Karyotype Analysis and Relationships Among Varieties of *Arachis hypogaea* L.¹

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The cultivated peanut, *Arachis hypogaea* L., is an allotetraploid species ($2n=40$) grown in tropical and warm temperate regions. Peanuts are rich in both oil and protein and make a major contribution to human nutrition. *Arachis hypogaea* is a highly variable species with a primary center of origin in southern Bolivia or northern Argentina (Gregory and Gregory 1976). Krapovickas (1969, 1973) described five secondary centers of diversity which are to some extent characteristic of peanut varieties. A tertiary center of diversity also exists in Africa (Gibbons *et al.* 1972). The species has been subdivided into two subspecies (Table 1) and evidence for some genetic isolation between the groups exists (Krapovickas 1973, Wynne 1974). Each subspecies has two varieties (Krapovickas 1969, 1973) as follows: subsp. *hypogaea* var. *hypogaea* and var. *hirsuta* and subsp. *fastigiata* var. *fastigiata* and var. *vulgaris*. In addition, var. *nambyquarae* was given varietal status by Gregory *et al.* (1973), but its classification as a unique group versus a type of var. *hypogaea* is still unresolved because subsequent publications by Gregory and Gregory (1976), Gregory *et al.* (1973) and Wynne and Gregory (1980) follow the convention of only four *A. hypogaea* varieties.

The chromosomes of *A. hypogaea* are small and mostly metacentric (Ghimpu 1930). Husted (1933, 1936) observed a distinctively small chromosome pair plus a chromosome pair with a secondary constriction. However, he was unable to differentiate the other *A. hypogaea* chromosomes. Babu (1955) later reported several types of secondary constrictions in peanut cultivars and D'Cruz and Tankasale (1961) cytologically distinguished four peanut cultivars. Singh and Moss (1982) also illustrated karyotypes of *A. monticola* and two *A. hypogaea* cultivars and showed differences among genotypes.

Arachis hypogaea probably originated from a wild allotetraploid species (Krapovickas and Rigoni 1957, Smartt and Gregory 1967). Since *A. monticola* Krap. et Rig. grows near the proposed center of origin and is the only other tetraploid species cross-compatible with *A. hypogaea*, it is the most likely progenitor of the cultivated peanut (Krapovickas 1973). Furthermore, diploid ancestors of *A. hypogaea* probably belong to section *Arachis* of the genus since only this group of species will hybridize with the cultigen (Gregory and Gregory 1976, Smartt *et al.* 1978a, b). *Arachis hypogaea* has an A and B genome which corresponds to the genomic make-up of diploid species of section *Arachis* (Smartt *et al.* 1978a, b).

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Most diploid species of section *Arachis* have the A genome while *A. batizocoi* is the only one known with a B genome (Smartt *et al.* 1978a, Stalker and Dalmacio 1981). Although several species have been suggested as progenitors of *A. hypogaea*, according to Smartt and Stalker 1982, the true ancestors have not been positively identified.

The objective of this investigation was to identify the morphology of *A. hypogaea* chromosomes to derive a standard karyotype for the cultivated species. Variability among varieties of *A. hypogaea* was then evaluated. The karyotypes of *A. hypogaea* and *A. monticola* were also compared to the chromosomes of diploid species of section *Arachis* to identify possible progenitors of the cultivated species.

Materials and methods

Nine cultivars of *A. hypogaea* ($2n=40$), including representatives of the four botanical varieties plus two collections of *A. monticola* ($2n=40$) were karyotyped (Table 1). Both cultivars grown in the USA and unimproved genotypes from South

Table 1. Subspecific classification of *A. hypogaea* L. after Krapovickas (1969) and germplasm cytologically analyzed

Name	Type	Differences	Collection analyzed	Origin
Subsp. <i>hypogaea</i>	—	Reproductive axis absent on main stem; alternating pairs of reproductive and vegetative axis on lateral branches		
var. <i>hypogaea</i>	Virginia	Short branches with little pubescence	NC 4 NC 6 Nambyquarae	USA USA NW Brazil
var. <i>hirsuta</i>	Peruvian runner	Long pubescent branches	PI 158850	China
Subsp. <i>fastigiata</i>	—	Reproductive axis on main stem, continuous runs of reproductive axis on lateral branches		
var. <i>fastigita</i>	Valencia	Little branched	Tennessee Red NC Acc. 17090	USA Paraguay
var. <i>vulgaris</i>	Spanish	More branched, small seeds	Argentina Spanhoma PI 262000	USA USA Paraguay
<i>A. monticola</i>	—	Biarticulated fruit	Coll. K7264 Coll. GKBSPPSc 30062	NW Argentina NW Argentina

America were analyzed to estimate variability within varieties. Root tips were collected from seedlings germinated in sand. Materials were pretreated 15 minutes in a saturated solution of paradichlorobenzene, hydrolyzed in 1 N HCl, stained in 2% aceto-orcein for 1 hour, squashed in an oil press at 211 kg/cm², dehydrated in an alcohol series after removal of cover slips, and permanently mounted with Per-

mount. Observations were from cells in the early metaphase stage of mitosis. Fifteen cells were analyzed for each genotype and standard errors calculated. Photomicrographs were taken of all cells and printed at $4000\times$. Chromosomes were then cut from photomicrographs, homologues paired based on heterochromatic regions, differential staining of chromosome arms, chromosome lengths and centromere positions, and then ordered from 1 to 20 based on chromosome morphology and total length. The longest chromosome was designated number 1 and the shortest chromosome was number 20. The length of the short and long chromosome arms was recorded and three centromere-position types designated as follows: 0.80 to 1.00=median, 0.65 to 0.79=slightly submedian, and 0.33 to 0.64=submedian. A principal component analysis of the nine *A. hypogaea* genotypes plus *A. monticola* was conducted using the calculated short/long chromosome arm ratios to better estimate similarities among collections.

Results

The homologues within single cells of most analyzed genotypes were relatively easy to pair based on absolute length, centromere position and differential staining of heterochromatic regions. Although absolute chromosome lengths appear nearly as a continuum from 1 through 20, the six to nine asymmetrical chromosomes interspersed throughout the genome made ordering the homologues possible. When two chromosomes had nearly identical lengths, they were arbitrarily, but consistently, ordered within each cultivar so that one was designated as the shorter chromosome of the set. However, several chromosome pairs had very similar lengths and morphology and they were nearly impossible to differentiate, for example, chromosomes 7 and 8 or 13, 14 and 15. Although care was taken to analyze cells at the same stage of mitosis, the average length of chromosome 1 ranged from $3.33\text{ }\mu\text{m}$ for Spanhoma to $4.16\text{ }\mu\text{m}$ for PI 158850. When the ratio of chromosomes 1 : 20 was calculated for the 10 genotypes, the range was 2.05 to 2.38 (avg=2.22). The variation in absolute lengths among the genotypes is believed to be attributable to different chromosome condensation rates among the genotypes. Karyotypes of representative collections of each botanical variety of *A. hypogaea* plus *A. monticola* are presented in Figs. 1, 2. Chromosome measurements, including absolute lengths and short/long arm ratios, are presented in Tables 2 and 3; and karyotypic variation among the 10 analyzed genotypes is illustrated in Table 4.

The karyotypes of the 9 *A. hypogaea* cultivars varied more among than within varieties (Fig. 3; Table 4). A principal component analysis (Fig. 3) illustrates the karyotypic relationships among cultivars and varieties. Varieties *vulgaris*, *fastigiata*, *hirsuta*, and *hypogaea* formed distinct groups. Intermediate to varieties *vulgaris* and *hypogaea* was the variety *hirsuta*; and *nambyquarae* clustered with var. *fastigiata* even though it is a member of subsp. *hypogaea*.

The three var. *vulgaris* genotypes (Argentine, Spanhoma and PI 262000) had very similar karyotypes except for the chromosome with a secondary constriction. Argentine had a secondary constriction on chromosome 11 while PI 262000 and Spanhoma had the secondary constriction on chromosomes 6 and 7, respectively

Table 2. Chromosome length measurements for *A. hypogaea* subsp. *hypogaea* and *A. monticola*

Genotype	1	2	3	4	5	6	7	8	9	10
PI 158850										
A (μm)*	4.16 $\pm$.20	3.60 $\pm$.16	3.57 $\pm$.14	3.42 $\pm$.11	3.34 $\pm$.14	3.33 $\pm$.15	3.23 $\pm$.10	3.18 $\pm$.12	3.09 $\pm$.10	3.01 $\pm$.12
S/L ratio	0.83 $\pm$.03	0.72 $\pm$.03	0.85 $\pm$.01	0.90 $\pm$.02	0.73 $\pm$.03	0.90 $\pm$.02	0.83 $\pm$.03	0.87 $\pm$.02	0.78 $\pm$.02	0.87 $\pm$.01
NC 6										
A	3.45 $\pm$.13	3.22 $\pm$.11	3.05 $\pm$.12	2.98 $\pm$.11	2.93 $\pm$.09	2.89 $\pm$.16	2.89 $\pm$.08	2.79 $\pm$.10	2.78 $\pm$.10	2.73 $\pm$.09
S/L	0.87 $\pm$.02	0.77 $\pm$.02	0.86 $\pm$.03	0.85 $\pm$.02	0.65 $\pm$.03	0.68 $\pm$.04	0.80 $\pm$.02	0.85 $\pm$.02	0.70 $\pm$.03	0.87 $\pm$.02
NC 4										
A	3.43 $\pm$.08	3.29 $\pm$.05	3.16 $\pm$.05	3.12 $\pm$.07	3.08 $\pm$.05	3.00 $\pm$.05	3.00 $\pm$.06	2.98 $\pm$.05	2.91 $\pm$.05	2.82 $\pm$.05
S/L	0.83 $\pm$.03	0.89 $\pm$.02	0.65 $\pm$.03	0.86 $\pm$.02	0.73 $\pm$.02	0.72 $\pm$.04	0.90 $\pm$.02	0.80 $\pm$.02	0.76 $\pm$.02	0.86 $\pm$.03
Nambyquarae										
A	3.34 $\pm$.12	3.00 $\pm$.10	2.95 $\pm$.09	2.92 $\pm$.11	2.91 $\pm$.11	2.87 $\pm$.10	2.80 $\pm$.13	2.78 $\pm$.10	2.74 $\pm$.09	2.65 $\pm$.09
S/L	0.80 $\pm$.03	0.62 $\pm$.03	0.88 $\pm$.02	0.85 $\pm$.02	0.68 $\pm$.02	0.67 $\pm$.03	0.81 $\pm$.02	0.82 $\pm$.01	0.71 $\pm$.02	0.90 $\pm$.02
<i>A. monticola</i>										
A	3.51 $\pm$.10	3.26 $\pm$.07	3.23 $\pm$.05	3.08 $\pm$.03	2.98 $\pm$.05	2.97 $\pm$.05	2.91 $\pm$.05	2.90 $\pm$.04	2.81 $\pm$.04	2.79 $\pm$.04
S/L	0.85 $\pm$.03	0.64 $\pm$.02	0.80 $\pm$.03	0.84 $\pm$.02	0.66 $\pm$.02	0.85 $\pm$.03	0.81 $\pm$.02	0.84 $\pm$.02	0.68 $\pm$.02	0.84 $\pm$.02
Genotype										
	11	12	13	14	15	16	17	19	19	20
PI 158850										
A	2.99 $\pm$.08	2.88 $\pm$.09	2.84 $\pm$.10	2.83 $\pm$.10	2.71 $\pm$.09	2.59 $\pm$.09	2.53 $\pm$.08	2.44 $\pm$.08	2.37 $\pm$.07	1.75 $\pm$.09
S/L	0.70 $\pm$.02	0.83 $\pm$.03	0.76 $\pm$.02	0.86 $\pm$.02	0.86 $\pm$.02	0.83 $\pm$.02	0.69 $\pm$.02	0.86 $\pm$.02	0.70 $\pm$.02	0.82 $\pm$.03
NC 6										
A	2.65 $\pm$.08	2.58 $\pm$.08	2.56 $\pm$.09	2.53 $\pm$.07	2.46 $\pm$.08	2.43 $\pm$.09	2.30 $\pm$.06	2.24 $\pm$.07	2.07 $\pm$.06	1.63 $\pm$.06
S/L	0.69 $\pm$.04	0.92 $\pm$.02	0.89 $\pm$.07	0.91 $\pm$.02	0.82 $\pm$.03	0.73 $\pm$.03	0.71 $\pm$.03	0.84 $\pm$.02	0.66 $\pm$.03	0.70 $\pm$.03
NC 4										
A	2.77 $\pm$.05	2.72 $\pm$.06	2.62 $\pm$.05	2.62 $\pm$.04	2.55 $\pm$.04	2.49 $\pm$.04	2.46 $\pm$.04	2.34 $\pm$.05	2.23 $\pm$.06	1.64 $\pm$.04
S/L	0.69 $\pm$.03	0.87 $\pm$.02	0.81 $\pm$.02	0.90 $\pm$.02	0.88 $\pm$.02	0.79 $\pm$.02	0.66 $\pm$.03	0.82 $\pm$.03	0.65 $\pm$.03	0.77 $\pm$.03
Nambyquarae										
A	2.56 $\pm$.06	2.56 $\pm$.08	2.45 $\pm$.07	2.38 $\pm$.08	2.35 $\pm$.09	2.30 $\pm$.09	2.17 $\pm$.07	2.11 $\pm$.09	1.95 $\pm$.05	1.50 $\pm$.07
S/L	0.69 $\pm$.02	0.87 $\pm$.02	0.76 $\pm$.02	0.88 $\pm$.02	0.89 $\pm$.02	0.77 $\pm$.02	0.69 $\pm$.02	0.88 $\pm$.02	0.62 $\pm$.04	0.84 $\pm$.03
<i>A. monticola</i>										
A	2.77 $\pm$.09	2.69 $\pm$.03	2.66 $\pm$.04	2.64 $\pm$.03	2.56 $\pm$.06	2.53 $\pm$.04	2.44 $\pm$.04	2.38 $\pm$.04	2.11 $\pm$.04	1.63 $\pm$.07
S/L	0.78 $\pm$.03	0.88 $\pm$.03	0.74 $\pm$.02	0.86 $\pm$.03	0.88 $\pm$.02	0.79 $\pm$.02	0.87 $\pm$.03	0.67 $\pm$.03	0.72 $\pm$.03	0.85 $\pm$.02

* A = absolute and S/L = short arm/long arm ratio.

Table 3. Chromosome length measurements for *A. hypogaea* subsp. *fastigiata*

Genotype	1	2	3	4	5	6	7	8	9	10
Argentine										
A (μm)*	3.58±.08	3.38±.09	3.25±.06	3.15±.06	3.05±.06	3.05±.07	2.98±.06	2.92±.05	2.87±.05	2.85±.05
S/L ratio	0.85±.02	0.65±.02	0.85±.03	0.80±.02	0.86±.02	0.86±.02	0.82±.02	0.90±.02	0.69±.03	0.88±.02
PI 262000										
A	3.53±.15	3.15±.12	3.10±.14	3.05±.12	3.01±.12	2.98±.15	2.90±.11	2.87±.10	2.80±.10	2.76±.10
S/L	0.86±.02	0.69±.03	0.87±.02	0.80±.02	0.90±.02	0.83±.03	0.86±.03	0.88±.02	0.73±.02	0.85±.02
Spanhoma										
A	3.33±.17	3.05±.13	3.01±.12	2.91±.11	2.82±.10	2.80±.10	2.69±.12	2.67±.09	2.67±.10	2.61±.08
S/L	0.87±.02	0.68±.02	0.85±.02	0.86±.02	0.84±.03	0.88±.02	0.81±.04	0.87±.02	0.69±.02	0.86±.02
NC Acc. 17090										
A	3.64±.16	3.63±.12	3.30±.11	3.27±.09	3.27±.09	3.14±.08	3.01±.08	2.98±.07	2.87±.06	2.81±.07
S/L	0.79±.03	0.81±.02	0.89±.02	0.83±.02	0.67±.03	0.77±.02	0.88±.02	0.80±.02	0.70±.03	0.89±.02
Tennessee Red										
A	3.88±.11	3.51±.10	3.42±.18	3.41±.10	3.31±.08	3.13±.06	3.04±.05	3.01±.06	2.91±.08	2.90±.05
S/L	0.80±.03	0.73±.03	0.74±.04	0.83±.02	0.69±.03	0.78±.01	0.87±.02	0.85±.02	0.68±.03	0.86±.01
Genotype	11	12	13	14	15	16	17	18	19	20
Argentine										
A	2.85±.10	2.79±.06	2.73±.05	2.64±.05	2.62±.05	2.47±.06	2.44±.06	2.34±.05	2.17±.07	1.60±.04
S/L	0.75±.04	0.91±.02	0.71±.03	0.87±.03	0.85±.03	0.80±.02	0.66±.02	0.85±.02	0.67±.02	0.84±.03
PI 262000										
A	2.64±.08	2.62±.10	2.58±.09	2.50±.08	2.50±.09	2.40±.07	2.35±.07	2.25±.07	2.13±.05	1.58±.08
S/L	0.88±.02	0.87±.02	0.73±.02	0.89±.02	0.84±.02	0.85±.02	0.71±.03	0.86±.03	0.67±.03	0.88±.02
Spanhoma										
A	2.50±.10	2.47±.08	2.40±.07	2.32±.08	2.30±.09	2.28±.08	2.22±.07	2.22±.09	1.95±.07	1.49±.06
S/L	0.84±.03	0.84±.02	0.75±.02	0.86±.02	0.81±.03	0.86±.02	0.68±.02	0.86±.03	0.68±.03	0.80±.03
NC Acc. 17090										
A	2.76±.06	2.75±.07	2.69±.05	2.60±.05	2.52±.08	2.50±.05	2.35±.05	2.24±.05	2.20±.06	1.61±.05
S/L	0.80±.03	0.81±.02	0.82±.02	0.85±.03	0.84±.03	0.80±.03	0.76±.02	0.86±.03	0.64±.02	0.84±.03
Tennessee Red										
A	2.82±.07	2.80±.06	2.72±.05	2.67±.06	2.57±.05	2.53±.05	2.40±.06	2.30±.06	2.17±.05	1.66±.05
S/L	0.74±.03	0.81±.02	0.84±.02	0.85±.02	0.79±.01	0.84±.02	0.69±.03	0.85±.02	0.65±.02	0.81±.03

* A = absolute and S/L = short arm/long arm ratio.

(Table 4). Argentina also had the most distinctive heterochromatic regions among all the genotypes analyzed. Chromosome 2 had a distinct heterochromatic region on the long arm. Two darkly stained regions were observed on the long arm of chromosome 3. Chromosomes 5 and 15 had distinct heterochromatic regions proximal to the centromere and chromosome 17 had one distinct region in the short arm

Fig. 1. Karyotypes of *A. hypogaea* subsp. *hypogaea* varieties *hypogaea* (NC 4 and nambyquarae) and *hirsuta* (PI 158850).

and two in the long arm. Argentina had 14 median chromosomes and six slightly submedian chromosomes (2, 9, 11, 13, 17 and 19). The centromere positions of PI 262000 and Spanhoma were very similar to those observed for Argentina, except that these two cultivars had a median chromosome number 11.

The karyotypes of the two var. *fastigiata* cultivars (Tennessee Red and NC Acc. 17090) were very similar (Fig. 3; Tables 3, 4), and the only significant differences were for chromosomes 1, 2 and 3. The two cultivars were the only ones observed with a secondary constriction on one of the longer chromosomes. NC Acc. 17090 had a secondary constriction on chromosome 1, while it was on chromosome 3 of Tennessee Red. NC Acc. 17090 had 13 median, 6 slightly submedian (1, 5, 6, 9, 11 and 17) and 1 submedian chromosome (19), while Tennessee Red had 12 median and 8 slightly submedian chromosomes (2, 3, 5, 6, 9, 11, 17 and 19).

Table 4. Chromosome variation among *A. hypogaea* cultivars and *A. monticola*

<i>A. hypogaea</i> variety	Cultivar/species	Individual chromosomes				
		With secondary constriction	Slightly submedian/submedian			
<i>vulgaris</i>	Argentine	11	2,	9, 11, 13,	17,	19
	PI 262000	6	2,	9, 13,	17,	19
	Spanhoma	7	2,	9, 13,	17,	19
<i>fastigiata</i>	NC Acc. 17090	1	1,	5, 6, 9, 11,	17,	19*
	Tennessee Red	3	2,	3, 5, 6, 9, 11,	17,	19
<i>hirsuta</i>	PI 158850	6	2,	5, 9, 11, 13,	17,	19
<i>hypogaea</i>	NC 6	6	2,	5, 6, 9, 11,	16, 17,	19, 20
	NC 4	6		3, 5, 6, 9, 11,	16, 17,	19, 20
	Nambyquarae	7	2*,	5, 6, 9, 11, 13, 16, 17,		19*
—	<i>A. monticola</i>	11	2*, 3, 5,	9, 11, 13, 16,		18, 19

* Chromosomes with * are submedian.

The only representative of subsp. *hypogaea* var. *hirsuta* analyzed was PI 158850. This cultivar had the longest genome length—59.9 μm —among all analyzed genotypes. However, the relative chromosome lengths, as measured by the ratio of the chromosomes to number 20, were very similar to all other cultivars analyzed. When the karyotype was compared to the other varieties, PI 158850 formed a unique group, but was more similar to the subsp. *fastigiata* cultivars than to members of subsp. *hypogaea* var. *hypogaea* (Fig. 3; Table 4). PI 158850 had 13 median and seven slightly submedian chromosomes (2, 5, 9, 11, 13, 17 and 19). Chromosome 6 had a secondary constriction.

The two subsp. *hypogaea* var. *hypogaea* cultivars, NC 4 and NC 6, had very similar karyotypes. Each cultivar had a secondary constriction on chromosome 6 and slightly submedian chromosomes 5, 6, 9, 11, 16, 17, 19 and 20. NC 4 and NC 6 had 11 median and 9 slightly submedian chromosomes (Tables 2, 4). The karyotypes of the two North Carolina cultivars were similar except chromosome 2 was median for NC 4, but slightly submedian for NC 6; and chromosome 3 was slightly submedian for NC 4 but median for NC 6. However, the standard errors of chromosomes 2 and 3 for NC 6 were relatively large (Table 2), and the order for this cultivar could possibly be reversed. NC 4 had two distinct heterochromatic regions in the long arm of chromosome 13, but the darkly stained regions were absent in the corresponding NC 6 chromosome. Distinct heterochromatic regions

were also observed in the long arm of chromosome 5 for both NC 4 and NC 6.

The karyotype of *nambyquarae* significantly differed from other subsp. *hypogaea* cultivars analyzed and karyologically grouped with var. *fastigiata* cultivars (Fig. 3; Table 4). Two distinctive heterochromatic regions were observed on the long arm of both chromosomes 2 and 13. Eleven chromosomes were median, 7 were slightly submedian and chromosomes 2 and 19 were submedian. Absolute

A. MONTICOLA

A. HYPOGAEA VAR. VULGARIS
(ARGENTINE)

A. HYPOGAEA VAR. FASTIGIATA
(TENNESSEE RED)

10 μ m

Fig. 2. Karyotypes of *A. monticola* and *A. hypogaea* subsp. *fastigiata* varieties *vulgaris* (Argentine) and *fastigiata* (Tennessee Red).

chromosome length measurements indicated that *nambyquarae* had a secondary constriction on chromosome 7. However, the standard deviation of chromosomes 6 and 7 were large (Table 2) and the order of the two chromosomes could possibly be reversed.

Both *A. monticola* genotypes had the same karyotype, thus only data for collection 7264 is presented in Tables 2 and 4. The *A. monticola* genotypes had continuous runs of reproductive axes on lateral branches, but no flowers on the main stem. It is thus intermediate between the *A. hypogaea* subsp. *hypogaea* and *fastigiata* for flowering patterns. *Arachis monticola* and Argentine were the only collections analyzed with a secondary constriction on chromosome 11. *Arachis monticola* had 11 median and 8 slightly submedian chromosomes and a submedian chromosome number 2. The species differed from *A. hypogaea* by having a median rather than a slightly submedian chromosome 17 and the reverse was true for chromosome 18. Chromosome 17 for *A. monticola* averaged $2.44 \pm .04 \mu\text{m}$ while chromosome 18 averaged $2.38 \pm .04 \mu\text{m}$, so the chromosome order is believed to be correct as presented and truly different from the cultivated species.

Fig. 3. Scatter diagram of nine *A. hypogaea* cultivars and *A. monticola* which were separated by principal components of chromosome arm ratios.

Discussion

The chromosomes of *A. hypogaea* are small, ranging from 1.49 to 4.16 μm in the cultivars analyzed. While most chromosome pairs are easily distinguishable when they are not highly condensed or overstained, several homologues are nearly identical. Most problems in identification were experienced when a series of metacentric chromosomes with nearly the same lengths were observed. For example, chromosomes 14 and 15 of Argentine were 2.64 and 2.62 μm long and had arm ratios of 0.87 and 0.84, respectively. The heterochromatic regions were also similar and chromosome identification is nearly impossible. A similar problem was observed for chromosomes 5 and 6 of Argentine because both chromosomes had a length of 3.05 μm and arm ratios of 0.86. However, chromosome 5 had one heterochromatic region proximal to the centromere on the short arm while chromosome 6 had one proximal and one distal heterochromatic region.

The karyological groups of cultivars (Fig. 3) generally correspond to the varietal classification proposed by Krapovickas (1969). Even though members of varieties *vulgaris*, *fastigiata* and *hypogaea* are cytologically unique, varieties *vulgaris* and *hypogaea* represent the karyological extreme groups while the other botanical varieties are intermediate. The karyological data agree with Gregory *et al.* (1973) who gave nambyquarae varietal status. The cultivars nambyquarae and PI 158850 are

karyologically most closely related to var. *fastigiata* and from this investigation it appears that cultivars of varieties *hirsuta* and *fastigiata* (and nambyquarae) may have been of hybrid origin between ancient var. *hypogaea* and var. *vulgaris* genotypes. Isleib (1982) reported a principal component analysis using 21 morphological characters and 28 cultivars, including nambyquarae and PI 158850. In his morphological study, nambyquarae was significantly different from all other genotypes while PI 158850 clustered with var. *vulgaris* genotypes. Additional collections need to be analyzed, especially of var. *hirsuta*, before true phylogenetic relationships of this group are verified. Although meiotic studies have previously indicated little genomic differentiation among varieties (Stalker 1980), the analysis has revealed a significant and unexpected amount of chromosomal variation among cultivars of *A. hypogaea*. Five different chromosomes with secondary constrictions were observed among the genotypes ranging from the largest chromosome for NC Acc. 17090 to one of the smaller chromosomes for Argentine. If all the genotypes had a common ancestor, as is commonly believed, then structural rearrangements probably were the result of translocations and subsequent varietal differentiation. A standard karyotype is not being proposed for the species as a whole because of the observed chromosome variation. However, in all nine cultivars the chromosomes 4, 8, 10, 12, 14, 15 and 18 were median and nearly identical, and chromosomes 9 and 19 were slightly submedian to submedian and very similar. Variation was observed among genotypes for the remaining 11 *A. hypogaea* chromosomes. Patterns of variation were apparent within botanical varieties and the cultivars illustrated in Figs. 1 and 2 are proposed as standard karyotypes for each group and *A. monticola*. Identification of the *A. hypogaea* chromosomes was sufficient to develop cytogenetic stocks (such as aneuploid series) for specific cultivars.

The primary center for domestication of peanuts is probably southern Bolivia or northern Argentina (Gregory and Gregory 1976, Krapovickas 1969). Members of subsp. *hypogaea* var. *hypogaea* are the predominant peanut type in this region and believed to be most closely related to the wild *Arachis* species because var. *hypogaea* usually has a runner habit, a similar branching pattern as other *Arachis* species and does not have compound spikes (Krapovickas 1969). Although Husted (1936) proposed that *A. hypogaea* is of direct amphidiploid origin, Krapovickas and Rigoni (1957) and Smartt and Gregory (1967) suggested that the cultivated species evolved from a pre-existing wild allotetraploid which probably arose from an annual $\times$ perennial interspecific hybrid within section *Arachis* (Gregory and Gregory 1976). Since *A. monticola* is the only other known tetraploid species in section *Arachis*, this is the most likely candidate for the direct progenitor of *A. hypogaea*. The *A. monticola* collections used in this investigation were collected from northern Argentina and no cultivars were observed in the area (Gibbons *et al.* 1972). While K 7264 has been in germplasm nurseries for many years, collection GKBSPSc 30062 was only grown in the greenhouse after introduction from South America. If *A. monticola* is the progenitor of the cultivated species and if var. *hypogaea* is the most closely related cultivated type, as previously suggested (Krapovickas and Rigoni 1957), then similar karyotypes should be observed for the two groups. However, the conclusion drawn from the karyological analyses of *A. monticola* and *A. hypogaea*

varieties is that *A. monticola* is most closely related to var. *vulgaris*. Thus, based strictly on the karyological data, the members of subsp. *fastigiata* var. *vulgaris* appear to be the ancestral *A. hypogaea* type.

Several diploid species of section *Arachis* have been suggested as possible progenitors to *A. hypogaea*, including *A. villosa* Benth. (Varisai Muhammad 1973), *A. duranensis* Krap. et Greg. *nom. nud.* (Seetharam *et al.* 1973), *A. batizocoi* Krap. et Greg. (Karpovickas *et al.* 1974), *A. duranensis* and *A. cardenasii* Krap. et Greg. *nom. nud.* (Gregory and Gregory 1976) and *A. cardenasii* and *A. batizocoi* (Smartt *et al.* 1978a, 1978b). When the karyotypes of above species as reported by Stalker and Dalmacio (1982) are compared with the karyotypes of *A. hypogaea*, several conclusions can be drawn. First, *A. hypogaea* has a symmetrical karyotype with mostly median chromosomes but several diploid species (*i.e.*, *A. batizocoi* and *A. cardenasii*) each has as many as seven slightly submedian or submedian chromosomes. Since the number of asymmetrical chromosomes in the tetraploid species ranged from 5 in PI 262000 and Spanhoma to 9 in NC 6 and *A. monticola*, it is unlikely that two diploid species such as *A. batizocoi* and *A. cardenasii* hybridized to eventually become the progenitors of *A. hypogaea*. The other species hypothesized as progenitors of cultivated peanuts, *A. villosa* or *A. duranensis*, have two and three submedian or slightly submedian chromosomes, respectively. The karyotype of either species can be superimposed over the karyotype of *A. hypogaea*. The karyotypic data so far collected have not confirmed or eliminated possible progenitors of the cultivated peanut. However, the data can be used to eliminate unlikely species combinations such as *A. cardenasii* $\times$ *A. batizocoi*. As additional genotypes are analyzed and more refined techniques utilized, the evolutionary patterns of the cultivated peanut and related species will become more apparent.

Summary

Mitotic chromosomes of nine *Arachis hypogaea* L. cultivars plus *A. monticola* Krap. et Rig. were analyzed cytologically. The cultivars included representatives of the four botanical varieties of *A. hypogaea*. The objective of this investigation was to identify the chromosomes of cultivated peanuts and determine the variation among subspecies and varieties. The *A. hypogaea* chromosomes ranged in length between 1.44 and 4.17 μm and were mostly metacentric. Variation was observed in the numbers of asymmetrical chromosomes per cultivar and in the chromosome with a secondary constriction. Based on heterochromatic regions, centromere positions, and chromosome lengths, homologues were identified and ordered from 1=longest to 20=shortest. Although each genotype had a unique karyotype, patterns of variation were observed and all *A. hypogaea* varieties were cytologically distinguished. Varieties *vulgaris* and *hypogaea* were karyologically the extreme groups with other varieties being intermediate. The cultivar nambyquarae is grouped karyologically with var. *fastigiata* even though it is a member of subspecies *hypogaea*. *Arachis monticola* was karyologically more similar to variety *vulgaris* than other groups of *A. hypogaea*. This suggests that the variety *vulgaris* cultivars may be the most ancient *A. hypogaea* group.

Literature cited

- Babu, C. N. 1955. Cytogenetical investigations on groundnuts I. The somatic chromosomes. *Indian J. Agr. Sci.* **25**: 41–46.
- D'Cruz, R. and Tankasale, M. P. 1961. A note on chromosome complement of four groundnut varieties. *Indian Oilseeds J.* **5**: 58–59.
- Ghimpu, V. 1930. Recherches cytologiques sur les genres: *Hordeum*, *Acacia*, *Medicago*, *Vitis* et *Quercus*. *Arch. d'Anat. Microsc.* **26**: 136–234.
- Gibbons, R. W., Bunting, A. H. and Smartt, J. 1972. The classification of varieties of groundnut (*Arachis hypogaea* L.). *Euphytica* **21**: 78–85.
- Gregory, W. C. and Gregory, M. P. 1976. Groundnut, *Arachis hypogaea* (Leguminosae—Papilionatae). In *Evolution of Crop Plants*. N. W. Simmonds, Ed. Longman, London and New York. p. 151–154.
- , —, Krapovickas, A., Smith, B. W. and Yarbrough, J. A. 1973. Structure and genetic resources of peanuts, Chapter 3. In *Peanuts—Culture and Uses*. Amer. Peanut Res. Educ. Assoc., Stillwater, Okla. p. 47–133.
- , Krapovickas, A. and Gregory, M. P. 1980. Structures, variation, evolution and classification in *Arachis*. In *Advances in Legume Science*, Vol. 1. R. J. Summerfield and A. H. Bunting, Eds. International Legume Conference, Royal Botanic Gardens, Kew. p. 469–481.
- Husted, L. 1933. Cytological studies of the peanut *Arachis* I. Chromosome number and morphology. *Cytologia* **5**: 109–117.
- 1936. Cytological studies of the peanut *Arachis* II. Chromosome number, morphology and behaviour and their application to the origin of cultivated forms. *Cytologia* **7**: 396–423.
- Isleib, T. G. 1982. Heterosis and early generation testing in testcrosses of exotic peanut (*Arachis hypogaea* L.) cultivars. Ph. D. Dissertation, N. C. State Univ., Raleigh. 84 p.
- Krapovickas, A. 1969. Origen, variabilidad y difusión del maní (*Arachis hypogaea*). In *The Domestication and Exploitation of Plants and Animals*. P. J. Ucko and G. W. Dimbleby, Eds. London. p. 427–441.
- 1973. Evolution of the genus *Arachis*. In *Agricultural Genetics, Selected Topics*. R. Moav, Ed. National Council for Research and Development, Jerusalem. p. 135–151.
- , Fernandez, A. and Seeligman, P. 1974. Recuperación de la fertilidad en un híbrido interspecífico estéril de *Arachis* (Leguminosae). *Bonplandia* **3**: 129–142.
- and Rigoni, V. A. 1957. Nuevas especies de *Arachis* vinculadas al problema del origen del maní. *Darwiniana* **11**: 431–455.
- Seetharam, A., Nayar, K. M. D., Sreekantaradhya, R. and Achar, D. K. T. 1973. Cytological studies on interspecific hybrids of *Arachis hypogaea* × *Arachis duranensis*. *Cytologia* **38**: 277–280.
- Singh, A. K. and Moss, J. P. 1982. Utilization of wild relatives in genetic improvement of *Arachis hypogaea* L. Part 2: Chromosome complements of species in section *Arachis*. *Theor. Appl. Genet.* **61**: 305–314.
- Smartt, J. and Gregory, W. C. 1967. Interspecific cross-compatibility between the cultivated peanut *Arachis hypogaea* L. and other members of the genus *Arachis*. *Oleagineux* **22**: 455–459.
- , — and Gregory, M. P. 1978a. The genomes of *Arachis hypogaea*. 1. Cytogenetic studies of putative genome donors. *Euphytica* **27**: 665–675.
- , — and — 1978b. The genomes of *Arachis hypogaea*. 2. The implications in interspecific breeding. *Euphytica* **27**: 677–680.
- and Stalker, H. T. 1982. Speciation and cytogenetics of *Arachis*. In *Peanut Science and Technology*. H. Pattee and C. Young, Eds. Am. Peanut Res. and Educ. Soc., Yoakum, Tex. p. 21–49.
- Stalker, H. T. 1980. Cytogenetic investigations in the genus *Arachis*. ICRISAT Proc. of the Internat. Workshop on Groundnuts, Oct. 13–17, 1980, Patancheru, A. P., India. p. 73–81.
- and Dalmacio, R. D. 1981. Chromosomes of *Arachis* species, section *Arachis* (Leguminosae). *J. Hered.* **72**: 403–408.

- Varisai Muhammad, S. 1973. Cytological investigations in the genus *Arachis* L. II. Triploid hybrids and their derivatives. Madras Agric. J. 60: 1414-1427.
- Wynne, J. C. 1974. Breeding potential of the subspecies of *Arachis hypogaea* L.: Inheritance of branching pattern, estimates of combining ability and photoperiodic response. Ph. D. Dissertation, N. C. State Univ., Raleigh. 103 p.
- and Gregory, W. C. 1981. Peanut breeding. Adv. in Agron. 34: 39-72.
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