PN-ARH-767

American Journal of Botany 78(2): 238–246. 1991.

CYTOLOGICAL AND INTERFERTILITY RELATIONSHIPS OF ARACHIS SECTION ARACHIS[1]

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Twenty-nine recently introduced diploid ($2n = 2x = 20$) accessions of section *Arachis* plus an *A. correntina* (Burk) Krap. et Greg. *nom. nud.* control were hybridized to the diploid A-genome species *A. duranensis* Krap. et Greg. *nom. nud.* (acc. 7988), the diploid B-genome species *A. batizocoi* Krap. et Greg. (acc. 9484), and with two subspecies of the A-B genome ($2n = 4x = 40$) *A. hypogaea* cultivars NC 4 and Argentine. Most attempted crosses were successful and the resulting plants were vigorous. However, *A. batizocoi* × accession 30008 hybrids died as seedlings and *A. batizocoi* × accession 30017 produced only dwarf plants. The 710 diploid F_1s from *A. batizocoi* were generally sterile, while those from *A. duranensis* had fertility ranges from 5% to 84%. Meiotic chromosome relationships in diploid crosses were cytologically evaluated in 185 plants plus tester accessions. Most taxa in section *Arachis* have an A genome, only *A. batizocoi* accessions have a B genome, a D genome is represented by accessions 30091 and 30099, and two other genomic groups, represented by accessions 30011 and 30033, may be present in the section. Most cytological differentiation was found among species originally collected in southern and eastern Bolivia. On the other hand, species collected at the extremes of the distribution of section *Arachis* species (northern Argentina to north-central Brazil) were cytologically very similar. Evidence is presented for speciation in *Arachis* being associated with both genetic differentiation and with translocated chromosomes. All taxa in the section except the D-genome species are believed to be cross-compatible with *A. hypogaea*, so germplasm introgression from most *Arachis* species should be possible.

Members of the genus *Arachis* are native to South America and consist of a diverse group of taxa. *Arachis* is distinguished from other genera by flowering above the ground but producing fruits and seeds below the soil surface. The possibly 50 or more species have been grouped into seven sections (Krapovickas, 1969, 1973; Gregory et al., 1973). Most species are diploid ($2n = 2x = 20$), while polyploid ($2n = 4x = 40$) species have evolved independently in sections *Arachis* and *Rhizomatosae* (Smartt and Stalker, 1981).

Species of section *Arachis* are of special interest to both botanists and geneticists because the cultivated peanut, *A. hypogaea* L. ($2n = 4x = 40$), is a member of the group. Other species of this section will hybridize with *A. hypogaea* (Smartt and Gregory, 1967; Gregory and Gregory, 1979; Pompeu, 1983), and the taxonomic closeness makes biosystematic information in section *Arachis* important for germplasm utilization. These taxa are widely distributed in central and southern Brazil, Argentina, Bolivia, Paraguay, and Uruguay (Valls et al., 1985). Distinguishing characters of section *Arachis* are tap roots but no adventitious roots, vertical pegs (modified gynophores), and flowers without red veins on the back of the standard (Gregory et al., 1973; Resslar, 1980). To date, four diploid species have been validly described, and eight additional species names without descriptions commonly appear in the literature (Resslar, 1980). However, many additional species are likely present in *Arachis* germplasm collections among the 60 annual and 98 perennial diploid accessions of section *Arachis* collected between 1936 and 1983 (Valls et al., 1985). A taxonomic revision is greatly needed for both section *Arachis* and for the genus as a whole (Stalker and Moss, 1987).

Based on the analysis of a limited number of accessions (see Smartt and Stalker, 1981; Stalker and Moss, 1987), genomes have been designated cytologically in section *Arachis*. Many species of this group have an A genome, *A. batizocoi* Krap. et Greg. has a B genome, and *A.* sp. (represented by accessions 30091

[1] Received for publication 19 March 1990; revision accepted 17 October 1990.

Paper No. 12108 of the Journal Series of the North Carolina Agricultural Research Service, Raleigh, NC 27695-2643. This research was partially funded by USAID Peanut CRSP grant DAN-4048-G-SS-2065-00.

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and 30099) has a D genome (Smartt and Stalker, 1981; Stalker, 1985). The cultivated peanut and its tetraploid progenitor species *A. monticola* Krap. et Rig. combine both the A and B genomes. Interspecific hybrids can be obtained in the greenhouse among all species in section *Arachis*, although reciprocal crosses can be difficult to make (Gregory and Gregory, 1979). First generation hybrids among diploid A-genome species range in fertility from 20% to 95%, and chromosomes usually associate as ten bivalents (Raman, 1976; Resslar and Gregory, 1979; Stalker and Wynne, 1979; Singh and Moss, 1984). However, when *A. batizocoi* is used as a parent with other taxa, interspecific hybrids are sterile, and a range of two to ten univalents and laggards is observed (Smartt, Gregory, and Gregory, 1978a; Stalker and Wynne, 1979; Singh and Moss, 1984).

The somatic chromosomes of *Arachis* species are small and mostly median or slightly submedian (Stalker and Dalmacio, 1981; Singh and Moss, 1982). Karyotype data generally support the A and B genomic groupings which were designated based on cross-compatibility data and the presence or absence of a very small chromosome pair (Smartt, Gregory, and Gregory, 1978a, b).

The purpose of this paper is to present results from a study of section *Arachis* taxa using a larger number of accessions than has been attempted previously. Morphological, cytological, and geographical information has been used to evaluate variability and systematic relationships of taxa in the group. The objective of this investigation is to further define species variation cytologically in section *Arachis* by analyzing fertility and meiotic relationships of F_1 hybrids.

MATERIALS AND METHODS

Thirty-two accessions of *Arachis* species that had been collected in South America (Gregory et al., 1973; Simpson and Higgins, 1984) (Table 1; Fig. 1) were used in crossing experiments. Twenty-one accessions were obtained from D. Banks, U.S. curator for *Arachis* species at Stillwater, Oklahoma in 1979 and 1981; six were obtained from C. Simpson of Texas A&M University, Stephenville, Texas during 1982; and two accessions (6536 and 7762) were collected in 1984 by the senior author. In addition, the previously identified perennial A-genome species *A. correntina* Krap. et Greg. *nom. nud.* (GKP 9530, PI 262808) plus annual species *A. duranensis* Krap. et Greg. *nom. nud.* K 7988, PI 219823 and *A. batizocoi*, K 9484, PI 298639 were already in the North Carolina collection

Fig. 1. Approximate locations in South America of original collection sites for 32 *Arachis* accessions.

prior to 1979. A cluster analysis was conducted on 27 seed-producing taxa using a SAS computer program based on the unweighted pair group method with arithmetic averages (Sneath and Sokal, 1973). Ten leaf, 11 leaflet, eight pod, six seed, two peg, seven flower, and ten branch or plant habit traits were scored on each of three plants at 90 to 95 days after planting.

The diploid species *A. duranensis* (A genome) and *A. batizocoi* (B genome) were used as female parents in crossing programs during the summers of 1980 to 1984 with the 30 accessions introduced to North Carolina from South America; *A. correntina* accession 9530; and each other, in reciprocal. Flowers were hand-emasculated during the afternoon between 3 and 5 p.m. and then pollinated the following morning between 8 and 10 a.m. Non-emasculated flowers were removed from female parents between 7 and 8 a.m. to prevent self-fertilization. Seedlings found at the time of harvest (60 days after the last pollination for a cross) were transplanted into a pot and maintained in the greenhouse. Seeds from crosses were harvested, dried, and planted the following spring in the greenhouse. Hybrids were confirmed by morphological observations and by estimating male fertility levels for at least 300 pollen grains of two flowers col-

TABLE 1. *Accessions of* Arachis *and their collection sites used in crosses and cytological analyses*

Entry	Collector[a]	Accession number	P.I. No.	Species	Latitude	Longitude	Locality
1.	VSGr	6331	476045	*A. helodes*	16.20	56.44	Mato Grosso, Brazil
2.	VSGr	6352	476109	*A. helodes*	15.56	57.47	Mato Grosso, Brazil
3.	VKRSv	6536	—	*A.* sp.	9.47	48.70	Goias, Brazil
4.	VSStGyW	7762	—	*A.* sp.	15.35	52.72	Mato Grosso, Brazil
5.	K	7988	219823	*A. duranensis*	22.19	63.43	Salta, Argentina
6.	K	9484	298639	*A. batizocoi*	20.50	63.14	Santa Cruz, Bolivia
7.	GK	9530	262808	*A. correntina*	27.33	58.46	Corrientes, Argentina
8.	GK	30001	468141	*A. diogoi*	17.40	57.45	Mato Grosso, Brazil
9.	GK	30005	468142	*A. diogoi*	17.47	57.40	Mato Grosso, Brazil
10.	GK	30006	468150	*A.* sp.	18.08	57.30	Mato Grosso, Brazil
11.	GK	30008	468152	*A.* sp.	19.02	56.39	Mato Grosso, Brazil
12.	GK	30011	468154	*A.* sp.	19.11	57.29	Mato Grosso, Brazil
13.	GK	30017	468159	*A.* sp.	20.21	55.51	Mato Grosso, Brazil
14.	GK	30031	468146	*A. helodes*	16.02	57.07	Mato Grosso, Brazil
15.	GK	30033	468166	*A.* sp.	16.18	57.28	Mato Grosso, Brazil
16.	GK	30034	468167	*A.* sp.	16.08	57.18	Mato Grosso, Brazil
17.	GK	30035	468168	*A.* sp.	16.05	57.15	Mato Grosso, Brazil
18.	GKBSPSc	30064	468200	*A.* sp.	24.23	65.70	Jujuy, Argentina
19.	GKBSPSc	30076	468322	*A. ipaensis*	21.00	63.24	Tarija, Boliva
20.	GKBSPSc	30081	468327	*A. batizocoi*	19.40	63.41	Santa Cruz, Bolivia
21.	GKBSPScZ	30084	468330	*A.* sp.	17.19	63.18	Santa Cruz, Bolivia
22.	GKBSPScZ	30085	468331	*A.* sp.	17.23	63.28	Santa Cruz, Bolivia
23.	GKSSc	30091	468336	*A.* sp.	16.29	62.05	Santa Cruz, Bolivia
24.	GKSSc	30099	468342	*A.* sp.	16.36	61.01	Santa Cruz, Bolivia
25.	GKSSc	30102	468345	*A.* sp.	16.10	61.02	Santa Cruz, Bolivia
26.	GKPSc	30106	468354	*A.* sp.	27.22	57.05	Misiones, Paraguay
27.	GKPSc	30108	468356	*A.* sp.	25.26	57.16	Central, Paraguay
28.	GKSPScGb	3500[illegible]	475873	*A.* sp.	17.19	63.18	Santa Cruz, Bolivia
29.	GKSPScGb	35003	475875	*A.* sp.	17.22	63.29	Santa Cruz, Bolivia
30.	KSSc	36031	476010	*A.* sp.	17.50	60.45	Santa Cruz, Bolivia
31.	KSSc	36032	476011	*A. cardenasii*	17.58	60.47	Santa Cruz, Bolivia
32.	KSSc	36035	476014	*A. cardenasii*	17.57	60.50	Santa Cruz, Bolivia

[a] Abbreviations for collectors are as follows: B = D. J. Banks, G = W. C. Gregory, Gb = R. W. Gibbons, Gr = A. Gripp, Gy = P. J. Godoy, K = A. Krapovickas, P = J. Pietrarelli, R = V. R. Rao, S = C. E. Simpson, Sc = A. Schinini, St = H. T. Stalker, Sv = G. P. Silva, V = J. F. M. Valls, W = W. L. Werneck, and Z = H. Zuritz O.

lected on different days for each plant (Pittenger and Frolik, 1951).

F_1 hybrids were grown in pots in the greenhouse during the summers of 1981 to 1988 to analyze meiotic chromosome relationships. Flower buds were collected between 8 and 9 a.m. and then fixed and stored in Carnoy's solution. Anthers were squashed in acetocarmine using plastic needles to avoid adding iron into solutions. Chromosome associations were confirmed by observing the early metaphase stage in microsporocytes.

Twenty-three diploid *Arachis* accessions were also hybridized with two *A. hypogaea* cultivars representing subspecies *hypogaea* var. *hypogaea* (cv. NC 4) and subspecies *fastigiata* var. *vulgaris* (cv. Argentine). Cultivars were initially used as the female parent, but reciprocal hybrids were also attempted. Interspecific hybrids were identified by observing plant morphology, by scoring male fertility as described above, and by observations of nonpegging plants.

RESULTS

Accessions were chosen for study that represented a range of variability among introduced taxa of section *Arachis*. Of the 32 entries used for cytological and crossing studies, 26 produced seeds and were measured for traits used in a cluster analysis (Fig. 2). Based on the grouping of accessions of species *A. batizocoi* (9484 and 30081) and *A. diogoi* (30001 and 30005), at least ten species may be represented among entries in the cluster analysis. Accessions of other species were more variable, however, and members of *A. cardenasii* and *A. helodes* did not form good groups. The six accessions for which seeds were not available (30031, 30033, 30035, 30102, 30106, and 30108) included a third entry of *A. helodes* (30031) and at least three additional unique taxa.

Crossing programs were conducted during a 7-yr period to acquire the F_1 hybrids listed in Tables 2 and 3. Many hybrids were difficult to

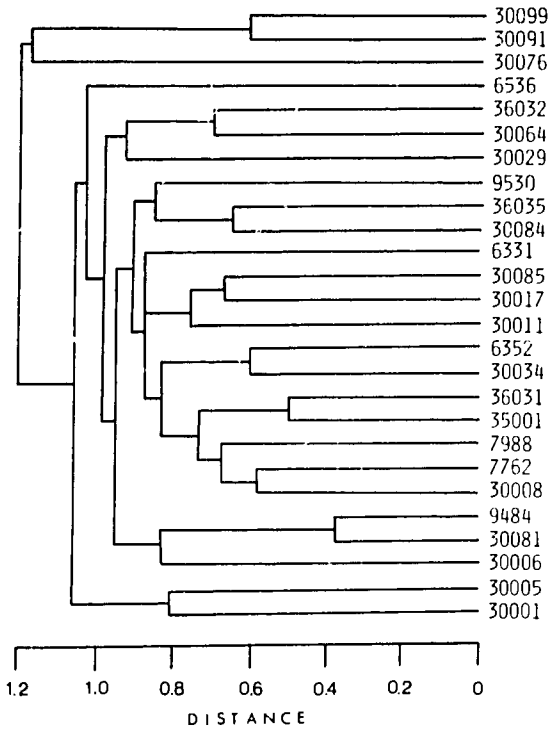

Fig. 2. Phenogram from cluster analysis of 26 section *Arachis* accessions based on taxonomic distance coefficients using 23 reproductive and 31 vegetative traits.

obtain because respective male parents had poor flower production when female plants were blooming. Further, even though care was taken to remove nonemasculated flowers, occasional plants resulting from selfed seeds were discarded. For most cross combinations the maturing hybrid seeds germinated by the time of normal harvest (about 60 days after pollination), whereas selfed seeds always remained dormant. The following results are reported according to genomic classification of the female parent used in crossing programs.

A-genome interspecific hybrids — Twenty-eight of 30 attempted interspecific hybrids between the A-genome species *A. duranensis* and other taxa in the section *Arachis* were obtained (Table 2). A total of 3,495 pollinations yielded 322 hybrids, or an overall success rate of 9.91%. The two accessions of *A. diogoi* (30001 and 30005) and an unnamed species accession 30017 were the easiest to cross with *A. duranensis* where more than 25% of pollinations resulted in hybrids. On the other hand, 51 and 81 pollinations with accessions 30035 and 30076, respectively, did not result in hybrids.

Pollen viability of the interspecific hybrids varied greatly among the individual cross combinations, but generally ranged between 35% and 60% (Table 2). When a plant had above 50% fertility, pegging was usually observed on hybrid plants. Very low fertility levels were observed for plants of several hybrid combinations, including crosses with accessions 9484, 30084, 30091, and 30099 (Table 2). Because 9484 (*A. batizocoi*) is the B-genome control in the crossing set, the results for this cross were expected. Surprisingly, 12 plants of a second *A. duranensis* × *A. batizocoi* (accession 30081) hybrid were semifertile and averaged 28.14% stained pollen. Since 600 pollen grains were scored for each of the 12 plants, the percentage has a high confidence level. The remaining three hybrids which were mostly sterile accessions included two entries (30091 and 30099) which are members of the same species and accession 30084 which represents a second taxa.

Meiotic chromosome pairing in F_1 plants was next analyzed to better evaluate species relationships. For comparisons, pollen mother cells (PMCs) of selfed *A. duranensis* averaged 0.05 I + 9.98 II and 19.63 chiasmata, whereas *A. duranensis* × *A. batizocoi* averaged 5.30 I + 7.35 II and 7.99 chiasmata per PMC. Thus, expectations are for a high number of bivalents and high chiasmata frequency in PMCs for closely related species and significantly lower frequencies for more distantly related taxa.

A low, but consistent, number of univalents was observed across the group of 90 F_1 hybrids between 28 accessions and *A. duranensis*. This indicates that at least some differentiation has occurred between the genome of *A. duranensis* and the other taxa. However, most of the hybrids had high chiasmata frequencies and must be considered to have the same A genomic complement as *A. duranensis* (Table 2).

Although most F_1 hybrids were cytologically very similar to the A genome of *A. duranensis*, several hybrids were observed with cytological abnormalities such as univalents and laggards—e.g., the two *A. batizocoi* (9484 and 30081) crosses. Although the *A. duranensis* × *A. batizocoi* (accession 30081) hybrid had 28% pollen stained, the five cytologically analyzed plants averaged 7.63 I + 6.17 II and 8.41 chiasmata per PMC. Stained pollen is thus an indicator, but not truly definitive of cytological homology in *Arachis*. Univalents and/or low chiasmata frequencies were also observed for hybrids between *A. duranensis* and accessions 30011, 30033, 30091, 30099, and 30102.

In addition to univalents and bivalents, multivalents were observed in the two hybrid combinations 7988 × 30008 and 7988 × 30085. The frequency of trivalents or quadrivalents was very low at 0.1 per PMC in the two crosses (Table 2). The most likely cause of these mul-

TABLE 2. *Pollen stainability and chromosome associations of first generation hybrids between A-genome* A duranensis *Krap. et Greg.* nom. nud. *and section* Arachis *accessions*

Cross (7988 ×)	Number		Pollen stainability (%)	Meiosis						
				Number		Average				
	Pollinations	Hybrids		Plants	Cells	I	II	III	IV	Chiasmata
⊗	—	—	98.0	6	171	0.05	9.98	—	—	19.63
6331	52	5	66.8	2	130	0.05	9.98	—	—	18.76
6352	45	1	49.8	1	50	0	10.00	—	—	19.20
6536	138	7	68.4	2	151	0.21	9.89	—	—	18.60
7762	134	7	43.7	3	187	0.04	9.98	—	—	19.52
9484	113	2	6.2	1	75	5.30	7.35	—	—	7.99
9530	46	7	83.5	4	204	0.06	9.99	—	—	18.41
30001	86	31	43.6	4	232	0	10.00	—	—	18.28
30005	85	22	39.0	4	148	0.34	9.83	—	—	17.16
30006	160	8	11.8	4	149	0.11	9.94	—	—	17.22
30008	198	14	23.3	4	150	0.21	9.86	0.01	0.01	18.76
30011	215	11	26.5	4	199	7.59	6.25	—	—	7.23
30017	156	31	63.4	4	164	0	10.00	—	—	17.24
30031	54	16	36.5	3	155	0.90	9.50	—	—	15.60
30033	35	1	33.4	1	50	2.76	8.62	—	—	11.82
30034	117	5	35.6	3	99	0.08	9.96	—	—	16.98
30035	51	0	—	—	—	—	—	—	—	—
30064	142	6	58.2	2	78	1.35	9.33	—	—	16.74
30076	81	0	—	—	—	—	—	—	—	—
30081	79	12	28.1	5	150	7.63	6.17	—	—	8.41
30084	139	11	5.7	2	49	0.78	9.61	—	—	17.10
30085	149	9	36.4	5	191	0.14	9.92	—	0.01	18.20
30091	132	4	10.8	1	75	10.00	5.10	—	—	5.80
30099	108	2	5.3	1	50	7.00	6.50	—	—	7.50
30102	187	11	46.3	6	231	0	10.00	—	—	13.85
30106	133	22	72.5	4	200	0.01	10.00	—	—	18.39
30108	192	32	54.7	4	307	0	10.00	—	—	17.27
35001	113	11	60.9	3	174	0.24	9.89	—	—	19.50
35003	186	18	57.0	4	156	0.15	9.92	—	—	17.93
36031	66	6	54.2	4	199	0.05	9.97	—	—	18.52
36032	23	2	47.3	1	50	0	10.00	—	—	19.98
36035	80	8	55.7	4	150	0.15	9.92	—	—	17.93
Totals	3,495	322		90	4,203					

tivalents is reciprocal translocations between *A. duranensis* and the other taxa.

A. batizocoi hybrids—There were 3,176 pollinations that resulted in 388 interspecific hybrids with *A. batizocoi*. This represented a 12.4% success rate which was slightly higher than when the same accessions were crossed with *A. duranensis*. One intraspecific hybrid was also obtained (9484 × 30081) at approximately the same success rate as interspecific hybrids (Table 3). Only accessions 6352 and 6536 failed to hybridize with *A. batizocoi* even though 94 and 76 pollinations, respectively, were made. Further, all F_1 hybrids with accession 30008 died as seedlings, and F_1 hybrids with accession 30017 were extremely small and failed to flower. At the other morphological extreme were plants of the cross 9484 × 30011 which had gigas plant parts, including branches that reached more than 7 m in length.

Pollen fertility was very low, approaching zero, in all interspecific hybrids with *A. batizocoi* (Table 3). Further, pegs were never observed on interspecific *A. batizocoi* hybrids which indicated complete reproductive sterility. However, the intraspecific hybrids with accession 30081 averaged 85.7% pollen stained, and many pegs were produced on all plants.

PMCs of selfed *A. batizocoi* plants all had ten bivalents and averaged 19.06 chiasmata. The B × A genome cross 9484 × 7988 averaged 7.14 I + 6.45 II, with a chiasmata frequency of 8.98. Although more than half of the chromosomes paired, many bivalents were only loosely associated. Most interspecific hybrids between *A. batizocoi* and the other 29 accessions had six to eight bivalents per PMC, but chiasmata frequencies fewer than 12 (Table 3). Thus, approximately half of the potential chromosome associations were observed when *A. batizocoi* was used as a female parent with other species in section *Arachis*.

The intraspecific hybrid 9484 × 30081 (B × B genomes) averaged 0.09 I + 9.75 II + 0.10 IV per PMC and had a high chiasmata

TABLE 3. *Pollen stainability and chromosome associations of first generation hybrids between B-genome* A. batizocoi *Krap. et Greg. and section* Arachis *species*

Cross (9484 ×)	Number		Pollen stainability (%)	Meiosis						
				Number		Average				
	Pollinations	Hybrids		Plants	Cells	I	II	III	IV	Chiasmata
⊗	—	—	99.1	3	83	0	10.00	—	—	19.06
6331	83	3	0.6	4	217	10.37	4.82	—	—	6.83
6352	9	0	—	—	—	—	—	—	—	—
6336	76	0	—	—	—	—	—	—	—	—
7762	38	7	1.3	5	156	6.46	6.77	—	—	10.91
7988	118	8	1.3	3	150	7.14	6.45	—	—	8.98
9530	116	10	1.6	7	225	7.38	6.31	—	—	9.51
30001	69	35	0.4	4	139	6.50	6.76	—	—	7.71
30005	60	37	0	1	50	5.70	7.14	—	—	7.40
30006	87	11	0.4	3	215	6.26	6.85	—	—	8.50
30008	307	5[a]	—	—	—	—	—	—	—	—
30011	128	21	6.8	4	150	3.35	8.32	—	—	10.18
30017	202	4[b]	—	—	—	—	—	—	—	—
30031	83	11	4.1	3	165	7.32	6.33	—	—	7.91
30033	—	—	—	—	—	—	—	—	—	—
30034	123	6	7.8	—	—	—	—	—	—	—
30035	165	3	0.5	2	101	6.77	6.61	—	—	8.85
30064	51	4	0	—	—	—	—	—	—	—
30076	106	7	0.8	2	57	4.42	7.79	—	—	12.47
30081	84	7	85.7	4	136	0.09	9.75	—	0.10	19.07
30084	9-	24	0.2	9	213	7.87	6.07	—	—	9.43
30085	119	14	0	4	150	9.27	5.36	0.01	—	7.70
30091	233	19	1.8	6	277	3.29	8.36	—	—	12.08
30099	52	4	2.1	2	101	2.38	8.81	—	—	10.61
30102	55	6	1.3	3	186	5.83	7.08	—	—	9.33
30106	111	39	0.9	6	150	6.35	6.83	—	—	11.00
30108	131	24	0.8	3	214	4.91	7.55	—	—	10.81
35001	30	16	1.7	4	150	8.23	5.87	—	—	9.63
35003	125	23	0.4	5	150	10.36	4.82	—	—	7.14
36031	66	11	0	3	140	8.24	5.84	0.02	—	8.33
36032	90	13	2.4	2	134	6.60	6.60	—	—	9.20
36035	80	15	1.0	6	150	6.85	6.57	—	—	9.69
Totals	3,176	388		95	3,776					

[a] Seedlings died.
[b] Dwarf seedlings without flowers.

frequency equal to that of selfed *A. batizocoi* (Table 3). Multivalents were also observed in the two interspecific hybrids 9484 × 30085 and 9484 × 36031, but at very low frequency (Table 3). Translocations are likely causes of the multivalents, at least in the intraspecific hybrids.

A. hypogaea hybrids—Cultivars NC 4 and Argentine were hybridized as female parents with 23 and 22 diploid accessions of section *Arachis*, respectively (Table 4). In addition, reciprocal crosses were made using 15 and 14 diploids with NC 4 and Argentine, respectively (Table 4). Hybrids were easily identified based on intermediate plant morphology and very low pollen stainability. Although interspecific hybrids were generally vigorous, seminecrotic F_1s were also observed for the hybrid combination Argentine × accession 30102. Reciprocal crosses were not attempted because this diploid accession does not set pegs or pods in the greenhouse in North Carolina. Similar phenotypes have also been observed in other cultivated × diploid species crosses, such as NC 4 × *A. chacoense* Krap. et Greg. *nom. nud.* (accession GKP 10602), but not for the reciprocal hybrid (unpublished data).

Although the cultivars were successfully used in crosses with most taxa, success rates varied significantly among the two cultivars and among reciprocal crosses. *Arachis hypogaea* is a 'good' female parent when used in crosses with accessions 9530, 30005, 30008, 30031, and 30108, but a 'poor' male parent. However, the diploid accessions 9484 and 30011 were much better female parents when hybridized with either cultivar. Overall, when NC 4 was used as a female parent, 3.01% of pollinations were successful, whereas only 1.07% of the reciprocal pollinations resulted in hybrids. A similar trend was observed for Argentine where

TABLE 4. *Number of hybrids produced between* Arachis hypogaea *L. and diploid accessions of secton* Arachis

Cross	No. pollinations	No. hybrids	% Hybrids
NC 4 × 6331	43	0	0
Argentine × 6331	65	16	16.8
NC 4 × 6352	39	2	5.1
Argentine × 6352	59	10	16.9
NC 4 × 6536	79	0	0
NC 4 × 7762	42	1	2.3
Argentine × 7762	18	0	0
Argentine × 7988	137	11	8.0
NC 4 × 7988	145	1	0.6
7988 × NC 4	113	3	2.7
7988 × Argentine	150	8	5.3
NC 4 × 9484	219	0	0
Argentine × 9484	158	3	1.9
9484 × NC 4	78	5	6.4
9484 × Argentine	99	3	3.0
NC 4 × 9530	127	2	1.6
Argentine × 9530	104	9	8.7
9530 × NC 4	299	0	0
9530 × Argentine	237	0	0
NC 4 × 30005	161	6	3.7
Argentine × 30005	115	5	4.3
30005 × NC 4	268	0	0
30005 × Argentine	216	1	0.5
NC 4 × 30006	86	9	10.5
Argentine × 30006	96	6	6.3
30006 × NC 4	144	6	4.2
30006 × Argentine	131	2	1.5
NC 4 × 30008	185	6	3.2
Argentine × 30008	203	12	5.9
30008 × NC 4	232	0	0
30008 × Argentine	237	1	0.4
NC 4 × 30011	172	0	0
Argentine × 30011	194	2	1.0
30011 × NC 4	185	13	7.0
30011 × Argentine	254	28	11.0
NC 4 × 30017	104	2	1.9
Argentine × 30017	97	5	5.1
30017 × NC 4	405	1	0.2
30017 × Argentine	395	1	0.3
NC 4 × 30031	74	2	2.7
Argentine × 30031	106	2	1.9
30031 × NC 4	294	0	0
30031 × Argentine	258	0	0
NC 4 × 30034	125	9	7.2
Argentine × 30034	82	5	6.1
30034 × NC 4	99	0	0
NC 4 × 30035	110	2	1.8
Argentine × 30035	109	8	7.3
30035 × NC 4	158	0	0
30035 × Argentine	81	1	1.2
NC 4 × 30076	76	0	0
NC 4 × 30081	182	1	0.5
Argentine × 30081	224	0	0
30081 × NC 4	66	0	0
30081 × Argentine	50	5	10.0
NC 4 × 30085	50	0	0
Argentine × 30085	53	1	1.9
NC 4 × 30091	161	0	0
Argentine × 30091	141	0	0
30091 × NC 4	98	0	0
30091 × Argentine	84	0	0
NC 4 × 30099	105	0	0
Argentine × 30099	125	0	0
30099 × NC 4	57	0	0
30099 × Argentine	54	0	0
NC 4 × 30102	52	13	25.0
Argentine × 30102	115	11[a]	9.5
NC 4 × 30106	78	11	14.1
Argentine × 30106	101	10	9.9
NC 4 × 30108	43	7	16.2
Argentine × 30108	24	7	29.1
30108 × NC 4	114	0	0
30108 × Argentine	111	0	0

[a] Seminecrotic plants.

5.25% of pollinations produced hybrids when it was used as the female, but only 2.12% when the diploid species were females. Although female plants of the subsp. *fastigiata* var. *vulgaris* cultivar appeared to be a better parent than the subsp. *hypogaea* var. *hypogaea* genotype, the higher percentage of 2.12 for Argentine males was largely due to the one cross 30011 × Argentine, where more than half of the total hybrid progenies came from this combination.

Hybrids were not obtained for any pollinations when accessions 30076, 6536, 30091, or 30099 were used as parents. For 30076 and 6536 the failure to obtain hybrids was likely due to using only cultivar NC 4 and not attempting reciprocals. However, 1,357 pollinations with the accessions 30091 and 30099 were made with both Argentine and NC 4, in reciprocal. Thus, the species represented by these two accessions can be considered as truly cross-incompatible with *A. hypogaea*.

DISCUSSION

A considerable range of morphological variation was observed among the 32 accessions of section *Arachis* species. Obvious differences were observed for leaf shapes, flower sizes, growth habits, presence or absence of flowers on the mainstem, sequential vs. alternate arrangement of flowers on branches, pod reticulation, and seed size and shape. Although four species have been botanically described and eight others named in section *Arachis* (Resslar, 1980), the observations indicated that many additional species should be named.

In general, *A. duranensis* had a high level of cytological homology with other taxa in section *Arachis*. Excluding the two *A. batizocoi* hybrids, 19 of 27 other successful crosses had chiasmata frequencies ranging between 17 and 20. Although these taxa could be considered the same biological species, most of the taxa are difficult to cross and the resulting hybrids

are semisterile. Genic, rather than cytological, differentiation is thus proposed as the mechanism for speciation in this group of A-genome taxa.

In addition to hybrids with very close cytological homology to *A. duranensis*, the three accessions 30033, 30102, and 30031 had chiasmata frequencies of 11.82, 13.85, and 15.60, respectively. Hybrids with accession 30033 had many univalents, and obvious differentiation among the genomes has occurred. Because plants of accession 30033 died before further analyses, the status of this taxon is unclear. However, the hybrids *A. duranensis* × 30031 and × 30102 averaged 0.09 and no univalents, respectively. The chromosomes have obviously differentiated, but pairing occurs between all homologues, and separation into individual genomic groups is unwarranted. Further, accession 30031 is a representative of species *A. helodes* Martius ex Krap. et Rig., but two other accessions of this taxon (6331 and 6352) had almost perfect homology when hybridized with *A. duranensis* (Table 2). Thus, cytological differentiation may have occurred between accessions of *A. helodes* as well as between *A. duranensis* and this species.

The only accessions crossed with *A. duranensis* that had chiasmata frequencies as low as *A. duranensis* (A genome) × *A. batizocoi* (B genome) hybrids were with the three unnamed species represented by 30091 or 30099, 30011, and 30033. Because accessions 30091 and 30009 produced sterile hybrids with both genome testers, chiasmata frequencies were low, and hybrids were not obtained with *A. hypogaea*; this species represents a unique genome as previously suggested by Stalker (1985). Although 30011 is cytologically different from both the A and B genome testers, hybrids were obtained with the tetraploid species *A. hypogaea*. The genomic classification is somewhat complicated if one assumes that *A. hypogaea* combines the A and B genomes, but accession 30011 is cytologically of equal distance from both the A- and B-genome diploids. Further, preliminary karyotypic data indicate that the chromosomes of this unnamed species (30011) are generally symmetrical, which corresponds to the 'normal' A genome. Perhaps the species represented by accession 30011 is a true intermediate of both the A and B genomic groups. All plants of accessions 30033 died, so further characterization was not possible during this investigation.

Most cytological variation was observed among species collected in Bolivia as seen by the presence of A-, B-, and D-genome species in this region. Although the accession of *A. duranensis* used as the A-genome tester was from northern Argentina (while several accessions came from the north-central part of Brazil), a high level of chromosome homology existed between the accessions (for example, the northernmost taxa of 6536). The conclusion drawn from these results is that members of section *Arachis* with an A genome, or their immediate ancestors, had an early and widespread distribution. Cytological differentiation among species then occurred in isolated areas—for example, in southern and eastern Bolivia.

Observations of multivalents were not expected in hybrids between diploid species and thus warrant further attention. Quadrivalents were observed between *A. duranensis* (7988) and accessions 30008 and 30085 and between *A. batizocoi* (9484) and accessions 30081, 30085, and 36031. Although the frequency of multivalents was low, they have also been observed in other diploid crosses by Singh and Moss (1984). The most likely explanation for quadrivalents in the interspecific crosses is the presence of reciprocal translocation heterozygotes. Even though a higher quadrivalent frequency from those observed would be expected in the heterozygotes, the small size of *Arachis* chromosomes could have restricted multivalent formation. Translocations are thus hypothesized as a mechanism by which cytological differentiation and subsequently speciation occurs among taxa of the genus.

In summary, analysis of F_1 interspecific hybrids indicated that cytological groups can be distinguished in section *Arachis*, where most have an A genome; only *A. batizocoi* accessions have thus far been found with a B genome; and a D genome is represented by accessions 30091 and 30099. Possibly additional genomic groups are represented by 30011 or 30033. Thus, introgression of germplasm from most species of section *Arachis* to the cultivated species should not be inhibited by genomic isolation of taxa in section *Arachis*.

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