follows HIDEUX and FERGUSON (1976), PUNT et al. (1994) and NOWICKE (pers. comm.).

6.3 RESULTS

6.3.1 Pollen morphological description

Brunellia pollen grains are small to medium-sized (P X E = 11.0-32.5 x 10.5-28.0 μm; Table 6.1). Pollen grain shape is prolate, subprolate, oblate spheroidal or occasionally suboblate (P/E = 0.88-1.48). The outline in polar view (amb) is (sub) circular. The grain is 3-colporate, with 8.0-25.0 μm long colpi. The endoapertures are mostly elliptic pores, lalongate or lolongate (2.0-8.0 x 1.0-8.0 μm). The exine is incompletely (Fig. 6.1a, b) to almost completely tectate (e. g. Fig. 6.2).

6.3. 2 Categories of exine ornamentation

Regarding the exine ornamentation in *Brunellia*, two extremes and three intermediate categories were distinguished. One extreme is a reticulate tectum
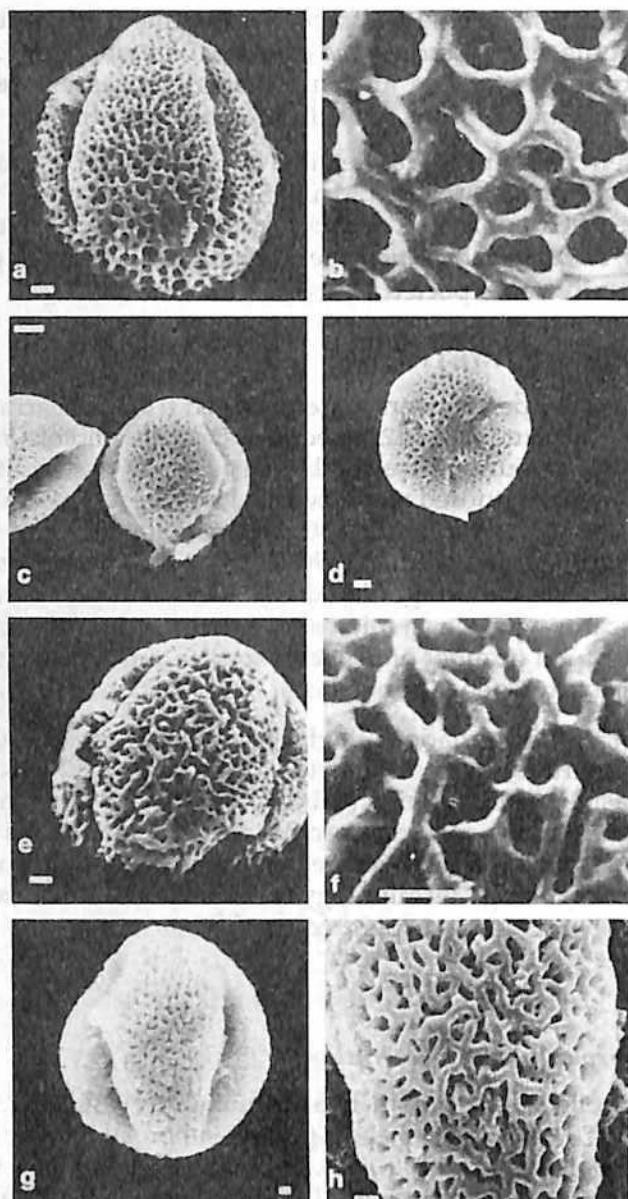
Fig. 6.1 Scanning electron micrographs of *Brunellia* pollen: **(a, b)** *B. comocladifolia* ssp. *comocladifolia*. **(a)** Equatorial view. **(b)** Exine of striate reticulate type. **(c, d)** *B. hygrothermica*. **(c)** Equatorial view, exine finely reticulate type. **(d)** Polar view showing circular amb. **(e, f)** *B. comocladifolia* ssp. *cundinamarcensis* **(e)**. Equatorial view. **(f)** Exine finely reticulate type. **(g,h)** *B. darienensis*. **(g)** Ecuatorial view. **(f)** Finely reticulate type. Scale bars 1µm.

with large lumina and high muri (Fig. 6.1 a, b). Due to the interwoven aspect the term, reticulate striate could be applied to this ornamentation (Nowicke pers. comm.). This type of ornamentation is the most incomplete (most open) and is observed in *B. comocladifolia* subsp. *comocladifolia*. Some species have a punctate tectum (Table 6.1) representing the other extreme, as the tectum is almost complete (Fig. 6.2). The lumina are smaller and circular to slit-shaped. This can be observed in *B. susaconensis* (Fig. 6.2 a, b; OROZCO in prep.), *B. brunnea* (Fig. 6.2 c, d) and *B. costaricensis* (Fig. 6.2 e, f).

Three groups of species show intermediate forms between the two extremes of ornamentation. One intermediate type is finely reticulate (Nowicke pers. comm.; Fig. 6.1 c-h, Table 6.1). The lumina can be recognized and the muri are joined at different levels, such as in *B. comocladifolia* subsp. *cundinamarcensis* (Fig. 6.1 e, f), *B. darienensis* (Fig. 6.1 g, h), *B. hygrothermica* (Fig. 6.1 c, d). Another intermediate group includes species with muri and lumina of variable sizes with both muri and lumina at various levels (Fig. 6.3 a-d). This type of sculpture is denoted as modified reticulate (Nowicke pers.comm.). The columellae are visible as in *B. glabra* and *B. goudotii* (Fig. 6.3 a, b). Columellae are not visible and the muri appear unevenly connected as in *B. propinqua* (Fig. 6.3 c, d; Table 6.1). In a third group of species, the lumina are not distinct, and the muri are expanded, irregular in shape, and are connected at different levels (Figs. 6.3 e, f; 6.4). This sculpture type, modified rugulate (Nowicke pers. comm.), is observed in *B. boliviana*, and in some populations of *B. sibundoya* (Fig. 6.3 f, Table 6.1). A similar ornamentation type is present in *B. inermis* (Fig. 6.4 a, b), *B. cayambensis* (Fig. 6.4 c -e) and *B. weberbaueri* (Fig. 6.4 f).

An overview of the ornamentation types of the exine in *Brunellia* is presented in Figure 6.5.

6.4 DISCUSSION

Pollen morphological characters in *Brunellia* indicate that despite variations of the exine ornamentation none of the grains is echinate or intectate, as was reported by BASAK (1967) and MARTICORENA (1970), respectively. Differences in the exine ornamentation of the pollen grains of *Brunellia* were observed.

SEM results of different populations of *Brunellia antioquensis*, *B. comocladifolia* and *B. sibundoya* (OROZCO 1991) partially covered the variation in exine ornamentation observed in the present study. Two additional groups of variation of the exine to those observed in OROZCO (1991) were seen: the punctate type with small, and circular to slit-shaped lumina (Fig. 6.2) and the modified reticulate type (Fig. 6.3 a-d). The variations shown in OROZCO (1991), regular, irregular reticulate and rugulate perforate type correspond respectively to Nowicke´s terms striate reticulate (Fig. 6.1 a, b), finely reticulate (Fig. 6.1 c, h), and rugulate ornamentation (Figs. 6.3 e, f; 6.4). Variations in the exine
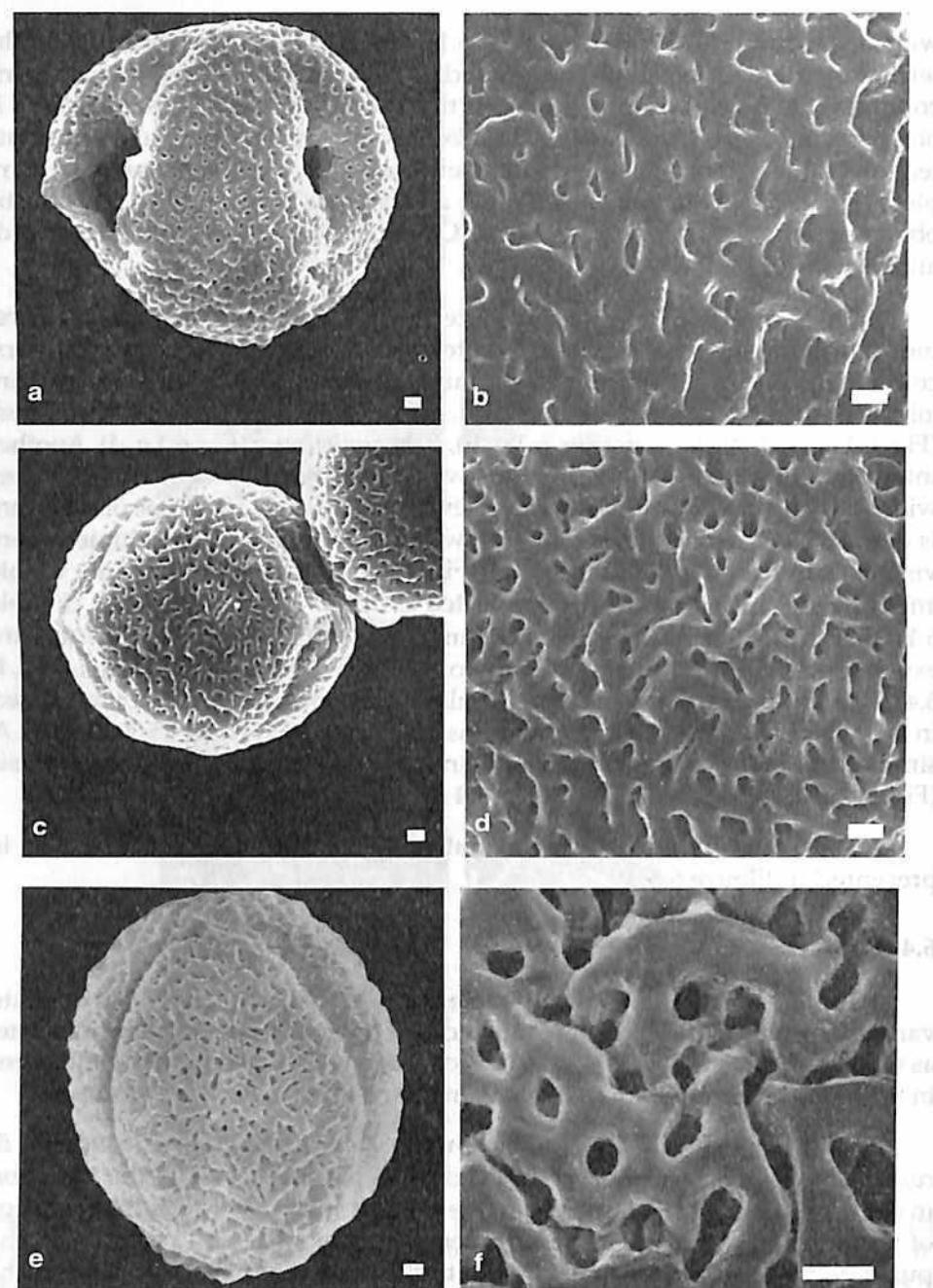
Fig. 6. 2 Scanning electron micrographs of *Brunellia* pollen. **(a, b).** *B. susaconensis* . **(a)** Equatorial view. **(b)** Exine punctate type. **(c, d).** *B. brunnea.* **(c)** Equatorial view. **(d)** Exine punctate type. **(e, f)** *B. costaricensis.* **(e)** Equatorial view. **(f)** Exine punctate type. Scale bars 1µm.

ornamentation were seen in populations of *B. sibundoya* and *B. comocladifolia* (OROZCO 1991). Finely reticulate (Fig. 6.1 c, h) and rugulate ornamentation (Figs. 6.3 e, f; 6.4) were observed in different populations of the widely distributed species of *B. sibundoya*, (OROZCO, 1991, Fig. 10, p. 982 versus Fig. 11, p. 983). In different populations of *B. comocladifolia*, striate reticulate (Fig. 6.1 a, b) and finely reticulate exine (Fig. 6.1 e, f) were observed (OROZCO, 1991, Fig. 8, p. 980 versus Fig. 9, p.981). According to SARMIENTO (1986) the humidity related to the altitude and temperature is an important factor in specific variation in the Colombian Andes. Thus, in the case of intraspecific variation in the ornamentation of the exine and in accordance with the distribution of *Brunellia* in humid montane forests, the humidity might be an influential factor. MULLER (1979) suggests that changes of exine sculpture in the same species are probably responses of the pollen grain to maintain a stable pollen function. Polymorphism in the ornamentation of the exine has been also found in other groups, as in Sapindaceae, *Dimocarpus longan* and species of *Artyera* (VAN DER HAMM 1993, VAN BERGEN ET AL. 1995). They consider that in both cases the intraspecific variation of the ornamentation is due to ecological causes and also related to the intraspecific variation of other characters.

The number of *Brunellia* species studied under LM and SEM allows a characterization of variation in shape and ornamentation of the pollen grains. However, relationships based on transmission electron microscopical data (TEM) are necessary for understanding the SEM results. The preliminary research under TEM analysis, applied to some species of *Brunellia* by NOWICKE (pers. comm.) indicates that the variation observed in SEM is due to variation in tectum thickness, and is not the result of longer columellae or of a thicker foot layer. The columellae in the grains of all taxa examined are uniformly short. Using TEM analysis, also a solid endexinous aperture membrane including a finely lamellate inner sublayer was observed. TEM studies in *Brunellia* are necessary due to the polymorphism found in some species indicating that ornamentation is probably not a very conservative character for some species.

Compared to the pollen grains of *Brunellia*, Cunoniaceae pollen grains show much more variation, but a reticulate exine pattern is usually present (HIDEUX and FERGUSON 1976). Pollen grains are 2-3-colporate and in some species as in *Geissois superba* (Fig. 6.6 d), 2-colpate grains are observed. In the available material of Cunoniacae, 2-colporate grains are present in *Lamanonia tomentosa* (Fig. 6.6 g) whereas, 3-colporate grains occur in *Aistopetalum viticoides*, *Caldcluvia paniculosa* and *Spiraeanthemun katakata* (Fig. 6.6 a, e, h), and 3-colporate grains are also present in *Acsmithia pubescens*. Dicolporate or 2-colpate pollen, presence of sexinous granules in some lumina of *Geissois ternata* (Fig.6.6 f), and the triangular (amb) with apertures in the angles as in *Aistopetalum viticoides*, (Fig. 6.6 h) are characters not present in *Brunellia*.
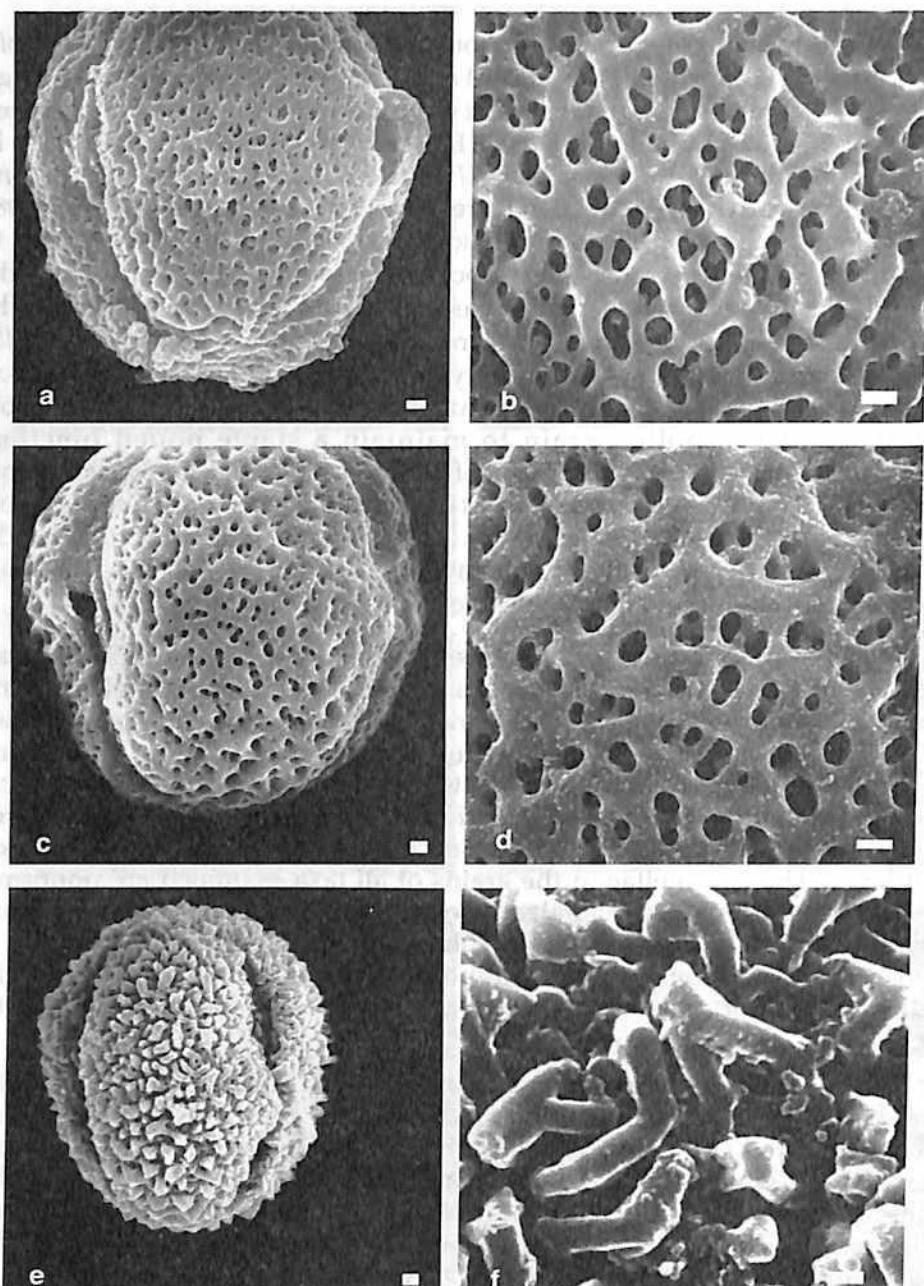
Fig. 6. 3 Scanning electron micrographs of *Brunellia* pollen. (a) *B. goudotii*, equatorial view (b) *B. glabra*, exine modified reticulate type. (c, d) *B. propinqua*. (c) Equatorial view. (d) Exine modified reticulate type. (e) *B. boliviana* equatorial view. (f) *B. sibundoya* ssp. *sibundoya*, exine rugulate type. Scale bars 1µm.

NOWICKE (pers. comm.), with basis on a study of 12 species, considers that the pollen of Cunoniaceae is fundamentally similar in sculpture. However, the presence of 3-colporate grains in *Aistopetalum, Caldcluvia, Spiraeantemum,* and *Acsmithia* and the generalized 2-colporate grains present in the remaining genera (except *Pullea glabra*) might be related to the lack of monophyly in the family as was mentioned by HICKEY and TAYLOR (1991) and also by OROZCO (1997). On the contrary, such variation might indicate polymorphism as could occur in other characters. The genera *Aistopetalum, Spiraeanthemum* and *Acsmithia* closely related to *Brunellia,* (HUFFORD and DICKISON 1992) present 3-colporate grains. Further research is necessary to establish whether the pollen morphology could be used as an informative character in relationships within Cunoniaceae. BRADFORD (pers. comm.) considered Cunoniaceae as a monophyletic group, based on molecular data.

Displaying the two states of pollen ornamentation used in the phylogenetic study of *Brunellia* (Chapter 7) in the five categories, and superposing them on the cladogram it can be seen that the punctate exine with circular to slit-like lumina (almost complete tectum) could be a plesiomorphic condition. This state is present in some species of *Brunellia* and *Spiraeanthemun,* with which *Brunellia* shares other characters (DICKISON 1971, 1980; EHRENDORFER ET AL. 1984, CUATRECASAS 1985). The other exine categories, incomplete tectum (striate reticulate, modified reticulate and modified rugulate) are derived conditions. The same could hold in Cunoniaceae and Connaraceae. A complex exine sculpture, such as a rugulate ornamentation, present in Cunoniaceae (*Vesselowskya* and *Pullea;* HIDEUX and FERGUSON 1976) and also in Connaraceae (DICKISON 1980), is considered a specialized character. In *Brunellia* the modified reticulate exine could be considered a synapomorphy of the Subsection *Simplicifoliae* (Chapter 7), in contrast to the homoplasic condition of the remaining categories of exine ornamentation in *Brunellia.* This modified reticulate condition is very often related to foliar and inflorescence reduction and the presence of a navicular shaped endocarp (Chapter 7).

6.5 ACKNOWLEDGMENTS

Fig. 6. 4 Scanning electron micrographs of *Brunellia* pollen: **(a, b)** *B. inermis.* **(a)** Equatorial view. **(b)** Exine rugulate type. **(c, d, e)** *B. cayambensis* **(c)** Equatorial view. **(d)** Exine rugulate type. **(e)** Equatorial view, showing the endoaperture. **(f)** *B. weberbaurei,* exine rugulate type. Scale bars 1μm.
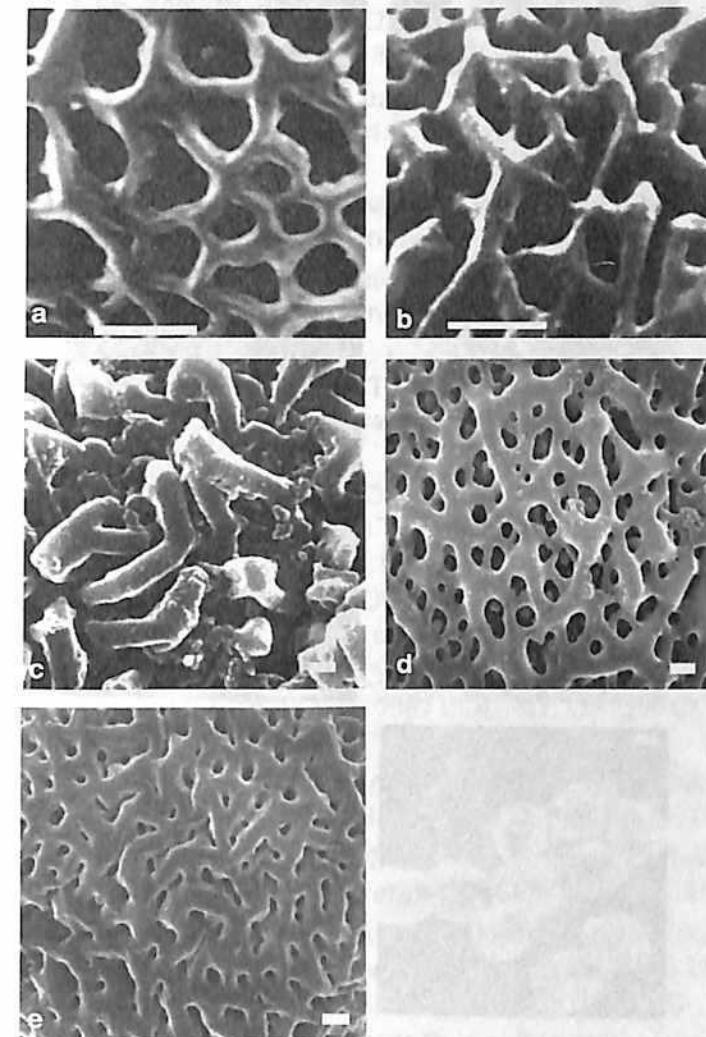
Fig. 6. 5 Summary of the exine ornamentation in *Brunellia.* **(a)** *B. comocladifolia* ssp. *comocladifolia* striate reticulate exine. **(b)** *B. comocladifolia* ssp. *cundinamarcensis,* finely reticulate exine. **(c)** *B. cayambensis,* rugulate exine. **(d)** *B. glabra,* modified reticulate exine. **(e)** *B. brunnea,* punctate exine. Scale bars 1 μm.
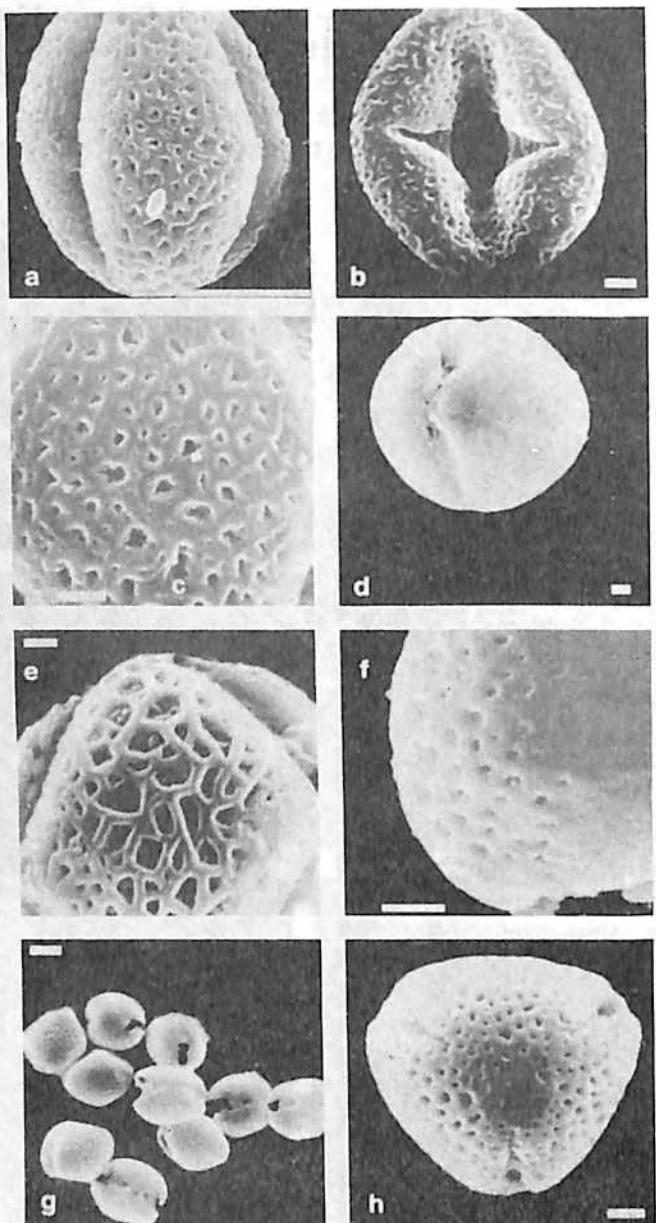
Fig. 6. 6 Scanning electron micrographs of *Cunoniaceae* pollen. (a, b, c). *Spiraeanthemum katakata.* (a) Equatorial view. (b) Endoaperture. (c) Exine punctate type. (d) *Geissois superba*, 2-colporate pollen. (e) *Caldcluvia paniculosa*, 3-colporate pollen. (f) *Geissois ternata*, granule in the exine punctate type. (g) *Lamanonia tomentosa*, 2-colporate pollen. (h) *Aistopetalum viticoides*, angulaperturate pollen. Scale bars 1 μm.

SPECIMENS INVESTIGATED

B. boliviana (Rusby) Cuatrec. BOLIVIA: La Paz, Nor-Yungas, La Paz 72 km toward Coroico, 28.8.1979, Beck 1832, US 2232 COL, US (LM, SM).

B. brunnea Macbride. PERU: Cuzco, Prov. Paucartambo, between Tambo of Tres Cruces and Tambomayo, 14. 5 Weberbauer 6969 US 2301 COL, US (LM, SM).

B. cayambensis Cuatrec. COLOMBIA: Cauca, Mnpo. del Timbio, group of houses Las Cruces, 23.9.1983, Orozco et al. 1187 COL 2330 COL, US (LM,SM).

B. comocladifolia H. B. K. ssp. *comocladifolia.* COLOMBIA: Valle del Cauca, Mnpo. del Dagua, road toward Pichendé, 22. 9. 1983, Orozco et al. 1158 COL, (LM,SM).

B. costaricensis Standley. COSTA RICA: San José, 3 km south of El Empaine on the Pan-American Highway, 11.7.1982, Todzia 1909 NY 2298 COL, US (LM,SM).

B. darienensis Cuatrec. & Porter. COLOMBIA: Antioquia, Mnpo. Frontino, road from Nutibara to La Blanquita, 17.2.1985 Henderson & Bernal 144 COL, NY 2300 COL, US (LM, SM).

B. dulcis Macbride. COLOMBIA: Chocó, Mnpo. San José del Palmar, toward Gálapago, 11.11.1985, Lozano et al. 4890 COL 2289 COL, US (LM,SM).

B. glabra Cuatrec. COLOMBIA: Cauca, Parque Nacional Munchique, kms. 42-47 NE of Uribe, 24.5.1979, Luteyn 7387 COL, NY, US 2290 COL, US (LM, SM).

B. goudotii Tulasne. COLOMBIA: Valle del Cauca, road between Tulua and Barragán, 20. 9.1983, Orozco et al. 1129 COL 2291 COL, US (LM, SM).

B. hygrothermica Cuatrec. COLOMBIA: Chocó, Mnpo. Quibdó, 2 km from S of Zuto, Pan-American road, Bernal et al. 1105 COL 2235 COL, US (LM, SM).

B. inermis Cuatrec. ECUADOR: Loja, Parque Nacional Podocarpus, along the road from Yangana to radio towers on Cerro Toledo, 29. 4. 1987, van der Werff and Palacios 9196 COL, US, MO 2331 COL, US (LM, SM).

B. latifolia Cuatrec. COLOMBIA: Valle, Los Farallones eastern slopes at Almorzadero, 30. 7.1944, Cuatrecasas 18109 COL, US 2297 COL, US (LM, SM).

B. propinqua H. B. K. COLOMBIA: Cundinamarca, Sibate, Vereda San Miguel, Cuchuco hill, 7. 9.1987, Orozco & Linares 1768 COL 2295 COL, US (LM, SM).

B. propinqua H. B. K. COLOMBIA: Cundinamarca, Choachí, Hoya del Río Blanco, Quebrada de Agua Dulce, in front of El Rajadero, 8. 2. 1969, Cuatrecasas and Llano 27329 COL, NY, US 2294 COL, US (LM,SM).

B. putumayensis Cuatrec. COLOMBIA: Nariño, south of Laguna de la Cocha, near Páramo de Santa Lucia, the source of Río Alisales, 2. 2. 1941, Cuatrecasas 11883 COL, F, US 2288 COL, US (LM, SM).

B. racemifera Tulasne. COLOMBIA: Cundinamarca, Cordillera Oriental, Mnpo. of San Bernardo, road between Paquiló and Santa Marta, 10. 7.1986, Jaramillo 7035 COL 2293 COL, US (LM, SM).

B. rhoides Rusby. BOLIVIA: Songo, 11. 1890, Bang 839 MO 2329 COL, US (LM, SM).

B. sibundoya Cuatrec. ssp. *antioquensis*. COLOMBIA: Antioquia, Medellín, Cerro del Padre Amaya, 1.30.1984, Orozco & Tobón 1320 COL (LM, SM).

B. sibundoya Cuatrec. ssp. *sibundoya*. COLOMBIA: Cundinamarca, Bojacá road, towards La Merced, 21. 4. 1983, Orozco & Lozano 1092 COL (LM, SM).

B. stuebelii Cuatrec. COLOMBIA: Cauca, El Tambo, Munchique, 24. 9.1983, Orozco et al. 1176 COL 2234 COL,US (LM, SM).

B. subsesillis Killip & Cuatrec. COLOMBIA: Medellín, road to San Cristobal, Cerro del Padre Amaya, 1. 11. 1985, Orozco et al. 1425 COL 2302 COL, US (LM,SM).

B. susaconensis (Cuatrec.) Orozco. COLOMBIA: Boyacá, Susacón, Santa Rosita to Onzaga road, 10. 1987, Orozco et al. 1810 COL 2296 COL, US (LM,SM).

B. tomentosa H. & B. ECUADOR: Pichincha, below San Juán towards Chiriboga, 2. 8. 1955, Asplund 17162 NY 2231 COL, US (LM, SM).

B. trianae Cuatrec. COLOMBIA: Antioquia, Medellín, Padre Amaya hill, 1.11.1985, Lozano 3969 COL 2230 COL, US (LM, SM).

B. trigyna Cuatrec. COLOMBIA: Norte de Santander, Parque Tama-Orocue, 27.3.1987, Lozano et al. 5535 COL 2233 COL, US (LM, SM).

B. weberbaueri Loesener. PERÚ: Huamalíes, Monzón, 8.3. 1903, Weberbauer 3551 NY, F 2287 COL, US (LM, SM).

Spiraeanthemum katakata Seem. FIJIE: A.C. Smith 1967 US 2232 COL, US (LM, SM).

6.6 LITERATURE

APG. 1998. An ordinal classification for the families of flowering plants. Ann. Missouri Bot. Gard. 85: 531-553.

BASAK, R. K. 1967. Studies on the pollen morphology of Simaroubaceae. Bull. Bot. Surv. India 9: 63-67.

BRADFORD, J. and BARNES, R. 2001. Phylogenetics and classification of Cunoniaceae (Oxalidales) using chloroplast DNA sequences and morphology. Syst. Bot. 26: 354-385.

CUATRECASAS, J. 1970. Brunelliaceae. Flora Neotropica Monograph 2: 1-189

CUATRECASAS, J. 1985. Brunelliaceae. Flora Neotropica Monograph 2 (suppl.): 29-103.

CRONQUIST, A. 1981. An integrated system of classification of flowering plants. Columbia Univ. Press, New York.

DAHLGREN, R. M. 1980. A revised system of classification of the angiosperms. Bot. J.Linn. Soc. 80: 91-124.

DICKISON, W. C. 1971. Anatomical studies in the Connaraceae. I. Carpels. J. Elisha Mitchell Sci.Soc. 87: 77-86.

DICKISON, W. C. 1975. Studies on floral anatomy of the Cunoniaceae. Amer. J. Bot. 62: 433-477.

DICKISON, W. C. 1979. A survey of pollen morphology of the Connaraceae. Pollen & Spores 21: 31-79.

DICKISON, W. C. 1980. Comparative wood anatomy and evolution of the Cunoniaceae. Allertonia 2: 281-321.

DICKISON, W. C. 1989. Comparisons of primitive Rosidae and Hamamelidae. In: Evolution, systematics, and fossil history of the Hamamellidae. Vol. 1. Introduction and "Lower" Hamamelidae (eds. P. CRANE & S. BLACKMore), pp. 47-73. Clarendon Press, Oxford.

ERDTMAN, G. 1952. Pollen morphology and plant taxonomy, Angiosperms. Almqvist & Wiksell, Stockholm.

ERDTMAN, G. 196-Pollen morphology and plant taxonomy. Angiosperms. Hafner Publ.Co., New York.

EHRENDORFER, F, MORAWETZ, W., and DAWE, J. 1984. The Neotropical angiosperm families Brunelliaceae and Caryocaraceae: First karyosystematical data and affinities. Pl. Syst. Evol. 145: 183-191.

GRAHAM, A. and JARZEN, D. M. 1969. Studies in neotropical paleobotany. I. The oligocene communities of Puerto Rico. Ann. Missouri Bot. Gard. 56: 308-357.

HICKEY, L. J. and TAYLOR, D. W. 1991. The leaf architecture of *Ticodendron* and the application of foliar characters in discerning its relationships. Ann. Missouri Bot. Gard. 78: 105-130.

HIDEUX, M. J. and FERGUSON, I. K. 1976. The stereostructure of the exine and its evolutionary significance in Saxifragaceae sensu lato. In: The evolu-

tionary significance of the exine (eds. I. K. FERGUSON and J. MULLER), pp. 327-377. Linn. Soc. Symp. Ser. 1. Academic. Press, London.

HUFFORD, L. and DICKISON, W. C. 1992. A phylogenetic analysis of Cunoniaceae. Syst. Bot. 17: 181-200.

MARTICORENA, C. 1970. Pollen morphology. In (ed. J. CUATRECASAS) Brunelliaceae. Flora Neotropica Monograph 2: 27-32.

MULLER, J. 1979. Form and function in angiosperm pollen. Ann. Missouri Bot. Gard. 66: 593-632.

OROZCO, C. I. 1991. Análisis multivariado de un complejo de especies de *Brunellia*. Ann. Missouri Bot. Gard. 78: 126-136

OROZCO, C. 1997. Sobre la posicion sistemática de *Brunellia* Ruiz & Pavón. Caldasia 19: 145-164.

PUNT, W., BLACKMORE, S., NILSSON S., and LE THOMAS, A. 1994. Glossary of pollen and spore terminology. LPP Foundation, Utrecht.

SALOMONS, J. B. 1986. Paleoecology of volcanic soils in the Colombian Central Cordillera. (Parque Nacional Natural de los Nevados). In: The Quaternary of Colombia, Vol. 13 (ed. T. VAN DER HAMMEN), pp. 17-212, Amsterdam.

SARMIENTO, G. 1986. Ecological features of climate in high tropical mountains. In: High altitude tropical biogeography (eds. F. VULLEUMIER and M. MONASTERIO), pp. 11-45. New York, Oxford.

TAKHTAJAN, A. L. 1981. Outline of the classification of flowering plants (Magnoliophyta). Bot. Rev. 46: 225-359.

THORNE, R. F. 1983. Proposed realignments in the angiosperms In: 13th Int. Bot. Congr. Sydney 1981. New evidence of relationships and modern systems of classifications of the angiosperms. Symp. Proc. (eds. F. EHRENDORFER and R. DAHLGREN). Nord. J. Bot. 3: 85-117.

VAN BERGEN, M. A., VAN DER HAM, R. and TURNER, H. 1995. Morphology and evolution of *Arytera* pollen (Sapindaceae-Cupanieae). Blumea 40: 195-209.

VAN DER HAM. 1993. Polymorphisme dans L'ornamentation du pollen de *Dimocarpus logan* (Sapindaceae): aspects morphologiques et geographiques. Palynosciences 2: 239-254.

WALKER, J. W. and DOYLE, J. A. 1975. The basis of angiosperm phylogeny: Palynology. Ann. Missouri Bot. Gard. 62: 664-723.

Chapter 7

A CLADISTIC ANALYSIS OF *BRUNELLIA* RUIZ & PAVÓN BASED ON MORPHOLOGICAL CHARACTERS[1]

CLARA INÉS OROZCO

ABSTRACT

Brunellia is a neotropical tree genus widely distributed in the Andes. Species of this genus are also found in Central America and in the Caribbean Antilles. Fifty four species are currently recognized. Most of the species are found in the high elevation and sub-Andean forest of Colombia. A phylogenetic study was performed to determine the relationships among the species, to evaluate the infrageneric clasification, to identify characters defining natural groups of species. Forty-four characters from conventional morphology and the exine sculpture of the pollen grain were coded for 31 *Brunellia* species and one outgroup taxon, *Spiraeanthemum* (Cunoniaceae) represents four species of Cunoniaceae. PAUP and NONA found the same three most parsimonious cladograms, of which one is recommended as the current working hypothesis for *Brunellia* relationships. The preferred tree was also found by HENNIG86, Pee-Wee (k=6), and majority rule consensus of trees obtained with Pee-Wee (K= 3, k=1). *Brunellia* is monophyletic, and the infrageneric clasification is discussed.

RESUMEN

Brunellia es un género, arbóreo, neotropical ampliamente distribuido en los Andes. Las especies del género se encuentran también en Centro América y

---

1   To be submitted to Cladistics.

las Islas del Caribe. Actualmente se reconocen 54 especies. La mayor parte de las especies se encuentran en los bosques Andinos y sub-Andinos de Colombia. Se practicó un estudio filogenético para determinar las relaciones de parentesco de las especies, como también para evaluar la división infragenérica y para identificar qué caracteres definen grupos naturales de especies. Cuarenta y cuatro caracteres de morfología convencional, anatomía de la hoja, nodal, inflorescencias, morfología floral, ornamentación de la exina del grano de polen, fueron codificados para 32 taxones de *Brunellia* y un grupo externo *Spiraeanthemum* (Cunoniaceae) el cual representa cuatro especies de la familia Cunoniaceae. PAUP y NONA encontró los mismos tres más parsimoniosos cladogramas, de los cuales uno es recomendado como la hipótesis de trabajo para las relaciones filogenéticas de *Brunellia*. La hipótesis recomendada fue también encontrada con HENNIG86, Pee-Wee(*K*=6), y el árbol de consenso de mayoría obtenido con Pee-Wee (*K*=1,*K*=3). *Brunellia* es monofilético. Se discute la clasificación infra-genérica.

7.1 INTRODUCTION

The genus *Brunellia* Ruíz & Pavón is presently placed in the monotypic family Brunelliaceae. Most species are found in the Andean Cordillera, some with a wide distribution in Colombia; a few species are present in Central America and the Greater Antilles. The species are an important element of humid, cloud forest ecosystems.

Engler (1897) regarded *Brunellia* as distinct from Cunoniaceae mainly because of the epitropic position of its ovules. Recent research (HUFFORD and DICKISON 1992, OROZCO 1997) have focused on whether *Brunellia* is better retained as a separate family or placed in the Cunoniaceae. Close relationships among *Brunellia, Spiraeanthemum,* and *Acsmithia* (Cunoniaceae) were early noted by DICKISON (1975). The same relationships were also observed in the phylogenetic study of Cunoniaceae by HUFFORD and DICKISON (1992) and BRADFFORD and BARNES (2001). According to HICKEY and TAYLOR (1992) and OROZCO (1997) Cunoniaceae is not monophyletic. *Brunellia, Spiraeanthemum* and *Acsmithia* share characters no present in the rest of genera of Cunoniaceae, and it is not clear that the remaining Cunoniaceae are monophyletic (OROZCO 1997). However, according to the latest research on molecular data (BRADFORD and BARNES 2001) Brunelliaceae " must be retained as a family, which shows close relationships with *Cephalotus* and to Cunoniaceae, but Brunelliaceae do not probably fall in the Cunoniaceae clade". Currently Brunelliaceae, and Cunoniaceae are placed in the order Oxalidales (APG, 1998).

All species of *Brunellia* are trees with opposite or whorled stipulate leaves; stipels are also obviously present, on the rachis of the compound leaves, and sometimes are easily observed on the petiole of the unifoliate leaves. The genus is also easily distinguished by the presence of leaf vestiture on the abaxial side,

of the leaves, the absence of petals, apocarpic ovaries, fruits with frequent presence of prickly hairs.

CUATRECASAS (1970, 1985) divided *Brunellia* in two larger sections and thirteen subsections. Subsequently four additional species were described giving a total of 64 species (CUATRECASAS 1985, PORTER and CUATRECASAS 1975, OROZCO 1986, 1991). CUATRECASAS changed his concept of subsections in 1985 and moved many species to other subsection (Table 7.1). He also presented a phylogeny of the subsections in which *B. boliviana* and *B. cuzcoensis* were considered as ancestral species within the genus. Of the 64 species finally recognized by CUATRECASAS, 10 are doubtfully distinct; 54 species are recognized here (Orozco in prep.).

A phylogenetic Hennigian hypothesis for the genus does not exist. The grounds of the phylogenetic study is the searching of natural groups (monophyletic groups). Six years ago I presented a tentative hypothesis (OROZCO unpublished) based only on macro-morphological characters and in which many outgroup states were unknown. The present analysis uses more character systems and more complete outgroup coding to estimate the relationships among the species, to evaluate the infrageneric classification, to determine synapomorphies for natural species groups, and also to contribute to the discussion about the relationships of the basal groups of Rosidae.

7.2 MATERIALS AND METHODS

7.2. 1 Taxa selection

The 31 terminal taxa included here (Tables 7.2, 7.3) represent twelve subsections considered by CUATRECASAS (1985). Taxa were selected to broadly represent *Brunellia,* but were constrained by material available for study. Subsection *latifoliae* (*B. latifolia*) could not be studied adequately because it is monotypic and known only from the type specimen. *B. foreroi* and *B. latifolia* present some features not present in other species of *Brunellia,* such as the reduction to an unifoliate leaf, two pairs of evident stipels on the petiole (Chapter 3), a U-shaped endocarp, and highly developed inflorescence (OROZCO and WEBERLING 1999). *B. latifolia* is represented here by *B. foreroi.*

For an outgroup I synthesized character data from four species, *Spiraeanthemum katakata, Acsmithia brongniartiana, A. pedunculata* and *A. reticulata* (HOOGLAND 1979). *Acsmithia* is a segregate genus of *Spiraeanthemum.* The selection of the outgroup was based prior cladisitic analysis (HUFFORD and DICKISON 1992, OROZCO 1997). The outgroup selection of this study was limited by the type of characters studied for *Brunellia.* Not all the possible outgroups have been studied in depth with respect to the type of characters used in this research. However, the information for the outgroup was displayed and the matrix was also run with these data (Tables 7.4, 7.5).

Table 7. 1 Infrageneric classification of *Brunellia*.

	SECTIONS (Cuatrecasas 1970)		SECTIONS (Cuatrecasas 1985)				PROPOSED INFRAGENERIC DIVISION*				
SUBSECTIONS	Brunellia	Simplicifolia	SUBSECTIONS	GROUPS	Brunellia	Simplicifolia	Brunellia	Simplicifolia	Ovalifolia	Pauciflorae	Stuebelia
Apopetalae	B. pinnata										
Bullatae	B. cuzcoensis, B. bullata, B. cayambensis						B. hygrothermica, B. latifolia, B. sibundoya, B. foreroi, B. velutina	B. cayambensis, B. dichapetaloides, B. ecuadorensis, B. rhoides	B. ovalifolia	B. pauciflora	B. stuebelii, B. briquetii
Bolivianae	B. boliviana, B. hexasepala, B. racemifera, B. weberbaueri, B. ecuadorensis, B. inermis, B. integrifolia		Propinquae		B. bullata, B. cayambensis, B. boliviana, B. hexasepala, B. cuzcoensis, B. weberbaueri, B. ecuadorensis		B. brunnea, B. mexicana, B. stenoptera, B. acostae, B. costaricensis, B. standleyana	B. colombiana, B. propinqua, B. susaconensis, B. trigyna, B. zamorensis			
Brunelliae	B. colombiana, B. elliptica, B. littlei, B. propinqua, B. trigyna		Comocladifoliae		B. inermis, B. integrifolia, B. carpishensis, B. colombiana, B. elliptica, B. littlei, B. propinqua, B. racemifera, B. trigyna		B. boliviana, B. cuzcoensis, B. hexasepala	B. comocladifolia			
Colombianae	B. comocladifolia, B. coroicana, B. costaricensis, B. hygrothermica, B. sibundoya			Acostae	B. antioquensis, B. comocladifolia, B. coroicana, B. gentryi, B. hygrothermica, B. sibundoya	B. foreroi	B. cutervensis, B. oliveri	B. weberbaueri			
Comocladifoliae	B. acostae, B. brunnea, B. dichapetaloides, B. mexicana, B. occidentalis, B. oliveri, B. rhoides, B. standleyana, B. stenoptera, B. subsessilis, B. ovalifolia		Brunellia	Bolivianea	B. acostae, B. brunnea	B. cutervensis	B. amayensis, B. elliptica, B. inermis, B. integrifolia, B. littlei, B. occidentalis, B. racemifera, B. subsessilis				
Forerae		B. forereo		Cuterva	B. dichapetaloides, B. mexicana, B. occidentalis, B. oliveri, B. rhoides, B. stenoptera, B. subsessilis, B. ovalifolia						
Occidentales	B. acutangula, B. boqueronensis, B. farallonensis, B. glabra, B. goudotii, B. ovalifolia, B. pallida, B. pendeniscana, B. putumayensis, B. tomentosa, B. latifolia		Simplicifoliae	Congestiflora		B. cutervensis	B. acutangula, B. farallonensis, B. glabra, B. goudotii, B. tomentosa, B. macrophylla, B. pallida, B. pendeniscana, B. pitayensis, B. putumayensis, B. rufa, B. trianae				
Ovalifoliae						B. acutangula, B. boqueronensis, B. farallonensis, B. glabra, B. goudotii, B. ovalifolia, B. tomentosa, B. latifolia					
Simplicifoliae	B. almaparensis, B. briquetii, B. dulcis, B. macrophylla, B. ovalifolia, B. pallida, B. pendeniscana, B. putumayensis, B. stuebelii					B. briquetii, B. stuebelii					
Stipellatae / Stuebeliae			Trianae			B. almaparensis, B. danenensis, B. cuatrecasana, B. macrophylla, B. ovalifolia, B. pallida, B. pendeniscana, B. pitayensis, B. putumayensis, B. spinalii					
Trianae		B. velutina	Velutinae		B. costaricensis, B. danenensis, B. diversifolia, B. hillyana, B. morii, B. standleyana, B. velutina						
Velutinae											

* New arrangements in preparation.

Table 7.2 Terminal taxa representing Sections and Subsections of Cuatrecasas 1985.

SUBSECTIONS	TERMINAL TAXA		Represented species*
	SECTIONS		
	Brunellia	Simplicifolia	
Bullatae	B. cayambensis		
Bolivianae	B. boliviana		B. cuzcoensis, B. hexasepala, B. weberbaueri
Brunelliae	B. ecuadorensis, B. inermis, B. integrifolia		B. amayensis
Colombianae	B. elliptica, B. propinqua, B. racemifera		B. colombiana, B. littlei, B. susaconensis, B. trigyna, B. zamorensis
Comocladifoliae	B. hygrothermica, B. sibundoya		B. comocladifolia
Foreroae		B. foreroi	B. latifolia
Occidentales	B. acostae		B. costaricensis, B. morii, B. standleyana
	B. brunnea, B. cutervensis, B. rhoides, B. stenoptera, B. subsessilis		B. oliveri, B. dichapetaloides, B. mexicana, B. occidentalis
Ovalifoliae	B. ovalifolia		
Simplicifoliae		B. acutangula, B. boqueronensis, B. farallonensis, B. glabra, B. tomentosa	B. goudotii
Stuebeliae	B. stuebelii	B. briquetii	
Trianae	B. dulcis, B.macrophylla, B. putumayensis, B. pitayensis, B. rufa		B. pendeniscana, B. trianae, B. pallida
Velutinae	B. velutina		

*Species represented by terminal taxa
B. pauciflora Orozco & Cuatrec. and B. amayensis Orozco were described after Cuatrecasas, 1985.

CUATRECASAS's species considered synonymous (1985) are not listed hear.

Table 7.3 Terminal taxa * representing taxa of the Sections and Subsections of Cuatrecasas 1985. Synonymous species (Orozco in prep.) represented by •.

Section I. Brunellia	**Subsection Velutinae**
Subsection Bullatae	B. costaricensis
B. bullata •	B. danenensis •
B. cayambensis *	B. diversifolia •
	B. hiltyana •
Subsection Bolivianae	B. moni
B. boliviana *	B. standleyana
B. hexasepala	B. velutina *
B. cuzcoensis	
B. weberbaueri	**Section II. Simplicifolia**
Subsection Brunelliae	**Subsection Foreroae**
B. carpishensis •	B. foreroi *
B. ecuadorensis *	
B. inermis *	**Subsection Stipellatae**
B. integrifolia *	B. latifolia
	Subsection Simplicifoliae
Subsection Colombianae	B. acutangula *
B. colombiana •	B. boqueronensis *
B. elliptica *	B. farallonensis
B. propinqua *	B. glabra *
B. racemifera *	B. goudotii
B. susaconensis	B. ovalifolia *
B. trigyna	B. spinalii •
B. zamorensis	B. tomentosa *
Subsection Comocladifoliae	**Subsection Stipellatae**
B. antioquensis	B. briquetii
B. comocladifolia •	B. stuebelii *
B. coroicana •	
B. gentryi •	**Subsection Trianae**
B. hygrothermica *	B. almagarensis •
B. sibundoya *	B. cuatrecasana •
	B. dulcis *
Subsection Occidentales	B. macrophylla *
B. acostae *	B. pallida
B. brunnea *	B. penderiscana
B. cutervensis *	B. pitayensis *
B. dichapetaloides	B. putumayensis *
B. mexicana	B. rufa *
B. occidentalis	B. trianae
B. oliveri	
B. rhoides *	
B. stenoptera *	
B. subsessilis *	
Subsection Ovalifoliae	
B. ovalifolia *	

Table 7.4. Character matrix of *Brunellia* using *Spiraeanthemum* as outgroup.

Table 7.5. Character matrix of *Brunellia* displaying the information of the synthetic outgroup of Table 7.4.

7.2. 2 Characters

The character matrix (Appendix 7.1, Tables 7.4, 7.5) includes 44 characters based on my own observations. Six characters are synapomorphies for *Brunellia* (characters 2, 8, 22, 24, 25 and 40). The character 31 is a clear autapomorphy in the context of this matrix, and for this reason it was treated as inactive.

Twentynine characters pertain to anatomy (10-24), morphology of inflorescences (27-34), floral morphology (35-37, 41), and pollen morphology (38 and 39); all were unavailable during the earlier phylogenetic study of *Brunellia* (OROZCO unpublished). Remaining characters concern vegetative characters of leaves (1, 3-7, 9, 25, 26), the stigma (40), and fruit morphology (42-44). Characters were scored from specimens deposited in AU, COL, F, S, MO, NY, and US.

Exine ornamentation of twelve species could not be examined. This character (39) was codified as missing data (?) (Table 7.4). Anatomy of leaves and petioles for all species were observed with exception of *B. brunnea* because a single specimen was available. For characters (10-24) this species was assumed to show the same character states as *B. cutervensis* based in other character systems.

Two characters were excluded from the analysis: the presence and number of stipels on the petiole. Most compound leaves species with a very few exceptions (e.g. *B. ecuadorensis*) with compound leaves have stipels on the petiole. Even in the unifoliate species (except *B. foreroi* and *B. latifolia*), where stipels are not evident, anatomical results indicate their presence in many species (absent in *B ecuadorensis*, *B. ovalifolia* and *B. pauciflora*, OROZCO and COBA, in prep.). The absence of reliable data for all species prevents confident use of this character in the cladistic analysis, especially given that stipels are suspected in *B. ecuadorensis*.

The matrix includes 10 multistate characters (6, 7, 9, 17, 23, 27, 37, 38, 41 and 44). All the characters were treated as unordered because no priori transformation series were obvious.

7.2. 3 Cladistic analysis

A search for most parsimonious cladograms (MPT´s) was performed with Hennig86 (FARRIS, 1988), PAUP version 3.1.1 (SWOFFORD 1990) and NONA version 1.15 (GOLOBOFF 1993a). The study of character evolution was done in MacClade, version 3.0 (MADDISON and MADDISON 1992) and Clados version 1.2 (NIXON 1992). NONA default (amb-) ignores "potential support" and collapses a branch if " there exists any optimization at all under which it has zero length" (CODDINGTON and SCHARFF 1994). The "amb =" option reports trees with nodes having only ambiguous support. To search among tree islands I used the "mult+ N" option in NONA and random addition sequences in PAU. PAUP is slower

Table 7.6 Length, Maximum Fit and cost (%) of the obtained trees with different concavity values.

Concavity (K)	Maximum Fit	Length	Cost	Number of trees with maximum fit
1	126.0	209	21%	7
2	166.3	208	25%	2
3	194.8	201	28%	84
4	216.1	198	31%	30
5	232.4	190	33%	2
6	246.3	187	36%	1

than NONA, but it collapses branches using the same criteria as HENNIG86 (CODDIGNTON and SCHARFF 1994). The equally weighted data were run several times using various search strategies with these programs. Pee-Wee version 2.15 was also used (GOLOBOFF 1993b), it implements a concept of maximum fit, that supposedly maximizes "explanotory power". The fit for each character is measured as a concave function of homoplasy, originally proposed by FARRIS (1969). The final weight of a character (the weight that the character "deserves"), is that "implied" by the optimal tree topology, itself determined by the data as a whole, given their implied weights. Highly homoplasious characters receive less weight and non homoplasious characters get a relatively high weight, thus a concave weight function is created The curvature of the concave homoplasy function depends on the parameter k, which can vary from 1 to 6. Lower k values assign relatively less weight to homoplasious characters than higher k values (GOLOBOFF 1993c). It is not clear which value of k should be used under what circumstances. TURNER and ZANDEE (1995) critized implied weights. Each phylogenetic analysis package analyzes the data in a different way, which helps to gain a more complete understanding of one´s results.

The mh+; and bb+ options were used in Hennig86. In PAUP, the heuristic option was used with stepwise addition and tree bisection reconnection (TBR) for 30 random addition sequences. In NONA the options search; max+ (TBR) and 10 random addition sequence (mult+ 10), were used at the beginning. Later different random number seeds, different values of mult+ and both support (amb-and amb =) were also used and combined with best, and max+ options. For Pee-Wee the same options as in NONA were used for all k values. Strict and majority rule consensus trees were obtained with PAUP.

To determine character step differences among the trees of NONA, commands ref, and cmp+ were used (GOLOBOFF 1993a). Successive weighting was also used with NONA using run swt. run mu+10 max+ (CODDINGTON pers. comm.). Successive weighting was also used with PAUP, using the rescaled consistency indices (FARRIS 1989) with base weight 1000.

7.3 RESULTS

NONA and PAUP both found three trees of 183 steps (C.I. = 0.32; R.I. = 0.58). Henning86 found only one most parsimonious (MPT), identical to one of the three found previously. Displaying the information of the outgroup in the four species (Table 7.6) and using NONA were obtained five trees of 193 steps (C.I. = 0.30; R.I. = 0.59); three of them are identically to the three trees found with the synthetic outgroup (Table 7.4) The two additional trees have same information of the relationships, however, the order of the branches change in some parts of the topology. Successive weighting with NONA produced the same three trees, which stabilized after two times weighting cycles. With $k=1$ (with a cost of 21% of homoplasy), I obtained seven trees 24 steps longer than minimal (Fig. 7.5 a, Table 7.5); with $k= 6$ (36% of homoplasy), one tree was obtained of length 184 which is three steps longer than minimal. An intermediate K value of 3 and a homoplasy cost of 28% yielded 84 trees, 14 steps longer than the minimal (Fig. 7.5 b). The same 3 topologies were found when the pollen ornamentation was assumed for some species (Table 7.5). However the trees were two steps longer.

Majority rule and strict consensus trees for the three minimal trees appear in Fig. 7.2. The greatest differences among tree topologies were two groups which changed position: the group *B. hygrothermica, B. sibundoya, B. velutina, B. foreroi* and the group *B. cayambesis*, and *B. rhoides* Fig. 7.3. Other changes occur in for *B. inermis, B. integrifolia, B. brunnea, B. acostae, B. boliviana,* and *B. cutervensis* were observed. Character steps difference of NONA´s trees are noted in Table 7.7.

7.3.1 Tree selection

Because of the criticisms on using consensus tree for studying the evolution of characters (ANDELBERG and THELER 1990, CODDINGTON and SCHARFF 1997). I prefer to select a working hypothesis among the three most parsimonious trees obtained with NONA and PAUP. The selection of one hypothesis phylogenetic represents the basis for future examination (corroboration or refutation). However, from one philosophycal point of view (KLUGE 1997), the selection of a hypothesis is seen as a verificacionist argument against Popper´s corroboration

Of the three hypotheses trees 2 and 3 offer better fit of some characters (Table 7.7) I chose tree 2 (Fig. 7.1) as working hypothesis of the *Brunellia* phylogeny for the following reasons: 1. Tree 2 is similar to the results of PAUP after successive weighting (Fig. 7.4), and similar to Pee-Wee, $k= 6$ except for *B. boliviana* with a different position. 2. Tree 2 is also similar to trees of figures 7.5a, b, majority rule consensus, Pee-Wee, $k= 1, k = 3$. 3. Tree 2 has the same topology than tree obtained with HENNIG86. 4. More nodes in tree 2 are supported by evolutionary novelties.

7.3.2 Character evidence

In the context of this matrix, the monophyly of *Brunellia* is supported by five unreversed synapomorphies: lateral inception of the stipules (2),

craspedodromous inervation (8), unilacunar node (22), presence of apotracheal axial parenchyma of the wood (24) and decurrent position of the stigma (40). Actinocytic stomata (23) and the presence of hairs in the abaxial part of the foliar lamina are also a synapomorphic but are lost in *B. cutervensis* and *B. glabra* respectively. With exception of the apotracheal axial parenquima of the wood (24) and the decurrent position of the stigma (40), the remaining characters are also present in other genera of Cunoniaceae (Chapter 2), but not present in the synthetic outgroup used in this study.

At node 58 (Fig. 7.1) two large groups are supported by the presence of compound leaves (6-1), and the complexity of ramification of the inflorescences (28-1). The presence of several leaflets per leaf (6-1), reduces to two or three leaflets per node, (6-0), in the group with navicular-shaped endocarp, node 40. Many leaflets per leaf (6-1), is also a reversion because it is present in the outgroup and in *B. ovalifolia, B. stuebelii* and *B. pauciflora*. Reduction to a unifoliate leaf, (6-2) also occurs in the group with U-shaped endocarp (e.g., *B. foreroi*, node 55), and in *B. acostae* (node 50), and in one species of the group with urceolar-shaped endocarp and congested inflorescences, *B. inermis* (node 45). The complexity of the branching of the inflorescence (28-1) reverses at node 33 (Fig. 7.1).

"Carpels number less than calyx merosity" (41-1) supports node 44. This character is shared by the group with reduced leaves (unifoliate group, (node 41) and navicular endocarp (44-0);node 42) but it reverses at node 36 (Fig. 7.1). "Carpels number less than calyx merosity" is also shared by species at nodes 43 and in *B. ecuadorneis*, (node 44) and *B. propinqua* (node 42). Of these species only *B. propinqua* and the group that it exemplifies (Tables 7.2, 7.3) are characterized by an evolutionary novelty, the autapomorphic presence of a short paracladium on the main axis of the inflorescence (32-1). Within the group with reduced leaves (node 41),the presence of a right internal side of the main vascular bundle of the foliar lamina (15-1) is sinaphomorphic at node 37. This synapomorphy is shared by the species with three unifoliate leaves per node (absent in *B. dulcis* with two unifoliate leaves per node). The presence of a right internal side of the vascular bundle (15-1) reverts to a convex or concave main vascular bundle 15(0) in *B. acuatangula* and *B. farallonensis* (Fig. 7.1).

The U-shaped endocarp (44-2) supports another large group at node 57 (absent in *B. boliviana*). This character reverts to an urceolar shape at node 48 (Figs. 1). Two autapomorphies, (37-1; 41-2), are present in *B. boliviana*, which also represents other species [no present in this matrix] (Tables 7.2, 7.3). Under the assumption of parsimony ACCTRAN and DELTRAN were used. Under ACCTRAN optimization maximum complexity of the paracladia (29-2; Fig. 7.1) supports, the monophyly at node 56, but it reverts to less complexity in *B. velutina* (29-0). Presence of one layer of palisade tissue (12-1) supports the monophyly of *B. foreroi* and *B. velutina* at node 55. A simple inflorescence with three or seven
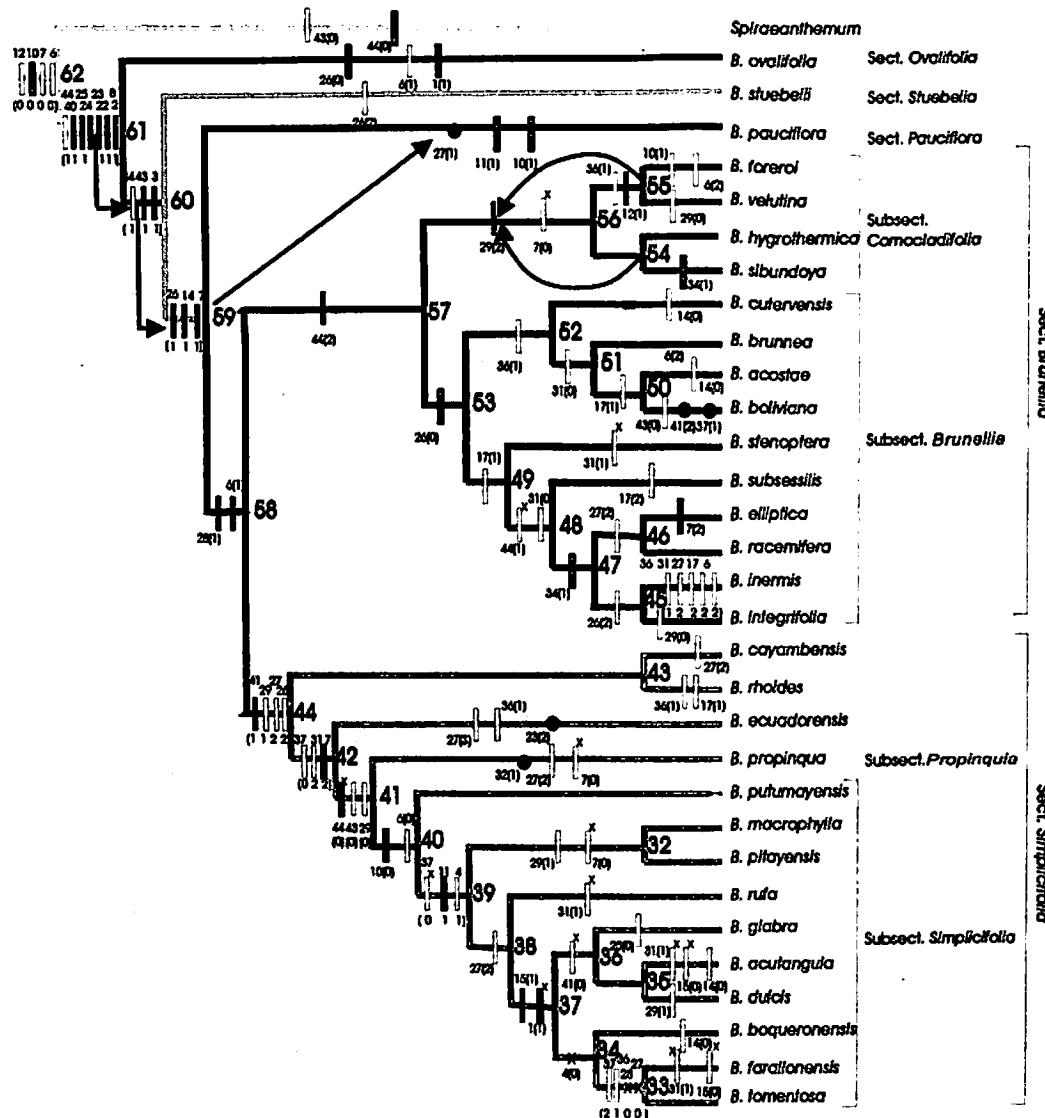
Fig. 7.1 Preferred topology of the three most parsimonious trees obtained with NONA and PAUP (length = 184, CI =0.31, IR =0.58). Black bars = unique characters (synapomorphies), black circles = autapomorphies, hatched bars = character present in a group and in one different phyletic line , white bars = character states arising once or no more than twice, the sign x = lost and reversals.
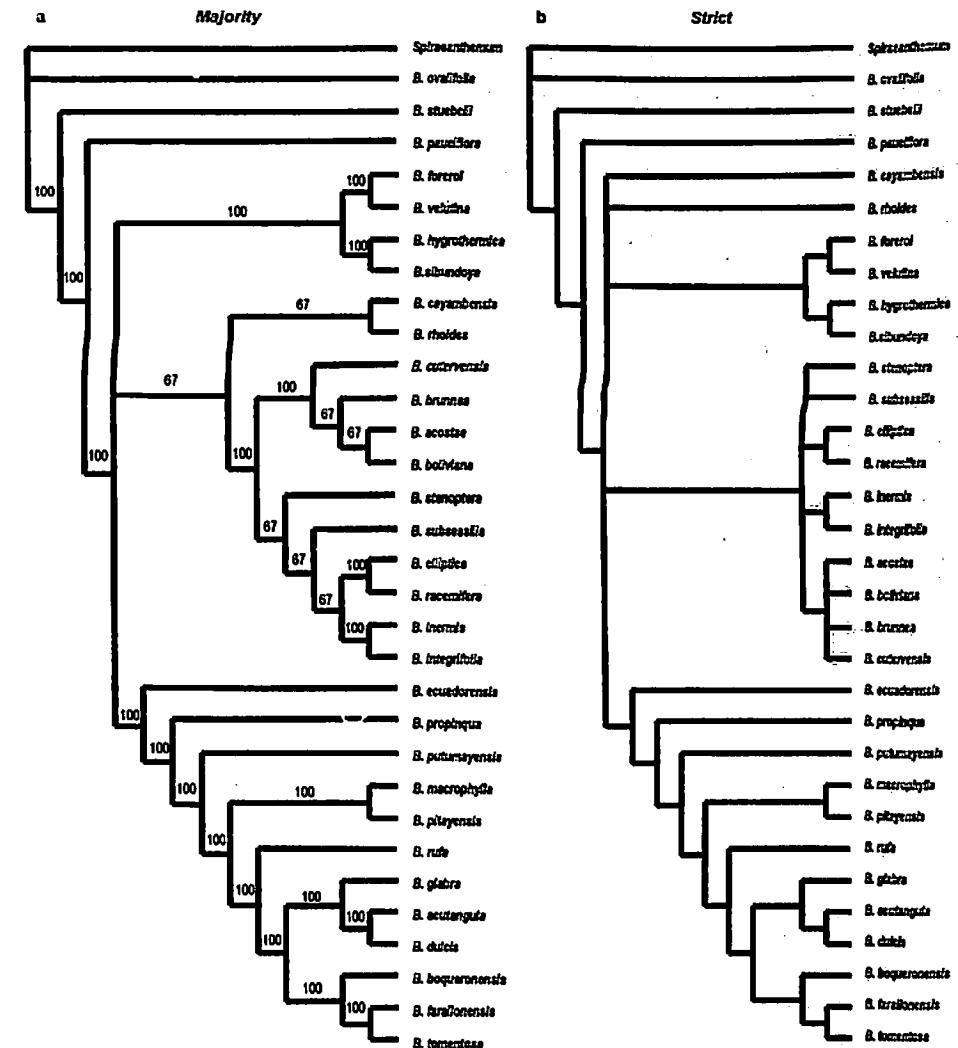
Fig. 7.2 Majority and strict consensus of the three most parsimonious trees obtained with NONA and PAUP.
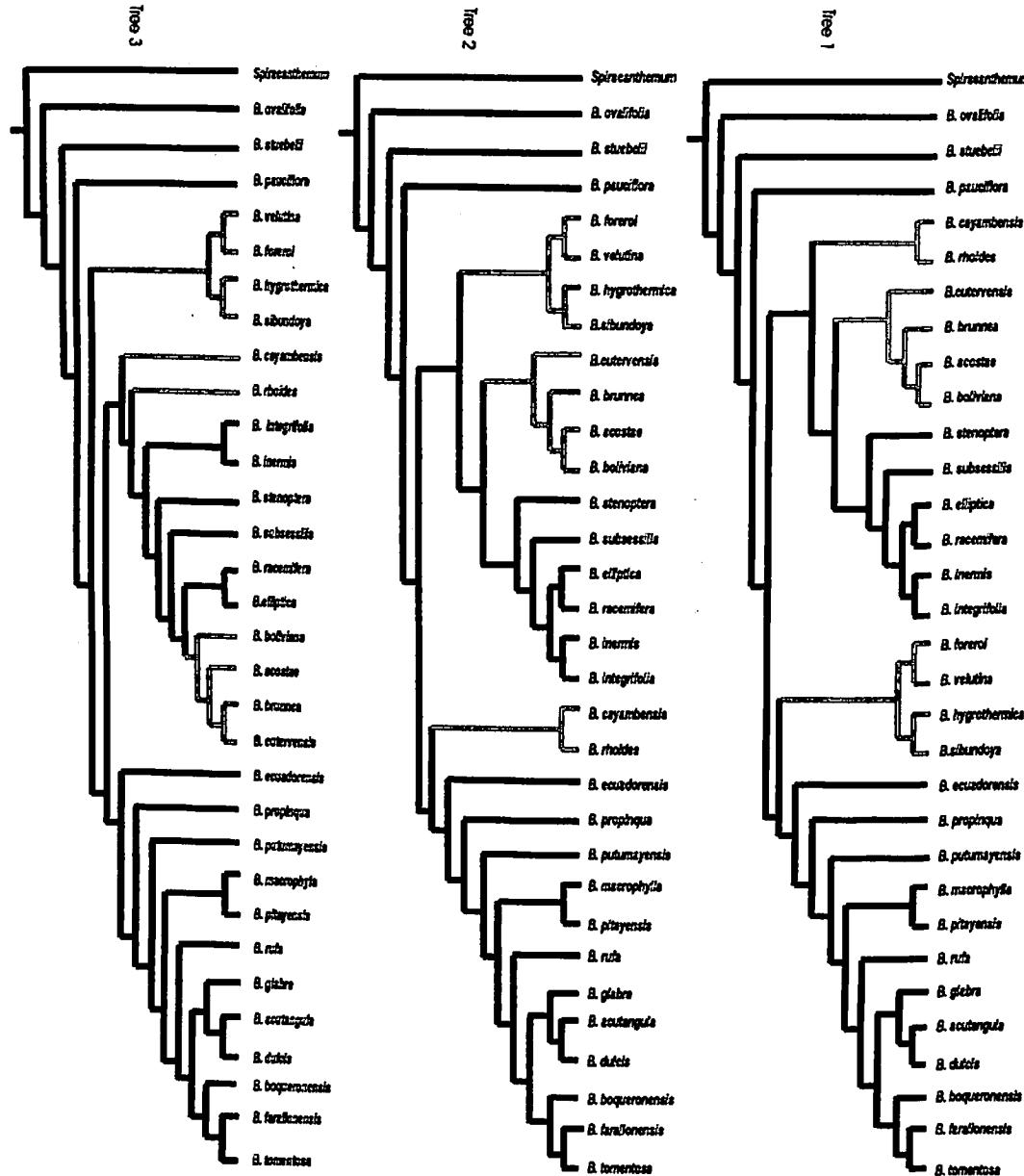
Fig. 7.3 Differences among three topologies of NONA and PAUP.

Table 7. 7 Character steps differences of the NONA trees.

Better / Worse Fit for character step differences		
Trees	Better	Worse
1	For no character	For no character
2	41(1) carpel number less than calyx merosity	13(1): presence of crypts or slight waves in the abaxial side of the lamina
	44(1) the urceolate endocarp	38(2): the prolate shape of the pollen grain
3	9(1) secondary veins, less than 25 pairs	5(1) two pairs of stipules
	17(2) the concave shape of the adaxial strand of the main vascular bundle	7(1) the narrowly-oblong shape of the leaflets
	26(1) erect abaxial indument	34(1): proliferation of inflorescence present
	38(1) the altered relationship between androecium and calyx merosity	

flowers in the floriferous paracladium (27-1; Fig. 7.1) is autapomorphic for *B. pauciflora* under DELTRAN optimization. Under DELTRAN optimization, the presence of erect hairs (26-1); Fig. 7.1 is apomorphic for the group of species with U-shaped endocarp and well developed inflorescences, (29-2) at node 56, which also includes *B. pauciflora*, a species with the smallest inflorescence of the genus. This character state is transformed to flat appressed hairs, (26-0), node 53 and woolly hairs, (26-2), node 45.

No other node is supported by unique evolutionary novelties but it is worthwhile to mention those characters which define groups but are also present in another clade (Fig. 7.1). This is seen for the presence of appressed hairs on the abaxial part of the lamina (26-0,Fig. 7.1) at node 53 (absent in *B. inermis* and *B. integrifolia*, node 45) and in *B. ovalifolia*, node 61. Similar observations were made for the characteres: whorled leaves(1-1) [present in *B. ovalifolia*, but reverting in the three unifoliolate leaves and navicular shaped-endocarp group, node 37]; the lanceolate-shape leaves (7-2) [present in species of naviculary shaped endocarp, at node 42, absent in *B. macrophylla*, *B. pitayensis* and *B. propinqua*, but it is also present in *B. elliptica* from the urceollary-shaped endocarp and appressed indument group (44-1), 26-0), node 46],two layers of hypodermis (11-1) [in species with unifoliolate leaves and navicular shaped-endocarp, node 39 and also present in the clade of *B. pauciflora*, node 59], of inflorescence proliferation (34-1), [in species of urceolar endocarp, node 47 and in *B. sibundoya*, node 54], navicular-shaped endocarp (44-0), in species at node 42 and in

Spiraeanthemum]. Remaining nodes [excepted 43 and 54 nodes] are supported by reversals or by parallel characters, which arising more than twice.

Implied weights with intermediate values of (k = 1), [majority rule consensus of 7 trees], and k= 3 [majority rule consensus of 84 trees] show also two large groups (Fig. 7.5 a, b, Table 7.5); but differ by the inclusion of *B. ovalifolia, B. pauciflora* and *B. stuebelii*. Other differences from Fig. 7.1 are the positions of *B. cayambensis, B. rhoides, B. ecuadorensis* and *B. boliviana*. The following groups are seen in the hypothesis of minimal length (Fig. 7.1) and consensus tree K= 1, 3 (Figs. 7.5): the group of *B. foreroi* (U shaped endocarp and very complex inflorescences), *B. elliptica* (urceolar endocarp and congested inflorescences) and the group of reduced inflorescences, navicular shaped endocarp are seen altogether, excepted *B. boliviana*.

7.4 DISCUSSION

HENNIG86 failed to find the most parsimonious trees because it cannot effectively search among islands. The data management program DADA (NIXON, 1999) will generate matrices with different taxa orders and submit them automatically to HENNIG86 which solves that problem. In the consensus tree of implied weights (Fig. 7.6), endocarp shape explains the two groups found under parsimony analysis, which are mainly different from the trees of implied weight with the exclusion of *B. ovalifolia, B. pauciflora* and *B. stuebelii*. Although the topology of the trees of implied weights did not change greatly from the selected MTP (Fig. 7.1), future research should focus on the problematic taxa, *B. rhoides* and *B. cayambesis*.

7.4.1 Infrageneric groups

Most of the resulting phylogenetic groups are defined either by unique evolutionary novelty or unique combinations of characters (Fig. 7.1, Table 7.1). The homoplasious synapomorphies typical of the latter could imply intergradation or reticulate process of sepeciation. Some proposed groups are also limited to a defined geographical area (OROZCO in prep.).

CUATRECASAS's ifragneric division does not correspond at all with that proposed by Table 7.1. His sections and most of the subsections are paraphyletic. In his sense, I recognize less inclusive groups such as *B. boliviana*, subsection *Bolivianae*, node 50, proposed in Sect. *Brunellia*, Subsect. *Occidentales* (Table 7.1). Within more inclusive groups as Section *Brunellia*, I recognize some of the CUATRECASAS's species (Fig. 7.1, Table 7.1).

Five sections are here proposed (Table 7.1) of which some are distinguished by a distinct set of characters such as Sect. *Ovalifoliae* (*B. ovalifolia*,
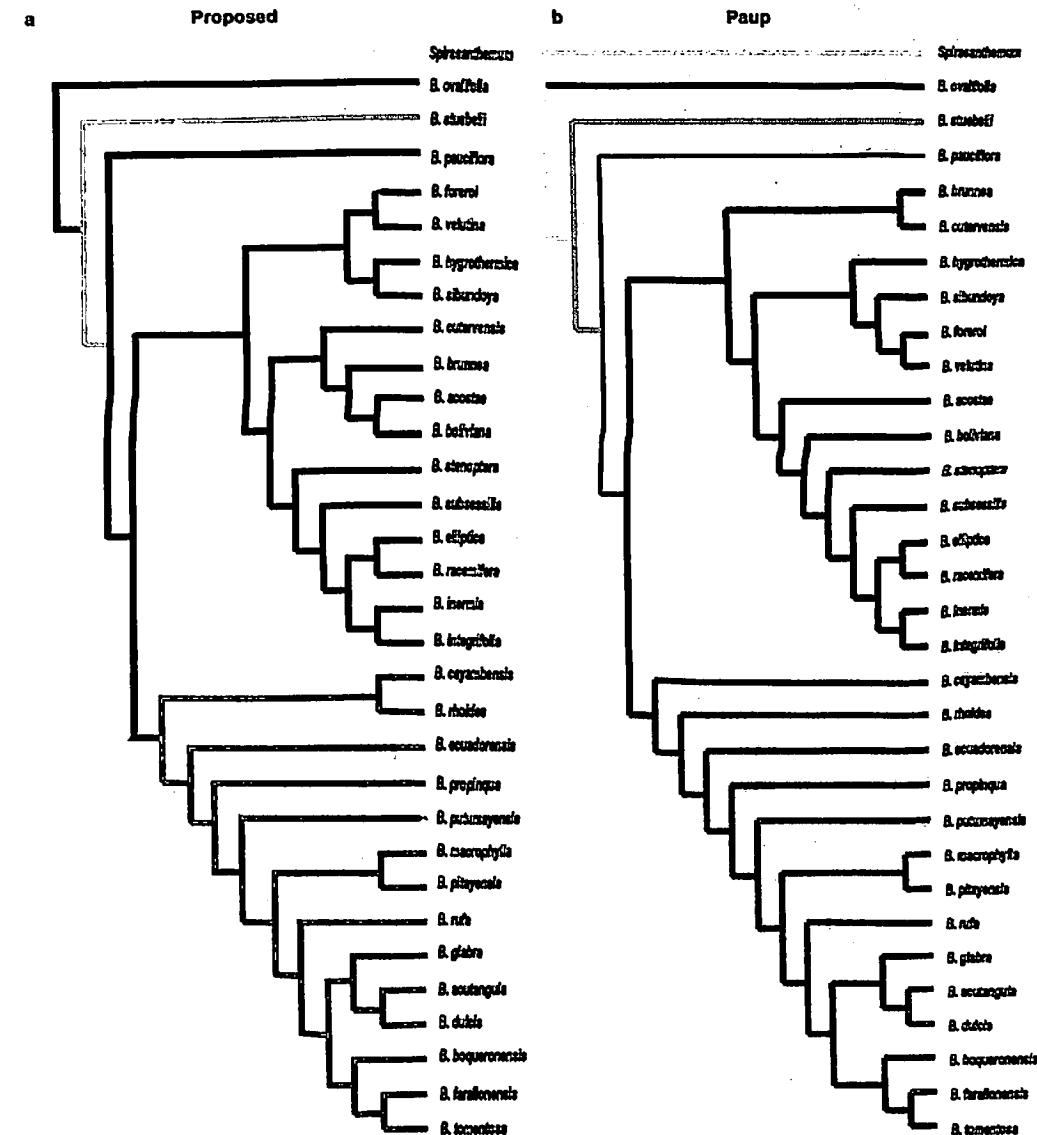
Fig. 7.4 Comparison of the tree obtained with PAUP, using successive weighting based on the rescaled consistency index (b) and the hypothesis proposed from NONA (a).
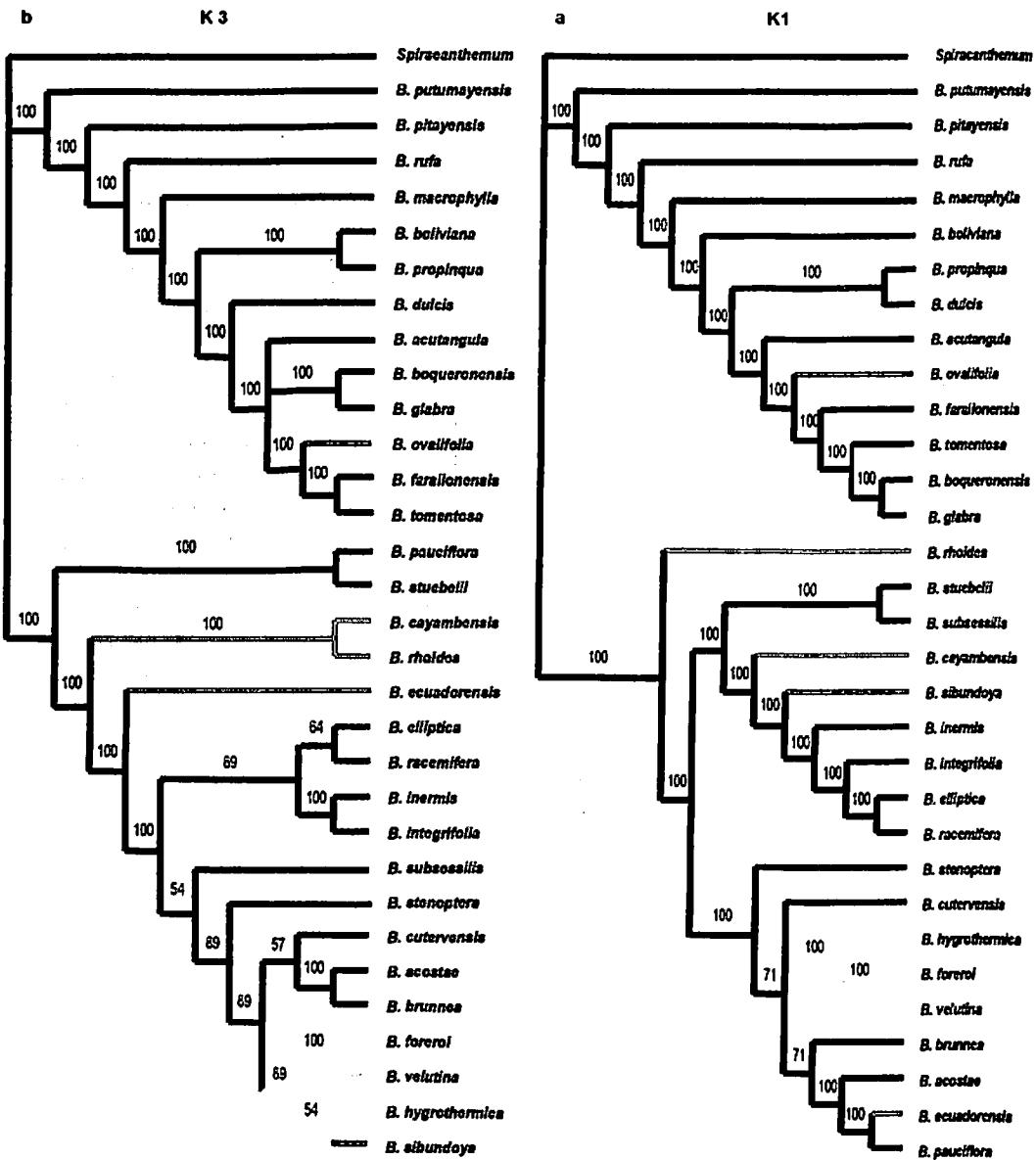
Fig. 7. 5 Majority consensus tree of trees obtained with Pee-Wee, k=1 (a), K=3 (b). a. Fit = 125.8, length =205. b. Fit = 194.8, length = 201.
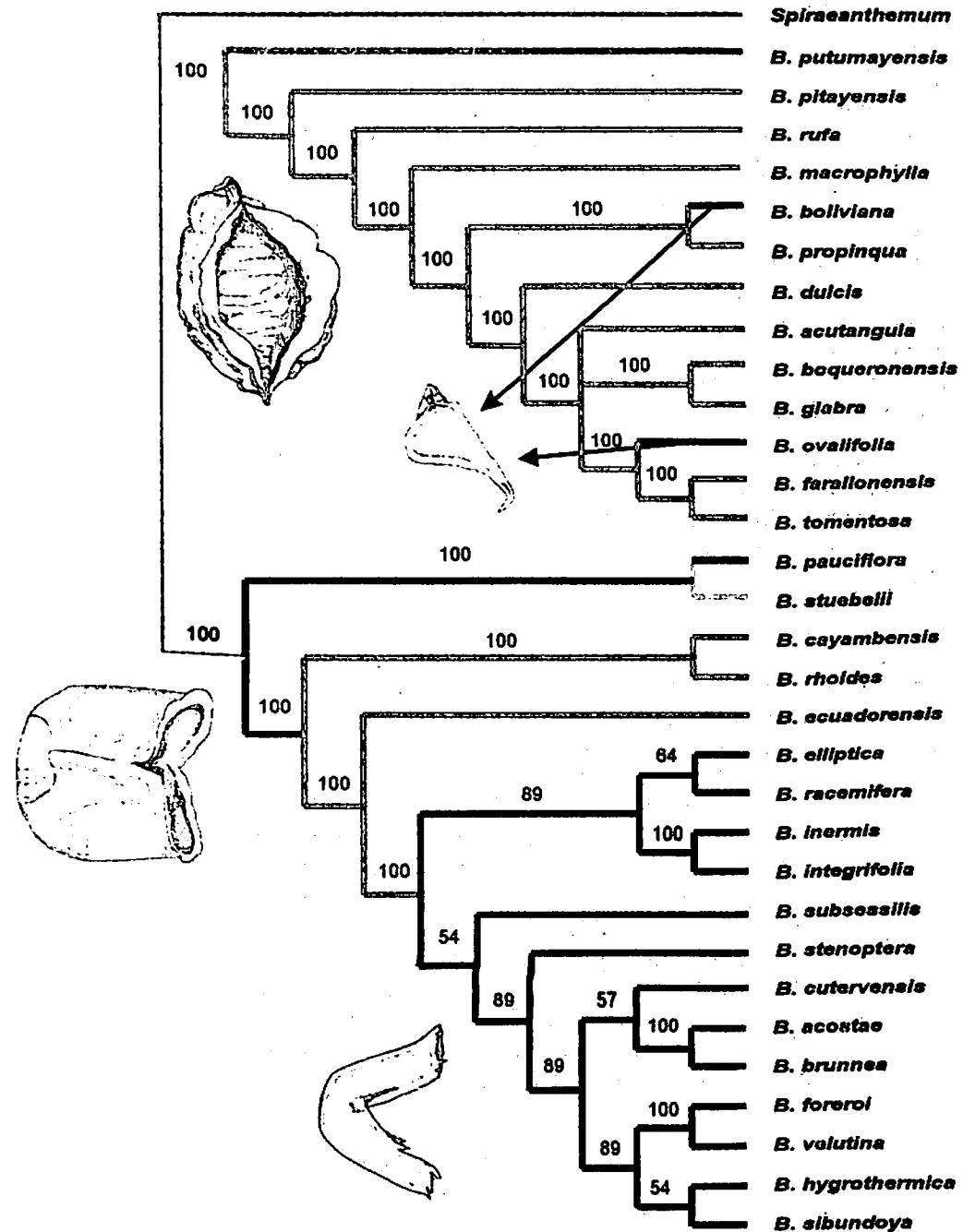
Fig. 7.6 Consensus of majority rule of 84 trees obtained with Pee-Wee, k=3, Fit = 194.8, length = 201. Behavior of the endocarp shaped (44).

node 61). This species is distinguished by the frequent presence of whorl, either unifoliate or compound, leaves per node,(1-1), (6-2), straight or flat appressed hairs (26-0). Sect. *Stuebelia* (*B. Stuebelii;*node 60) is defined by two unifoliate leaves per node, (1-0), woolly hairs (26-2), endocarp urceolar, (43-1; 44-1). Sect. *Pauciflorae* (*B. pauciflora;* node 59), defined by the simplest inflorescences found in the genus, (27-1). Section *Brunellia* (node 58), includes the majority of Cuatrecasas 's species'(Table 7.1) except *B.ecuadorensis, B. ovalifolia, B. propinqua, B. cayambensis* and *B. rhoides* This section is now best defined by the presence of small U endocarps, 44(2), and urceolar shaped endocarp, (44-1; absent in *B. boliviana*), the frequent presence of compund leaves, (6-1), and inflorescence complexity (28-1;Fig. 7.1). Section *Simplicifoliae* is defined by reduction of the carpel number (44-1) and includes the majority of species of the Cuatrecasas's section *Simplicifoliae*, excepted *B. ovalifolia, B. cayambensis, B. rhoides, B. propinqua* and *B. ecuadorensis* (Table 7.1).

Four subsections are proposed. Subsection *Comocladifoliae* (node 57;Table 7.1; Fig. 7.1) shares the presence of a U-shaped endocarp, (44-2), and the most complex branching inflorescences (29-2). Within this subsection *B. hygrothermica* and *B. velutina* form a less inclusive group defined by the synapomorphy of thinner leaves (12-1).

Subsection, *Brunellia* is the sister group of subsection *Comocladifoliae* node 57, is defined by the homoplasious synapomorphy, of straight and appressed indument on the abaxial side of the leaf, (26-0), absent in *B. inermis* and *B. integrifolia*. In this subsection four groups, are recognized, (Fig. 7.1; Table 7.1): *acostae* represented by *B. acostae, boliviana* represented *B. boliviana, cuterva* represented by *B. cutervensis,*(at nodes, 52 and 50). The *congestiflorae* group of urceolar shaped endocarp (44-1),node 48) congested inflorescence (31-0), the presence of a proliferating inflorescence (34-1) (WEBERLING 1989, OROZCO and WEBERLING 1999). This group is represented by *B. elliptica, B. inermis, B. integrifolia, and B. racemifera* (Fig. 7.1, Table 7.1). The sister group of *B. boliviana*, represented by *B. acostae* (Table 7.2), is still not defined by an evolutionary novelty, however, the frequent presence of lenticels on the branches in each of the species helps to distinguish the group and could be considered as a novelty. *B. cutervensis* is another subgroup within subsection *Occidentales* with imperceptible papillae, *B. oliveri* omitted from this matrix shares the endocarp shape and the kind of hairs on the abaxial side of the leaves with its relative *B. cutervensis*.

In the morphologically homogeneous section *Simplicifoliae*, subsection *Simplicifoliae* is distinguished at node 40, defined by the presence of hypodermis, (10-0; Fig. 7.1; Table 7.1). These species also share floral and inflorescence reduction (41-1; 29-0), woolly hairs on the abaxial side of the leaves (26-2), unifoliate leaves, (6-0), and navicular-shape endocarp (44-0). No drastic changes are proposed within subsection *Simplicifoliae* except that

B. ovalifolia is transferred to the section *Ovalifoliae* (Table 1). Within section *Simplicifoliae* another group of species with three leaves per node is distinguished by the synapomorphy of the straight internal side of the adaxial main vascular bundle (15-1; node 37).

Subsection *Propinquae* is part of section *Simplicifoliae* (Fig.7.1, Table 7.1)and is the sister group of subsection *Simplicifoliae* (node 41). It is defined by the presence of a short paracladium on the main axis of the inflorescences (32-1). This subsection share with the remaining species of the Section, floral or inflorescence reduction (41-1), woolly hairs in the abaxial side of the leaves (26-2), and navicularly-shaped endocarp (44-0). Most of the species of this subsection was previously considered paraphyletic as part of subsection *Colombianae* (Tables 7.1, 7.3; Fig. 7.1). *B. ecuadorensis* and the group *B. cayambesis- B. rhoides,* (nodes 42, 44), are recognizable by unique combination of characters, but no unique evolutionary novelties are yet known (Table 7.1). These species share with the remaining species of section *Simplicifolia* floral reduction (41-1) and a less complex inflorescence (29-1).

7.4.2 Behavior of some characters within the genus

CUATRECASAS's infrageneric clasification was based on the presence of compound vs. simple leaves. Cladistic analysis showed that simple leaves are a secondary novelty; compound leaves are primitive for *Brunellia*. In most species of subsection *Simplicifoliae* (simple leaves) vascular bundles were observed as little wings in the central vascular bundle of the petiole (OROZCO and COBA in prep.). The same condition occurs in *B. stuebelii* (Fig. 7.1), but it is apparently absent in the anatomical preparations of the other monotypic secciones (*Pauciflorae, B. pauciflora; Ovalifoliae, B. ovalifolia*), but it could be due to a very reduced size of the wings in the central vascular bundle. In very recent observations of the herbarium material of AU, by loupe, tiny stipels were detected on the petiole of *B. ovalifolia*. In most of the proposed groups of the section *Brunellia* reduction to a simple foliar lamina occurs.

Of fifteen anatomical characters used in the analysis, five characters are synapomorphies such as nodal anatomy, stomata, apotracheal axial parenchyma in the wood, the straight side of the adaxial main vascular bundle of the leaf, and the number of layers of palisade. Even though the presence of a hypodermis was found to be homoplasious, it was associated with reduction of the exposed area due to one leaflet per leaf. Several species have crypts (13) in the leaves, which correlate with humidity and cloud forests.

It seems that reduction of floral parts correlates with the presence of larger fruits and navicular-shaped endocarp, e.g. Sect. *Simplicifoliae*, whereas the more frequent presence of U-or urceolar-shaped endocarp, Sect. *Brunellia*,

correlates with an increase in or constant number of floral parts (except for *B. boliviana* with a modified navicular endocarp and *B. cayambensis, B. rhoides, B. ecuadorensis* of Sect. *Simplicifoliae,* with urceolar endocarp). Reduction of branching in the inflorescences is also seen in section *Simplicifoliae* as opposed to the very developed inflorescence branching in section *Brunellia.* Foliar and inflorescence reduction are the most remarkable reversals present in the genus.

Even though the pollen grains were not seen for all the species, "punctate" and, more often, modified reticulate sculpture exine (OROZCO 2001) is present in section *Simplicifoliae* and in one of the most basal sections of the genus, section *Stuebelia.* Rugulate and finely reticulate exines were often found in the section *Brunellia.*

7.4.3 *Species concept*

NIXON and WHEELER (1990) recently discussed the autapomorphic and phylogenetic species concepts. However, is not the case for *Brunellia,* they state that the autapomorphic concept, fails in the monophyletic concept of species (DE QUEIROZ and DONOGHUE 1990) for species with asexual reproduction due to lack, in the case of asexual reproduction, of relationships between tokogenetic and phylogenetic identites. A small number of species of *Brunellia* can be defined by autapomorphic characters, but the remainder lack apomorphic character. However, these clades can be diagnosed by unique combinations of characters (DAVIS and NIXON 1992). These combinations could be autapomorphic, even though each contributing character is in itself homoplasious. In summary, most *Brunellia* species are best defined by "the smallest aggregation of population (sexual) or linages (asexual) diagnosable by a unique combination of character states in comparable individuals (semaphoronts)" (NIXON and WHEELER's concept 1990). Unique combinations of characters also account for most group of species (Table 7.1).

7.5 ACKNOWLEDGMENTS

7.6 LITERATURE

APG. 1998. An ordinal classification for the families of flowering plants. Ann. Missouri Bot. Gard. 85: 531-553.

ANDERBERG, A. and THELER, A. 1990. Consensus trees, a necessity in taxonomic practice. Cladistics 6: 399-402.

BRADFORD, S. and BARNES, R. 2001. Phylogenetics and classification of Cunoniaceae (Oxalidales) using chloroplast DNA sequences and morphology. Syst. Bot. 26: 354-385.

CODDINGTON, J. and SCHARFF, N. 1994. Problems with zero-length branches. Cladistics 10: 415-423.

CODDINGTON, J. and SCHARFF, N. 1997. A phylogenetic anlysis of the orb-weaving spider family Araneidae (Arachnida, Aranae). Zool. J. Linn. Soc. 120 : 355-434.

CUATRECASAS, J. 1970. Brunelliaceae. Flora Neotropica. Monograph 2 : 1-189.

CUATRECASAS, J. 1985. Brunelliaceae. Flora Neotropica Monograph 2 (suppl.) : 29-103.

DAVIS, J. I. and NIXON, K. C. 1992. Populations, genetic variation, and the delimitation of phylogenetic species. Syst. Biol. 41: 421-435.

DE QUEIROZ, K. and DONOGHUE, M. 1990. Phylogenetic systematics and the species revisited. Cladistics 6: 83-90.

DICKISON, W. C. 1973. Anatomical studies in the Connaraceae. III. Leaf anatomy. J. Elisha Mitchell Sci. Soc. 89: 121-138.

DICKISON, W. C. 1975. Leaf anatomy of Cunoniaceae. Bot. J. Linn. Soc. 71: 275-294.

DICKISON, W. C. 1980. Diverse nodal anatomy of the Cunoniaceae. Amer. J. Bot. 67: 975-981.

EYDE, R. 1979. Anatomy. In: (ed. J. CUATRECASAS) Flora Neotropica. Monograph 2: 32-43.

ENGLER, A. 1897. Brunelliaceae. In Die natürlichen Planzenfamilien, 2 nd (eds. A . ENGLER and K. PRANTL), pp. 182-184,. Leipzig, Engelmann.

FARRIS J. S. 1969. A succesive approximations approach to character weigthing. Syst. Zool. 18 : 374-385.

FARRIS, J. S. 1988. Hennig86. Version 1.5. Port Jefferson Station, New York.

FARRIS, J. S. 1989. The retention index and the rescaled consistency index. Cladistics 51: 417-419.

KLUGE, A. G. 1997. Testability and the refutation and corroboration of cladistic hypothesis. Cladistics 13: 81-96.

Goloboff, P. 1993a. *NONA. Nona ver. 1.15 (a bastard son of Pee-Wee) 32-bit version* Program and documentation available from Jonathan Coddington. Dept. of Entomology, Smithsonian Institution. Washington D.C.

Goloboff, P. 1993b. *Pee-Wee. Version 2.15 (32-bit version).* Program and documentation. Computer program available from Jonathan Coddington. Dept. of Entomology, Smithsonian Institution. Washington D.C.

Goloboff, P. 1993c. Estimating character weights during tree search. Cladistics 9: 83-91.

Hennig, W. 1966. Phylogenetic Systematics. Univ. of Illinois Press, Urbana.

Hickey, L. J. and Taylor, D. W. 1991. The leaf architecture of *Ticodendron* and the application of foliar characters in discerning its relationships. Ann. Missouri Bot. Gard. 78 : 105-130.

Hoogland, R. D. 1979. Studies in the Cunoniaceae. II. The genera *Caldcluvia, Pullea, Acsmithia,* and *Spiraeanthemum.* Blumea 25: 481-505.

Hufford, L. and Dickison, W. C. 1992. A phylogenetic analysis of Cunoniaceae. Syst. Bot. 17: 181-200.

Maddison W. P. and Maddison, D. R. 1992. MacClade: Interactive analysis of phylogeny and character evolution. Version 3.0. Sunderland, Massachusetts: Sinauer Associates.

Nixon, K. C. and Wheeler, Q. D. 1990. An amplification of the phylogenetic species concept. Cladistics 6: 211-224.

Nixon, K. C. 1992. *Clados ver. 1.2.* Program and documentation. Distributed by the author, P. O. Box 270, Trumansburg, NY 14886, USA.

Orozco, C.I. 1981. Contribución al estudio de las Bruneliáceas de Colombia I. Mutisia 50 : 4-8.

Orozco, C.I. 1985. Dos especies nuevas de *Brunellia* para Colombia. In (ed. J. Cuatrecasas). Flora Neotropica Monograph 2 (suppl.): 20-24.

Orozco, C. I. 1986. Contribución al estudio de Brunelliaceae de Colombia III. Caldasia 15: 177-185.

Orozco, C. I. 1991. Contribución al estudio de Brunelliaceae IV. Dos especies del Ecuador. Caldasia 16: 453-458.

Orozco, C. I. 1997. Sobre la posición sistemática de *Brunellia* Ruiz & Pavón. Caldasia 19: 145-164.

Orozco, C. I. 1999. Indumento del envés foliar en *Brunellia* Ruiz & Pavón y su relativa importancia en la filogenia. Rev. Acad. Colomb. Cienc. 23 (Suplemento especial): 97-102.

Orozco, C. I. and Weberling, F. 1999. A comparative study of inflorescences in *Brunellia* Ruiz & Pav. (Brunelliaceae) and related taxa. Beitr. Biol. Pflanzen 71: 261-279.

Orozco, C. I. 2001. Pollen morphology of *Brunellia* (Brunelliaceae) and related taxa in the Cunoniaceae. Grana 40: 245-255.

Porter, D. and Cuatrecasas, J. 1975. Brunelliaceae. Flora de Panama. Ann. Missouri Bot. Gard. 62: 11-14.

Rutishauser, R. and Dickison. W. C. 1989. Developmental morphology of stipules and systematics of the Cunoniaceae and presumed allies I. Taxa with interpetiolar stipules. Bot. Helvetica 99: 147-169.

Swofford, D. 1990. PAUP: Phylogenetic analysis using Parsimony Version 3.0. Computer program, Illinois Natural History Survey, Illinois.

Turner, H. and Zandee, R. 1995. The behaviour of Goloboff's tree fitness measure F. Cladistics 11: 57-72.

Weberling, F. 1989. Morphology of flowers and inflorescences. Cambridge Univ. Press, Cambridge. 405 pp. English translation of the German edition 1981 Ulmer, Sttutgart.

APPENDIX. CHARACTERS USED IN THE CLADISTIC STUDY OF *BRUNELLIA.*

1. *Leaf arrangement*: decussate(0); whorled (1).
2. *Stipule position*: interpetiolar (0) in *Spiraeanthemum*; lateral (1) in *Brunellia*. (Rutishauser and Dickison 1989)
3. *Stipule basal*: round (0); straight (1).
4. *Stipule body*: pyramidal (0); ovate-oblong (1).
5. *Stipule number*: one pair (0); two pairs (1). In some cases an additional stipule is present between the two larger stipules.
6. *Number of leaflets* (in late stages of development): one leaflet (unifoliate) per leaf (0); several leaflets per leaf (1); unifoliolate and multifoliate leaves at the same node (2). (Orozco 1981,1985, 1986), (Chapter 3).
7. *Lamina shape*: broadly oblong or ovate and widely elliptic (0); narrowly oblong (1); lanceolate (2).
8. *Secondary venation*: brochidodromous (0); craspedodromous (1) (Hufford and Dickison 1992, Hickey and Taylor 1991).
9. *Number of secondary veins*: less than 16 pairs (0); no more than 25 pairs (1); more than 25 pairs (2).
10. *Foliar hypodermis*: present (0); absent (1). (Dickison 1973, Chapter 3).
11. *Layers of the hypodermis*: one layer (0); more than one layer (1).(Dickison 1973, Chapter 3).
12. *Number of layers of palisade parenchyma*: more than one layer (0); one layer (1).
13. *Abaxial side of lamina*: right or plane (0); with crypts or slights waves (1). It is unknown for the outgroup.
14. *External side of the adaxial main vascular bundle of the lamina*: right or plane (0); depressed or concave (1). It is unknown for the outgroup.
15. *Internal side of the adaxial main vascular bundle of the lamina*: convex or concave (0); flat (1). (Chapter 3). It is unknown for the outgroup.
16. *Continuity of the abaxial strand of central vascular bundle in the lamina*: continuous (0); discontinuous (1). (Dickison 1973, Chapter 3).
17. *Adaxial strand of the main vascular bundle*: depressed (0); discontinuous or folded (1); concave (2). (Chapter 3).

18. *Abaxial strand of the main vascular bundle of the petiole:* continuous (0); discontinuous (1). (DICKISON 1973, Chapter 3).

19. *Accessory vascular tissues in the petiole:* present (0); absent (1). (DICKISON 1973; Chapter 3).

20. *Accessory vascular tissues in the petiole:* medullary bundles (0);cortical bundles (1). (DICKISON 1973, Chapter 3).

21. *Support tissues around the main vascular bundle of the petiole:* sclerenchyma (0); collenchyma (1). (DICKISON 1973, Chapter 3).

22. *Nodal anatomy:* trilacunar (0); unilacunar (1). (DICKISON 1980, Chapter 3).

23. *Stomata:* paracytic stomata (0); actinocytic stomata (1); anomocytic stomata (2). (HUFFORD and DICKISON 1992, Chapter 3).

24. *Apotracheal axial parenchyma of the wood:* present (0); absent (1). (EYDE 1979).

25. *Abaxial leaf indument:* absent (0); present (1). (HOOGLAND 1979, OROZCO 1999).

26. *Abaxial leaf indument:* straight or appressed (0); erect (1), or woolly (2).

27. *Complexity of the inflorescences:* monothyrsoid inflorescences as in *B. tomentosa* (0); The simplest inflorescence with three or seven flowers in the floriferous paracladium, as in *B. pauciflora* (1); thyrsoid with subthyrsoid partial inflorescence, (complexity at the proximal branches) (2); thyrsoid with subthyrsoid partial inflorescence, (complexity in proximal and distal inflorescence (3). (HOOGLAND 1979, OROZCO and WEBERLING 1999).

28. *Complexity order of paracladia:* absent (0); present (1).

29. *Complexity order of the paracladia:* second-third order (0); third-fourth order (1); more than fourth order (2). (OROZCO and WEBERLING 1999).

30. *Shortening of the hypopodium and epipodium:* inflorescence lax (0); congested (1).

31. *Acrotonic growth of the distal paracladia overlapping the main axis:* absent (0); present (1).(OROZCO and WEBERLING 1999).

32. *Presence of a short paracladium on the main axis of the inflorescences:* absent (0); present (1). (OROZCO and WEBERLING 1999).

33. *Fertile portion of the inflorescence (%) in relation to the total inflorescence:* more than 50% (0); less than 50% (1). (OROZCO and WEBERLING 1999).

34. *Proliferating of inflorescence:* absent (0); present (1). (OROZCO and WEBERLING 1999).

35. *Floral symmetry:* asymmetric flower (0); actinomorphic flower (1). (Chapter 5).

36. *Relation between androecium and calyx merosity:* obdiplostemonous (0); altered obdiplostemonous plan (1) (Chapter 5).

37. *Carpel number:* 4-5 carpels (0); more than 5 carpels (1); less than 4 carpels (2). (HOOGLAND 1979, Chapter 5).

38. *Pollen shape:* suboblate (0); oblate (1); prolate (2); subprolate (3). (Chapter 6).

39. *Exine ornamentation:* reticulate (0); rugulate (1). (Chapter 6).

40. *Stigma position:* terminal (0); decurrent (1). (CUATRECASAS 1970).

41. *Ratio carpel /calyx merosity:* carpel merosity equal to calyx merosity (0); less than calyx merosity (1);more than calyx merosity (2). Unknown for the outgroup. (Chapter 5).

42. *Seed shape:* ellipsoid (0); long ellipsoid (1).

43. *Endocarp proportions:* wider than long (0); longer than wide (1).

44. *Endocarp shape:* navicular (0); urceolate (1); U-shaped (2). These terms are used by CUATRECASAS (1970, 1985). A modified navicular endocarp was seen in *B. ovalifolia* and *B. boliviana* (3).

Chapter 8

PHYTOGEOGRAPHY[1]

CLARA INÉS OROZCO

ABSTRACT

The absence of phylogentic studies for groups with Andean and Caribbean distribution is a limiting factor in producing a historical biogeographical hypothesis from of point of view of vicariance biogeography. However, given the importance of a starting point for a biogeographic study, a scenario on the origin of the distribution of *Brunellia* is presented here, based on its current distribution, the related taxa and the geological formation of the Andes, Central America and Caribbean Islands. A diversification and speciaiton area is defined for *Brunellia* in terms of phylogenetic lineages. A Gondwanan origin by mass migration from south to the north is discussed as well as the origin of *Brunellia* in Central America and Greater Antilles by land connections with the north of South-America, during Eocene-Miocene. The highest speciation and diversification of *Brunellia* took place in the Mid-Miocene with the main upheaval of the Andes. *Brunellia* is well adapted to high altitudes and some species have developed special anatomical leaf characters as a response to these altitudes. Colombia has the highest number of *Brunellia* species and the highest rate of endemism. Patterns of geographic distribution are presented.

RESUMEN

La ausencia de estudios filogenéticos en grupos con distribución andina y distribución en las Islas del Caribe es una limitante para la generación de

hipótesis de biogeografía histórica. Otras ideas de biogeografía histórica se apartan de la biogeografía vicariante. Como un punto de partida para un estudio o planteamiento biogeográfico, se presenta un escenario del origen de la distribución de *Brunellia*. Este planteamiento es basado en la actual distribución, en los taxones relacionados con *Brunellia* y la información geológica sobre la formación de los Andes, Centro América y las Islas del Caribe. Áreas de especiación y diversificación son definidas para *Brunellia* en términos filogenéticos. Es discutido el origen Gondwanico de *Brunellia* por migración de masas de tierra desde el sur de América. El origen de *Brunellia* en Centro América y las Islas del Caribe es también discutido con base en las conexiones de tierra con el norte de Sur América durante el Eoceno y el Mioceno. La mayor especiación y diversificación de *Brunellia* ocurrió a mediados del Mioceno, época del mayor levantamiento de los Andes. *Brunellia* es bien adaptada a altas altitudes. Algunas especies han desarrollado algunas estructuras anatómicas en respuesta a las altas elevaciones. Colombia tiene el mayor número de especies y el más alto endemismo. Son también discutidos los patrones de distribución geográfica.

8.1 INTRODUCTION

According to ROSEN (1985) and other researchers such as NELSON and PLATNICK (1981), the knowledge of the current distribution of modern biota is not enough to understand the distribution of the organisms. For a good understanding, it is also necessary to know the relationships between the organism, the geological and paleo-environmental reconstruction and the fossil record. PAGE and LYDEARD (1994) consider that despite the difficulties in extracting relevant information from geological literature, the precision of geological reconstructions should not be overestimated.

In the case of the Andean angiosperms, HUMPHRIES and PARENTI (1986) consider that South America is a complex area where multiple factors have influenced the current distribution. TAYLOR (1995) considers their distribution as a consequence of ecological factors such as climate and historic geographical connections to Antarctica, Australia, Africa, Meso-and North America, as well as of the formation of the Andes. GRAHAM (1995) states that North Latin American biota is viewed as a mosaic composed of elements arriving by different routes and at different times throughout the Late Mesozoic and the Tertiary. HOOGHIEMSTRA and CLEEF (1995) also consider that Andean montane ecosystems had a highly dynamic history during the Late Pliocene and as a result diversification in the Andes is not only a product of the migration of biota but also due to periodic, climatic, and environmental changes.

For a long time, explanations about the distribution of the organism were based on the current environmental causes, sometimes also accompanied by geological arguments. This discipline known as ecological biogeography has a marked difference with historical biogeography. Historical biogeography results
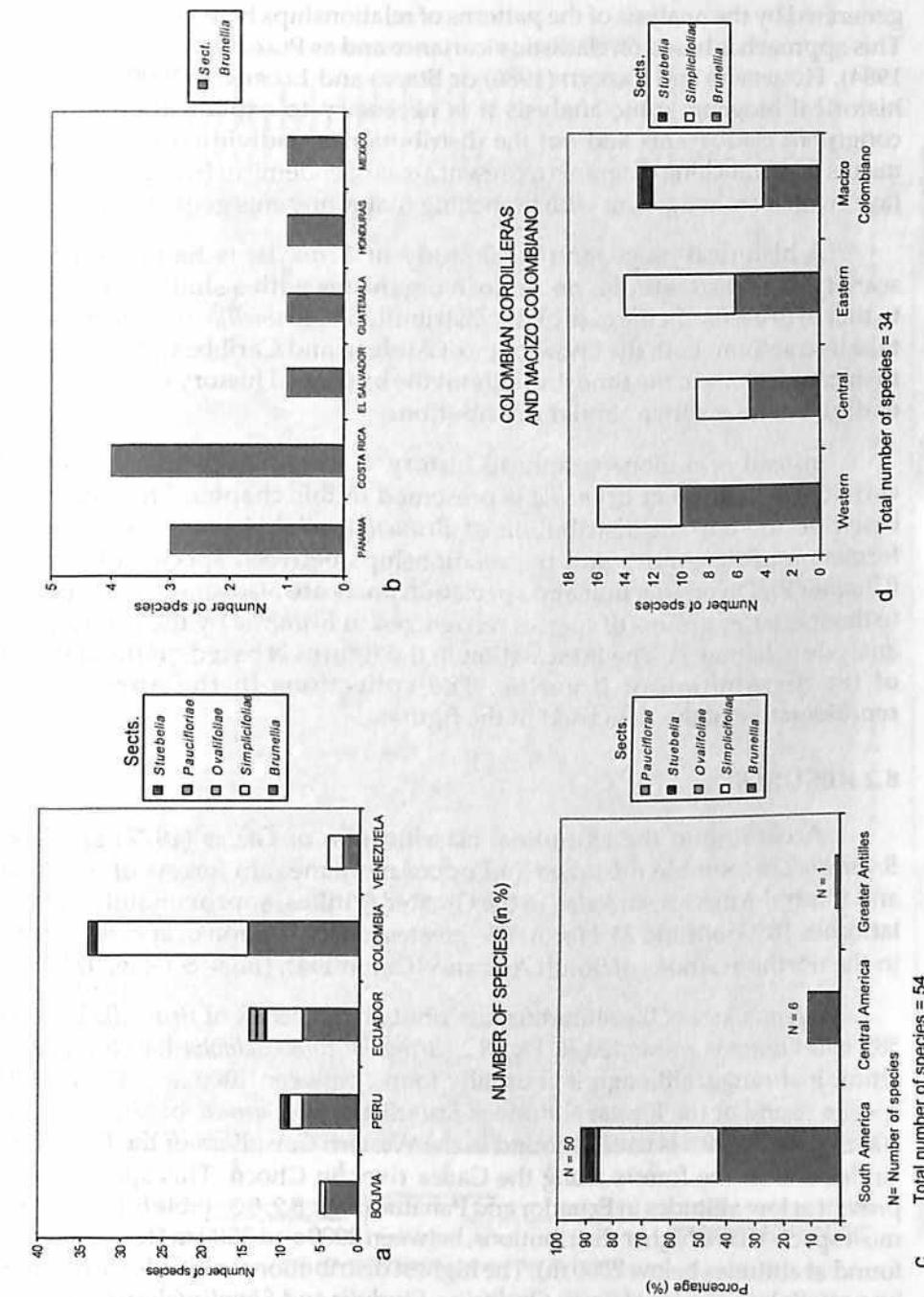
Fig 8. 1 Species of *Brunellia* according to geographical distribution.

from the study of relationships between areas where the evolutionary history of the taxa has occurred. This understanding results from a hypothesis of areas generated by the analysis of the patterns of relationships between different taxa. This approach is based on cladistic vicariance and as PLACTNIK and NELSON (1978, 1984), HUMPHRIES and PARENTI (1986) or BUENO and LLORENTE (2000) state, for a historical biogeographic analysis it is necessary to explain the patterns of congruent cladograms and not the distribution of individual groups. It also means that branching diagrams represent areas of endemism (shared by different taxa) which are congruent with branching that represents geological events.

A historical biogeographical study of *Brunellia* is hampered by the scarcity of cladistic studies on Andean organisms with a similar distribution to that of *Brunellia*. In the case of the distribution of *Brunellia*, this study should take into account both the knowledge of Andean and Caribbean histories, and their connections in the time throughout the biological history of *Brunellia* and of different taxa with a similar distribution.

Instead of a biogeographical history, a scenario on the origin of the current distribution of *Brunellia* is presented in this chapter. This scenario is based on the current distribution of *Brunellia*, related taxa, the geological formation of the Andes and the relationships between species of *Brunellia* (Chapter 7). Diversification and speciation areas are also analyzed according to the species or groups of species recognized in *Brunellia* by the phylogenetic analysis (Chapter 7). The information in the figures is based on the data base of the distribution of *Brunellia*. The collections in the appendix are representative of the data used in the figures.

8.2 RESULTS

According to the altitudinal classification of GRUBB (1977) species of *Brunellia* are found in the lower and upper montane rain forests of the Andes and Central America, and also in the Greater Antilles, approximately between latitudes 16⁰ South and 23⁰ North. The greatest concentration of species is found in the northern Andes of South America (Colombia), (Figs. 8.1-8.6, Table 8.1)

An analysis of the altitudinal distribution of species of *Brunellia* based on 585 collections is presented in Fig. 8.2. *Brunellia comocladifolia* has the greatest altitudinal range, although it is usually found between 1000 and 1700 m. The species found at the lowest altitude is *Brunellia hygrothermica*, between 400 and 600 m. This species is usually found in the Western Cordillera of the Colombian Andes and in the forests along the Cauca river, in Chocó. This species is also present at low altitudes in Ecuador and Panama (Figs. 8.2, 8.3, Table 8.1). Currently most species have higher distributions, between 2000 and 3500 m (few species are found at altitudes below 2000 m). The highest distributional altitudes are reached by unifoliolate species of Sects. *Ovalifoliae*, *Stuebelia* and *Simplicifoliae* (Chapter 7).
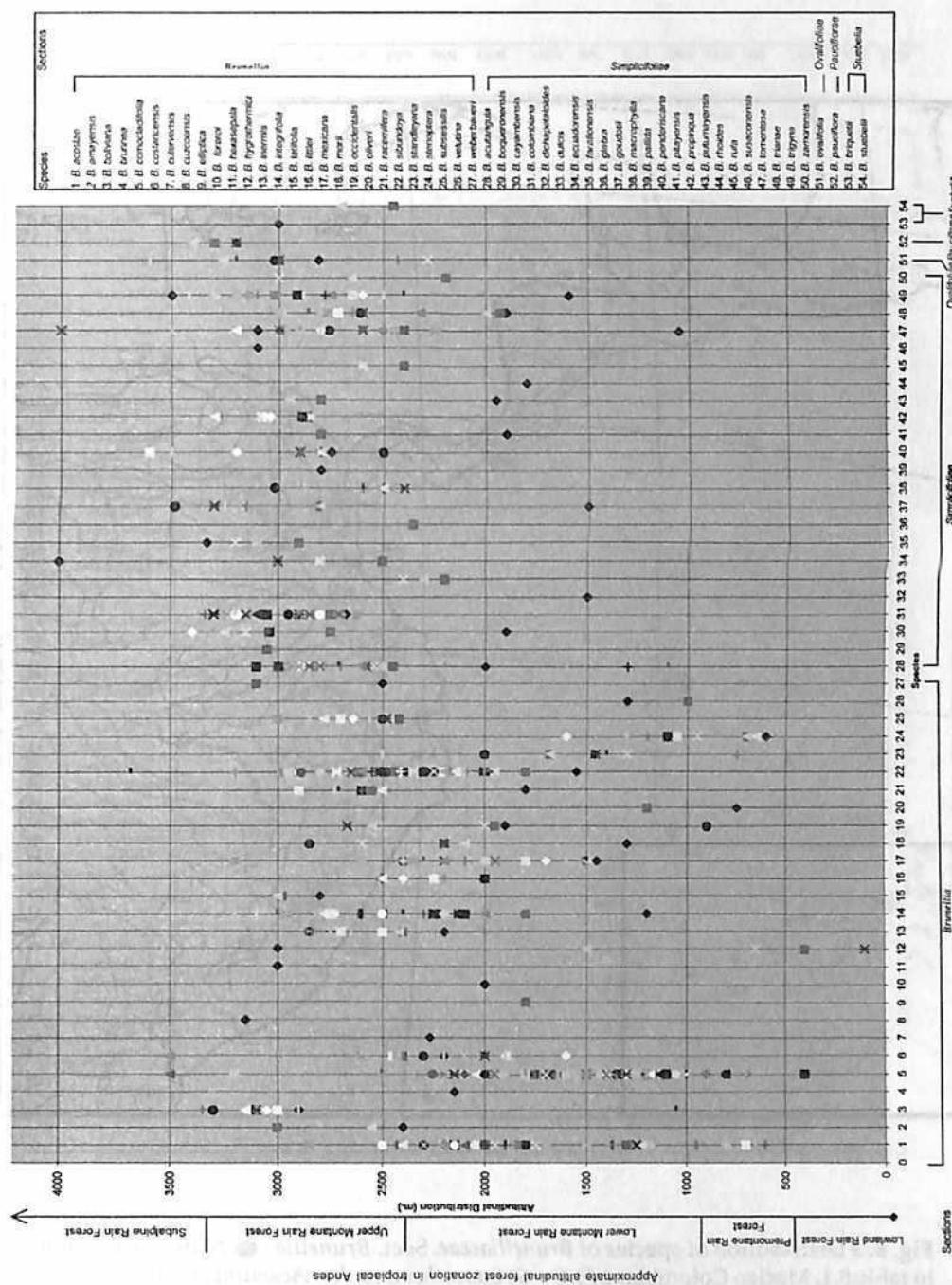
Fig. 8. 2 Altitudinal distribution of species of *Brunellia* based on collections.
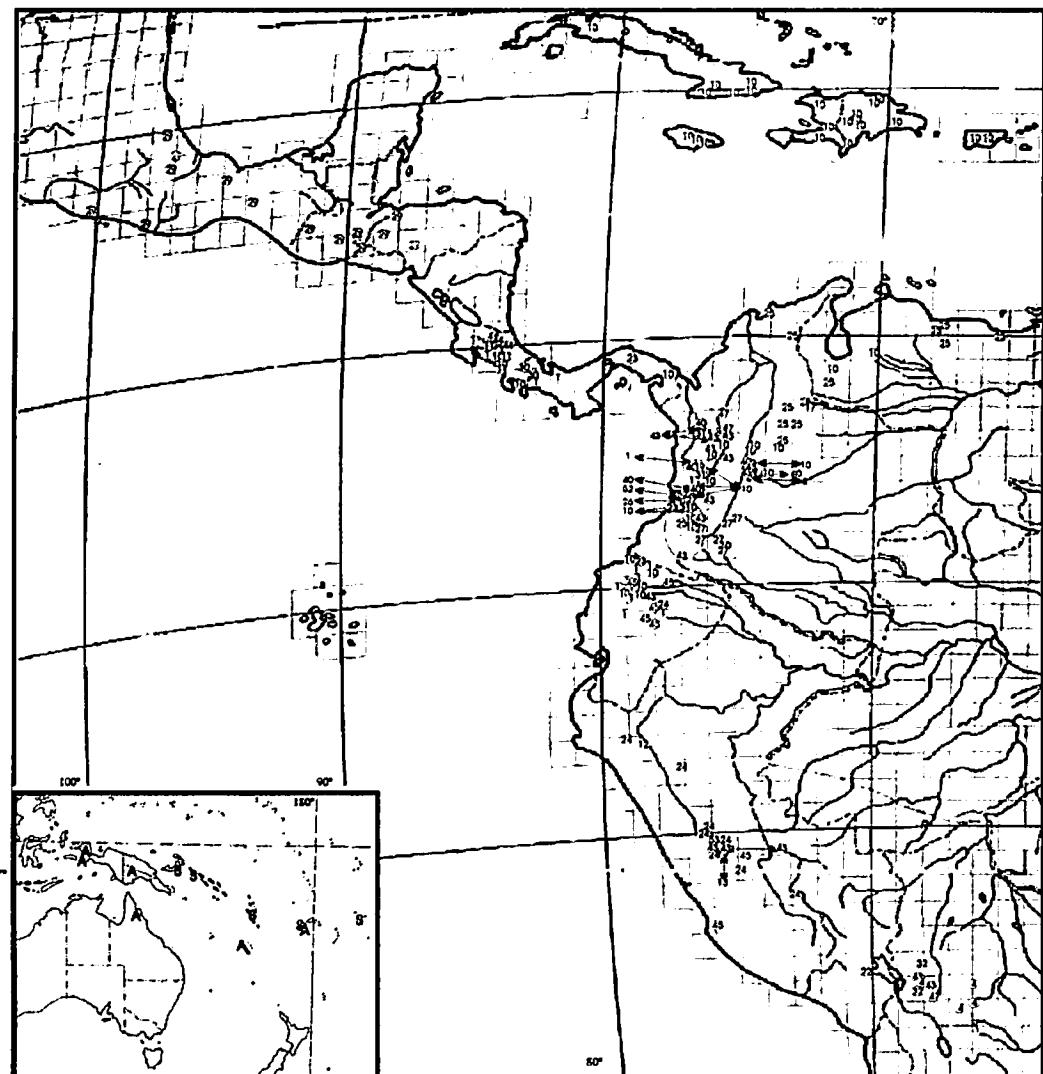
Fig. 8. 3 Distribution of species of *Brunelliaceae*. Sect. *Brunellia* ●. Names of species in table 8.1. Macizo Colombiano ◯. S = *Spiraeanthemum* A = *Acsmithia* (Cunoniaceae). Two genera close related to *Brunellia*.

Table 8. 1 Species of *Brunellia* per country.

	SPECIES	BOL	PER	ECU	COL	VEN	PAN	COS	HON	SAL	GUA	MEX	CUB	HAI	JAM	PUE	DOM
1	B. acostae			+	+												
2	B. acutangula				+	+											
3	B. amayensis				+												
4	B. boliviana	+															
5	B. boqueronensis				+												
6	B. briquetii		+														
7	B. brunnea		+														
8	B. cayambensis			+	+												
9	B. colombiana				+												
10	B. comocladifolia			+	+	+	+	+	+				+	+	+	+	+
11	B. costaricensis							+									
12	B. cutervensis		+														
13	B. cuzcoensis		+														
14	B. dichapetaloides		+														
15	B. dulcis		+		+												
16	B. ecuadorensis			+													
17	B. elliptica				+												
18	B. farallonensis				+												
19	B. foreroi				+												
20	B. glabra				+												
21	B. goudotii				+												
22	B. hexasepala		+														
23	B. hygrothermica			+	+	+											
24	B. inermis		+	+													
25	B. integrifolia				+	+											
26	B. latifolia				+												
27	B. littlei				+												
28	B. macrophylla				+												
29	B. mexicana										+	+	+	+			
30	B. morii						+	+									
31	B. occidentalis				+												
32	B. oliveri	+															
33	B. ovalifolia			+													
34	B. pallida				+												
35	B. pauciflora			+													
36	B. penderiscana				+												
37	B. pitayensis				+												
38	B. propinqua				+												
39	B. putumayensis				+												
40	B. racemifera				+												
41	B. rhoides	+			+												
42	B. rufa				+												
43	B. sibundoya	+	+	+	+												
44	B. standleyana				+						+						
45	B. stenoptera		+	+													
46	B. stuebelii				+												
47	B. subsesilis	+			+												
48	B. susaconensis				+												
49	B. tomentosa			+	+												
50	B. trianae				+												
51	B. trigyna			+	+	+											
52	B. velutina			+	+												
53	B. weberbaueri			+													
54	B. zamorensis			+													
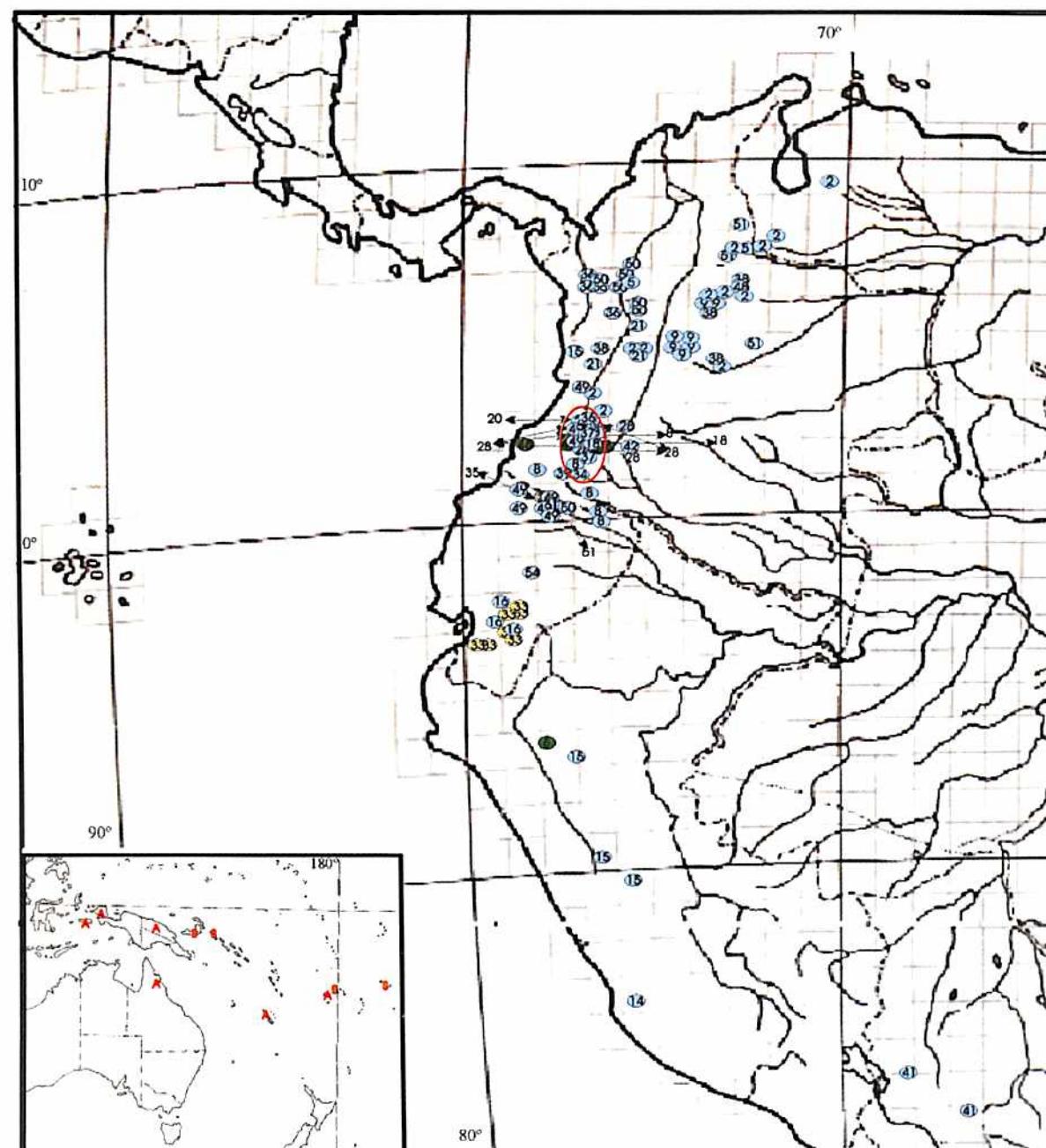
Fig. 8. 4 Distribution of species of Brunelliaceae. Sect. Simplicifoliae ●, Sect. Ovalifoliae ●, Sect. Pauciflorae ●,
Sect. Stuebelia ●. Names of species in table 8.1. Macizo Colombiano ◯. S = Spiraeanthemum A = Acsmithia (Cunoniaceae).
Two genera close related to Brunellia.

The number of species for each country is shown in Tables 8.2, 8.3. *Brunellia* has its maximum specific diversity in the Colombian Andes where 34 out of the 54 species occur, including 25 endemic species. Ecuador has 14 species including 4 endemic; Peru has 6 endemic species from a total of 11 species ; Bolivia has 2 endemic species from a total of 5 species. Number of species of *Brunellia* according to geographical distribution is presented in Fig. 8.1. With regard to Colombia, the greatest number of species is found in the Western Cordillera (16 species) and the Macizo Colombiano (located in southern Colombia before the branching point of the Colombian Andes, 13 species). Fourteen species are present in the Eastern and 9 in the Central Cordilleras (Table 8.3, Figs. 8.3, 8.4). These Cordilleras present a similar number of endemic species. Four endemic species are found in the Eastern Cordillera, three of which belong to Subsect. *Propinquae*, three endemic species in the Western and Central Cordilleras (Table 8.3). Two endemic species are present in the small area of the Macizo Colombiano.

Species of the five sections proposed in the phylogenetic study (Chapter 7) concur in a small area, from the Macizo Colombiano through Ecuador-to northern Peru (Figs. 8.5, 8.6). The largest number of species is found in the northern Andes (Colombia). For Sect. *Simplicifoliae*, the Macizo Colombiano and northern Andes, and for Sect. *Brunellia* Western of the North Central and West of Northeastern cordilleras are areas with the highest number of species. (Figs. 8.3, 8.4).

The Sects. *Brunellia* and *Simplicifoliae* have a broader distribution (Figs. 8.3, 8.4). Section *Brunellia* (26 species) is distributed from 20^0 south to 20^0 north in Central America and also in the Greater Antilles.

Table 8.2 Distribution, number of species and endemic species of *Brunellia*.

COUNTRY	Number of species	Endemic species
BOLIVIA	5	2
COLOMBIA	34	25
COSTA RICA	4	
CUBA	1	
DOMINICAN REPUBLIC	1	
ECUADOR	14	4
GUATEMALA	1	
HAITI	1	
HONDURAS	1	
JAMAICA	1	
MEXICO	1	
PANAMA	3	
PERU	10	6
PUERTO RICO	1	
EL SALVADOR	1	
VENEZUELA	4	

In the Sect. *Simplicifoliae* (24 species), most of its species are distributed in the northern Andes between 0° and 8° north (Fig. 8.4, 8.6). Few species of this section are present between 12° south and 0°. The three remaining subsections have few species and very restricted distributions. Sect. *Ovalifoliae* (1 species) distributed at the equatorial latitude, Sect. *Pauciflorae* (1 species) at 4° south and Sect. *Stuebelia* (2 species) at 5° south and 2° north (Figs. 8.4-8.6).

Table 8. 3 Species of *Brunellia* in the Colombian Cordilleras and Macizo Colombiano.

Species	Macizo Colombiano	Western Cordillera	Central Cordillera	Eastern Cordillera
B. acostae		+		
B. acutangula		+	+	+
B. amayensis			+	
B. boqueronensis			+	
B. cayambensis	+			
B. colombiana				+
B. comocladifolia	+	+	+	+
B. dulcis		+		
B. elliptica				+
B. farallonensis	+	+		
B. foreroi		+		
B. glabra	+			
B. Goudotii		+	+	
B. hygrothermica		+		
B. integrifolia	+			+
B. latifolia		+		
B. littlei	+	+		+
B. macrophylla	+			+
B. occidentalis		+	+	
B. pallida	+			
B. penderiscana		+		
B. pitayensis	+			
B. propinqua				+
B. putumayensis	+			+
B. racemifera		+		+
B. rufa				+
B. sibundoya	+	+	+	+
B. stuebelii	+			
B. subsessilis			+	
B. susaconensis				+
B. tomentosa	+	+		
B. trianae			+	
B. tingyna				+
B. velutina		+		

8.3 DISCUSSION

8.3. 1 *Origin of the current distribution of* Brunellia

It seems that the first mountains of the Andes appeared in southern South America during the Middle Cretaceous, however, fossil records from the southern Andes indicate that flora was present in the Late Cretaceous whereas northern flora elements arrived in the middle of Tertiary (TAYLOR 1995)

The presence of *Brunellia* could either date back to the uplift of the Andes in the Middle and Late Cretaceous, to before the separation of South America and Africa in the Early Cretaceous, or even to when the supercontinent, Gondwana, existed. Tracks of Gondwanan lineages could exist in the evolutionary history of *Brunellia* as southern South America was still connected with Antarctica and Australia during the Tertiary (VAN DER HAMMEN 1989). These assumptions are based on the close relationships of *Brunellia* and the Australian sister groups *Cephalothus* (Cephalothaceae) and two genera of Cunoniaceae from Australia and the Pacific Islands, *Spiraeanthemum* and *Acsmithia* (BRADFORD and BARNES 2001, OROZCO 1997). RAVEN and AXELROD (1974) consider the family Cunoniaceae to have a Gondwanan distribution. Other sources of data on the current distribution of closely related groups to Brunelliaceae, for example the distribution of *Davidsonia* in northeast Australia and *Eucryphia* in Australasia and South America, could corroborate the Gondwanan aspects of the origin of Brunelliaceae.

Brunellia is well adapted to high altitudes. Currently most species have high altitude distributions, between 2000-3500 m. However, the low altitudinal distribution of *B. hygrothermica* between 400-600 m could indicate that *Brunellia* was originally distributed at low altitudes in an early period of the formation of the Andes. This low altitudinal distribution could represent an ancient distribution for species of *Brunellia*. Despite the ramification of the Colombian Andes taking place during the Eocene (TAYLOR 1995), the adaptation to higher altitudes and consequent speciation could have taken place when the upheaval of the northern Andes was completed giving mountains over 1000 m. The high speciation of *Brunellia* in the northeast of the Andes must be related to the main uplift of the Andes, which according to KROONENBERG et al. (1990) took placed during the Mid-Miocene. However, this upheaval affected the whole Andean chain (KROONENBERG et al. 1990), high speciation of *Brunellia* was favored in the northeast of the Andes (Colombia), (Figs. 8.3-8.6). Upheaval of the Andes to over 3000-3500 m, occurred 9-12 m. y. ago in the three cordilleras of Colombia (KROONENBERG et al. 1990). This upheaval favored the distributional range of unifoliolate species of *Brunellia*, most of them in Sect. *Simplicifoliae*, and probably the distribution of the rest of the species (Fig. 8.2). The presence of *B. integrifolia* in the Santa Marta massif and the Eastern Cordillera of Colombia could indicate that this massif was also part of the Eastern

land Cordillera. Santa Marta, as with the Cocuy and Merida massifs were the result of the most recent uplift of the Andes around 4-6 m. y. ago, their formation coincides with the accretion of the Panamanian Isthmus to the continent (KROONENBERG et al. 1990).

The absence of phylogenetic studies for most of the Caribbean taxa is also a limiting factor to constructing a biogeographic hypothesis. Among the various attempts to explain Caribbean biogeography, Rosen´s studies (1975, 1985) indicate that biotic data provide a direct means to the corroboration or falsification of geological hypotheses. The current distribution of *Brunellia* in Central America and the Greater Antilles is related to the theories of Caribbean geological history of ROSEN (1985). The Caribbean hypotheses of ROSEN (1985) are based on the geological theories and cladograms of areas where each branching represents cladogenesis of different groups. It is in accordance with the available distribution pattern data of *Brunellia* in Panama and eastern Costa Rica, Honduras, Nicaragua, Mexico, and the Greater Antilles-Cuba, Jamaica, Puerto Rico, and the Dominican Republic (Figs. 8.1, 8.3, 8.4-8.6,Tables 8.1, 8.2).

Both theories, latitudinal and longitudinal displacement (SALVADOR and GREEN 1980, ANDERSON and SCHMIDT 1983) are rather similar and coincide in the sequence of land connection and disconnection but differ in the parental origin of the biota (ROSEN 1985). The two theories take into account both north and west South America, and either can explain the presence of *Brunellia* in Central America and the Greater Antilles.

According to GRAHAM (1985) climatic and physical conditions favored the interchange of tropical elements across the North Atlantic from the Cretaceous to the early Eocene but this interchange became less from the Middle Eocene to later times. The formation of the Panamanian isthmus of 3.6 m. y. ago B. P. (KEIGWIN 1978) in the Pliocene, and the upheaval of the Andes in the Miocene and Pliocene have been considered as barriers for the interchange of tropical elements. In this interchange, tectonic displacement would have been implied as well as connections forming between different geographical features and thus transforming them into other configurations (ROSEN 1985). The finding of *Brunellia* pollen grains from the Oligocene in Puerto Rico by GRAHAM and JARZEN (1969) indicates that such interchange was happening before the Eocene when it was favored by the connections of the Panama-Costa Rican Arc with the northwest of South America, and the Proto-Greater Antilles plates with northern South America. However, KROONENBERG et al. (1990) reported a very close approximation of the island arc to Colombia at the end of Miocene around 7 m. y. ago. Despite this discordance the presence of *Brunellia* in Central America and the Greater Antilles could have been due to land connections of northern South-America with proto-Central America and the Greater Antilles.

Fig. 8. 5 Geographic distribution of Sections of Brunellia. Macizo Colombiano O.

Section *Brunellia*
Section *Ovalifoliae*
Section *Pauciflorae*
Section *Simplicifoliae*
Section *Stuebelia*
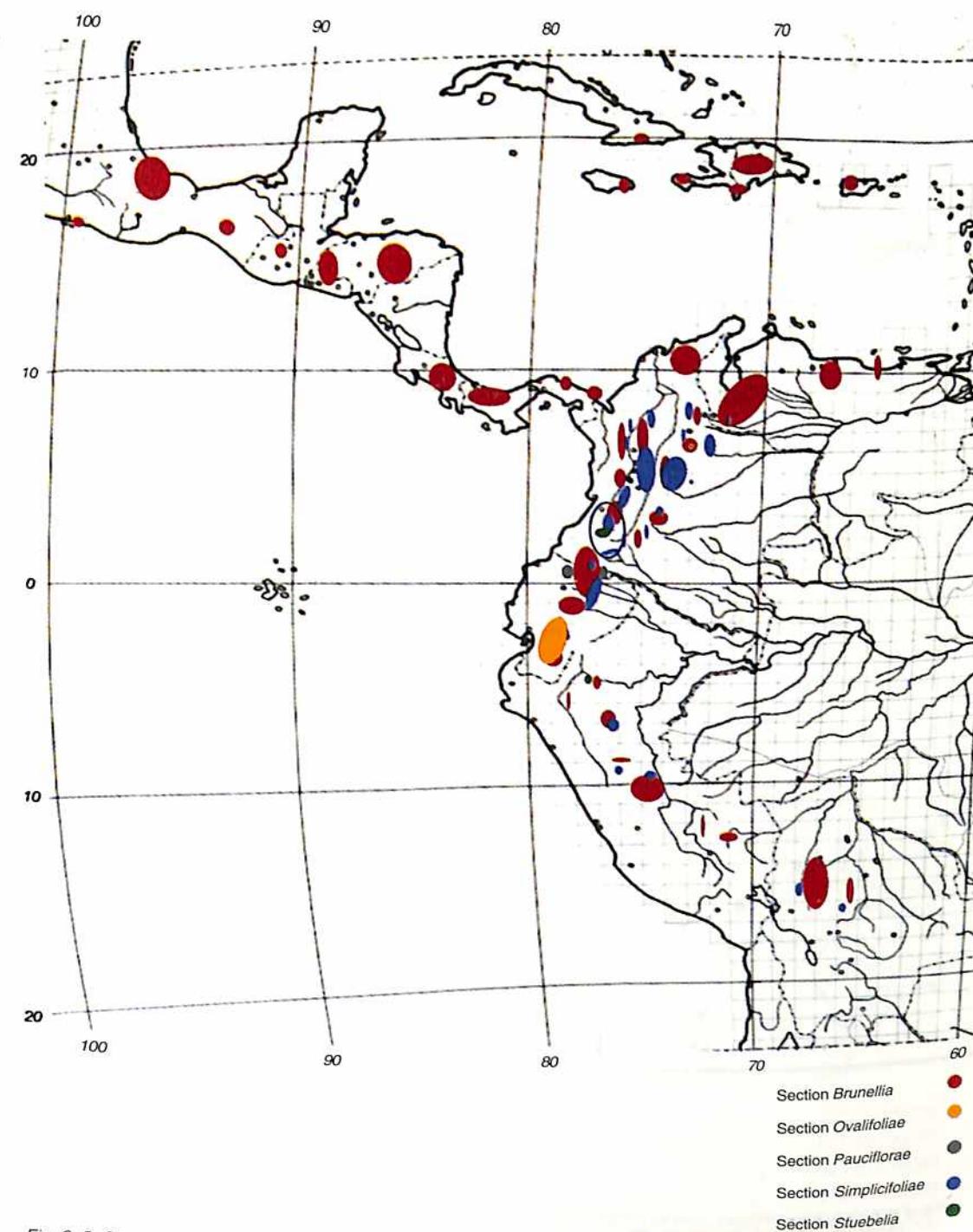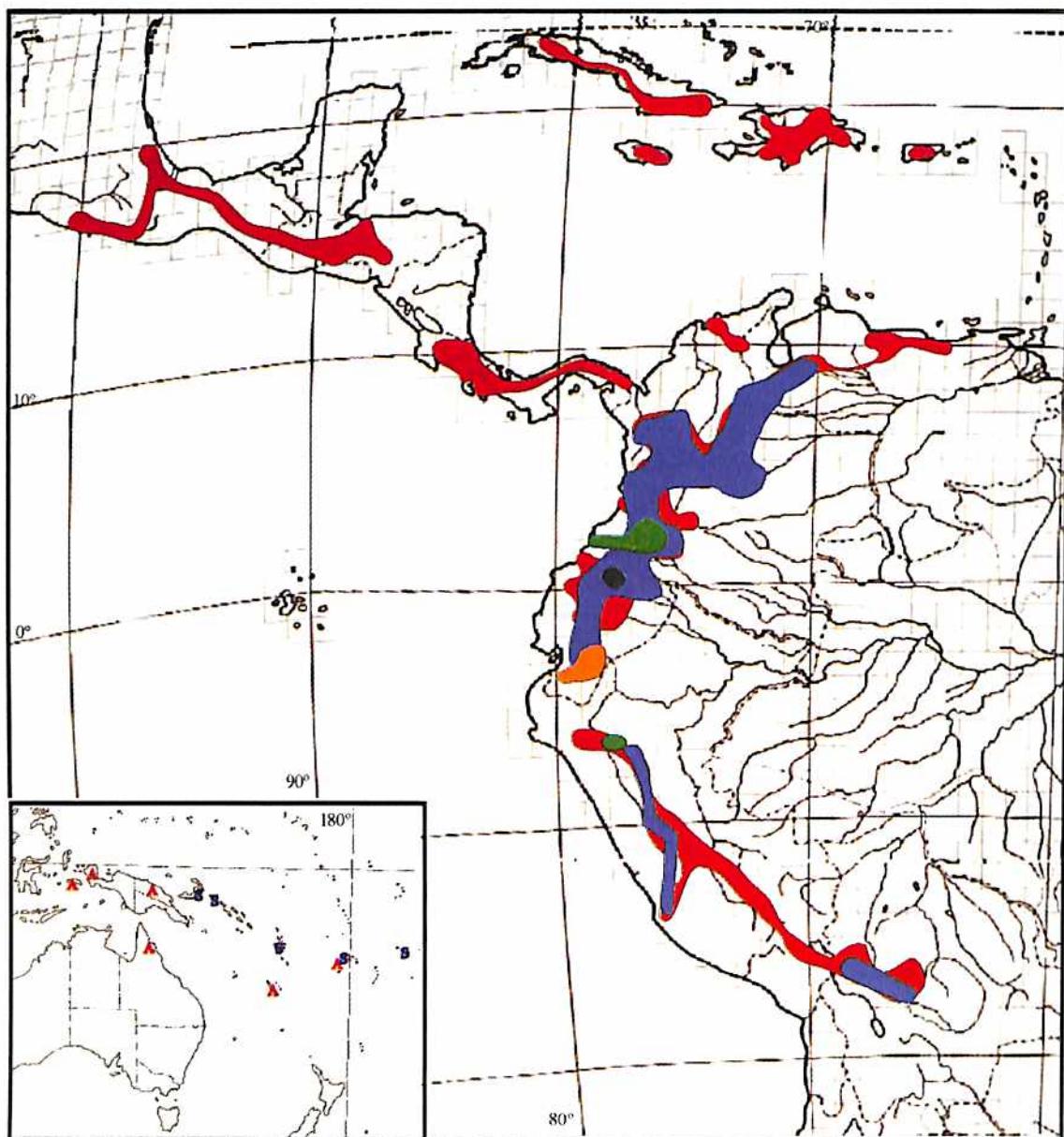
Fig. 8. 6 Approximate distribution area for Brunellia based on data from Figs 8.3 and 8.4. Sect. Brunellia ▢ , Sect. Ovalifoliae ▢ , Sect. Pauciflorae ▢ , Sect. Simplicifoliae ▢ , Sect. Stuebelia ▢ . S = *Spiraeanthemum* A = *Acsmithia* (Cunoniaceae). Two genera close related to *Brunellia*.

The aforementioned evidence indicates that is more plausible to accept that *Brunellia* came from the south-north migration of Gondwana biota mass than from North America as was considered by CUATRECASAS (1970). CROIZAT (1952) believed that *Brunellia* was present in the New World before the Andes began to rise, entered from the south, and migrated from Bolivia and Peru to the northern Andes, Central America, and the Greater Antilles. However, there is no evidence that the species first appeared in Bolivia. It is known that the current distribution of several groups does not represent the original distribution. For example, climatic changes could have reduced the primary distribution. In some groups of Cunoniaceae such as *Spiraeanthemum* and *Acsmithia,* fossils from the Eocene and Oligocene in southern and southwestern Australia occur in dryer places than where extant species are found; fossil pollen of *Gillbeea* from Late Paleocene, Eocene and Oligocene indicate the extinction of this genus in South America (BRADFORD and BARNES 2001).

8.3. 2 Diversification area or ancient area

Center of origin, or ancestral area as it is now known, has been a matter of study since the time of Linnaeus and Darwin. It was a popular method to explain current distributions until the 1970´s (BREMER 1992). Under this concept only dispersion was used to explain the patterns of geographical distribution of an individual organism. Hennig´s progression rule functions under the same concept as center of origin and has been widely used by evolutionary researchers such as, SIMPSON (1952), MAYR (1982) and also WILEY (1981). PAGE and LYDEARD (1994) consider that the strict adherence to dispersal as the unique cause of distribution was due to the geological concept of stable areas, for example, the permanence of continents and ocean basins.

As stated in a recent review by BUENO and LLORENTE (2000) center of origin was criticized by CAIN as far back as 1944. Ten criteria and their corresponding inconsistencies were indicated in their review. Thus, according to the current discussion on historical biogeography and the available information of *Brunellia*, it is most appropriate to talk about diversification area instead of ancient area. I consider diversification area as the place where different morphological lineages co-occur and where the most important evolutionary changes occurred. In this case, species of the five sections recognized by the phylogenetic analysis concur in a small area, from the Macizo Colombiano (southern Colombia before the branching point of the Colombian Andes) through Ecuador-to northern Peru (Figs. 8.5, 8 6).

From the altitudinal data on the cladogram (Fig. 8.7), and using ACCTRAN optimization (FARRIS 1970) in an early cladogenesis of *Brunellia* an altitudinal range between 2000 and 3000 m was found for the diversification area. Regarding the cladogram the highest altitude is seen at the basal cladogenesis of Sects. *Brunellia* and *Simplicifoliae*, but a reversion at the latest

cladogenesis of Sect. *Simplicifoliae* is also seen (Fig. 8.7). This result possibly can be related to the glacial and interglacial periods in which masses of vegetation extended or contracted their original distribution area.

8.3. 3 Speciation and speciation area

Speciation in *Brunellia* has been favored by the uplift of the Andes. In northern South America, (Colombia), the three Cordilleras offer different
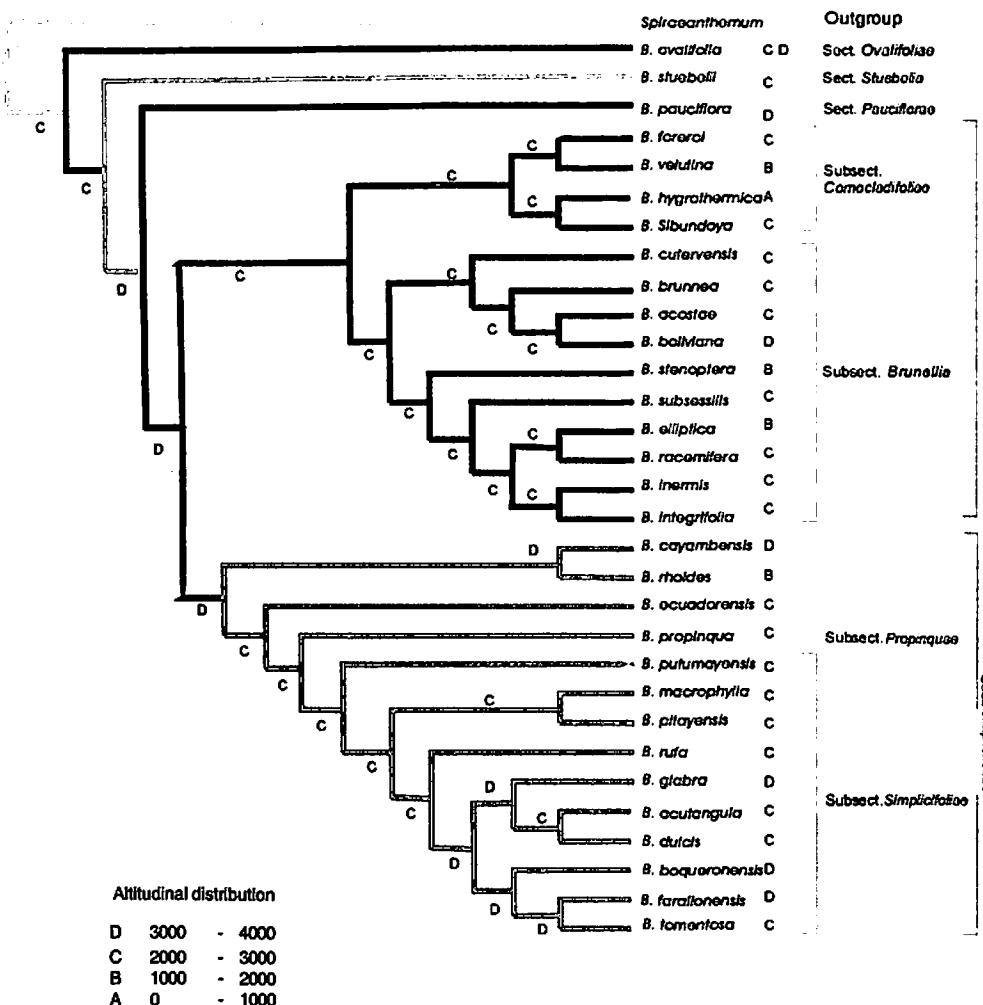
Fig. 8.7 Altitudinal distribution for *Brunellia* based in ACCTRAN optimization (Farris 1970).

climates in which *Brunellia* has adapted (Table 8.3). According to SARMIENTO (1986), temperature and humidity changes as a result of altitude influence variation and speciation in the Andes. He also considered that the climate of high tropical mountains creates different environments to those of extra-tropical temperate mountains and tropical lowlands.

Speciation area is here defined as the area with the highest number of species of any lineage. According to this statement the Macizo Colombiano, northern Andes in Colombia, Western cordillera and the northwest of Central and northwest of Eastern cordilleras are areas of high speciation for Sects. *Brunellia* and *Simplicifoliae*.

8.3. 4 Patterns of geographical distribution

The study presented here allows the identification of geographic distribution patterns (Fig. 8.6) which are correlated to a unique type of morphological diversification. These areas could be considered as natural and correspond to the distribution of natural groups (monophyletic groups) which were found by the phylogenetic study in chapter 7. Some of these areas are of very restricted distribution and are occupied by few monophyletic groups such as Section *Ovalifoliae*, Sect. *Pauciflorae* and Sect. *Stuebelia*, distributed between 4^0 north and 8^0 south (Figs. 8.4-8.6). Sect. *Ovalifoliae* is distinguished by the combination of three whorled, either unifoliate or compound leaves per node, straight or flat hairs and an urceolate endocarp (see chapter 1, Figs. 1. 1f, g, 1. 3 b). Sect. *Pauciflorae* by the autapomorphic character of the simplest inflorescences found in the genus (see chapter 4, Fig.4. 3a), Sect. *Stuebelia* is defined by the combination of two unifoliate leaves per node, woolly hairs and an urceolate endocarp shape (see chapter 1, Figs. 1.1 e, 1. 3 b). The two remaining Sections, Sects. *Brunellia* and *Simplicifoliae* have a broader distribution in Colombia and high speciation (Figs. 8.3, 8.4). Section *Brunellia* is widely distributed from 20^0 South to 20^0 North in Central America and also in the Greater Antilles. This Section is represented by 26 species which present a small U-shaped or urceolate endocarp this character is combined with compound leaves and complex inflorescence (see chapter 1, Figs.1. 2 d-f, 1.3 a, b,). Most of the species of the Sect. *Simplicifoliae* have a wide distribution in the northern Andes, between $0^0$and 8^0north (Fig. 8.4, 8.6). This section is defined by reduction of the carpel number and includes the majority of species with reduced leaves covered by an arachnoid or woolly indument and a large endocarp (navicular endocarp) (see chapter 1). Species with reduced leaves are not present in Central America and the Greater Antilles. The diversification of *Brunellia*, Sect. *Simplicifoliae* in the northern Andes is somewhat related to foliar and flower reduction and the presence of a navicular endocarp. These species have developed some anatomical leaf adaptations in response to the high altitudes where they frequently occur (Chapter 3).

8.4 ACKNOWLEGMENTS

Thanks to HENRY HOOGHIEMSTRA and PAUL J.M. MAAS from Amsterdam and Utrecht Universities, respectively. I am also thankful to FERRY BOUMAN for his comments and specially to ANTOINE CLEEF from Amsterdam University for his suggestions and his advice to present additional information about distribution of *Brunellia*. Thanks also to JAN WOLF for his comments and to SANDRA CASTRO, student of biology, who drew up the figures that accompany this chapter.

8.5 LITERATURE

ANDERSON, T. H. and SCHMIDT, V. A. 1983. The evolution of Middle America and the Gulf of Mexico-Caribbean Sea region during Mesozoic time. Bull. Geol. Soc. Amer. 94: 941-966.

BRADFORD, S. and BARNES, R. 2001. Phylogenetics and classification of Cunoniaceae (Oxalidales) using choloroplast DNA sequences and morphology. Syst. Bot. 26: 354-385.

BREMER, K. 1992. Ancestral areas : A cladistic reinterpretation of the center of origin concept. Syst. Biol. 41: 436-445.

BUENO, H. A. and LLORENTE, J. B. 2000. Una revisión histórica de la biogeografía dispersionalista con críticas a sus fundamentos. Caldasia 22: 161-184.

CAIN, S. A. 1944. Foundations of plant geography. Harper and Brothers. New York and London.

CROIZAT, L. 1952. Manual of phytogeography, or an account of plant-dispersal throughout the world. W. Junk, The Hague.

CUATRECASAS, J. 1970. Brunelliaceae. Flora Neotropica. Monograph 2: 1-189.

FARRIS, J. 1970. Methods for computing Wagner trees. Syst. Zool. 19: 83-92.

GRAHAM, A. and D. M. JARZEN. 1969. Studies in neotropical paleobotany. I. The oligocene communities of Puerto Rico. Ann. Missouri Bot. Gard. 56: 308-357.

GRAHAM, A. 1995. Development of affinities between Mexican/ Central American and Northern South American lowland and lower montane vegetation during the Tertiary. In : Biodiversity and Conservation of Neotropical Montane forests (eds. STEVEN P. CHURCHILL et al.), pp. 11-22. The New York Botanical Garden, New York.

GRUBB, J. P. 1977. Control of forest growth and distribution on wet tropical mountains. Ann. Rev. Ecol. Syst. 8: 83-108.

HOOGHIEMSTRA, H. and CLEEF, A. 1995. Pleistocene climatic change and environmental and generic dynamics in the northern Andean montane forest and páramo. In: Biodiversity and Conservation of Neotropical Montane forests (eds. S. P. CHURCHILL et al.), pp. 35-49. The New York Botanical Garden, New York.

HUMPHRIES, C. J. and PARENTI, L. R. 1986. Cladistic biogeography. Clarendon Press, Oxford.

KEIGWIN, L. D. 1978. Pliocene closing of the isthmus of Panama, based on biostratigraphic evidence from nearby Pacific Ocean and Caribbean Sea cores. Geology 6: 626-629.

KROONENBERG, S., BAKKER, J. G. M., and WIEL M. VAN DER. 1990. Late Cenozoic uplift and paleogeography of the Colombian Andes: constrains on the development of high-andean biota. Geol. Mijnbouw 69: 279-290.

MAYR, E. 1982. The growth of biological thought. Diversity, evolution and inheritance. Harvard University Press, Harvard, Massachusetts

NELSON, G. and PLATNICK, N. I. 1981. Systematics and biogeography. Cladistics and vicariance. Columbia Univ. Press, New York.

OROZCO, C. I. 1991. Análisis multivariado de un complejo de especies de *Brunellia*. Ann. Missouri Bot. Gard. 78: 105-130.

OROZCO, C. I. 1997. Sobre la posición sistemática de *Brunellia* Ruiz & Pavón. Caldasia 19: 145-164.

PAGE, R. D. M. and LYDEARD, C. 1994. Towards a cladistic biogeography of the Caribbean. Cladistics 10: 21-42.

PLATNICK, N. I. and NELSON G. 1978. A method of analysis for hystorical biogeography. Syst. Zool. 27: 1-16.

PLATNICK, N. I. and NELSON, G. 1984. Composite areas in vicariance biogeography. Syst. Zool. 33: 328-335.

RAVEN, P. and AXELROD, D. 1974. Angiosperm biogeography and past continental movements. Ann. Missouri Bot. Gard. 61: 539-673.

ROSEN, D. 1975. A vicariance model of Caribbean Biogeography. Syst. Zool. 24: 431-464.

ROSEN, D. 1985. Geological hierarchies and biogeographic congruence in the Caribbean. Ann. Missouri. Bot. Gard. 72: 636-659.

SALVADOR, A. and GREEN, A. R. 1980. Opening of the Caribbean Tethys (origin and development of the Caribbean and the Gulf of Mexico). In: Geology

des chaines Alpines Issues de la Tethys. Mem Bur. Rech. Geol. Minières 115: 224-229.

SARMIENTO, G. 1986. Ecological features of climate in high tropical mountains. In: High altitude tropical biogeography (eds. F. VULLEUMIER and M. MONASTERIO), pp. 11-45. New York, Oxford.

SIMPSON, G. G. 1952. Probabilities of dispersal in geologic time. Bull. Amer. Mus. Nat. Hist. 99: 163-176.

TAYLOR, D. W. 1995. Cretaceous to Tertiary geologic and angiosperm paleobiogeographic history of the Andes. In: Biodiversity and Conservation of Neotropical Montane forests (eds. S.P. CHURCHILL et al.), pp. 3-9. The New York Botanical Garden.

VAN DER HAMMEN, T. 1989. History of the montane forests of the northern Andes. Pl. Syst. Evol. 162: 109-114.

WILEY, E. O. 1981. Phylogenetics: the theory and practice of phylogenetic systematics. J. Wiley & Sons, New York.

Appendix. Reference collections of the distributional data. Names of species number in Table 8. 1.

SOUTHAMERICA

Collector	Collection number	Species number	Collector	Collection number	Species number	Collector	Collection number	Species number
BOLIVIA								
Bang, M.	839	41	Lewis, M.	37351	47	Solomon, J.C.	14418	4
Beck, St. G.	1832	4	Smith, D.N. & J. F. Smith	13077	43	Solomon, J.C.	16403	4
Hartshorn, G. & Meneses	2039	41	Solomon, J.C.	9317	43	Solomon, J.C.	17686	32
Krukoff's, B. A.	10361	32	Solomon, J.C.	14249	4	Steinbach, R. F.	575	4
Lewis, M.	40829	4						
COLOMBIA								
Alston, A. H. G.	7491	10	Dan Hartman	296	26	Idrobo & Fernández	272	10
Barclay	3413	40	Dan Hartman	620	10	Karsten	s.n.	10
Betancur, J. et al.	986	43	Dan Hartman	733	49	Little, E.	7253	10
Black, J. A.	46-681	9	Díaz, S.	2822	40	Little, E.	7383	10
Brant, A.E. et al.	1139	47	Devia, W.	2035	21	Little, E.	7506	27
Callejas, R. et al.	3739	43	Duque, A. A.	69	10	Lozano, G. et al.	3970	4
Callejas, R. & R. Fonnegra	7275	10	Duque-Jaramillo, J. M.	3390	2	Lozano, G.	4860	52
Cuadros, H.	1293	10	Duque-Jaramillo, J. M.	3795	10	Lozano, G.	5518	51
Cuadros, H.	N0.707	10	Echeverry, R.	3233	21	Lozano, G.	5535	51
Cuatrecasas, J.	1963	38	Espinal, C.	2613	38	Lozano, G.	5557	51
Cuatrecasas, J.	9811	38	Espinal, S.	3189	25	Luteyn, J.	7387	20
Cuatrecasas, J.	13532	9	Espinel, S.	3287	18	McPherson, G.	13145	38
Cuatrecasas, J.	13857	10	Forero, E. et al.	2639	19	McPherson, G.	13259	50
Cuatrecasas, J.	13881	10	Franco, P. & D. Rivera	2449	2	McPherson, G. et al.	13382	50
Cuatrecasas, J.	15187	10	Fuentes, M.	s.n.	43	Moldenke, H.N.	18V.C.012	10
Cuatrecasas, J.	17278	23	García-Barriga, H.	20379	10	Orozco, C.I. et al.	256	40
Cuatrecasas, J.	22218	31	Gentry, A.	1984	38	Orozco, C.I. et al.	259	43
Cuatrecasas, J.	23524	43	Gentry, A.	17064	43	Orozco, C.I. et al.	966	43
Cuatrecasas, J.	23953	52	Gentry, A.	47790	37	Orozco, C.I. et al.	996	21
Cuatrecasas, J. et al.	24264	50	Gentry, A.	55548	25	Orozco, C.I. et al.	998	9
Cuatrecasas, J.	26889	10	Gutierrez	290	2	Orozco, C.I. et al.	1003	25
Cuatrecasas, J.	27329	9	Hilty Ja.	3	52	Orozco, C.I. et al.	1010	25
Cuatrecasas, J.	27588	2	Huertas & Camargo	5617	2	Orozco, C.I. et al.	1078	9

Appendix. Reference collections of the distributional data. Names of species number in Table 8.1.

COLOMBIA

Collector	Collection number	Species number	Collector	Collection number	Species number	Collector	Collection number	Species number
Orozco, C.I. et al.	1100	40	Orozco, C.I. et al.	1595	51	Orozco, C.I. et al.	2807	27
Orozco, C.I. et al.	1159	31	Orozco, C.I. et al.	1596	25	Orozco, C.I. et al.	2850	42
Orozco, C.I. et al.	1175	49	Orozco, C.I. et al.	1679	25	Orozco, C.I. et al.	2869	27
Orozco, C.I. et al.	1176	46	Orozco, C.I. et al.	1715	25	Orozco, C.I. et al.	2901	37
Orozco, C.I. et al.	1187	8	Orozco, C.I. et al.	1745	51	Orozco, C.I. et al.	2914	2
Orozco, C.I. et al.	1201	39	Orozco, C.I. et al.	1754	9	Orozco, C.I. et al.	2970	17
Orozco, C.I. et al.	1217	49	Orozco, C.I. et al.	1753	9	Orozco, C.I. et al.	2974	51
Orozco, C.I. et al.	1219	43	Orozco, C.I. et al.	1758	4	Pennell, F.W.	7472	46
Orozco, C.I. et al.	1247	2	Orozco, C.I. et al.	1768	38	Pérez, A.	70	50
Orozco, C.I. et al.	1251	10	Orozco, C.I. et al.	1771	43	Rentería, E.	4074	43
Orozco, C.I. et al.	1256	10	Orozco, C.I. et al.	1775	9	Rentería, E.	5130	43
Orozco, C.I. et al.	1276	43	Orozco, C.I. et al.	1812	38	Rentería, E.	5231	35
Orozco, C.I. et al.	1297	47	Orozco, C.I. et al.	1854	48	Rentería, E.	7901(7)	35
Orozco, C.I. et al.	1306	50	Orozco, C.I. et al.	1956	9	Romero-Castañeda	7553	25
Orozco, C.I. et al.	1321	50	Orozco, C.I. et al.	1959	2	Sarria, S.	999	2
Orozco, C.I. et al.	1341	10	Orozco, C.I. et al.	2031	9	Silverstone, S. et al.	2756	15
Orozco, C.I. et al.	1353	50	Orozco, C.I. et al.	2032	9	Stein, B.	3513	43
Orozco, C.I. et al.	1402	40	Orozco, C.I. et al.	238	43	Stein, B.	3756	8
Orozco, C.I. et al.	1423	9	Orozco, C.I. et al.	2534	27	Stein, B.	3478	2
Orozco, C.I. et al.	1426	3	Orozco, C.I. et al.	2553	43	Torres, J.& G. Lozano	674	2
Orozco, C.I. et al.	1427	50	Orozco, C.I. et al.	2627	43	Zarucchi, J.L.	4373	47
Orozco, C.I. et al.	1458	5	Orozco, C.I. et al.	2707	27	Zarucchi, J.L.	4460	50
Orozco, C.I. et al.	1482	43	Orozco, C.I. et al.	2783	27	Zarucchi, J.L.	5151	50
Orozco, C.I. et al.	1505	36	Orozco, C.I. et al.	2785	28	Zarucchi, J.L.	5237	50
Orozco, C.I. et al.	1539	1	Orozco, C.I. et al.	2791	10	Zarucchi, J.L.	7139	1

Appendix. Reference collections of the distributional data. Names of species number in Table 8.1.

ECUADOR

Collector	Collection number	Species number	Collector	Collection number	Species number	Collector	Collection number	Species number
Acosta Solís, M.	14761	54	Freire Fierro, A.	1151	35	Rubio, D. et al.	591	49
Alvarez, A. et al.	301	24	Gentry, A. et al.	54960	45	Rubio, D. et al.	2278	43
Asplund	18698	45	Gentry, A. et al.	69966	1	Stein, B.	2617	49
Aulestia, M. & T. Pennington	2965	8	Gudiño, E. et al.	1479	45	Steyermark, J.	53576	33
Balslev, H. & W.C. Steere	3099	1	Jaramillo, J.	8858	33	Steyermark, J.	53814	53
Camp, W.H.	E-4837	33	Josse, C. et al.	392	33	Steyermark, J.	54435	45
Camp, W.H.	E-5116	33	Lehmann, F.H.	5610	16	Steyermark, J.	54903	54
Cazalet, P.C.& T. Pennington	5601	8	Little, E.	6776	16	Tipaz, G.	12	51
Cerón, C.E. & S. Villavicencio	2783	1	Lugo, S. Her.	4665	45	Tipaz, G.	23	35
Cuamacás, B. & E. Gudiño	243	10	Madison et al.	4482	10	Tipaz, G.	55	35
Dodson, C.	11846	1	Neill, D.& B. Castro	9789	1	Tipaz, G. et al.	75	51
Dodson, C. et al.	13692	10	Neill, D.	10044	45	Tipaz, G. et al.	1288	23
Dodson, C. et al.	14879	45	Ortiz, L.	231	54	V. Zak	1183	1
Dodson, C. et al.	15150	49	Palacios, W. & E. Freire	5025	43	Werff, H. van der & et al.	9198	24
Dodson, C. et al.	15230	1	Palacios, W. & v. der Werff	3700	35	Werff, H. van der et al.	10704	49
Duque-Jaramillo, J.	7824	1	Palacios, W. & v. der Werff	3705	16	Werff, H. van der & et al.	10760	10
Duque-Jaramillo, J.	9252	49	Palacios, W. & D. Rubio	5273	35	Werff, H. van der & et al.	10810	1
Duque-Jaramillo, J.	9262	49	Palacios, W. & et al.	6978	8	Werff, H. van der & et al.	10826b	45
Fosberg, F.R. & F. Prieto	22783	33	Palacios, W. & D. Rubio	7162	16	Werff, H. van der & et al.	12311	10
Foster, R.B.	10292	24	Romeroux, K.	210	1			

PERU

Collector	Collection number	Species number	Collector	Collection number	Species number	Collector	Collection number	Species number
Aronson, J. & P. Berry	565	24	Matheus	s.n.	6	Vasquez, E	137	15
Díaz, C. et al.	2625	24	Nuñez, P. & F. Motocanchi	8750	12	Vasquez, E	150	15
Foster, R. et al.	7815	43	Smith, D.N. & A. Pretel	7811	43	Weberbauer, A.	734	22
Gentry, A. et al.	35951	53	Smith, D.N.	48264	7	Weberbauer, A.	6781	53
Gentry, A. et al.	39928	43	Stein, B. & C. Todzia	2291	24	Werf, H. van der et al.	8822	45
Gentry, A. et al.	39945	24	Stein, B.	2407	45	Young, K.	3342	15
Gentry, A. et al.	44878	24	Stein, B. & C. Todzia	2419	24	Young, K. & B. León	4975	24
Gentry, A. et al.	61486	12	Vargas, C.	13667	13			

Appendix. Reference collections of the distributional data. Names of species number in Table 8. 1.

Collector	Collection number	Species number	Collector	Collection number	Species number	Collector	Collection number	Species number
VENEZUELA								
Banting, G.	3438	25	Luteyn, J.	5330	51	Steyermark, J. A	127860	25
Benitez, C. E.	1336	25	Luteyn, J. et al.	9976	25	Werff, H. van der et al.	5869	10
Berry, P.	3429	2	Steyermark, J.	100838	2	Werff, H. van der et al.	6089	2
Davidse, Gt. & A. González	19848	25	Steyermark, J.	118621	25	Werff, H. van der et al.	7526	10
			CENTRAL AMERICA					
COSTA RICA								
Brenas, A.	6767	44	Lems, K.	5016	11	Skutch, A. F.	3429	44
Burger, W. C.	5225	11	Molina, A. et al.	17836	11	Steven, R. Hill	17896	11
Davidse, G. L. et al.	25489	30	Molina, A. et al.	18078	10	Stevens, W. D.	13452	10
Haber	3395	11						
EL SALVADOR								
Allen	7143	29	Molina, A.	16718	29			
GUATEMALA								
Steyermark, J. A.	30998	29	Steyermark, J. A.	48747	29	Steyermark, J. A.	43261	29
Steyermark, J. A.	43788	29						
HONDURAS								
Allen, P. H. et al.	6125	29	Molina	12743	29	Molina, A.	8270	29
MEXICO								
Avedaño, S. & Castillo	500	29	Marquez, W. & J. Dorantes	113	29	V. Vaszquez	21001	29
Laurence, D. A. Garcia	930	29	Miller, J. S. & G. Campos	2909	29			
PANAMA								
Cuadros, H.	3955	10	Nevert, de G.	4445	23	Miller, J. S.	874	23
Knapp S. & R. Schmalzel	4760	30						

Appendix. Reference collections of the distributional data. Names of species number in Table 8. 1.

Collector	Collection number	Species number	Collector	Collection number	Species number	Collector	Collection number	Species number
GREATER ANTILLES								
CUBA								
Clemente & Alain, Bros.	6704	10	Ekman, E.	10269	10	Webster, G.	4151	10
Blücher, G.	183	10	Morton, C. V.	9345	10			
Ekman, E.	10269	10						
			DOMINICAN REPUBLIC					
Augusto, B.	1098B	10	Fuertes, M.	1601	10	R. A. & E. S. Howard	9143	10
Gentry, A. et al.	50523A	10	Mejía, M. et al.	533	10			
Howard	12328	10	R. A. & E. S. Howard	8521	10			
JAMAICA								
Proctor, G. T.	22267	10	Hart, J.	1042	10	William T. Gillis	14999	10
HAITI								
Emercy, C.	14533	10	Leonard & Leonard	14533	10	Nash, G. V.	729	10
PUERTO RICO								
Britton & Brown	5436	10	Sargent, F. H.	3153	10			

Chapter 9

GENERAL DISCUSSION AND CONCLUSIONS

Much of the biological research carried out today furthers our knowledge of the relationships between the wide diversity of species. Observable attributes, or characters, the products of the evolutionary process, are used to establish these relationships. Therefore, the study of these characters is the most important activity of those working in systematics in their search for relationships among the diversity of species. Although each pool of characters identifies species or groups of species, not all of them are informative from the point of view of evolutionary relationships.

This phylogenetic study of 32 taxa and 45 morphological characters indicates that *Brunellia* is a monophyletic group (with 54 species). These species are grouped in five sections, three of them, defined by evolutionary novelties (synapomorphies, or autapomorphies) are: *Brunellia* (26 species) widely distributed throughout the Andean region from Bolivia to Colombia and in Central America from Panama to Mexico, *Simplicifoliae* (24 species) has a narrower distribution than Sect. *Brunellia*, but with a wide diversification towards the northeast of the Colombian Andes, and *Pauciflorae* (1 species) with a restricted distribution in southern Ecuador. The remaining two sections also have a restricted distribution and are each defined by a unique combination of characters: *Stuebelia* (2 species) one species in southern Colombia and the second one in northeastern Peru, *Ovalifoliae* (1 species) in the northeastern Ecuador. Subsections and less inclusive groups were also recognized, some of them defined by synapomorphies or by a unique combination of characters. Unique combinations of characters, as was mentioned in the text (chapter 7), function as autapomorphies even though each contributing character is itself

homoplasious. The hypothesis selected from the three MPTs obtained contains homoplasious characters arising no more than twice. Thus the information contained is less conflictive but at the same time has less power of explanation about the real evolution of the characters. Although BRADFORD and BARNES (2001) criticizes the use of morphological characters in establishing the systematic position of *Brunellia* (due to the absence of information for many clades), in this case the kind of morphological data used is informative for defined groups of species and species as well. However, additional study is needed for the groups, *B. cayambensis, B. rhoides, B. brunnea,* and B. *cutervensis* due to their varying positions in the three MPTs. The appropriate outgroup has been also criticized but as BRADFORD and BARNES (2001) stated Brunelliaceae is closely related to Cephalothaceae, Cunoniaceae, and Elaeocarpaceae (Oxalidales), thus the outgroup(s) could be selected from these taxa or any other taxon. The outgroup selection of this study was limited by the type of characters studied for *Brunellia* and not all the possible outgroups have been studied in depth with respect to the type of characters used in this research. After an unsuccessful attempt to obtain a phylogenetic hypothesis for *Brunellia* in 1993 (OROZCO unpublished) owing to the lack of possible informative morphological characters, these are the first results about relationships within the genus.

Of all the characters used in the phylogenetic study of *Brunellia*, most research was carried out on what were considered as systems of characters, such as the leaf anatomy, typology of synflorescences, and floral and pollen morphology. Furthermore, some of these characters, such as floral morphology, had not been adequately defined in previous studies, and consequently, information resulting from their use was very contradictory, making this in depth intra-population study very important. The most outstanding result of this study was the finding of an asymmetric flower, the protandrous condition, and the apparent polyandry of some species, although the diplostemonous floral organ arrangement is the basic condition in *Brunellia*. The flower asymmetry is due to changes in sepal number influencing the apparent polyandry and possibly the carpel numbers as well. This apparent polyandry, due to extra-whorls of stamens resulting from sepal number variation within the species, was only found in a few species, for example, in the *boliviana* group. Apparent polyandry is less perceptible in the remaining species although it is seen in some individual flowers despite a clear underlying diplostemonous floral organ arrangement. The floral reduction and increase in the number of floral parts define the sect. *Simplicifoliae* and the B. *boliviana* group.

Anatomical characters are useful for studying relationships above the species level. In *Brunellia,* the anatomical results contribute to establishing inter-family relationships as well as defining groups of species within the genus. It was made clear in this study that unifoliolate species had been wrongly considered as simple-leafed species in earlier studies. Nodal anatomy is a very

important character for determining family relationships but further research is necessary to establish whether or not *Brunellia* has unilacunar or multilacunar nodes. Some of the anatomical leaf characters are evolutionary novelties at subsectional or infra-subsectional level. Among these characters, the presence of a hypodermis, the number of layers of palisade parenchyma, and some characteristics of the main vascular bundle of the central vein of the leaf are specially important in the unifoliolate species group, for example, Subsect. *Simplicifoliae* and Sect. *Pauciflorae.* The presence of medullary bundles is also important at inter-family level as this character was only observed in B. *foreroi* and it is also present in some genera of Cunoniaceae, however, the homology of this character in the two families is doubtful. The presence of crypts in the abaxial foliar lamina of some species is a response to high altitude distribution, being the case in most species of the sect. *Simplicifoliae.* It seems that the presence of crypts is related to extreme environmental conditions as they have also reported also for some groups living in dry environments. Actinocytic stomata were present in most of the species examined but anomocytic stomata were only observed in *B. ecuadorensis.* As a result of a study of the indument that covers the foliar lamina of species of *Brunellia,* OROZCO (1999) found it to be of informative value for defining groups and in some cases it correlates with other unique character combinations.

A comparative study of inflorescences was used as another system of characters. This study was carried out according to TROLL and WEBERLING's typologic concept of synflorescence, making this information available for studying relationships. The position of the inflorescence in the vegetative system and its structure are informative characters for familial, intergeneric or even intrageneric relationships. A monotelic proliferating synflorescence was observed for the first time in *Brunellia* and in *Spiraeanthemum* and *Acsmithia,* two closely related genera of Cunoniaceae. This character is an evolutionary novelty shared by these taxa. Other inflorescence characters are also informative at sectional or subsectional level. The simpler monotelic proliferate synflorescence of triadic floriferous paracladia, representing an autapomorphy, is present in the Sect. *Pauciflorae.* For sections *Brunellia* and *Simplicifoliae* the degree of complexity of the floriferous paracladia is also informative. The position in the synflorescence of a short paracladium is also an evolutionary novelty in the Subsect. *Propinquae.* Congested and proliferate floriferous paracladia are also important in infra-subsectional groups such as in *B. congestiflora.*

Pollen grain analysis provides greater knowledge of the exine sculpture variation than had previous studies. Interest in this character had been expressed by CUATRECASAS since 1970 with MARTICORENA's work. Five categories of exine ornamentation were found in this study which had not previously been observed at all. From the point of view of establishing relationships the exine ornamentation is not much informative in the whole cladogram. The

reason being that the exine ornamentation is variable at inter and intraspecific level. All species in the unifoliolate group having three leaflets per node share a modified reticulate ornamentation of the exine. Punctate exine is a plesiomorphic character while modified reticulate, striate reticulate and rugulate exine sculptures are considered apomorphies for *Brunellia*. Although the latter two exine sculptures are apomorphies, these have a homoplasic behavior and are usually present in Sect. *Brunellia*. I consider TEM analysis as the most appropriate method for studying family relationships and it is expected to be informative at sectional level in *Brunellia* as well.

Current molecular research in Cunoniaceae (BRADFORD and BARNES 2001) shows that *Brunellia* (Brunelliaceae) is not a taxon of Cunoniaceae and must be considered as a separate family. This opinion is different to that of HUFFORD and DICKISON (1992) who include *Brunellia* as a genus of Cunoniaceae. It is also different to that of OROZCO (1997, chapter 2), who proposed that *Brunellia* and *Spiraeanthemum* and *Acsmithia* of Cunoniaceae, to be placed in a family different from Cunoniaceae. As BRADFORD and BARNES (2001) found through molecular analysis, *Spiraeanthemum* and *Acsmithia* form a sister group apart from the rest of the taxa (of Cunoniaceae), OROZCO (Chapter 2) observed the same group but including *Brunellia*. OROZCO also considers that there is a lack of monophyly in Cunoniaceae due to the inclusion of *Eucryphya* and *Davidsonia*, each one previously placed in different families and also by the inclusion of *Spiraeanthemum* and *Acsmithia*. As a result of this research *Davidsonia* is also included for the first time in Cunoniaceae and the relationship of *Connarus* (Connaraceae) with Cunoniaceae and Brunelliaceae is made clear. These families are now included in the order Oxalidales (AGP 1998).

The relationship of Brunelliaceae with the Australian family Cephalothaceae and genera of Cunoniaceae from Australia and west Pacific Islands, including *Davidsonia* and *Eucryphia*, and also with South American genera of Cunoniaceae, indicates that at some time they shared a common evolutionary history. Southern South American land connections during the Tertiary with Australia and Antarctica support the supposed relationships of Brunelliaceae with the Australian groups (Cephalothaceae and some genera of Cunoniaceae) It is assumed that *Brunellia* has been present in the Andes at low altitudes since the Late Cretaceous. The high diversification of *Brunellia* is related to the ramification of the Colombian Andes during the Eocene and to their adaptation to high altitudes during the upheaval of the Andes which was completed in the Pliocene. The most important upheaval of the Andes over 3000m, 9-12 m.y., influenced the distribution range of species of *Brunellia*. Thirty four species are present in Colombia, 14 in Ecuador, 5 in Bolivia, 9 in Peru, 4 in Costa Rica and Venezuela, and 3 in Panama. Two species are present in other regions, *B. mexicana* in Mexico, El Salvador and Guatemala and *B. comocladifolia* in Cuba, Haiti, Jamaica, Puerto Rico, and the Dominican Republic.

Brunellia is widely distributed in lower and upper montane rain forest, usually between 1500 and 3400 m above sea level. Only one species, *B. hygrothermica*, is frequently found at altitudes lower than 1000 m, except for one collection of *B. comocladifolia* that was recorded at less than 500 m. The distribution of *B. hygrothermica* ties in with the possible scenario of the ancient distribution of *Brunellia* at lower altitudes before the upheaval of the Andes. With the exception of two collections of *B. ecuadorensis* and *B. tomentosa* (both species from sect. *Simplicifoliae*), *Brunellia* is not found higher than 3500 m. Species of the Sect. *Brunellia* are very often distributed between 1000 and 3000 m, while the most frequent altitudinal range for sections *Ovalifoliae, Pauciflorae, Simplicifoliae,* and *Stuebelia* is between 2000 and 3400 m.

In an early cladogenesis of *Brunellia* an altitudinal range between 2000-3000 m is observed. Species of Sect. *Simplicifoliae* reach the highest altitudes. In this section, at latest cladogenesis level, altitudinal reversals were observed in the cladogram and these reversals are attributed to glacial and interglacial periods. It could mean that the most recently speciation process is found in *Simplicifoliae*.

There is little fossil evidence regarding which groups in *Brunellia* are primitive or derived. To establish this, it is necessary to find historical relationships between geographic areas, in the same way that phylogenetic study provides this information among the species. Biogeographical hypotheses about ancestral areas and vicariance events associated with speciation are necessary to make any assumption on primitive taxa and areas. Any attempt therefore implies assumption that speciation events of the relationship patterns can only be explained by dispersal biogeography. However, under dispersal biogeography any explanation can be possible, and by consequence, the scenario can neither be corroborated or refuted. As is mentioned in the text, it is necessary to know the phylogenies of other groups with a similar distribution in the Andes to obtain a working historical biogeographic hypothesis that can be corroborated or refuted.

Brunellia is considered to have been present in the Andes since the Middle and Late Cretaceous. This consideration is based on the scenario resulted by the current distribution of *Brunellia* and the geological formation of the Andes. The Gondwanan origin of *Brunellia* is also assumed based on the ancient geomorphology of the continents, and its relationships with groups of Australian and West Pacific distribution such as Cephalothaceae, *Spiraeanthemum* and *Acsmithia* (Cunoniaceae). A diversification area instead of center of origin and speciation area were defined for *Brunellia*. Diversification area is an area in which species of different morphological linaeges co-occur. This area is localized between 3 °North and 7 °South (from the Macizo Colombiano (in southern Colombia) to northern Peru). In this area species of the five sections recognized in *Brunellia* are present. Speciation area is here defined by

the presence of highest number of species of any morphological lineage. Colombia is the country with widest speciation. For Sect. *Brunellia* Western, West of the Central and East of Eastern Cordilleras is the area of most speciation, whereas the Macizo Colombiano and northern Central and Eastern Cordilleras is the area of highest speciation for Sect. *Simplicifoliae.* According to the cladogram, Sects. *Simplicifoliae* and *Brunellia* have a later cladogenesis compared with the early cladogenesis of Sects. *Ovalifoliae, Pauciflorae,* and *Stuebelia.*

PUBLICACIONES DE LA
ACADEMIA COLOMBIANA DE CIENCIAS EXACTAS, FÍSICAS Y NATURALES

COLECCIÓN JORGE ÁLVAREZ LLERAS

Volumen 1 - Mora-Osejo, L.E. 1987. Estudios morfológicos, autoecológicos y sistemáticos en Angiospermas. 1/16. 196 pp, 75 figs.

Volumen 2 - Murillo, M.T. & M.A. Harker. 1990. Helechos y plantas afines de Colombia. 1/16. 326 pp, 145 figs.

Volumen 3 - Lozano-Contreras, G. 1994. Las Magnoliaceae del Neotrópico 1/16, 148 pp, 46 figs.

Volumen 4 - Eslava Ramírez, J.A. 1994. Aspectos relacionados con la erupción del volcán Nevado del Ruiz. 1/16, 174 pp, 46 figs.

Volumen 5 - Rocha de Campos, M. 1994. Diversidad en Colombia de los cangrejos del género *Neostrengeria.* 1/16 iv + 144 pp, 47 figs.

Volumen 6 - Mora-Osejo, L. E. & H. Sturm. 1994. Estudios ecológicos del páramo y del bosque altoandino, cordillera Oriental de Colombia. Tomos 1 y 2, 1/16, 716 pp, 190 figs.

Volumen 7 - Díaz, J.M., J. Garzón-Ferreira & S. Zea. 1995. Los arrecifes coralinos de la isla de San Andrés, Colombia, estado actual y perspectivas para su conservación. 1/16, 152 pp, 15 figs, 14 tablas, 9 fotografías.

Volumen 8 - Eslava Ramírez, J.A. 1995. Régimen de la presión atmosférica en Colombia. 1/16, 152 pp, 94 figs, 59 tablas.

Volumen 9 - Donato, J.Ch., L. E. González & C. L. Rodríguez. 1996. Ecología de dos sistemas acuáticos de páramo. 1/16, 168 pp, 53 figs, 14 tablas, 9 fotografías.

Volumen 10 - Andrade-C. M., G. Amat & F. Fernández. (eds.) 1996. Insectos de Colombia - Estudios escogidos. 1/16, 544 pp, 145 figs, tablas y mapas.

Volumen 11 - González, F. 1998. Inventario preliminar de gases de efecto invernadero en Colombia, 1990, Fuentes y sumideros. 1/16, xiv + 174 pp, 108 figs, tablas y gráficas.

Volumen 12 - Díaz-Piedrahita, S. & J. Cuatrecasas. 1999. Asteráceas de la flora de Colombia, Senecioneae I, géneros *Dendrophorbium y Pentacalia.* 1/16, 392 pp, 110 figs, 24 mapas.

Volumen 13 - Amat, G., M. C. Andrade, M. G. & F. Fernández (eds.) 1999. Insectos de Colombia. 1/16, 438 pp. + tablas, figuras y mapas.

Volumen 14 - Rodríguez, H. & F. González. Opciones para la reducción de emisiones de gases de efecto invernadero (GEI) en Colombia. 1/16 (262 pp.)

Volumen 15 - Rodríguez, H. & González, F. 2000. Portafolio colombiano de proyectos para el MDL –sector en energía–. 1/16 (164 pp.)

Volumen 16 - Espinosa, Baquero A. 2000. Erupciones históricas de los volcanes colombianos, 1/16. (292 pp.)

Volumen 17- Mora Osejo, L. E. 2001. Contribuciones al estudio comparativo de la conductancia y de la transpiración foliar de especies de plantas del páramo. 1/16. xii + 282.

Volumen 18- Roldán, G., J. A. Posada & J. C. Gutiérrez. 2001. Estudio Limnológico de los Recursos Hídricos del Parque de Piedras Blancas. 1/16. x + 146.

Volumen 19- Donato-Rondón, J. Ch.. 2001. Fitoplancton de los Lagos Andinos del Norte de Sudamérica (Colombia). 1/16, xii+222.

Volumen 20- Aristizábal García, H. 2002. Los hemípteros de la película superficial del agua en Colombia. 1/16. xv+242, 331 figs.

Volumen 21- Pinto, M. 2002. Estudio sistémico del género *Cavia.* 1/16. 216 pp. 38 figs. 31 láminas, 59 tablas.

COLECCIÓN ENRIQUE PÉREZ ARBELÁEZ

Volumen 1 - Memorias del seminario en conmemoración del centenario de Erwin Schrödinger. 1987, 1/16, 221 pp.

Volumen 2 - Díaz-Piedrahita, S. & A. Lourteig. 1989. Génesis de una flora. 1/16, vii. + 362 pp, 35 figs.

Volumen 3 - Cubillos, G., F.M. Poveda & J.L. Villaveces. 1989. Historia epistemológica de la Química. 1/16, 128 pp.

Volumen 4 - Hernández de Alba, G. & A. Espinosa. 1991. Tratados de minería y estudios geológicos de la época colonial, 1616-1803, 1/16 xii + 92 pp, 1 fig.

Volumen 5 - Díaz-Piedrahita, S. (ed.) 1991. José Triana, su vida, su obra y su época. 1/16, viii + 188 pp, 73 figs.

Volumen 6 - Díaz-Piedrahita, S. 1991. La Botánica en Colombia, hechos notables en su desarrollo. 1/16, x + 126 pp. 30 figs, (reimpreso 1997).

Volumen 7 - Mantilla, L.C. & S. Díaz-Piedrahita. 1992. Fray Diego García, su vida y su obra científica en la Expedición Botánica. 1/16, xv + 284, 14 figs. (reimpreso 1995).

Volumen 8 - Arias de Greiff, J. 1993. Historia de la Astronomía en Colombia. 1/16, 200 pp, 32 figs.

Volumen 9 - Lértora Mendoza, C. 1995. Fuentes para el estudio de las ciencias exactas en Colombia. 1/16, 316 pp.

Volumen 10 - Gauss, C. F. 1995. Disquisitiones Arithmeticae. Traductores: H. Barrantes, M. Josephy & A. Ruiz Zúñiga. 1/16, 540 pp.

Volumen 11 - Murillo Quinche, L.M. 1997. Obra selecta. 1/16, x + 188 pp, 29 figs, 4 mapas.

Volumen 12 - Romero Beltrán, A. 1997. La investigación de las dolencias infecciosas en la historia. 1/16, xiv + 540 pp.

Volumen 13 - Barrantes, H. & A. Ruiz. 1998. La historia del Comité Interamericano de Educación Matemática. 1/16, x + 198 pp. (español e inglés). Edición electrónica paralela.

Volumen 14 - Díaz-Piedrahita, S. 2000. Matís y los dos Mutis, 1/16, x 346 pp, 41 figs.

COLECCIÓN JULIO CARRIZOSA VALENZUELA

Volumen 1 - Castillo, G. 1992. Física Cuántica, teoría y aplicaciones. Tomo I, 1/16, xxxii + 410, 77 figs.

Volumen 2 - Bernal de Ramírez, I. 1993. Análisis de alimentos. 1/16, xviii + 314 pp, 28 figs. (reimpreso 1998).

Volumen 3 - Castillo, G. 1994. Física Cuántica, teoría y aplicaciones. Tomo II. 1/16, xx + 406 pp., 49 figs.

Volumen 4 - Cáceres, D. (ed.). 1995. Creando ciencia crean docencia. 1/16, 140 pp, 38 figs.

Volumen 5 - Romero, C.M. & L.H. Blanco. 1996. Tópicos de Química Básica. 1/16, 240 pp, 56 figs.

Volumen 6 - Peña, G. & M. Pinto. 1996. Mamíferos más comunes en sitios precerámicos de la sabana de Bogotá. 1/16, 98 pp, 1 mapa, 6 dibujos a color, 18 láminas y 48 gráficos.

Volumen 7 - López, C. 1998. Mecánica Newtoniana. 1/16, 280 pp, 69 figs.

Volumen 8 - Charris, C. J., De Castro K. & J. Varela B. 2000. Fundamentos del análisis complejo de una variable. 1/16, 384 pp.

Volumen 9 - Varela, J. de D. 2000. Principios de Cristalografía. 1/16, xx + 250 pp., 141 figs.

Volumen 10 - Kairuz de Civetta, A. 2002. Introducción al estudio de la composición de los alimentos. 1/16. xx + 166 pp. 16 figs., 30 tablas.

COLECCIÓN MEMORIAS

Volumen 1 - (1994) Memorias del Seminario Nacional "El quehacer teórico y las perspectivas holista y reduccionista. 1/16, viii + 184 pp.

Volumen 2 - (1994) Memorias del Seminario Konrad Lorenz sobre Etología. 1/16, iv + 38 pp.

Volumen 3 - (1995) Memorias del Seminario-taller sobre Alta Montaña Colombiana. 1/16, 116 pp.

Volumen 4 - (1996) Memorias del Primer Congreso Nacional de Neurocomputación. 1/16, xiv + 184 pp.

Volumen 5 - (1996) Memorias del Primer Congreso Nacional de sobre Cambio Climático. 1/16, 138 pp, 9 figs.

Volumen 6 - (1996) Memorias del Coloquio "Ciencia, tecnología y cultura". 1/16, xii + 198 pp.

Volumen 7 - (1996) Memorias de la III Escuela de verano en Geometría Diferencial, ecuaciones diferenciales parciales y análisis numérico. 1/16, xiv + 174 pp.

Volumen 8 - (1996) Memorias del Seminario de Ciencia Teórica. 1/16, xv + 170 pp.

Volumen 9 - (1997) Memorias del Seminario en conmemoración de los 440 años del nacimiento de René Descartes. 1/16, xii + 174 pp.

Volumen 10 - (2001) Memorias del Simposio Pueblos y Ambientes del pasado precolombino 1/16, viii + 312 pp.

Volumen 11 - (2001) Reflexiones sobre la ciencia y la tecnología: Colombia al iniciarse el siglo XXI, 1/16, x+166 pp.

EDICIONES ESPECIALES

- Francisco José de Caldas, su vida, su personalidad, su obra y el descubrimiento de la hipsometría. 1958. 1/16 refilado. 94 pp.

- Alexander von Humboldt en Colombia. Extractos de sus diarios. 1982. Arias de Greiff, J. (ed.). 1/8 refilado. 286 pp., 20 figs.

- Status and problems of Science in Latin America and the Caribbeean. 1990. Guardiola, M.L., J.L. Villaveces & G. Violini (eds.). 1/16, 542 pp.

- Science in Latin America and the Caribbean ad its role in regional development. 1992. Hamende, A., M.H. Hassan, J.L. Villaveces & G. Violini (eds.) 1/16, 2 vols. 824 pp.

- El cambio global, ciencia para entender el mañana. 1998. Lozano, J. (ed.), carta, 322 pp. Figs., mapas y gráficos.

- Tradición Académica. Diccionario Biográfico y Bibliográfico de la Academia Colombiana de Ciencias Exactas, Físicas y Naturales, 1/16, x + 434 pp.

- Díaz Piedrahita, S. & L.C. Mantilla R. 2001. La terapéutica en el Nuevo Reino de Granada. Un recetario franciscano del siglo XVIII. Publicación especial No. 7 208 pp. 502 figs.

Las anteriores publicaciones pueden ser solicitadas directamente a la Academia, Apartado 44763, Bogotá, D.C., Fax (571) 2443186 Tel. (571) 3414805 Trans. 27 No. 39A - 63/67 – E.Mail: accefyn@org.co Se ofrecen en venta o mediante intercambio por publicaciones similares. Mayor información a través de Internet: URL: http://www.accefyn.org.co