

# Mexican species of the genus *Stevia* (Eupatorieae, Asteraceae): Chromosome numbers and geographical distribution

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## Abstract

Chromosome numbers from 244 accessions representing 73 species, five varieties and two putative species hybrids in Mexican *Stevia* are reported with comments on agamospermy, cytogeography and chromosomal evolution. The chromosome numbers of 47 species and two varieties were counted for the first time. Counts of 12 species and two varieties were new cytotypes, or differed from all previously reported numbers for the same species. B chromosomes were found in 16 species and two varieties. All species of shrubs or shrublets were sexual diploids ( $2n=24$ ). Perennial herbs were diploids or polyploids based on  $x=11$  or 12 and the widespread prevalence of agamospermous polyploids was revealed. Chromosome numbers of annual herbs varied from  $2n=24$  to  $2n=8$ . The cytogeographical survey revealed five distributional patterns of agamic complexes based on the relative abundance of polyploids compared to their diploid ancestors. The wider distributional range (extending to northern and inland areas) of some agamospermous polyploids have been achieved by recent colonization accompanying climatic warming after the last glaciation. This has occurred even in lower latitudes such as Mexico, due to more successful founder population reproduction and the acceleration of seed dispersal, as well as the creation of new and open habitats by human activities.

*Keywords:* agamospermy, cytogeography, Mexico, polyploidy, *Stevia*.

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## Introduction

*Stevia* is one of the most distinctive genera of the Eupatorieae. All species of *Stevia* have flower heads with five-tube florets and dense pubescence on the inner surface of the corolla throat, and distinctive obovate to elliptical anther appendages with a crenulate distal margin. *Stevia* is a large genus containing approximately 230 species and is widely distributed from the southwestern USA southward through Mexico and Central America, usually in montane regions from 1000 to 3000 m in elevation. The genus also occurs in non-Amazonian South America southward to central Argentina.

Approximately 100 species of *Stevia*, from shrub to annual herb, are native to Mexico (Grashoff 1972, 1974;

McVaugh 1984; King & Robinson 1987) and occur in oak–pine and fir–alder forests, which are essentially distributed along montane chains with a cool and semihumid climate. Turner (1997) listed 85 *Stevia* species in *The Comps of Mexico*, Vol. 1, and Soejima *et al.* (2001a) described 13 new species and elevated two varieties to species. Therefore, Mexico seems to be one of the centers of diversification within this genus. In the majority of herbaceous *Stevia* examined, agamospermous reproduction is prevalent. The mode of agamospermy is obligate diplospory by means of pseudohomeotypic division (Grashoff 1972). The morphological resemblance between species and morphological and cytological variation associated with agamospermy make species recognition in this genus very difficult and the distributional range of each species remains very ambiguous. Grashoff (1972) estimated the distributional ranges of sexual and agamospermous plants of 2380 herbarium specimens for 49

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species, based on morphology and size of pollen. Pollen morphology is a fairly reliable indicator of apomixis but 'sexual' plants with good pollen grains are not always diploids because incipient agamosperous polyploids often produce as many good pollen grains as diploids, as described by Watanabe *et al.* 1982. In addition, the diad formation and subsequent good pollen production have been observed frequently in polyploid *Stevia*. Thus, chromosome counts are needed to elucidate the taxonomy (based on character comparisons between diploids in the complex) and the distribution of Mexican species of *Stevia*.

Chromosome counts provide useful information to plant taxonomists for treatment of the agamic complex, and to evolutionary biologists interested in the origin and spread of apomixis, relating agamosperous forms to sexual diploids (Babcock & Stebbins 1938; Grashoff 1972). Employing this approach, many genera including agamic complexes have been studied, such as *Crepis* (Babcock & Stebbins 1938), *Taraxacum* (Valentine & Richards 1967; Morita 1980; den Nijs & Sterk 1980), *Eupatorium* (Sullivan 1976; Watanabe 1986; Kawahara *et al.* 1989; Watanabe *et al.* 1998) and *Ixeris* (Pak *et al.* 1997).

The present paper reports the chromosome numbers and geographic distributions of Mexican species of the genus *Stevia*, and discusses the circumscription of species and the origin and spread of apomixis in this genus.

## Materials and methods

Materials for the present study consisted of rootstocks collected during field trips to Mexico in 1992–1998 and grown in the greenhouse at Kobe University. Cytological observations were made mainly on mitotic cells of root tips. For the cultivation of plants and the preparation of materials for cytological work, the procedures of Watanabe *et al.* (1990, 1995) were followed. Voucher specimens have been deposited in MEXU, KYO and TEX.

## Results and discussion

### Chromosome number

All chromosome number determinations are presented in Appendix I. Figure 1 consists of photographs of somatic chromosomes of eight representative species. Chromosome number determinations from 244 accessions attributed to 73 species, five varieties and two putative hybrids of Mexican *Stevia* were reported. First reports were given for 47 species and two varieties (Appendix I). Counts of 14 species and three varieties, *Stevia aschenborniana* ( $2n=24+0-2B$ ), *Stevia caracasana* ( $2n=22+1-3B$  and  $33+0-2B$ ), *Stevia connata* ( $2n=22+0-2B$  and 33), *Stevia elatior* ( $2n=44$ ), *Stevia iltisiana* ( $2n=33$  and 44), *Stevia isomeca* ( $2n=33$

$+0-3B$  and  $44+1-4B$ ), *Stevia jorullensis* ( $2n=24, 36$  and 48), *Stevia latifolia* ( $2n=33, 44$  and 55), *Stevia micrantha* ( $2n=24$ ), *Stevia monardifolia* ( $2n=22+0-3B$ ), *Stevia pilosa* ( $2n=33$ ), *Stevia plummerae* var. *plummerae* ( $2n=22+1-2B$  and 44), *Stevia porphyrea* ( $2n=22, 33, 44$  and 55), *Stevia seemanii* ( $2n=33+0-1B, 44$  and  $55+2B$ ), *Stevia serrata* var. *arguta* ( $2n=22$  and 33), *Stevia serrata* var. *serrata* ( $2n=44, 55$  and 66), and *Stevia tephra* ( $2n=33+0-3B$ ) were new cytotypes or differed from all previously reported numbers for the same species. Remaining counts confirmed chromosome numbers reported previously in some species or, in other species, were consistent with one of a variety of numbers reported by several authors (Appendix II).

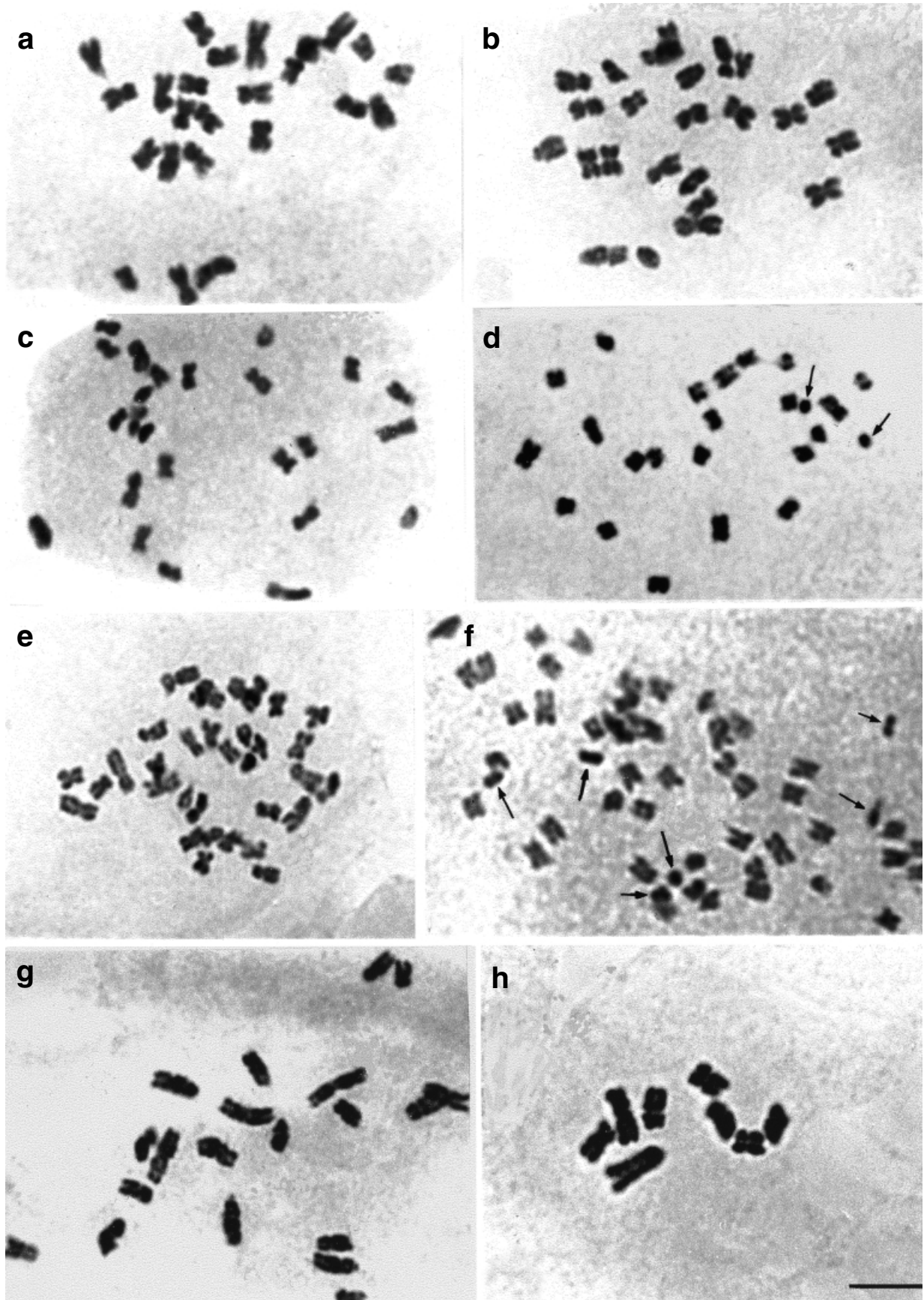
The presence of B chromosome(s) was reported for the first time in *Stevia ambryolepis*, *Stevia aschenborniana*, *Stevia caracasana*, *Stevia connata*, *Stevia constricta*, *Stevia deltoidea* (Fig. 1f), *Stevia hypomalaca* (Fig. 1d), *Stevia incongnita*, *Stevia isomeca*, *Stevia lehmannii*, *Stevia monardifolia*, *Stevia pilosa*, *Stevia plummerae* var. *plummerae*, *Stevia rotundifolia*, *Stevia seemanii*, *Stevia serrata* var. *serrata*, *Stevia stolonifera*, *Stevia tephra* and *Stevia tomentosa*.

The genus *Stevia* contains four basic habit types: (i) woody shrubs; (ii) plants with woody caudex and herbaceous stems; (iii) rhizomatous perennials; and (iv) annuals. These four types can be easily distinguished in the field and usually in the herbarium as well (Grashoff 1972). Rhizomatous perennials and plants with woody caudex and herbaceous stems were treated as 'perennial' in the present paper. These habit types corresponded well with the following chromosome numbers and modes of reproduction.

All eight species and two varieties of shrub or shrublet thus far reported in *Stevia* have had chromosome numbers of  $2n=24$  or  $n=12$ . Chromosome numbers of 12 shrub or shrublet species were reported for the first time. Our counts (Fig. 1a,b) agreed completely with these numbers in this regard. B chromosomes were found in only *S. salicifolia* var. *salicifolia* among shrubs or shrublets thus far examined.

Chromosome numbers of 29 perennial species and two varieties were reported for the first time. They were diploids or polyploids based on  $x=11$  or 12, although  $x=11$  was predominant (Fig. 1c–f).

Chromosome numbers of five annual species were reported for the first time. Chromosome number,  $2n=24$  in *S. aschenborniana* differed from the previous report,  $n=11$  by Keil & Stuessy (1977). *Stevia occidentalis* (Grashoff) Soejima, Yahara *et* Watanabe (Soejima *et al.* 2001a) was previously treated as a variety of *S. aschenborniana*; however, its chromosome number ( $2n=20$ ) differs from that of *S. aschenborniana* and they were distinct from each other both morphologically and geographically. Thus, the treatment of *S. occidentalis* as a new species should be supported. Chromosome numbers in annuals varied from



**Fig. 1** Photomicrographs of somatic metaphase chromosomes in Mexican *Stevia*. (a) *S. lemmonii* ( $2n=24$ ); (b) *S. phlebophylla* ( $2n=24$ ); (c) *S. ambryolepis* ( $2n=24$ ); (d) *S. hypomalaca* ( $2n=22+2B$ ) (arrows); (e) *S. serrata* var. *arguta* ( $2n=33$ ); (f) *S. deltoidea* ( $2n=44+6B$ ) (arrows); (g) *S. lita* ( $2n=20$ ); (h) *S. ephemera* ( $2n=8$ ). Bar=5  $\mu$ m.

$2n=24$  to  $2n=8$  (Fig. 1g,h) with the last number being the lowest chromosome number in the Eupatorieae (Watanabe *et al.* 1995). B chromosomes were found only in *S. aschenborniana* among annuals thus far examined.

Powell and Turner (1963) and Grashoff *et al.* (1972) reported three different chromosome base numbers,  $x=11$ , 12 and 17 in *Stevia* (Appendix II). We, however, could not find any chromosome numbers based on  $x=17$ , but found for the first time  $x=10$  and  $x=4$  in *Stevia*. We also frequently found B chromosome(s) in perennial *Stevia* and these were smaller and showed more dense condensation at prometaphase than autosomes. The previous miscounts of chromosome numbers in perennials may be largely due to the inclusion of B chromosome(s) as autosome(s).

### Cytogeography

Among the shrubs or shrublets of *Stevia*, some occur widely such as *S. salicifolia* and *S. lucida* but others are only known at the one location such as in *S. macvaughii*, *S. phlebophylla*, *S. vernicosa* and *S. zacatecana*. The shrubby species so far examined were all entirely sexual diploids with  $2n=24$ .

The distributional range of each annual *Stevia* is restricted to a small region, except for *S. occidentalis* and *S. micrantha*, which occur widely along the Sierra Madre Occidental, Trans-Mexican Volcanic Belt and Sierra Madre del Sur (Fig. 2). This indicates that there are many seasonally dry habitats scattered in the Mexican oak forests.

These morphotectonic provinces with distinct physiographic and geologic-tectonic features have been discussed by Ferrusquia (1993). All of the annual species so far examined were entirely sexual diploids.

A widespread prevalence of agamosperous polyploids has been found in perennial *Stevia*. Morphological similarities between sexual diploids and agamosperous polyploids in most Mexican *Stevia* suggest that the latter have been largely derived by autopolyploidy. This was confirmed by the comparative analyses between them of nucleotide sequences in internal transcribed spacer regions (Yahara *et al.* in preparation).

The relative abundance of polyploids as compared to their diploid ancestors can be classified into the following five types of distribution among perennials.

Type 1. Only sexual diploids were found in *S. ambryolepis* ( $2n=24$ ), *S. chilapensis* ( $2n=22$ ), *S. filodecalloana* ( $2n=22$ ), *S. hypomalaca* ( $2n=22+0-2B$ ), *S. lasioclada* (Grashoff 1972), *S. lehmannii* ( $2n=22$ ), *S. neurophylla* (Grashoff 1972), *S. oaxacana* ( $2n=24$ ), *S. ovalis* (Grashoff 1972), *S. pelophylla* (Grashoff 1972), *S. perfoliata* ( $2n=22$ ), *S. seemanioides* ( $2n=22$ ), *S. triangularis* ( $2n=24$ ), *S. trifida* ( $2n=22$ ), *S. velutinella* ( $2n=24$ ) and *S. viejoana* ( $2n=22$ ) (Fig. 3). Although Grashoff (1972) noted that all seven specimens of *S. lehmannii* had irregular pollen grains, our two accessions were  $2n=22$  and were sexual diploids. It will be necessary to count further samples to establish the chromosome number in the whole distributional range of this species. Most sexual diploid species in this type have scattered distributions in one or several isolated populations along

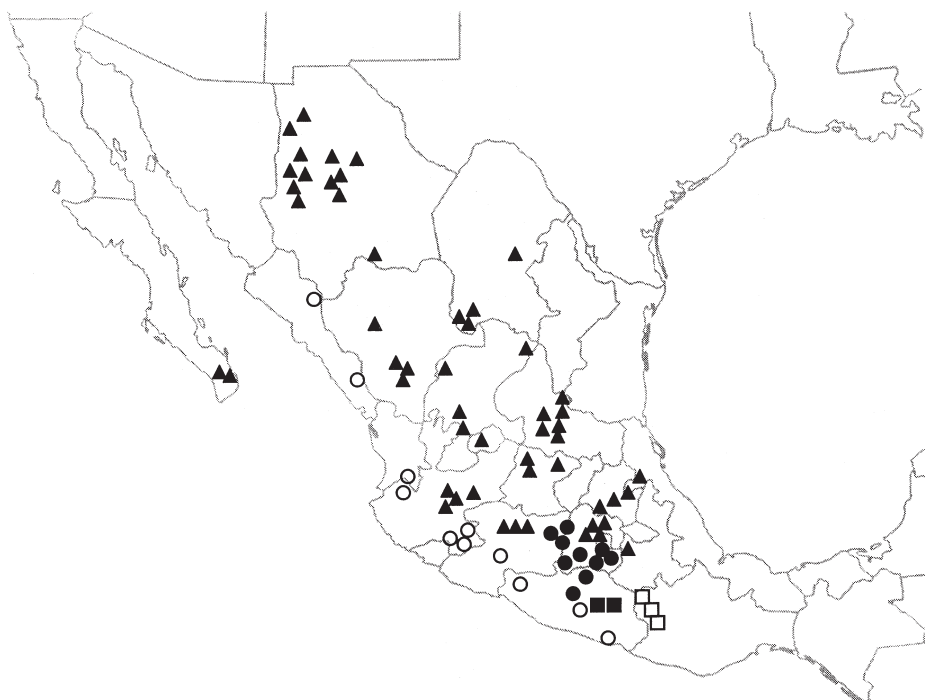
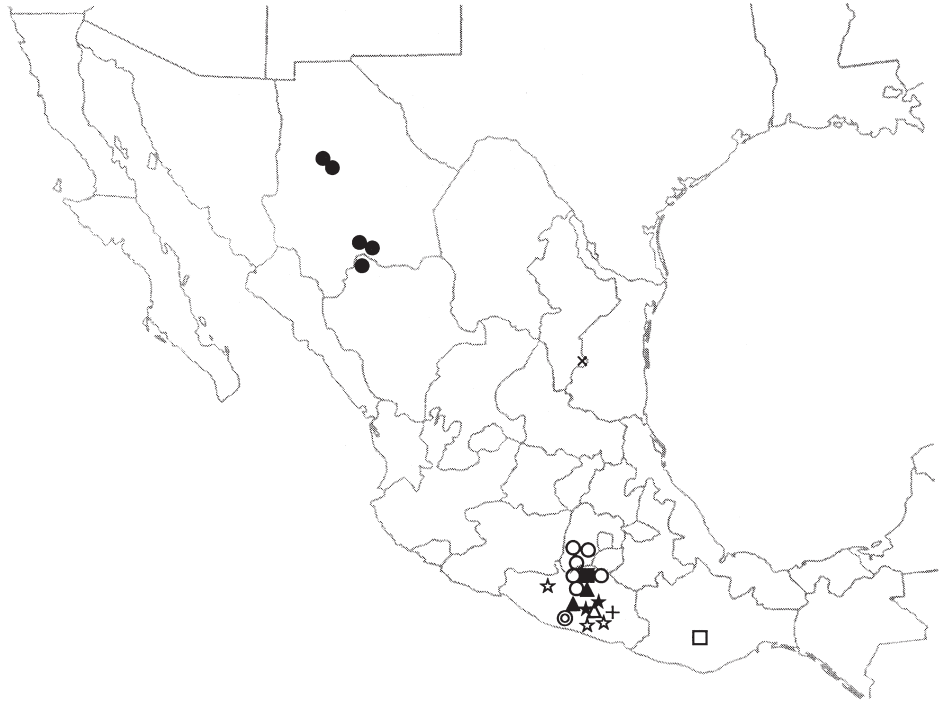


Fig. 2 Distribution map of Mexican *Stevia*. Five sexual annuals: ●, *S. aschenborniana* ( $2n=24$ ); ○, *S. occidentalis* ( $2n=20$ ); ▲, *S. micrantha* ( $2n=22$ ); ■, *S. lita* ( $2n=20$ ); □, *S. ephemera* ( $2n=8$ ).



**Fig. 3** Distribution map of Mexican *Stevia*. Eleven sexual diploid perennials: ●, *S. ambryolepis* ( $2n=24$ ); +, *S. chilapensis* ( $2n=22$ ); ▲, *S. filodecaballoana* ( $2n=22$ ); ○, *S. hypomalaca* ( $2n=22$ ); ★, *S. lehmannii* ( $2n=22$ ); □, *S. oaxacana* ( $2n=24$ ); ☆, *S. perfoliata* ( $2n=22$ ); ■, *S. seemanioides* ( $2n=22$ ); △, *S. triangularis* ( $2n=24$ ); ⊙, *S. velutinella* ( $2n=24$ ); ×, *S. viejoana* ( $2n=22$ ).

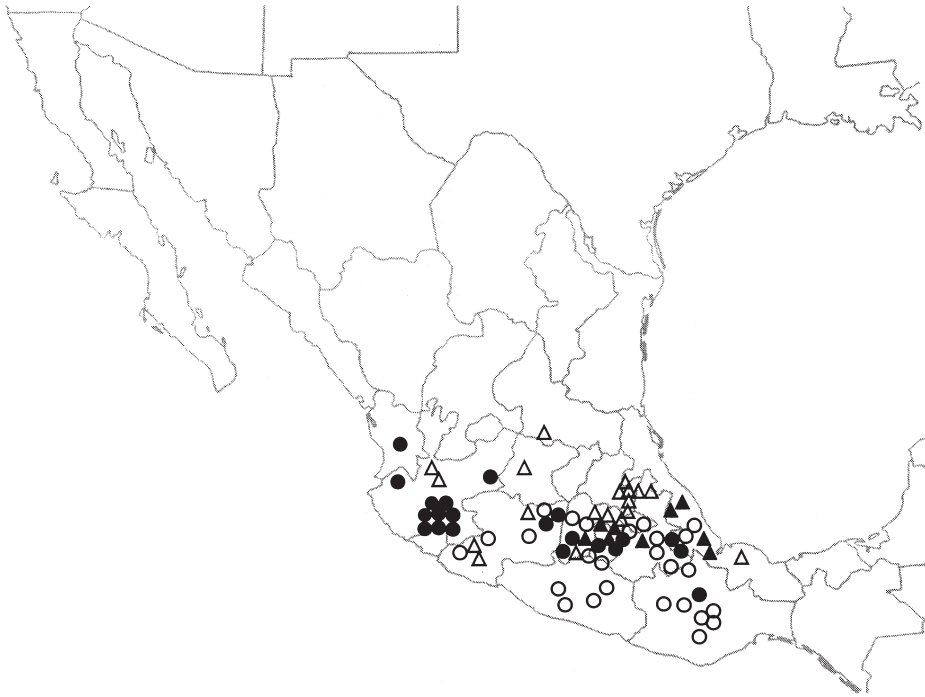
Sierra Madre Occidental, Sierra Madre Oriental, Trans-Mexican Volcanic Belt, Sierra Madre der Sur and Sierra Madre de Chiapas, particularly between Chilpancingo and Talpa in the State of Guerrero (Fig. 3). Exceptionally, *S. trifida* has a wide distributional range from the States of Sonora to Guerrero along the western and southern montane ranges. According to Grashoff (1972), this species is a fibrous rooted annual. However, we confirmed that it was not an annual but a rhizomatous perennial, with a chromosome number entirely  $2n=22$  or  $n=11$ , maintaining the diploid state throughout the distributional range. This species is probably autogamous and a colonizer of disturbed environments along stream margins.

Type 2. Relatively abundant sexual diploid populations with fewer polyploids were found at considerably wide geographic ranges including *S. monardifolia* (Fig. 4), *S. palmeri*, *S. pilosa*, *S. origanoides* and *S. stolonifera*. *Stevia monardifolia* occurs from the States of Jalisco to Guerrero, Oaxaca and Nuevo Leon in pine-oak or fir forests at altitudes considered to be high among Mexican *Stevia*. The abundance of sexual diploids may be due to its high altitude habitat and less disturbance. The agamospermous polyploids show a north-biased distribution. *Stevia pilosa* occurs from the States of Nuevo Leon to Morelos. Polyploids were found in the northern and southern-most regions of its distributional range. Diploids had smaller achenes than those of polyploids in this species. The detailed cytogeography for *S. origanoides* will be described in a separate paper (Soejima

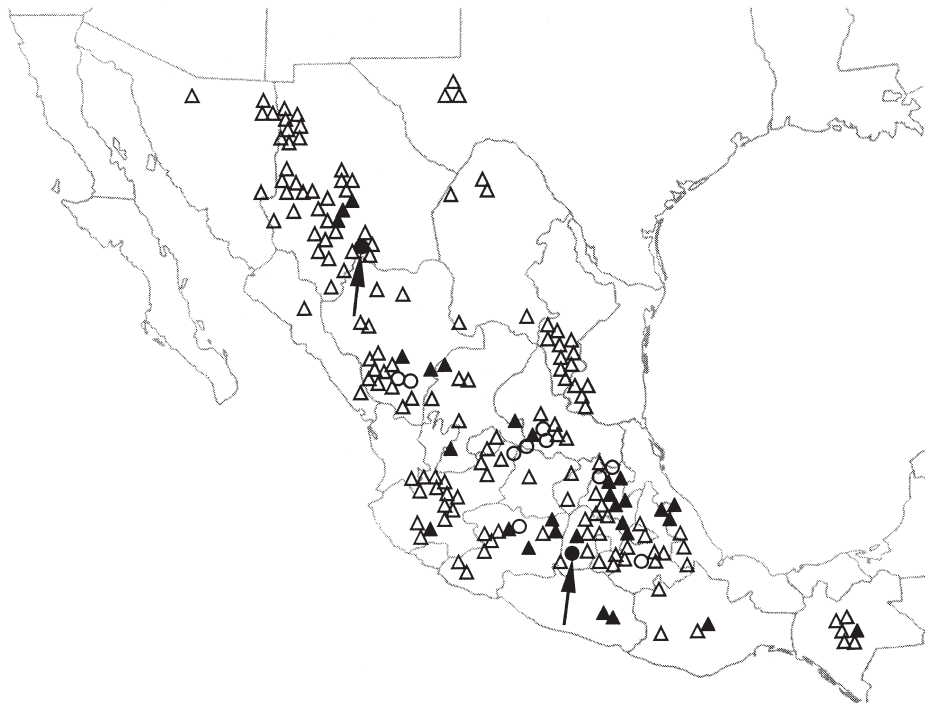
*et al.* 2001b). Sexual diploids were found in the southwestern Pacific and on more pluvial mountain slope, while agamospermous polyploids more north-biased and in inland areas.

Type 3. Widespread prevalence of agamospermous polyploids with only one or a few sexual diploid populations were very restricted in size and distribution. They include *S. alatis* ( $2n=22, 33$  and  $44$ ), *S. caracasana* ( $2n=22$  and  $33$ ), *S. connata* ( $2n=22, 33$  and  $44$ ), *S. elatior* ( $2n=22, 33$  and  $44$ ), *S. eupatoria* ( $2n=24, 36$  and  $48$ ), *S. jorullensis* ( $2n=24, 36, 48$ ), *S. oligophylla* ( $2n=22, 33, 44$  and  $55$ ; Soejima *et al.* 2001b), *S. ovata* ( $2n=22, 33, 44, 55$ , and  $66$ ; Soejima *et al.* 2001b), *S. nepetifolia* ( $2n=22$  and  $33$ ; Soejima *et al.* 2001b), *S. oligophylla* ( $2n=22$  and  $33$ ; Soejima *et al.* 2001b), *S. plummerae* ( $2n=22, 33$  and  $44$ ), *S. porphyrea* ( $2n=22, 33$  and  $55$ ), *S. serrata* ( $2n=22, 33, 44, 55$  and  $66$ ) (Fig. 5), *S. triflora* ( $2n=22, 33$  and  $44$ ; Soejima *et al.* 2001b) and *S. viscida* ( $2n=22, 33$  and  $44$ ).

The polyploid complex of the weedy *S. serrata* was found to be widespread in relatively level highland pastures and roadsides throughout Mexico, and was composed of diploids, triploids, tetraploids, pentaploids and hexaploids based on  $x=11$  (Fig. 5). The survival of diploids was confirmed from only one locality each for *S. serrata* var. *arguta* (State of Mexico) and for *S. serrata* var. *serrata* (State of Chihuahua), respectively, although Grashoff (1972) described many voucher specimens with a high production of good pollen grains. Polyploids are largely sympatric with other species at many sites throughout Mexico and have produced many hybrids



**Fig. 4** Distribution map of Mexican *Stevia*. Sexual and agamosperous perennial *S. monardifolia*: ●, sexual diploid; ○, possibly sexual diploid (Grashoff 1972); ▲, polyploids; △, possibly polyploid.



**Fig. 5** Distribution map of Mexican *Stevia*. Sexual and agamosperous perennial *S. serrata*: ●, sexual diploid; ○, possibly sexual diploid (Grashoff 1972); ▲, agamosperous polyploid; △, possibly agamosperous polyploid (Grashoff 1972).

with the intermediate morphology between putative parents [i.e. *S. serrata* × *S. plummerae*, *S. serrata* × *S. purpurea* in Grashoff (1972); *S. serrata* × *S. ovata*, *S. tephra* × *S. serrata*, *S. viscida* × *S. serrata* in Grashoff *et al.* (1972); *S. serrata* × *S. porphyrea* in the present paper]. Some of the putative

hybrids were confirmed to be polyploids (Grashoff *et al.* 1972; Soejima *et al.* 2001b; present study). This indicates that apomictic polyploids can rarely reproduce sexually.

Grashoff (1972) showed the distributional map of *S. viscida* with a few sexual populations near Guadalajara

in the State of Jalisco, although this species occurred widely from southern Arizona and Texas to western Guatemala. We revisited some populations reported as sexual by Grashoff, but have yet to document a diploid.

The detailed cytogeography for *S. nepetifolia*, *S. oligophylla*, *S. ovata* and *S. triflora* will be treated in a separate paper (Soejima *et al.* 2001b). In *S. oligophylla* and *S. triflora*, sexual and relictual diploids were found in the south-western Pacific and wetter montane slopes, while agamospermous polyploids were found more north-biased and in inland areas.

Although the diploid chromosome number had never been counted, Grashoff (1972) reported the restricted occurrences of putative sexual (based on pollen observation) populations in *S. incognita*, *S. latifolia*, *S. micradenia*, *S. seemanni*, *S. tephra* and *S. tomentosa*. In addition, the distributional ranges of the following species are not as widespread, but their populations are composed predominantly of agamospermous polyploids and a few diploids: *S. decumbens* ( $2n=24$  and  $36$ ), *S. karwinskiana* ( $2n=22$  and  $44$ ) and *S. reticulata* ( $2n=22$  and  $33$ ). The distributional patterns of all these species are tentatively grouped into Type 3.

Type 4. Only agamospermous polyploids were found throughout the distributional ranges of *S. deltoidea* ( $2n=33$ ,  $44$  and  $55$ ), *S. iltisiana* ( $2n=33$  and  $44$ ) and *S. isomeca* ( $2n=33$  and  $44$ ). *Stevia deltoidea* occurs widely from Jalisco to Honduras. No sexual populations were found in these areas (Grashoff 1972). However, Sundberg *et al.* (1986) reported a diploid plant with  $n=11$  in Argentina. It will be necessary to re-examine their identification and the occurrence of this diploid there.

Type 5. Only agamospermous polyploids were found in one or a few small populations of *S. clinipodioides* ( $2n=36$  and  $48$ ), *S. coahuilensis* ( $2n=33$ ), *S. cruzii* (Grashoff 1972), *S. liebmannii* ( $2n=33$ ), *S. mascotensis* ( $2n=44$ ), *S. potosina* ( $2n=33$ ) and *S. rosei* ( $2n=44$ ). *Stevia orizabensis* and *S. villaregallis* are also represented only by apomictic plants, although their chromosome numbers have not been counted (Grashoff 1972). The restricted distribution of some agamospermous polyploid species may be due to their in situ origins.

Agamospermous polyploids in the distribution types 2, 3 and 4 may have arisen many times from sexual diploids at several locations.

The widespread distribution of agamospermous polyploids compared with the restricted range of their diploid ancestors has been noted in various taxonomic groups (Babcock & Stebbins 1938; Gustaffson 1947; Müntzing 1954; Stebbins 1971; Sullivan 1976; Grant 1981; Watanabe 1986). Bayer and Stebbins (1981, 1983) and Wolf (1980) explained this distributional pattern (which was confirmed in the genera *Antennaria* and *Arnica* growing in

areas severely affected by successive advances and recessions of Pleistocene ice sheets) in the following way. The ancestral diploids were probably restricted to small southern areas at the end of the last glaciation. Accompanying the glacial recession, a large number of open northern habitats developed that were suitable for colonizers such as agamospermous polyploids. The agamospermous polyploids colonized those open and northern habitats successfully, but the sexual diploids were not good colonizers due to their fertilization loads, and they migrated and colonized to a lesser extent than the other successful colonizers from these southern refuges.

Soreng and Devender (1989) investigated fossils and interpreted the range dynamics of sexual and apomictic *Poa fendleriana* from 20 490 to 1820 years BP in south-western Arizona and south-eastern Utah. During the Late Wisconsin full-glacial period, the northern limit of *P. fendleriana* was displaced southward into Arizona and New Mexico. Sexual populations were present in southern areas such as the Santa Catalina Mountains. Climatic shifts since the Pleistocene appear to have restricted 'marginal' habitats and rearranged 'good' habitats for sexual populations to the south and increased marginal habitats to the north, favoring expansion of apomicts, but inhibiting the spread of sexuals. Apomicts expanded onto the Colorado Plateau in south-western Utah by 11 310 years BP. Sexuals probably did not move into that area until 3400 years BP. Since the last glacial maximum, apomicts advanced northward first, followed about 5000–7000 years later by sexuals. Relatively lower colonization capacities and upward constriction of suitable habitat appears to have slowed the migration of sexuals. The abundance of apomicts in colder and previously glaciated areas testifies to their ability to disperse rapidly as appropriate habitat becomes available with favorable climatic shifts.

In Mexico, which is located at a much lower latitude, climatic vacillation since the Pliocene has also affected the expansion and contraction of various vegetation zones, including temperate forests (Martin & Harrell 1957; Sarukhan 1968; McDonald 1993). Present distribution patterns of the temperate montane flora in the Sierra Madre del Sur, Sierra Madre Occidental, Sierra Madre Oriental and Trans-Mexican Volcanic Belt can be attributed to the pluvial and cooler environments of the Pleistocene when suitable habitats were much more extensive (Martin & Harrell 1957; Rzedowski 1981, 1993). The distribution areas of Mexican *Stevia* correspond to these temperate flora. Therefore, they seem to have been considerably well-maintained during the Pleistocene.

In the genus *Stevia*, most diploids are also restricted to the south-western Trans Volcanic Belt and Sierra Madre del Sur, which are warmer and wetter than the northern and inland areas, even though they occur at much lower

latitudes. However, several northern, higher and wetter areas in Sierra Madre Occidentalis and Sierra Madre Orientales have also served sporadically as refuges for some diploids such as *S. pilosa* and *S. ovata* (Soejima *et al.* 2001b). The agamospermous polyploids, with their much greater colonizing ability, have spread to more northern and wider areas accompanying climatic improvement. In addition, since the last glacial epoch, the temperate zone in Mexico has been disturbed most severely by agricultural and pastoral development. These areas were inhabited by early corn farmers approximately 5000 years ago (Fowler & MacNeish 1972). Livestock grazing, agriculture and human activities resulting in erosion have altered the Mexican landscape and threaten its botanical diversity (Bye 1993). Such extensive human activity has promoted both seed and livestock dispersal, thereby leading to sympatry for various pairs of taxa, and hybridization. According to a survey carried out by the Mexican Agricultural Department in 1983 (Anonymous 1983), only 19% of Mexico was uneroded. Human activities break down reproductive barriers of different plant species and also create intermediate habitats suitable for the survival of hybrid offspring (Anderson 1948). Creation of new and open habitats promotes the establishment of new elements that migrate into the local flora. Three to six *Stevia* species have been found existing sympatrically in many places. For example, 11–15 km north-east of the junction in Oaxaca City, on Route 175 to Valle National at 1930–2260 m altitude, we found eight sympatric species of *Stevia* (*S. crassifolia*, *S. decumbens*, *S. deltoidea*, *S. liebmairii*, *S. organoides*, *S. ovata*, *S. seemanii* and *S. nepetifolia*). Most of the ‘populations’ of these species were apomicts and were able to reproduce agamospermously. During their distributional range expansions, they have come together sympatrically and hybridized. Alteration of plant populations and habitats has produced marked changes in local flora, vegetation composition and the genetic structure of plants (Bye 1993). The wider distributional range of some agamospermous polyploids have been achieved by recent colonization accompanying climatic warming since the last glaciation. Their success is due to their efficient reproduction in founder populations, and the acceleration of seed dispersal and the creation of new and open habitats by human activities.

Except for the genus *Mikania*, large genera in the tribe Eupatorieae (e.g. *Ageratina*, *Chlomoraena*, *Eupatorium* and *Stevia*) have displayed bursts of agamospermy in disturbed habitats. However, agamospermy could not be found in shrubs or shrublets in these genera, and the reason why agamospermy was not found in the genus *Mikania* may be due to its woody vein habit. The prevalence of agamospermy for many perennial taxa in these genera might be due to their genetic background/tendencies to become agamospermic very easily and their

favoring of this successful reproductive mode in present habitats.

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Appendix I Chromosome number determinations in Mexican *Stevia*

Species	Locality	Voucher specimen	Chromosome number	
<i>*alatipes</i> B. L. Rob.	Jalisco: 4 km SE of JCT from Jilotlan via San Ishidro	Y & S 488	2n=22	
	Jalisco: 19.5 km N of Mascota	Y & S 421	2n=33	
	Jalisco: 22 km N of Mascota	Y & S 425	2n=33	
<i>*ambryolepis</i> (B. L. Rob.) B. L. Rob. <i>anadenotricha</i> (B. L. Rob.) Grashoff	Chihuahua: western hill of Parral	Y 731	2n=24+1-3B	
	Durango: 96.6 km NE from Vila Union to Durango	Y 784	2n=24	
<i>aschenborniana</i> Sch.-Bip.	Guerrero: 11.7 km W of JCT from Taxco to Ixcateopan	Y & S 243	2n=24	
	Guerrero: 39 km SW of Filo de Caballo	Y 869	2n=24+1-2B	
<i>caracasana</i> DC.	Jalisco: 4 km SE of JCT on the way to Jilotlan via San Ishidro	Y 944	2n=22+1-3B	
	Guerrero: 15.9 km E of Tixtra de Guerrero	Y & S 248 & 249	2n=33	
	Jalisco: 12.5 km S of Autlan	Y & S 474	2n=33	
	Oaxaca: 15.6 km S of Juchatengo	Y 1132	2n=33	
	Mexico: 8.8 km S of Valle de Bravo	Y 1292	2n=33+0-2B	
<i>chilapensis</i> Soejima et Yahara	Guerrero: 33.5 km E of Chilapa on the way from Chilpancingo to Tlapa (Mex 93)	Y & S 265, Y 1154	2n=22	
<i>*clinipodioides</i> Greenm.	Veracruz: 7 km SE from JCT with Route 140 in Perote, on the way to Cofre de Perote	Y & S 541	2n=48	
<i>*coahuilensis</i> Soejima et Yahara	Coahuila: N slope of El Morro Sierra del Marta	Y 1484	2n=33	
<i>connata</i> Lag.	Guerrero: 13.8 km E of Chilapa	Y & S 257	2n=22	
	Guerrero: 33.5 km E of Chilapa	Y 1160	2n=22+0-2B	
	Guerrero: 29 km W of Taxco	Y 43	2n=33	
	Guerrero: Cerro Alquitran	Y 854	2n=33	
	Mexico: 4.9 km N of Tejupilco	Y 1007	2n=33	
	Oaxaca: 20 km S JCT on Mex 190 on the way to Santa Maria Ecatepec	Y 1118	2n=33	
	Oaxaca: 4.2 km W of Ixtran to Valle Nacional	Y 1087	2n=44	
<i>*constricta</i> (Grashoff) Soejima, Yahara et Watanabe	Sinaloa: 61.3 km NE on Mex 40 from Vila Union	Y 1252	2n=22+4-8B	
	Sinaloa: 78.1 km NE from Vila Union to Durango	Y 772	2n=44	
<i>*crassifolia</i> Soejima et Yahara	Oaxaca: 3.6 km N of El Punto	Y & S 345	2n=33	
<i>*decumbens</i> (B. L. Rob. et Greenm.) Greene	Oaxaca: 2.8 km W of Huitzo	Y 1057	2n=24	
	Oaxaca: 3.6 km N of El Punto	Y & S 343	2n=36	
	Oaxaca: 18.5 km SW of Huitzo	Y 1059	2n=36	
<i>*deltoidea</i> Greene	Guerrero: 42 km SW of JCT on the way from Chilpancingo to Filo de Caballo	Y 860	2n=33	
	Hidalgo: 4.6 km NE from JCT to El Chico	Y 16	2n=33+0-3B	
	Guerrero: 49 km SW from JCT the Hwy 95 Libre to Filo de Caballo	Y & S 273	2n=33+0-3B	
	Oaxaca: 13.5 km NE from JCT on Route 175 to Valle Nacional	Y & S 326	2n=33+0-3B	
	Oaxaca: 17.8 km NE from JCT on Route 175 to Valle Nacional	Y & S 332	2n=33+0-3B	
	Oaxaca: 3.6 km N of El Punto	Y & S 341 & 342	2n=33+0-3B	
	Oaxaca: Microondes station just W of El Tejocote	Y 1061	2n=33	
	Zacatecas: 28.2 km from JCT on the way from Jalpa to Tlaltenango	Y 940	2n=44	

Appendix I *Continued*

Species	Locality	Voucher specimen	Chromosome number
	Guerrero: 49 km SW from JCT of Filo de Caballo	Y & S 274 & 275	2n=44+0-6B
	Chiapas: 16 km NW of Motozintla de Mendoza	Y 630	2n=55+0-5B
<i>deltoidea</i> × <i>nepetifolia</i>	Guerrero: 49 km SW from JCT on the Hwy 95 Libre to Filo de Caballo	Y & S 276	2n=33
* <i>dictyophylla</i> B. L. Rob.	Jalisco: 1.6 km W of El Pueblito	Y 801	2n=24
* <i>ecatepecana</i> Soejima, Yahara et Watanabe	Puebla: 14 km NE of Teotitlan	Y 1038	n=12II
<i>elatior</i> H. B. K. var. <i>elatior</i>	Michoacan: 11 km S of Morelia	Y 77	2n=33
	Oaxaca: 75 km S from Puebla-Oaxaca border	Y & S 309	2n=33
	Chiapas: 16 km NW of Motozintla de Mendoza	Y 631	2n=33
	Veracruz: 13.2 km W of Ciudado Mendoza	Y 836	2n=33
	Michoacan: 10 km W of Ucareo	Y 984	2n=33
	Oaxaca: 34.5 km N from Oaxaca to Valle Nacional	Y 1078	2n=33
	Oaxaca: 32.5 km of Huajuapán	Y & I 61	2n=44
	Oaxaca: 43.2 km of Nochixtlan	Y & I 101	2n=44
	Chiapas: 11 km W of San Cristobal de Las Casas	Y 608	2n=44
*var. <i>podophylla</i> B. L. Rob.	Oaxaca: 75 km S of Puebla-Oaxaca border	Y & S 307	2n=33
	Michoacan: 24.8 km E of La Cumbre on Mex 15	Y 818	2n=33
* <i>ephemera</i> Grashoff	Oaxaca: 75 km S from Puebla-Oaxaca border	Y & S 313	2n=8
	Oaxaca: 5.7 km SW of Suchixtlahuaca	Y 1043	n=4II
<i>eupatoria</i> (Spreng.) Willd.	Hidalgo: 11 km from Pachuca to El Chico	Y 6-1	2n=24
	Michoacan: 52 km W of Maravatio	Y 85	2n=36
	Hidalgo: 11 km from Pachuca to El Chico	Y 6-2	2n=48
	Mexico: 107 km NW of Mexico City	Y 667	2n=48
* <i>filodecaballoana</i> Soejima, Yahara et Watanabe	Guerrero: 25 km SW of Filo de Caballo	Y 51	2n=22
	Guerrero: 8.5 km SW of Filo de Caballo	Y & S 287	2n=22
* <i>glandulosa</i> Hook. & Arn.	Sinaloa: 43 km NE on Mex 24 from Badiraguato	Y 761	2n=24
* <i>hintoniorum</i> B. L. Turner	Nuevo Leon: Cerro Potosi, on the way from Diezocho de Marzo to the top	Y 1446	2n=44
* <i>hypomalaca</i> B. L. Rob.	Mexico: 11.3 km NE of Tejupilco	Y 1010	2n=22+1B
	Guerrero: 14.2 km N of Taxco	Y 1163	2n=22+0-2B
<i>iltisiana</i> Grashoff	Hidalgo: 8 km N from JCT to Mineral del Chico	Y 12 & 14	2n=33
	Morelia: 4 km N of Tres Marias	Y 29	2n=44
* <i>incongnita</i> Grashoff	Morelos: On the way from Tres Morias to Mt Zempoala	Y & S 215 & 217	2n=33
	Mexico: 2 km NE on Mex 134 from JCT to Sultepec	Y 987	2n=33+2B
	Coahuila: N slope of El Morro, Sierra del Marta	Y 1482	2n=33+0-2B
<i>isomeca</i> Grashoff	Veracruz: Dos Hermonas, 6 km NW of Acajete on Route 140	Y 829	2n=33
	Mexico: 9 km W of Rio Frio	Y & S 113	2n=33+1-3B
	Veracruz: 11 km SE from JCT in Perote	Y & S 538	2n=44+1-4B
	Veracruz: 9 km SE from JCT in Perote	Y & S 822	2n=44+4B

## Appendix I Continued

Species	Locality	Voucher specimen	Chromosome number	
<i>jaliscensis</i> B. L. Rob.	Jalisco: 32 km NW of Guadalajara	Y & S 404	2n=24	
	Jalisco: 2 km E of Villa Guerrero	Y 1240	2n=24	
<i>jurullensis</i> H. B. K.	Oaxaca: 75 km S from Puebla-Oaxaca border on Hwy 135 Cuota	Y & S 308	2n=24	
	Hidalgo: 4.6 km NE from JCT to El Chico	Y 17	2n=36	
	Michoacan: 3 km W of Zitacuaro	Y 68	2n=36	
	Michoacan: 24.8 km E of La Cumbre on Mex 15	Y & S 817	2n=36	
	Nuevo Leon: 26.5 km NW of Galeana	Y 1440	2n=36	
	Hidalgo: Entrance of Eloxochitlan	Y 24	2n=48	
	Oaxaca: 24.8 km NE from JCT in Oaxaca, 3.8 km N of La Cumbre	Y & S 337	2n=48	
	Oaxaca: 71 km NE from JCT in Oaxaca, on Route 175 to Valle Nationale	Y & S 350	2n=48	
	<i>*karwinskiana</i> Steud.	Veracruz: Tenampa	Y & S 555	2n=22
		Veracruz: Dos Hermonas, 6 km NW of Acajete on Route 140	Y & S 523 & 524	2n=33
Veracruz: 29 km SE from JCT on Route 140 in Guadalup, Victoria		Y & S 544	2n=33	
<i>latifolia</i> Benth.	Guerrero: 25 km SW of Filo de Caballo	Y 48	2n=33	
	Guerrero: 10 km N of JCT to Ixcateopan	Y & S 236	2n=33	
	Jalisco: 12 km E from JCT of Talpa de Allende-La Cuesta Route	Y & S 463	2n=33	
	Michoacan: 24.8 km E of La Cumbre	Y 816	2n=33	
	Mexico: 13.5 km N of Temascaltepec, 4 km S of JCT to Valle de Bravo	Y 1015	2n=33	
	Guerrero: 8.5 km SW of Filo de Caballo	Y & S 286	2n=44	
	Oaxaca: 40 km SW of Tlaxiaco	Y & S 362 & 363	2n=44	
	Michoacan: 24 km E of Zacapu	Y 805	2n=44	
	Oaxaca: 24.8 km NE from JCT of 3.8 km N of La Cumbre	Y & S 333	2n=55	
	<i>*lehmannii</i> Hieron.	Guerrero: 30.8 km S of Petaquillas	Y & S 299	2n=22
Guerrero: 13.6 km E of Chilapa		Y 1149	2n=22 + 1B	
<i>*lemmonii</i> A. Gray	Sinaloa: 92.3 km NE on Mex 40 from Vila Union	Y 1267	2n=24	
<i>*liebmannii</i> Sch.-Bip.	Oaxaca: 13.5 km NE from JCT on Route 175 to Valle National	Y & S 324	2n=33	
<i>*lita</i> Grashoff	Guerrero: 19.5 km E of Tixtla de Guerrero	Y & S 250	2n=20	
	Guerrero: 13.6 km E of Chilapa	Y 1150	2n=10II	
<i>lucida</i> Lag.	Durango: 130.2 km NE from Vila Union to Durango	Y 786	2n=24	
<i>*macvaughii</i> Grashoff	Jalisco: E slope of Nevado de Collima	Y 1216	2n=24	
<i>*mascotensis</i> Soejima et Yahara	Jalisco: 19.5 km N of Mascota	Y & S 420	2n=44	
	Jalisco: 44 km W of Talpa de Allende	Y & S 466	2n=44	
<i>*mexicana</i> Soejima, Yahara et Watanabe	Mexico: 21.5 km NE of Tejupilco	Y 1011 & 1527	n=11II, 2n=22	
<i>micradenia</i> B. L. Rob.	Guerrero: 42 km SW from JCT on the way from Chilpancingo to Filo de Caballo	Y 862	2n=33	
<i>micrantha</i> Lag.	Chihuahua: 1.6 km N on a lumber road from junction of Mex 24 at 6.4 km E from another junction to Santa Barbara	Y 730	2n=24	
<i>monardifolia</i> H. B. K.	Jalisco: 30 km N of Mascota	Y & S 435 & 437	2n=22	
	Jalisco: 9 km E from JCT of Talpa de Allende-La Cuesta Route	Y & S 456	2n=22	
	Jalisco: 10–11 km SE of JCT on the way to Jilotlan via San Ishidro	Y & S 492	2n=22	

Appendix I *Continued*

Species	Locality	Voucher specimen	Chromosome number
	Jalisco: 20 km SE of JCT on the way to Jilotlan via San Ishidro	Y & S 495	2n=22
	Jalisco: 13 km W of JCT on a lumber road to Nevado de Colima	Y & S 499	2n=22
	Jalisco: 22 km W of JCT on a lumber road to Nevado de Colima via Microondas Station	Y & S 502	2n=22
	Veracruz: near state border on the way from Esperanza to Orizaba	Y & S 515	2n=22
	Veracruz: 25.8 km W of Ciudado Mendoza	Y 839	2n=22
	Mexico: 6.5 km NE of San Francisco de los Ranchos	Y 1002	2n=22
	Oaxaca: 16.7 km N from Ixtran (W edge) to Valle Nacionale	Y 1095 & 1096	2n=22
	Morelos: Zempoala	Y 1200	2n=22
	Jalisco: E slope of Nevado de Collima, 12.7 km W of JCT on Route 110	Y 1210	2n=22
	Jalisco: E slope of Nevado de Collima, 16 Km W of JCT on Route 110	Y 1217	2n=22+2-3B
	Jalisco: 11 km SE of JCT on Route 110, on the way to Jilotran	Y 1222	2n=22
	Nayarit: NW slope of Valcan Sanganguey	Y 1248	2n=22
	Michoacan: above Los Azufres	Y 1281	2n=22
	Michoacan: Sierra de San Andres, 11 km from Campamento	Y 1286	2n=22
	Mexico: 21 km S from JCT of 134 and Sultepec route	Y 1303	2n=22
	Morelos: San Juan Tlacotenco	Y 1307	2n=22
	Mexico: 7.7 km E of Amecameca	Y 1311	2n=22
	Veracruz: 6 km NW of Acajete	Y & S 526	2n=33
	Veracruz: 11 km SE from JCT in Sierra de Agua	Y & S 531 & 539	2n=33
	Veracruz: 9 km SE from JCT in Perote	Y & S 823	2n=33
	Morelos: 4 km N of Tres Marias	Y 842	2n=33+0-1B
	Mexico: 2 km NE from JCT on the way from Toluca to Temascaltepec	Y 991 & 994	2n=33
	Mexico: 2.4 km W of exit to La Marquesa	Y 1017	2n=33
	Puebla: 31.2 km S of Acatepec	Y 1027	2n=33
<i>*myricoides</i> McVaugh	Michoacan: 26.8 km NW of Uruapan	Y 962	n=12II, 2n=24
<i>*nelsonii</i> B. L. Rob.	Jalisco: 22 km W of JCT on a lumber road to Nevado de Colima via Microondas station	Y & S 501	2n=24
<i>*oaxacana</i> Soejima et Yahara	Oaxaca: 34.8 km N of Juchatengo	Y 1143	2n=24
<i>*occidentalis</i> (Grashoff) Soejima, Yahara et Watanabe	Sinaloa: 25 km S of El Palmito	Y 917 & 1517	2n=20
<i>*palmeri</i> A. Gray	Sinaloa: 96.6 km NE of Vila Union	Y & S 780	2n=44
<i>*perfoliata</i> Cronquist	Guerrero: near El Fresco Microondas Station	Y 63	2n=22
	Guerrero: 91 km NE of Zihuatanejo	Y 67	2n=22
<i>*phlebophylla</i> A. Gray	Jalisco: 11 km N on Route 54 from JCT with Periferico	Y & S 512	2n=24
<i>pilosa</i> Lag.	Hidalgo: 10 km from Pachuca to El Chico	Y 3	n=11, 2n=22
	Hidalgo: 0.4 km N from JCT to El Chilo	Y 4	n=11
	Hidalgo: 4.6 km NE from JCT to El Chico	Y 15	2n=22+0-4B
	Nuevo Leon: 12.3 km SE of Zaragoza	Y 1415	2n=22
	Morelos: 4 km N of Tres Marias	Y 28	2n=33
	Coahuila: N slope of El Morro, Sierra del Marta	Y 1488	2n=44

## Appendix I Continued

Species	Locality	Voucher specimen	Chromosome number
<i>plummerae</i> A. Gray			
*var. <i>plummerae</i>	Chihuahua, 17.8 km E of Guadalupe y Calvo	Y 743	2n=22+1-2B
	Durango, 19.8 km W of El Salto	Y 788	2n=44
<i>polycephala</i> Bertol	Oaxaca: 7 km N of Diaz Ordes	Y & S 132	n=12II, 2n=24
	Oaxaca: 7.8 km N of Diaz Ordes	Y 1068	2n=24
<i>porphyrea</i> McVaugh	San Luis Potosi: 4 km N of JCT to El Realejo	Y 659	2n=22
	Jalisco: 10 km N of Ixtahuacan	Y & S 398	2n=33
	Jalisco: 10 km N of Ixtahuacan	Y & S 397 & 399	2n=44
	Mexico: 2 km NE on Mex 134 from JCT to Sultepec	Y 995	2n=55
* <i>potosina</i> Soejima, Yahara et Watanabe	Nuevo Leon: 37.4 km NW of Galeana	Y 1432	2n=33
	Nuevo Leon: Cerro Potosi, on the way from Diezoch de Marzo to the top	Y 1441	2n=33
	Coahuila: El Coahuilon	Y 1489	2n=33
* <i>reticulata</i> Grashoff	Nayarit: NW slope of Volcan Sanguey	Y 1246	2n=22
	Durango: 96.6 km NE from Vila Union to Durango on Mex 40	Y 779	2n=33
* <i>revoluta</i> B. L. Rob.	Puebla: 29.1 km S of Acatepec on road through San Juan Acatitlan to San Simon	Y 1026	2n=24
* <i>rosei</i> B. L. Rob.	Durango: 5.8 km W of La Ciudad	Y 905-1	2n=44
* <i>rotundifolia</i> Soejima, Yahara et Watanabe	Zacatecas: 28.8 km from JCT on the way from Jalpa to Tlaltenango	Y 933	2n=33+0-2B
* <i>rzedowskii</i> McVaugh	Zacatecas: 28.8 km from JCT on the way from Jalpa to Tlaltenango	Y 930 & 939	n=12II, 2n=24
* <i>scabrella</i> Benth.	Durango: 42.1 km SW of San Juan de Michis	Y 892	2n=24
* <i>seemannii</i> Sch.-Bip.	Oaxaca: 27.1 km S from JCT on Mex 190 on the way to Santa Maria	Y 1114	2n=33
	Oaxaca: 34.8 km N of Juchatango	Y 1141	2n=33+0-1B
	Guerrero: 49 km SW from JCT of Hwy 95 Libre to Filo de Caballo	Y & S 272	2n=44
	Guerrero: 42 km SW from JCT of Hwy 95 on the way from Chilpancingo to Filo de Caballo	Y 861	2n=44
	Oaxaca: 34.5 km from Oaxaca to Valle Nacional	Y 1079	2n=44
	Oaxaca: 29.4 km N of Mixtepec	Y 1126	2n=44
	Oaxaca: 27.1 km S from JCT on Mex 190 on the way to Santa Maria	Y 1113	2n=55+2B
* <i>seemannioides</i> Grashoff	Guerrero: 14.8 km N of JCT to Ixcateopan	Y 237	2n=22
<i>scleriana</i> B. L. Rob.	Oaxaca: 10 km SE of La Camaron	Y 147	n=12II
<i>serrata</i> Cav.			
var. <i>arguta</i> B. L. Rob.	Mexico: 21.5 km NE on Mex 134 from Tejupilco de Hidalgo	Y 1013	2n=22
	Zacatecas: 76 km SW of San Luis Potasi	Y 880 & 882	2n=33
var. <i>serrata</i>	Hidalgo: 0.4 km N from JCT to El Chico	Y 5	2n=33+0-2B
	Guerrero: Cerro Alquitran, on the way from Mazatlan to Microondas station	Y 853	2n=33+0-1B
	San Luis Potosi: 24.5 km SW of San Luis Potosi	Y 877	2n=33
	Zacatecas: 4 km E of Sombrerete	Y 884	2n=33+1-3B
	Durango: 14.7 km W of Suchil	Y 886	2n=33+1-2B
	Guerrero: 13.6 km E from Chilapa	Y 1152	2n=33
	Hidalgo: Entrance to Eloxochitlan	Y 22	2n=44
	Michoacan: 52 km W of Maravatio	Y 82	2n=44

Appendix I *Continued*

Species	Locality	Voucher specimen	Chromosome number
	Veracruz: 7 km SE from JCT on Route 140 in Perote	Y & S 542	2n=44
	Chiapas: 17 km W of San Cristobal de Las Casas	Y 607	2n=44
	Zacatecas: 28.8 km of JCT on the way from Jalpa to Tlaltenango	Y 932	2n=44
	Jalisco: 5.2 km SE of JCT on the way to Jilotlan via San Ishidro	Y 947	2n=44
	Oaxaca: 11 km SE of Tejuapan on Mex 190	Y 1050	2n=44
	Michoacan: 3 km W of Zitacuaro	Y 70	2n=55
	Guerrero: Cerro Alquitran on the way from Mazatlan to Microondas station	Y 847	2n=55
	Mexico: 11.5 km N of Temascaltepec	Y 1014	2n=55
	Michoacan: 11 km S of Morelia	Y 78	2n=66
<i>serrata</i> × <i>porphyrea</i>	Durango: 28.5 km W of La Ciudad	Y 910	2n=44
* <i>stolonifera</i> Yahara et Soejima	Jalisco: 20.5 km SE of JCT on the way to Jilotlan via San Ishidro	Y 953	2n=22
	Jalisco: 20 km SE of JCT on the way to Jilotlan via San Ishidro	Y 1223	2n=22+2B
	Jalisco: 20.5 km SE of JCT on the way to Jilotlan via San Ishidro	Y 955	2n=44
<i>subpubescens</i> Lag.	Oaxaca: 14 km N of Diaz Ordes	Y & S 133	n=12II
	Durango: 45.6 km W of La Ciudad	Y 916	2n=24
	Nayarit: NW slope of Volcan Sanganguey	Y 1247	2n=24
<i>ephra</i> B. L. Rob.	Zacatecas: 76 km SW of San Luis Potosi	Y 881	2n=33
	San Luis Potosi: 4 km of JCT to El Realejo	Y 656	2n=33+0-3B
<i>tomentosa</i> H. B. K.	Michoacan: 52 km W of Maravatio	Y 84	2n=33
	Mexico: 65 km NW from Mexico City along Hwy 57 Cuota	Y 875	2n=33+0-3B
	Durango: 14.7 km W of Suchil	Y 885	2n=33
	Mexico: 6-6.5 km NE on Mex 134 from San Francisco de los Ranchos	Y 998	2n=33
	Oaxaca: 1 km E from San Pedro Teposcolula on Mex 125	Y 1052	2n=33
	Nuevo Leon: 36.5 km NW from Galeana	Y 1434	2n=33
* <i>triangularis</i> Grashoff	Guerrero: Cerro Alquitran	Y 845	2n=24
<i>trifida</i> Lag.	Guerrero: 13 km W of Taxco	Y 40	2n=22
	Mexico: 21.5 km NE from Tejupilco	Y 1012	2n=22
	Guerrero: 17.6 km N of Taxco	Y 1165	2n=22
* <i>velutinella</i> Grashoff	Guerrero: 64 km SW from Filo de Caballo	Y 55	2n=24
* <i>viejoana</i> Soejima, Yahara et Watanabe	Nuevo Leon: 12.3 km SE of Zaragoza	Y 1416	n=11, 2n=22
<i>viscida</i> H. B. K.	Michoacan: 1 km S of Morelia	Y 74	2n=33
	Michoacan: 7 km S of Morelia	Y 81	2n=33
	Jalisco: 23 km W of Ameca	Y & S 40	2n=33
	Durango: 42.1 km SW of San Juan de Michis	Y 895	2n=33
	Jalisco: 8 km W of Guadalajara	Y 922	2n=33
	Zacatecas: 28.8 km from JCT on the way from Jalpa to Tlaltenango	Y 931	2n=33
	Michoacan: 10 km W of Ucareo	Y 985	2n=44
* <i>zacatecana</i> McVaugh	Zacatecas: 27.3 km from JCT on the way from Jalpa to Tlaltenango	Y 923	n=12II, 2n=24
* <i>zephyrantha</i> Grashoff	Guerrero: 12.3 km SW from Filo de Caballo	Y 867	2n=24

\* First chromosome count; Y, Yahara *et al.*; Y & I, Yahara & Ito; Y & S, Yahara & Soejima.

Appendix II Summary of chromosome numbers, habits and references of Mexican *Stevia*

Habit	Species	<i>n</i>	<i>2n</i>	References
Shrub or shrublet	<i>anadenotricha</i> (B. L. Rob.) Grashoff	–	24	Grashoff <i>et al.</i> 1972;
		12	–	Keil & Stuessy 1975
	–	24	Present study	
	<i>berlandieri</i> A. Gray var. <i>podadenia</i> B. L. Rob. var. <i>berlandieri</i>	–	24	Grashoff <i>et al.</i> 1972
		12	–	King <i>et al.</i> 1976
		12	–	Keil & Stuessy 1977
	12	–	Pinkava & Keil 1977	
	<i>chiapensis</i> Grashoff	–	–	
	<i>dictyophylla</i> B. L. Rob.	–	24	Present study
	<i>glandulosa</i> Hook. & Arn.	–	24	Present study
	<i>jalisensis</i> B. L. Rob.	12	–	Turner & Flyr (1966)
		–	24	Present study
	<i>lemmonii</i> A. Gray	–	24	Present study
	<i>lucida</i> Lag.	12	–	Powell & Turner 1963
		12	–	Powell & King 1969
		12	–	Grashoff <i>et al.</i> 1972
		12	–	King <i>et al.</i> 1976
		12	–	Powell & Powell 1978
		–	24	Present study
		–	24	Present study
	<i>macvaughii</i> Grashoff (= <i>baccharifolia</i> B. L. Turner)	–	24	Present study
	<i>microchaeta</i> Sch.-Bip.	–	24	Grashoff <i>et al.</i> 1972
		–	24	Watanabe <i>et al.</i> 1995
		12	24	Present study
	<i>myricoides</i> McVaugh	12	24	Present study
	<i>nelsonii</i> B. L. Rob.	–	24	Present study
	<i>phlebophylla</i> A. Gray	–	24	Present study
	<i>polycephala</i> Bertol.	12	–	Breedlove in Grashoff 1972
		12	–	King <i>et al.</i> 1976
		12II	24	Present study
	<i>pyrolifolia</i> Schlecht.	–	–	
	<i>revoluta</i> B. L. Rob.	–	24	Present study
	<i>rzedowskii</i> McVaugh	12	24	Present study
	<i>salicifolia</i> Cav. var. <i>salicifolia</i>	12	–	Powell & Turner 1963
		12	–	Cronquist in Grashoff 1972
		12	–	Grashoff <i>et al.</i> 1972
		12	–	Keil & Stuessy 1975, 1977
		–	24 + 0–1B	Watanabe <i>et al.</i> 1995
	var. <i>stenophylla</i> (A. Gray) B. L. Rob.	12	–	Turner & Flyr 1966
	12	–	Pinkava & Keil 1977	
	12	24	Present study	
	<i>scabrella</i> Benth.	–	24	
<i>scabrelloides</i> Soejima <i>et</i> Yahara	–	–		
<i>seleriana</i> B. L. Rob.	12	–	Turner & King 1964	
	12II	–	Present study	
<i>subpubescens</i> Lag.				
var. <i>intermedia</i> Grashoff	12	–	Turner & Flyr 1966	
	12	–	Grashoff <i>et al.</i> 1972	
var. <i>subpubescens</i>	12	–	Turner & Flyr 1966	
	12	–	Grashoff <i>et al.</i> 1972	
	12	–	King <i>et al.</i> 1976	
	12	–	Keil & Stuessy 1977	
	12II	24	Present study	
<i>tephrophylla</i> Blake	12	–	Turner & Flyr 1966	
<i>urceolate</i> Grashoff	–	–		
<i>vernicosa</i> Greenm.	–	–		
<i>zacatecana</i> McVaugh	12II	24	Present study	
<i>zephyrantha</i> Grashoff	–	24	Present study	

Appendix II *Continued*

Habit	Species	<i>n</i>	<i>2n</i>	References
Perennial herb	<i>alatipes</i> Sch.-Bip.	–	22	Present study
		–	33	Present study
	<i>ambryolepis</i> (B. L. Rob.) B. L. Rob.	–	24+1~3B	Present study
	<i>caracasana</i> DC.	–	22+1~3B	Present study
		–	33+0~2B	Present study
		<i>ca34I</i>	–	Turner & King 1964
		<i>ca34I</i>	–	Grashoff <i>et al.</i> 1972
	<i>chilapensis</i> Soejima <i>et</i> Yahara	–	22	Present study
	<i>clinipodioides</i> Greenm.	–	48	Present study
	<i>coahuilensis</i> Soejima <i>et</i> Yahara	–	33	Present study
	<i>connata</i> Lag.	–	22+0~2B	Present study
		–	33	Present study
		–	44	Watanabe <i>et al.</i> 1995
		–	44	Present study
	<i>constricta</i> (Grashoff) Soejima, Yahara <i>et</i> Watanabe	–	22+4~8B	Present study
		–	44	Present study
	<i>cordifolia</i> Benth.	–	–	
	<i>crassifolia</i> Soejima <i>et</i> Yahara	–	33	Present study
	<i>cruzii</i> Grashoff	–	–	
	<i>decunmbens</i> (B. L. Rob. <i>et</i> Greenm.) Greene	–	24	Present study
		–	36	Present study
	<i>deltoidea</i> Greene	–	33+0~3B	Present study
		–	44+0~6B	Present study
		–	55+0~5B	Present study
	<i>elator</i> H. B. K.			
	var. <i>elator</i>	11	–	King <i>et al.</i> 1976
		12II	–	Keil & Stuessy 1977
		<i>ca23</i>	–	King <i>et al.</i> 1976
		–	33	Grashoff <i>et al.</i> 1972
		–	33	Present study
		34I	–	Powell & Turner 1963
		34I	–	Turner & King 1964
		<i>ca35I</i>	–	Keil & Stuessy 1977
		–	44	Present study
		<i>ca46II</i>	–	Breedlove in Grashoff 1972
	var. <i>podophylla</i> B. L. Rob.	–	33	Present study
	<i>eupatoria</i> (Spreng.) Willd.	–	24	Grashoff <i>et al.</i> 1972
		–	24	Present study
		<i>ca33I</i>	–	Keil & Stuessy 1977
		34I	–	Powell & Turner 1963
		–	34	Grashoff <i>et al.</i> 1972
		–	36	Watanabe <i>et al.</i> 1995; present study
		–	48	Watanabe <i>et al.</i> 1995; present study
	<i>filodecalloana</i> Soejima, Yahara <i>et</i> Watanabe	–	22	Present study
	<i>hintonii</i> (Grashoff) B. L. Turner (= <i>Metastevia</i> <i>hintonii</i> Grashoff)	–	–	
	<i>hintoniorum</i> B. L. Turner	–	44	Present study
	<i>hypomalaca</i> B. L. Rob.	–	22+0~2B	Present study
	<i>iltisiana</i> Grashoff	–	33	Present study
		34	–	Turner <i>et al.</i> 1961
		–	44	Present study
	<i>incongnita</i> Grashoff	–	33+0~2B	Present study
	<i>isomeca</i> Grashoff	35II	–	Sundberg <i>et al.</i> 1986
		–	33+0~3B	Present study
	–	44+1~4B	Present study	

Appendix II *Continued*

Habit	Species	<i>n</i>	<i>2n</i>	References
	<i>lorullensis</i> H. B. K.	–	24	Present study
		–	34	Grashoff <i>et al.</i> 1972
		–	36	Present study
		–	48	Present study
	<i>karwinskiana</i> Stend.	–	22	Present study
		–	33	Present study
	<i>lasioclada</i> Grashoff	–	–	
	<i>latifolia</i> Benth.	–	33	Present study
		–	44	Present study
		–	55	Present study
		45II	–	Ralston <i>et al.</i> 1989
	<i>lehmannii</i> Hieron.	–	22+0–1B	Present study
	<i>liebmannii</i> Sch.-Bip.	–	33	Present study
	<i>mascotensis</i> Soejima <i>et</i> Yahara	–	44	Present study
	<i>micradenia</i> B. L. Rob.	33I	–	Turner & Flyr 1966
		–	33	Present study
	<i>monardifolia</i> H. B. K.	–	22+0–3B	Present study
		–	33	Watanabe <i>et al.</i> 1995
		–	33+0–2B	Present study
		–	34	Grashoff <i>et al.</i> 1972
	<i>nepitifolia</i> H. B. K. (= <i>suaveolens</i> Lag.)	–	22+3B	Soejima <i>et al.</i> 2001b
		–	33+0–2B	Soejima <i>et al.</i> 2001b
		–	34	Grashoff <i>et al.</i> 1972
		–	35	Grashoff <i>et al.</i> 1972
	<i>neurophylla</i> Robinson <i>et</i> Greenman	–	–	
	<i>oaxacana</i> Soejima <i>et</i> Yahara	–	24	Present study
	<i>oligophylla</i> Soejima <i>et</i> Yahara	–	22	Soejima <i>et al.</i> 2001b
		–	33	Soejima <i>et al.</i> 2001b
	<i>origanoides</i> H. B. K.	11	–	Keil & Stuessy 1975, 1977
		–	22+0–3B	Soejima <i>et al.</i> 2001b
		–	33	Soejima <i>et al.</i> 2001b
		–	34	Grashoff <i>et al.</i> 1972
		–	ca43±1	Grashoff <i>et al.</i> 1972
	<i>orizavensis</i> B. L. Rob.	–	–	
	<i>ovalis</i> (B. L. Rob.) B. L. Rob.	–	–	
	<i>ovata</i> Willd.	11	–	Robinson <i>et al.</i> 1989
		–	22+0–3B	Soejima <i>et al.</i> 2001b
		ca33I	–	Powell & Powell 1978
		–	33	Watanabe <i>et al.</i> 1995
		–	33	Soejima <i>et al.</i> 2001b
		33–34I	–	King <i>et al.</i> 1976
		34I	–	Powell & Turner 1963
		–	34–36	Grashoff <i>et al.</i> 1972
		34I	–	King <i>et al.</i> 1976
		36	–	King <i>et al.</i> 1976
		–	44	Soejima <i>et al.</i> 2001b
		–	51	Grashoff <i>et al.</i> 1972
		–	55	Soejima <i>et al.</i> 2001b
		–	66	Soejima <i>et al.</i> 2001b
	<i>palmeri</i> A. Gray	–	44	Lane & Li 1993
	var. <i>palmeri</i>	–	44	Present study
	<i>pelophila</i> Blake	–	–	
	<i>perfoliata</i> Cronquist	–	22	Present study
	<i>pilosa</i> Lag.	11	22	Watanabe <i>et al.</i> 1995
		11	22+0–4B	Present study
		–	33	Present study

Appendix II *Continued*

Habit	Species	<i>n</i>	<i>2n</i>	References
		34	–	Powell & Turner 1963 (as <i>S. purpurea</i> )
		–	34	Grashoff <i>et al.</i> 1972
		–	44	Present study
	<i>plummerae</i> A. Gray var. <i>plummerae</i>	–	22+1~2B	Present study
		–	34	Grashoff <i>et al.</i> 1972
		–	42	Grashoff <i>et al.</i> 1972
	var. <i>durangensis</i> B. L. Rob.	–	44	Present study
	<i>porphyrea</i> McVaugh	44I	–	Keil & Stuessy 1975
		–	22	Present study
		–	33	Present study
		–	34	Grashoff <i>et al.</i> 1972
		–	44	Present study
		–	55	Present study
	<i>potosina</i> Soejima, Yahara <i>et</i> Watanabe	–	33	Present study
	<i>purpusii</i> B. L. Rob.	–	34	Grashoff <i>et al.</i> 1972
	<i>quiexobra</i> B. L. Turner	–	–	
	<i>reticulata</i> Grashoff	–	22	Present study
		–	33	Present study
	<i>roscii</i> B. L. Rob.	–	44	Present study
	<i>rotundifolia</i> Soejima, Yahara <i>et</i> Watanabe	–	33+0~2B	Present study
	<i>seemannii</i> Sch.-Bip.	–	33+0~1B	Present study
		–	44	Present study
		–	55+2B	Present study
	<i>seemannioides</i> Grashoff	–	22	Present study
	<i>serrata</i> Cav.	44I	–	Lane & Li 1993
	var. <i>arguta</i> B. L. Rob.	–	22	Present study
		–	33	Present study
		34	–	Powell & Turner 1963
		–	34	Grashoff <i>et al.</i> 1972
		44	–	Powell & Turner 1963
		–	48	Grashoff <i>et al.</i> 1972
	var. <i>serrata</i>	–	22	Grashoff <i>et al.</i> 1972
		<i>ca27</i>	–	King <i>et al.</i> 1976
		–	31–37	Grashoff <i>et al.</i> 1972
		–	33	Grashoff <i>et al.</i> 1972
		–	33+0~3B	Watanabe <i>et al.</i> 1995; present study
		34I	–	Powell & Turner 1963
		–	34	Grashoff <i>et al.</i> 1972
		–	44	Present study
		–	55	Present study
		–	66	Present study
	<i>stolonifera</i> Yahara <i>et</i> Soejima	–	22+0~2B	Present study
		–	44	Present study
	<i>talpensis</i> Grashoff	–	–	
	<i>tephra</i> B. L. Rob.	–	33+0~3B	Present study
		–	48	Grashoff <i>et al.</i> 1972
	<i>tomentosa</i> H. B. K.	–	33	Watanabe <i>et al.</i> 1995
		–	33+0~3B	Present study
		–	40–47	Grashoff <i>et al.</i> 1972 (as <i>S. incanescens triangularis</i> Grashoff)
		–	24	Present study
	<i>trifida</i> Lag.	11	–	Turner & King 1964
		11	–	Breedrove in Grashoff 1972
		11	–	King <i>et al.</i> 1976
		11	–	Keil & Stuessy 1977
		–	22	Present study

Appendix II *Continued*

Habit	Species	<i>n</i>	<i>2n</i>	References
	<i>triflora</i> DC.	–	22, 33, 44	Soejima <i>et al.</i> 2001b
		–	34–36	Grashoff <i>et al.</i> 1972
	<i>velutinella</i> Grashoff	–	24	Present study
	<i>viejoana</i> Soejima, Yahara <i>et</i> Watanabe	11	22	Present study
	<i>villaregalis</i> McVaugh	–	–	
	<i>viscida</i> H. B. K.	11	–	Powell & Turner 1963
		–	22	Grashoff <i>et al.</i> 1972
		22II + 11I	–	Turner & Flyr 1966
		–	33	Grashoff <i>et al.</i> 1972
		–	33	Present study
		–	44	Grashoff <i>et al.</i> 1972
Annual herb				
	<i>aschenborniana</i> Sch.-Bip.	11	–	Keil & Stuessy 1977
		–	24 + 0–2B	Present study
	<i>caltepecana</i> B. L. Turner	–	–	
	<i>ecatepecana</i> Soejima, Yahara <i>et</i> Watanabe	12II	–	Present study
	<i>ephemera</i> Grashoff	4II	8	Present study
	<i>lita</i> Grashoff	10II	20	Present study
	<i>mexicana</i> Soejima, Yahara <i>et</i> Watanabe	11II	22	Present study
	<i>micrantha</i> Lag.	11II	–	Grashoff <i>et al.</i> 1972
		–	24	Present study
	<i>mitopoda</i> B. L. Rob.	–	–	
	<i>occidentalis</i> (Grashoff) Soejima, Yahara <i>et</i> Watanabe	–	20	Present study