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Potential Sources of Peanut Seed Proteins and Oil in the Genus *Arachis*

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Fat-free and full-fat seed meals from 37 selected wild species had crude protein concentrations ranging from 43.4 to 63.5% and 17.1 to 30.8%, respectively. Similar preparations from 21 selected *Arachis hypogaea* L. cultivars had 44.8 to 57.4% and 20.6 to 29.1% protein. Percentage oil in seed meals of wild species ranged between 46.5 and 63.1% while those of cultivars were 43.6 to 55.5%. Further characterization of proteins on polyacrylamide disc electrophoretic gels showed that most of the arachin and nonarachin components remained constant among cultivars while many new and variable patterns of these constituents were noted among wild *Arachis*. The gel data corroborated the known classification of wild *Arachis* into seven coherent sections; cultivated varieties could also be classified into seven groups. Quantities of protein and oil were randomly distributed among wild species and cultivars and could not be used to develop a chemotaxonomic relationship in *Arachis*.

Arachis hypogaea L. is a potential source of food-grade protein as it is readily cultivated commercially and has attained widespread acceptability because of its economic value to industry and dietary interest by the consumer. In addition, continued improvement of this commodity through genetic programs, updated agronomic practices, and modern food technology has helped to expand its food use. For example, research has shown that peanut seeds have potential as a source of food-grade protein for fortification of food products, and, through industrially applicable techniques, for recovery of meals, flours, flakes, grits, concentrates, and isolates (Ziemba, 1975; Quinn et al., 1975; Khan et al., 1975; Ayres et al., 1974; Mattil, 1973; Rhee et al., 1972, 1973; Harris et al., 1972; Smith, 1971; Mitchell and Malphrus, 1968; Altschul, 1967; Arthur, 1953; Pominski et al., 1952). However, only limited progress has been made in expanding utilization of peanuts specifically as a source of high protein food and research results have been much less extensive than those on soybeans and cereals (Pulle and Ino, 1975; Fan and Sosulski, 1974;

Bushuk, 1974; MacRitchie, 1973; Wolf and Cowan, 1971).

Other studies have suggested that peanut seeds may be useful sources of proteins (Cherry et al., 1974, 1975; Cherry, 1974; McKinney et al., 1973; Goldblatt, 1971). In addition, studies have shown that peanut seeds are composed of groups of proteins including arachin (major storage globulin) and conarachin or nonarachin components (Basha and Cherry, 1976; Shetty and Rao, 1974; Cherry et al., 1973; Cherry and Ory, 1973; Singh and Dieckert, 1973a,b; Dawson, 1968, 1971; Neucere, 1969). These fractions were also shown to differ in amino acid composition (Basha and Cherry, 1976; Singh and Dieckert, 1973a,b; Dawson, 1968, 1971; Neucere, 1969). Thus, these protein fractions may differ not only in nutritional value but in functional properties. Partitioning these components may lead to expanded utilization of peanuts as protein ingredients in new foods as well as other more conventional peanut products.

Research in protein quality has only recently been reemphasized in the exploration for new sources of utilizable protein. Among these sources are the approximately 50 to 70 known wild species collections of the genus *Arachis* (Gregory et al., 1973; Cherry, 1975; Neucere and Cherry, 1975). Wild species contain new sources of germ plasm which can be used to increase variability in the genetic base of cultivated varieties (Simpson and Haney, 1973; Miller, 1973; Kamra, 1971; Frankel and Bennett, 1970; Nelson, 1969). Once the cross-compatibility of

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species within the genus is understood, studies can begin to introduce the new genetic information into cultivated varieties. Separation of seed proteins of available wild species of the genus *Arachis* into discrete bands using gel electrophoretic techniques (Cherry, 1975) has given support to the botanical classification of the genus (Gregory et al., 1973) into the following taxonomic sections: Axonomorphae, Erectoides, Caulorhizae, Rhizomatosae, Extranervosae, Pseudoaxonomorphae, and Triseminalae. The present study was conducted using small quantities of seed because they were obtained with great difficulty from many wild collections through very tedious breeding programs. However, a more detailed biochemical assay than in the past has been completed on protein and oil in meals of peanut seeds from cultivated and wild species of *Arachis* in a continuing effort to characterize these components in the genus.

MATERIALS AND METHODS

A group of 37 collections comprising various wild species accessions of *Arachis*, some identified as species, others not, together with a sampling of 21 cultivars of *A. hypogaea* L. were examined in this study. The taxonomic classification and list of species collections and cultivars included in this study are presented in Table I (see Gregory et al. (1973) for a detailed description of the collection sites for wild species). Preliminary studies suggest that there are additional species among the undescribed collections; however, taxonomists have not assigned specific names to them and, therefore, they are labeled as *Arachis* species (*A. sp.*). Of far more importance and concern is the fact that a number of the *A. sp.* accessions duplicate species already known and named.

Two samples each containing three to six seeds from each species collection or cultivar were examined for protein and oil composition. Plump, assumed to be mature, seed from these sources was available in small quantities only, thus limiting extensive quantitative analyses. The samples were weighed in tared vials, lyophilized, ground into meals, and defatted completely with successive extractions of diethyl ether. Percentage oil of these peanut meals was determined by weight difference. Protein analyses were conducted on fat-free meals using the standard macro-Kjeldahl technique and a nitrogen conversion factor of 5.46. Protein values of full-fat meals were determined by calculation.

Samples from the preparations described above prior to defatting were used for gel electrophoretic evaluation of protein. The meals were ground with a mortar and pestle in pH 7.9 sodium phosphate buffer (I, 0.01) and centrifuged at 39 000g for 30 min. The partially purified extracts diluted to approximately 6.0 mg of protein/ml were subjected to disc gel electrophoresis on 10% polyacrylamide gels as described previously by Canalco (Canalco Catalog, 1973) and Cherry et al. (1970). Levels of protein per gel ranged between 200 and 600 μ g to ensure that all possible components in the extracts were distinguished; gels containing approximately 300 μ g of protein were used as examples of typical patterns for presentation in this paper.

RESULTS AND DISCUSSION

Quantitative Comparisons of Protein and Oil. In general, percentage protein of full-fat meals from wild species collections (Table I) could be grouped between 17.1 to 19.9% (section Erectoides, series Tetrafoliolatae, species *A. benthamii* P.I. (plant introduction no.) 2622859; *A. sp.* P.I. 276228; and section Rhizomatosae, *A. sp.* P.I. 262286) and 28.0 to 30.8% (Rhizomatosae, *A. sp.* P.I. 262794, *A. burkartii* "unpublished" P.I. 338325; Caulorhizae, *A. pintoii*

"unpublished" P.I. 338447; Erectoides, *A. sp.* P.I. 338302; Axonomorphae, series Perennes, *A. cardensii* "unpublished" P.I. 262141; and Triseminalae, *A. pusilla* P.I. 338449). Species "names" not officially published when mentioned for the first time in the text are followed with the notation "unpublished". Cultivars from *A. hypogaea* could be grouped between 2.06 (Tarapoto P.I. 259747) and 28.3 to 29.1% (Georgia Jumbo Necrotic Etch; Georgia Jumbo; Japan Jumbo; Croft Jumbo, Chalimbana). In protein and oil comparisons among wild species collections and cultivars throughout the text of the paper, percentages are arranged in order of increasing value and those included in the low group are not statistically different from the lowest number (e.g., 19.9% is not different from 17.1% in analyses of protein of full-fat meals from species comparisons) while those included in the high class are not different from the highest value (e.g., 28.0 to 30.8%).

Examination of oil content in meals prepared from seeds of individual species and collections of *Arachis* (Table I) showed percentages ranging between 46.5 to 49.5% (section Extranervosae, *A. villosulicarpa* P.I. 336985; Triseminalae, *A. pusilla* P.I. 339449; Axonomorphae series Perennes, *A. cardensii* P.I. 262141; Rhizomatosae Prorhizomatosae, *A. burkartii* P.I. 338325; Caulorhizae, *A. pintoii* P.I. 338447) and 59.7 to 63.1% (Erectoides, series Tetrafoliolatae, *A. sp.* P.I. 267228, *A. sp.* P.I. 276209, *A. paraguariensis* P.I. 331188, P.I. 262842, *A. sp.* P.I. 276225, *A. benthamii* P.I. 262859, series Procumbensae, *A. rigonii* P.I. 262142; Rhizomatosae, Eurhizomatosae, *A. glabrata* P.I. 262797, *A. sp.* P.I. 262286). Many of the meals containing the highest percentage oil were from species and collections of section Erectoides. Percentage oil of meals from samples of *A. hypogaea* ranged between 43.6 to 46.2% (Chalimbana; NC5; Georgia Jumbo Necrotic Etch) and 51.8 to 55.5% (Valencia Bunch; F-334-A-B-14; Florida 393-7-1; Tifspan; Spancross; Florigiant; Tarapoto P.I. 259747).

Mean protein percentages of full-fat meals by section showed that values for Axonomorphae (25.2%), Caulorhizae (28.2%), Rhizomatosae (24.4%), Extranervosae (23.2%), and Pseudoaxonomorphae (27.5%) were not significantly different (Table I). The average value for Erectoides (22.2%) was significantly less than all other sections except Extranervosae, and that of Triseminalae (30.8%) was significantly greater than all but Pseudoaxonomorphae and Caulorhizae. Further comparisons made with average protein percentages among series showed no significant differences within sections (Table I). Perennes (24.5%) was the only series within Axonomorphae having a percentage protein value not different from both series (Tetrafoliolatae, 22.4%; Procumbensae, 20.5%) of Erectoides. Annuae (25.7%) and Amphiploides (25.3%) of Axonomorphae, Prorhizomatosae (28.3%) and Eurhizomatosae (23.7%) of Rhizomatosae, Caulorhizae, Extranervosae, and Pseudoaxonomorphae had percentages which were not significantly different. Section Triseminalae was not different from sections Pseudoaxonomorphae and Caulorhizae and series Eurhizomatosae of section Rhizomatosae.

Arachis duranensis "unpublished" P.I. 219823 and *A. sp.* P.I. 263133 of series Annuae in section Axonomorphae contained an average percentage protein value (57.1%) for fat-free meal which was significantly greater than those of the other two series, Perennes (51.8%) and Amphiploides (50.7%) in this section (Table I); these values for full-fat meals were not significantly different (25.7, 24.5, and 25.3%, respectively). The percentage fat-free protein of Annuae was not different from any other series or

Table I. Percentage Protein and Oil Values^a of Peanut Seed Meals from a Sampling of 37 Accessions of Wild *Arachis* Species Materials and 21 Cultivars of *Arachis hypogaea* L.

Species/cultivars ^b	P.I. no. ^c (accession no.)	Protein, %		Oil, %
		Full-fat	Fat-free	
Section I: Axonomorphae		25.2 ^b	51.3 ^c	51.3 ^{cd}
Series 1. Annuae		25.7 ^b	57.1 ^a	54.7 ^{b-d}
<i>A. duranensis</i>	219823	24.7 ^{f-p}	52.4 ^{h-q}	53.0 ^{i-m}
<i>A. sp.</i>	263133	26.7 ^{b-f}	61.7 ^{ab}	56.4 ^{e-i}
Series 2. Perennes		24.5 ^{bc}	51.8 ^b	52.8 ^{de}
<i>A. correntina</i>	262137	23.3 ^{q-r}	47.3 ^{q-v}	50.6 ^{l-r}
<i>A. correntina</i>	262134	23.4 ^{f-r}	49.7 ^{l-u}	52.4 ^{j-p}
<i>A. correntina</i>	262808	23.1 ^{h-s}	54.2 ^{d-m}	57.4 ^{c-g}
<i>A. chacoense</i>	276235	22.6 ^{k-s}	48.2 ^{o-v}	54.6 ^{g-k}
<i>A. villosa</i>	298636	26.3 ^{b-h}	55.6 ^{c-k}	52.8 ⁱ⁻ⁿ
<i>A. cardensis</i>	262141	28.5 ^{a-d}	55.6 ^{c-k}	48.8 ^{p-s}
Series 3. Amphiploides		25.3 ^b	50.7 ^b	50.3 ^{d-f}
<i>A. monticola</i>	219824	23.4 ^{q-r}	49.9 ^{l-u}	53.1 ^{i-m}
<i>A. hypogaea</i> cultivars:				
F334A-B-14	T1385	22.4 ^{l-t}	46.9 ^{q-v}	52.3 ^{j-p}
Tarapoto	259747	20.6 ^{q-t}	46.4 ^{s-v}	55.5 ^{f-j}
Tifton-8	T2034	22.6 ^{j-s}	44.8 ^{uv}	49.5 ^{m-s}
Nambyquarae	221068	23.5 ^{f-r}	47.3 ^{q-v}	51.3 ^{k-q}
White Genotype I-Pearl	T894	24.2 ^{f-p}	47.6 ^{q-v}	49.1 ^{o-s}
Florida 393-7-1	T902	25.5 ^{c-m}	53.5 ^{g-o}	52.4 ^{j-p}
White Genotype II	T2002	25.5 ^{c-m}	49.7 ^{l-u}	48.7 ^{p-s}
Georgia Jumbo	PS2301	28.6 ^{a-d}	54.1 ^{d-m}	47.1 ^{rs}
Croft Jumbo	T1498	29.1 ^{ab}	57.4 ^{b-h}	49.2 ^{n-s}
Japan Jumbo	T1469	28.7 ^{a-c}	55.9 ^{c-j}	49.7 ^{p-s}
Valencia Bunch	268704	25.1 ^{e-o}	52.1 ^{h-t}	51.8 ^{j-q}
Tennessee Red		26.6 ^{b-g}	50.5 ^{j-s}	47.4 ^{rs}
Conagins Macrocarpa	T1468	23.8 ^{f-q}	49.1 ^{m-u}	51.5 ^{k-q}
Florunner		25.7 ^{c-k}	50.2 ^{k-t}	48.9 ^{o-s}
Florigiant		24.5 ^{f-p}	53.9 ^{f-n}	55.4 ^{f-j}
Dixie Spanish		26.1 ^{b-i}	51.2 ^{i-s}	49.1 ^{o-s}
Tifspan		23.3 ^{g-r}	50.6 ^{j-s}	53.9 ^{g-l}
Spancross		24.0 ^{f-p}	51.8 ^{i-r}	54.5 ^{g-k}
Chalimbana		29.1 ^{ab}	51.6 ^{i-s}	43.6 ^t
NC5		26.1 ^{b-i}	48.5 ^{n-v}	45.9 st
Ga. Jumbo Necrotic Etch		28.3 ^{a-e}	52.6 ^{h-q}	46.2 st
Section II: Erectoides		22.2 ^c	53.3 ^{bc}	58.4 ^a
Series 2. Tetrafoliolatae		22.4 ^c	53.3 ^{ab}	58.2 ^{ab}
<i>A. sp.</i>	276209	20.6 ^{q-t}	51.6 ^{i-s}	60.1 ^{a-d}
<i>A. sp.</i>	276225	22.1 ^{n-t}	56.3 ^{c-i}	60.6 ^{a-c}
<i>A. benthamii</i>	262859	17.1 ^u	45.1 ^{t-v}	62.1 ^{ab}
<i>A. sp.</i>	276229	25.9 ^{b-j}	59.2 ^{a-f}	57.1 ^{c-h}
<i>A. sp.</i>	262278	22.3 ^{l-t}	52.1 ^{h-r}	57.2 ^{c-h}
<i>A. sp.</i>	276228	19.2 ^{tu}	48.1 ^{p-v}	59.8 ^{a-e}
<i>A. sp.</i>	261877	23.2 ^{h-r}	53.1 ^{q-p}	56.3 ^{e-i}
<i>A. sp.</i>	262308	21.9 ^{o-t}	50.7 ^{j-s}	56.8 ^{e-i}
<i>A. paraguariensis</i>	331188	22.1 ^{n-t}	55.4 ^{c-k}	60.2 ^{a-d}
<i>A. paraguariensis</i>	331187	21.4 ^{p-t}	50.8 ^{j-s}	57.3 ^{c-h}
<i>A. paraguariensis</i>	262842	22.2 ^{m-t}	58.5 ^{b-g}	62.0 ^{ab}
<i>A. oteroi</i>	338287	23.8 ^{f-q}	51.4 ^{i-s}	53.9 ^{g-l}
<i>A. sp.</i>	338302	28.2 ^{a-e}	59.5 ^{a-d}	52.5 ^{j-o}
<i>A. sp.</i>	262140	22.9 ^{i-s}	54.7 ^{d-l}	58.7 ^{b-f}
Series 3. Procumbensae		20.5 ^c	53.1 ^{ab}	61.4 ^a
<i>A. rigonii</i>	262142	20.5 ^{r-t}	53.1 ^{g-p}	61.4 ^{ab}
Section III: Caulorhizae		28.2 ^{ab}	55.9 ^{bc}	49.5 ^{cd}
Series		28.2 ^{ab}	55.9 ^{ab}	49.5 ^{d-f}
<i>A. pintoii</i>	338447	28.2 ^{a-e}	55.9 ^{c-j}	49.5 ^{m-s}
Section IV: Rhizomatosae		24.4 ^b	55.1 ^{bc}	55.5 ^b
Series 1. Eurhizomatosae		23.7 ^{bc}	55.0 ^{ab}	56.8 ^{a-c}
<i>A. sp.</i>	262794	28.0 ^{a-e}	60.4 ^{a-c}	53.7 ^{h-l}
<i>A. glabrata</i>	262797	21.4 ^{p-t}	53.8 ^{f-n}	60.2 ^{a-d}
<i>A. sp.</i>	262286	19.9 ^{s-u}	54.1 ^{c-m}	63.2 ^a
<i>A. sp.</i>	262801	25.4 ^{d-n}	54.6 ^{d-l}	52.5 ^{j-o}
<i>A. sp.</i>	276233	<i>d</i>		
<i>A. sp.</i>	262817	23.6 ^{f-r}	52.0 ^{j-r}	54.7 ^{g-k}
<i>A. sp.</i>	262848			
Series 2. Prohizomatosae		28.3 ^{ab}	55.5 ^{ab}	48.9 ^{d-f}
<i>A. burkartii</i>	338325	28.3 ^{a-e}	55.5 ^{c-k}	48.9 ^{o-s}
Section V: Extranervosae		23.2 ^{bc}	43.4 ^d	46.5 ^d
Series		23.2 ^{bc}	43.4 ^c	46.5 ^f
<i>A. villosulicarpa</i>	336985	23.2 ^{h-r}	43.4 ^v	46.5 st
Section VI: Pseudoaxonomorphae		27.5 ^{ab}	60.1 ^a	54.2 ^{bc}
Series		27.5 ^{ab}	60.1 ^a	54.2 ^{c-e}
<i>A. sp.</i>	338452	26.0 ^{b-i}	56.7 ^{b-i}	54.2 ^{g-l}
<i>A. sp.</i>	338453	29.1 ^{ab}	63.5 ^a	54.1 ^{g-l}

Table I (Continued)

Species/cultivars ^b	P.I. no. ^c (accession no.)	Protein, %		Oil, %
		Full-fat	Fat-free	
Section VII: Triseminalae		30.8 ^a	59.4 ^{ab}	48.2 ^{cd}
Series		30.8 ^a	59.4 ^{ab}	48.2 ^{ef}
<i>A. pusilla</i>	338449	30.8 ^a	59.4 ^{a-e}	48.2 ^{q-s}

^a Values having no common roman superscript letter in each column (full-fat and fat-free protein, or oil) of percentages of sections, series, or species collections and cultivars are significantly ($P \leq 0.05$) different by analysis of variance. ^b Species collections and cultivars are ranked within each series according to dissimilarities and similarities of polyacrylamide disc gel electrophoretic patterns in Figures 1-4. ^c Plant introduction (P.I.) numbers are from Gregory et al. (1973). The taxonomic arrangement follows that published in this reference. "T" designates an accession in the Coastal Plain Station, Tifton, Ga. ^d Lack of seed material prevented quantitative analyses on these collections.

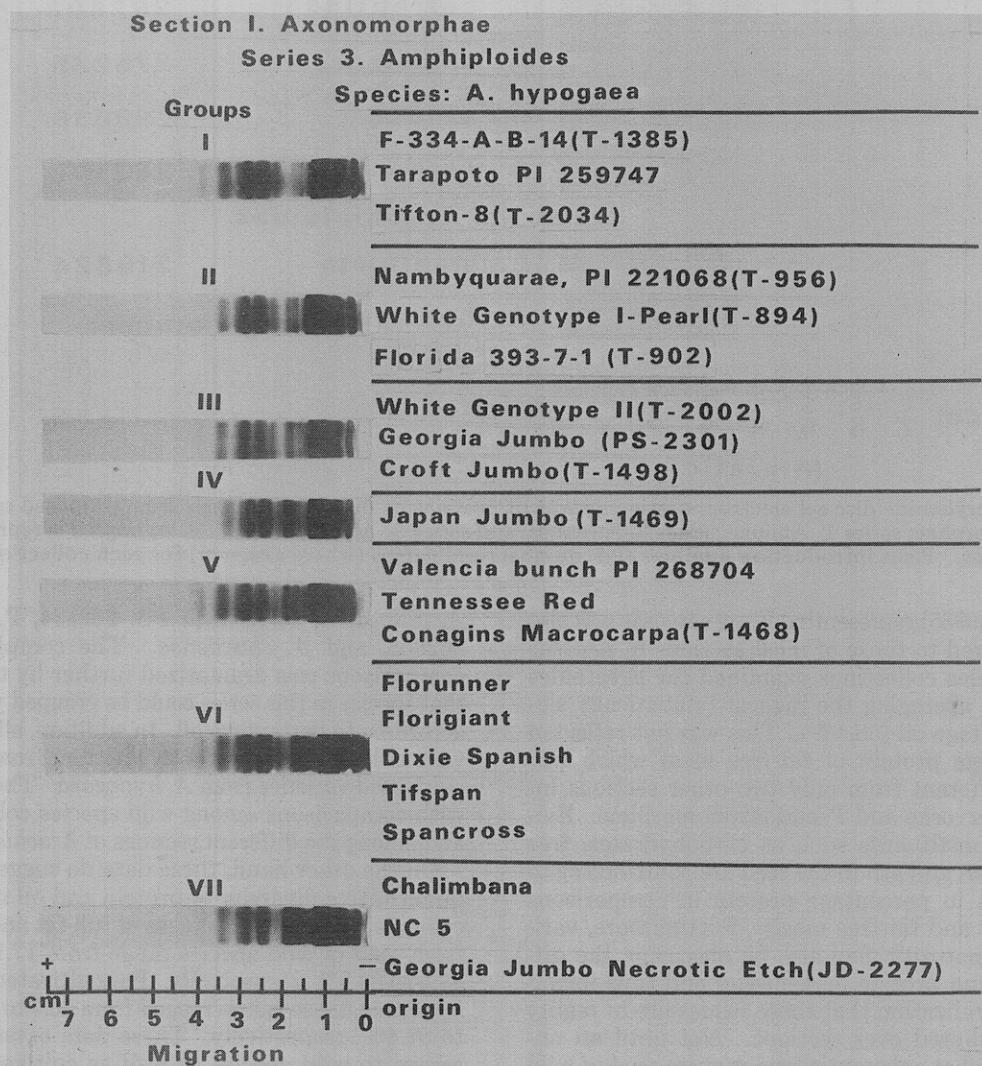


Figure 1. Polyacrylamide disc gel electrophoretic patterns of proteins of full-fat seed meals from selected cultivars of *A. hypogaea*, series 3, Amphiploides, section Axonomorphae. Each cultivar within *A. hypogaea* was subclassified into one of seven classes (groups I-VII) according to the gel pattern of their respective full-fat meal.

section in *Arachis* except section Extranervosae (43.3%). In addition to this latter section containing the least amount of protein in any samples of fat-free meal examined in the genus, it also had a very low oil content (46.5%). However, this percentage oil value was not significantly different from average values of species collections in sections Caulorhizae (49.5%) and Triseminalae (48.2%) and series Amphiploides (50.2%) and Eurhizomatosae (48.9%) of Axonomorphae and Rhizomatosae, respectively (Table I). Further examination of percentage oil shows that average values of species collections examined from the two series of Erectoides, Tetrafoliolatae (58.2%) and Procumbensae (61.4%), are

among the highest sampled in the genus. The complexity of this latter comparison is further exemplified where protein content of fat-free meal is also very high; however, in full-fat material it is among the lowest.

Statistical relationships among average values of species collections of sections were different from those of full-fat seeds. For example, percentage protein of full-fat meals from section Extranervosae was not statistically different from those of all other groups in *Arachis* except Triseminalae (Table I). On the other hand, percentage protein of fat-free meal from Extranervosae was significantly less than all sections compared. Interestingly, the percentage oil in the meal from the only available species (*A. villosa*)

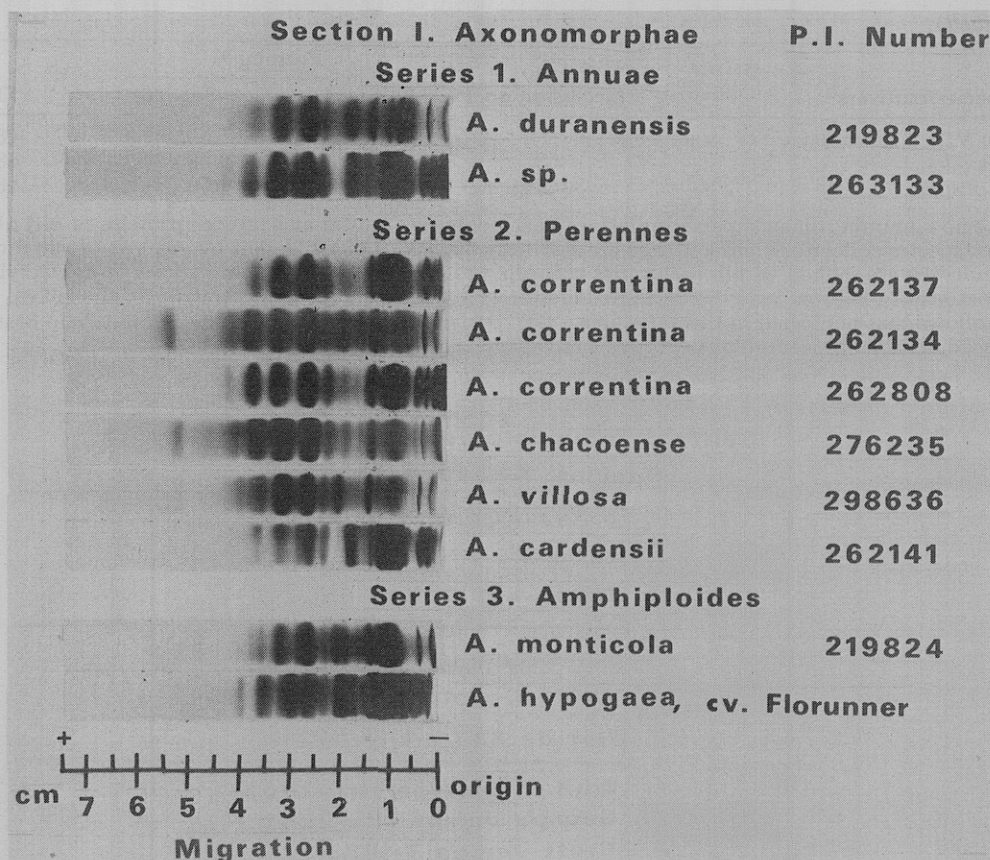


Figure 2. Polyacrylamide disc gel electrophoretic patterns of proteins of full-fat seed meals from proposed species of section Axonomorphae, series 1, Annuae; series 2, Perennes; and series 3, Amphiploides. *A. hypogaea* is represented by the cultivar, Florunner. Plant introduction numbers (P.I. number) and names (where assigned) for each collection are presented.

sulicarpa, P.I. 336985) representing Extranervosae was also very low compared to those of most sections in *Arachis* (Table I). Species collections examined for Erectoides produced meals averaging the highest (statistically significant) percentage oil (58.4%). This was not reflected in the percentage protein of fat-free meal which was significantly different from only two other sections including Extranervosae and Pseudoaxonomorphae. Evidently, other constituents such as carbohydrates, free amino acids, fiber, and ash in the seed are contributing to these variations in percentage protein in comparisons between full-fat and fat-free meals. Furthermore, variations in seed maturity may also be obscuring the differences in oil and protein percentages and thus invalidating the generalization that these values are in reality randomly distributed over sections. Not until an understanding of what substantiates a mature seed in wild species collections is developed will a completely valid answer to this question be obtained.

The data show that it is difficult to determine specific taxonomic relationships of species collections and cultivars within and among samples from sections of *Arachis* based only on percentages of meal protein and oil (Table I). In most cases, values of species collections within a section did not differ significantly from those of other groups. For example, based on statistical analysis of protein values from fat-free meals, species within Perennes, section Axonomorphae, could be grouped as follows: *A. villosa* P.I. 298636 (55.6%), *A. cardensii* P.I. 262141 (55.6%), *A. correntina* "unpublished" P.I. 262808 (54.2%), *A. correntina* P.I. 262134 (49.7%), *A. chacoense* "unpublished" P.I. 276235 (48.2%), and *A. correntina* P.I. 262137 (47.3%). Percentage proteins of full-fat meals grouped these species in a different order as follows: *A. cardensii*,

A. villosa, *A. correntina* P.I. 262134, P.I. 262137, P.I. 262808, and *A. chacoense*. The complexity of these comparisons was dramatized further by the observation that species in this series could be grouped yet another way according to percentage oil. In addition, all of these values were at least not significantly different from many of those cultivated varieties from *A. hypogaea*. The same was true with comparisons among wild species collections within and among the different sections of *Arachis* in this sample.

On the other hand, these data do suggest that there is quantitative diversity of protein and oil in *Arachis*. For example, protein percentages of full-fat and fat-free meals from seed of wild species range from 17.1 to 30.8% and 43.4 to 63.5%, respectively. For cultivated varieties of *A. hypogaea*, these values ranged from 20.7 to 29.1% and 44.8 to 57.4%, respectively. These data extend biochemical assays to wild species as well as cultivars and provide preliminary data on protein and oil diversity for use by geneticists in future breeding programs.

Gel Electrophoresis of Full-Fat Meal Proteins. Gel electrophoretic patterns of seed meals from selected cultivated varieties of *A. hypogaea* are shown in Figure 1, while those of a few wild species collections of *Arachis* are presented in Figures 2, 3, and 4. According to these protein profiles, the cultivated varieties may be separated into seven major groups (I to VII, Figure 1). Differences among groups were noted in the protein bands located in the region 1.0–2.5 cm. For example, group I produced gel patterns containing only one minor or light staining band in region 1.5 cm, group II showed three distinct bands in the region 1.2–1.8 cm while groups III, IV, V, VI, and VII showed increasing amounts of protein in the region 1.0–2.0 cm when compared to one another in this order. In region 2.3 cm, gel patterns of groups I, II, III, and VI showed a

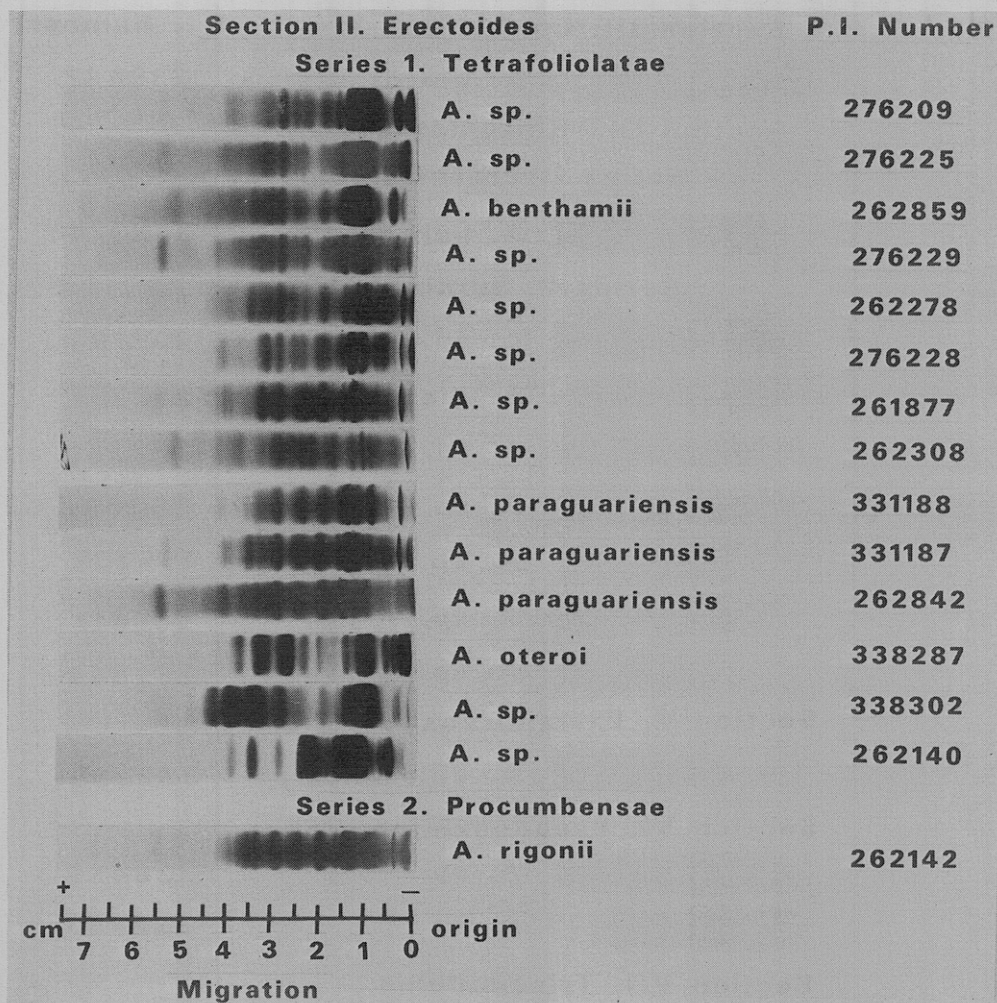


Figure 3. Polyacrylamide disc gel electrophoretic patterns of proteins of full-fat seed meals from proposed species of section Erectoides, series 1, Tetrafoliolatae, and series 2, Procumbensae. Plant introduction numbers (P.I. number) and names (where assigned) for each collection are presented.

distinct protein component whereas those of other groups had only minor bands. Some variability in the amount of protein in bands of region 3.0–4.0 cm was noted among all groups.

These data suggest that there is protein variability among cultivated varieties of *A. hypogaea*. However, most of the major components including arachin (region 0.5–1.5 cm) and nonarachin components (region 2.3–3.5 cm) which make up the majority of peanut seed proteins lack this variability; i.e., although protein quantity in full-fat seed meals of diverse cultivars ranged from 20.6 to 29.1% (or 44.8 to 57.4% for fat-free meals), this variability is not reflected in the quality of major proteins as distinguished by gel electrophoretic techniques. Therefore, proteins such as arachin which lack certain essential amino acids (e.g., lysine, methionine, threonine; Basha and Cherry, 1976) make up the major portion of peanut seed constituents of diverse cultivars of *A. hypogaea* and during breeding programs the result has been the maintenance of nutritional imbalance in commercial cultivated varieties.

New sources of protein material for genetic breeding programs in peanuts are needed to increase diversity in *A. hypogaea*. The wild species of *Arachis* may be an excellent source of new and diverse genetic material. The gel patterns of proteins in extracts from full-fat meals of 37 collections of *Arachis* species which were available for this study show many differences compared to those typical of the *A. hypogaea* sampled (cf. Figures 1–4). For example, many of the gel patterns of species samples in

sections Erectoides (series Tetrafoliolatae), Extranervosae, and Pseudoaxonomorphae contain low amounts of protein in region 0.5–2.5 cm where arachin is normally located. In all sections, except Axonomorphae, most species collections contained protein bands in region 3.5–7.0 cm which were not clearly shown in gels of *A. hypogaea* cultivars in the sample presented in this paper. This was especially true for the following species collections: *A. pinto* P.I. 338447, *A. sp.* P.I. 338452 and P.I. 338453, and *A. sp.* P.I. 338302.

The typical gel pattern of *A. hypogaea*, C.V. Florunner, was similar to those of its closest wild relatives in section Axonomorphae, series Annuae and Perennes. Similar patterns were especially noted for *A. hypogaea* C.V. Florunner and the wild relative *A. monticola* P.I. 219824 which are both taxonomically placed in the series Amphiploides.

In summary, by utilizing gel electrophoretic techniques this study shows chemotaxonomically that storage proteins from full-fat meals of a number of *Arachis* species collections further support the thesis that the samples of related species material have been properly classified by conventional taxonomic methods into coherent sections and series as presented in Figures 2, 3, and 4 (Gregory et al., 1973; Cherry, 1975). Species collections of Axonomorphae are typified by high arachin composition in the region 0.5–1.5 cm and nonarachin proteins in the region 2.5–4.0 cm (Basha and Cherry, 1976). Most gel patterns of species collections in section Erectoides contain a number of equally distinct bands throughout the region

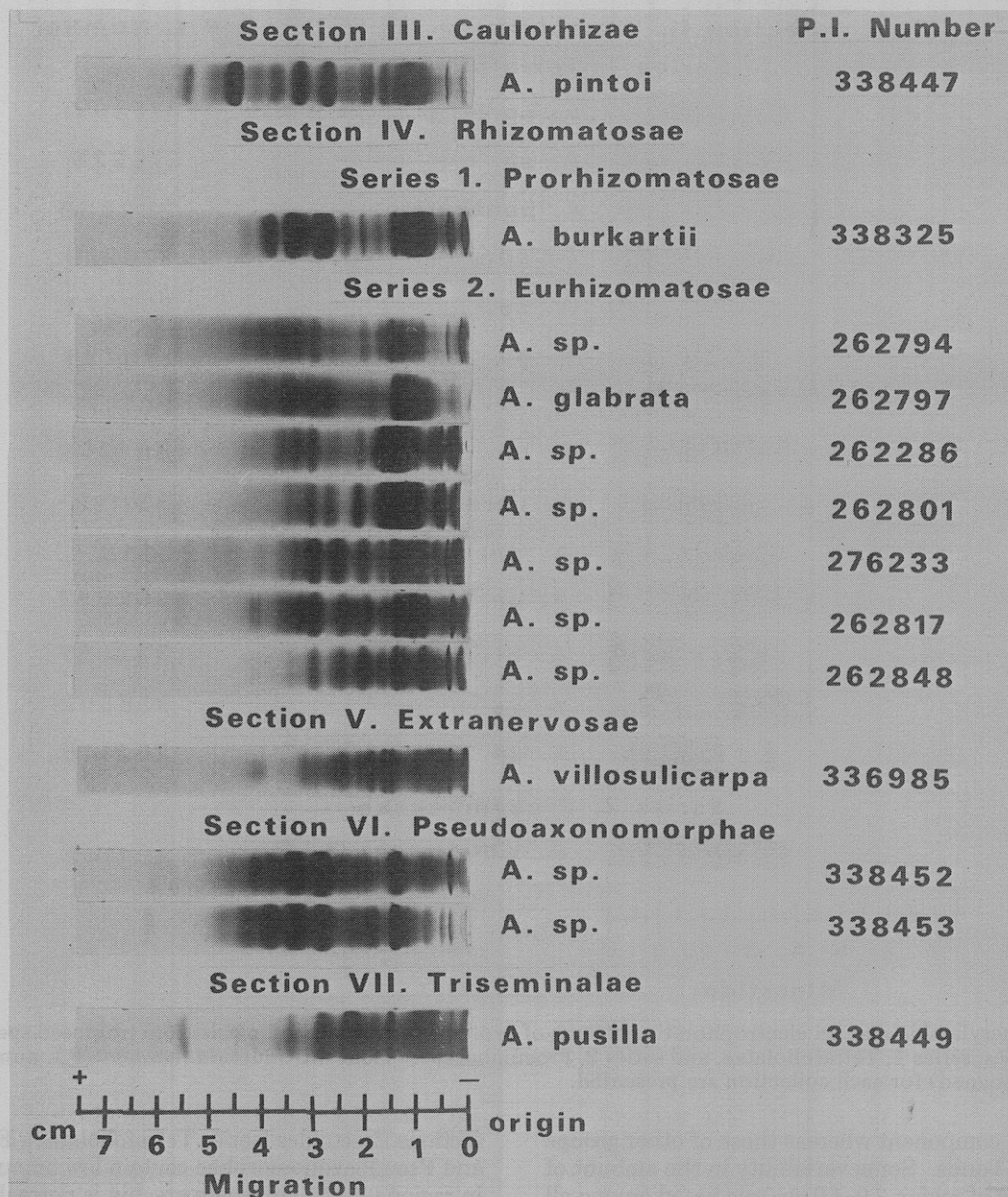


Figure 4. Polyacrylamide disc gel electrophoretic patterns of proteins of full-fat seed meals from proposed species of sections Caulorhizae, Rhizomatosae, series 1, Prorhizomatosae, and series 2, Eurhizomatosae, Extranervosae, Pseudoaxonomorphae, and Triseminalae. Plant introduction numbers (P.I. numbers) and names (where assigned) for each collection are presented.

0.5–3.5 cm. Some of the species collections within this section contain higher amounts of protein in the arachin region of the gel than others. *Arachis* sp. P.I. 338202 and *A. oteroi* “unpublished” P.I. 338287 were represented by gel patterns closely resembling those in section Axonomorphae; *A. sp.* P.I. 262140 seems to contain mainly proteins having similar mobilities on electrophoretic gels as components of arachin. Most of the collections (*A. sp.*) sampled from section Rhizomatosae contained a number of major or dark staining components in the region 0–3.0 cm. Protein patterns of Caulorhizae, Extranervosae, Pseudoaxonomorphae, and Triseminalae distinguish each of these sections in *Arachis*. Interestingly, the gel patterns of the two collections of Pseudoaxonomorphae were very similar.

Contrary to observations made with the sample of cultivated varieties of *A. hypogaea*, seed meals of wild species collections had qualitative differences in addition to much quantitative variation in proteins. Thus, the potential economic value of the wild relatives of *A. hypogaea* is apparent not only because they offer important

sources of diverse germ plasm for improving agronomic characteristics but also protein material with the potential of expanding functional and nutritional properties of peanuts when utilized as food ingredients.

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Rapeseed Protein Isolates by Countercurrent Extraction and Isoelectric Precipitation

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A countercurrent procedure for the extraction of protein from defatted rapeseed meal is described. Using 0.02 N NaOH and a meal to solvent ratio of 1:25 as much as 94% of meal nitrogen is extracted. A two-step precipitation, first at pH 6.0 and then at pH 3.6, the two isoelectric points of the protein in the extract, affords two protein fractions, which contain 69.4 and 24.3% of the meal protein, respectively. After washing the curds with water and drying with acetone two highly pure isolates are obtained. Protein isolate I, light grey, contains 92.9% protein and protein isolate II, white, contains 98.6% protein, both on a dry weight basis.

In view of the world-wide protein shortage, oilseeds are becoming of increasing importance as sources of edible proteins (Altschul, 1974). Rapeseed, which is a major oilseed crop of the temperate zones, has so far found little application in the production of protein.

Factors which favor the use of rapeseed protein in human nutrition include: (a) rapeseed is cultivated in countries which do not have suitable agricultural or environmental conditions for growing other oilseeds (Ohlson, 1972); (b) the concentration of essential amino acids in rapeseed protein compares favorably with the amino acid requirements of human adults (FAO, 1957; El Nockrashy et al., 1975a); (c) rapeseed protein is characterized by

relatively high contents of lysine (Leslie and Summers, 1975; El Nockrashy et al., 1975b) which is the first limiting amino acid in cereals and other vegetable proteins; and (d) rapeseed protein concentrates having high nutritional value and satisfactory functional properties are well accepted (Ohlson and Tear, 1974).

Problems involved in the use of rapeseed in nutrition include the presence of glucosinolates, which, on enzymatic hydrolysis, yield undesirable and even toxic factors such as isothiocyanates, nitriles, and oxazolidinethione (van Etten, 1969) and the relatively high cellulose content of the seeds (Theander and Åman, 1974). The breeding of new varieties of rape has been only partially successful with regard to reducing the glucosinolate and cellulose contents of the meal (Röbbelen and Leitzke, 1974).

The present investigation was carried out with the aim to develop a process for the preparation of protein isolates free of such undesirable substances. Seeds of two new varieties of rape were used as starting materials, viz., *Brassica napus*, Erglu, a summer variety which is low both in erucic acid and glucosinolates, and *Brassica napus*,

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