Variability in Fatty Acid Composition Among *Arachis* Genotypes: A Potential Source of Product Improvement[1]

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ABSTRACT

Research on peanut (*Arachis hypogaea* L.) genotypes has shown a high degree of genetic variability in fatty acid composition. The two major oil fatty acids, oleic and linoleic, range between 36-69% and 14-40%, respectively, and together make up 75-85% of total fatty acids. The very long chain (C_{20}-C_{24}) fatty acids make up 4-9%, palmitic acid 7-13%, and stearic acid 2-5% of total fatty acids. Stability of oil samples as measured by length of autoxidation induction period at 60 C shows variable but statistically significant ($P<0.01$) correlations with levels of linoleic acid; peanut butter samples show similar patterns of stability. Selection for lower levels of linoleic acid in the development of new varieties of peanuts should result in products with significantly improved shelf life. Some genotypes show consistent differences in oil stability patterns that are not related to oil linoleic acid content. Analysis of entries from 16 wild *Arachis* species collections revealed levels of oil linoleic acid higher than those found in *A. hypogaea*. One species, *A. villosulicarpa*, contained 49% linoleic acid and 21% very long chain acids. The range in linoleic acid within *A. hypogaea* and availability of suitable techniques for measuring selection progress give scope for product improvement through breeding.

INTRODUCTION

Peanut (*Arachis hypogaea* L.) oil has for many years been one of the world's major vegetable oils and in 1974 ranked third in importance after soy and sunflower oil. In contrast to other areas of the world where the bulk of the peanut crop is crushed for oil, more than half of the US production is consumed as peanut butter and other full fat peanut products. These products contain approximately 50% oil and are therefore susceptible to oxidative deterioration unless preventive measures are taken in handling, packaging, and storage.

Through the years various workers have observed differences in iodine value and susceptibility to oxidative rancidity in products derived from different varieties and types of peanuts, but prior to the advent of gas liquid chromatography (GLC), data on the fatty acid composition of oils were obtained with considerable difficulty using various combinations of physical and chemical methods. The data obtained by these techniques indicated considerable differences in the fatty acid composition of oils obtained from different localities and from different varieties of peanuts (1-3). Crawford and Hilditch (1) investigated peanut oils obtained from different locations in Africa and found large differences in levels of linoleic acid, but were unable to ascribe these differences to genetic or environmental factors. They pointed out, however, that those oils highest in linoleic acid could reasonably be expected to be most susceptible to oxidative rancidity. The stability of oils has subsequently been shown to be related to linoleic acid content (3) as well as to other undefined factors (2,4,5). It would seem therefore that one fruitful approach in the improvement of the shelf life of peanut products would be through breeding and selection for lower levels of linoleic acid or for other characteristics that would improve product stability. An assessment of genetic diversity is of necessity the first step in a program of this type.

In this paper we present fatty acid and oil stability data obtained over a period of several years from a substantial number of genotypes of *A. hypogaea*, together with fatty acid data from other species of the genus *Arachis* that are of interest to peanut breeders. The stability of peanut butter samples prepared from several *A. hypogaea* genotypes was also determined.

Many of the genotypes of *A. hypogaea* examined in this study were described in an earlier paper by Young and Hammons (6), in which these authors presented a color photograph showing variations in seed size, shape, and testa color together with genotype code numbers, trivial names, plant introduction numbers, and country of origin. Statistical relationships between oil linoleic acid content and oil stability have been published elsewhere (4) as have fatty acid correlations and yearly fatty acid values for some of the varieties (7).

MATERIALS AND METHODS

Genotypes of *A. hypogaea* were grown and processed as described previously (4). Seed samples from other *Arachis* spp. were grown in 1970 at one location or were made available by the Southern Regional Plant Introduction Station at Experiment, GA. Oil stability measurements, reported in oven days, were made by holding samples of ca. 200 mg in a forced air oven at 60 C and determining the number of days until first definite gain in weight (7). The stability of peanut butter samples, prepared as described by Cecil (8) was evaluated in a similar manner except that 0.5 g samples were used. Fatty acid methyl esters were prepared and determined by GLC on either 10% diethylene

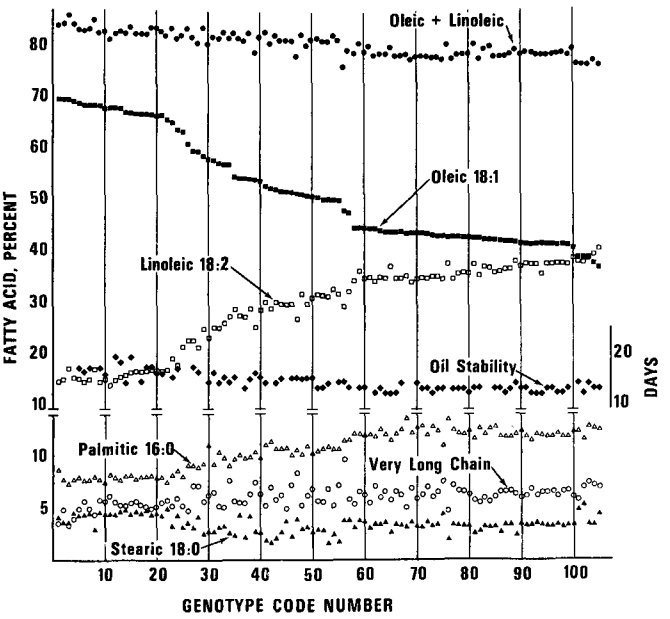

FIG. 1. Variations in oil fatty acid and stability values among genetically diverse peanut genotypes.

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glycol succinate (DEGS) (4) or 8% butanediol succinate (BDS) columns (9).

RESULTS AND DISCUSSION

The genetic variability in fatty acid composition within *A. hypogaea* is shown by arranging genotypes sequentially from highest to lowest in levels of oleic acid (18:1) as percent of total fatty acids (Fig. 1). The code numbers assigned in Figure 1 are cross-referenced in Table I with serial code numbers previously assigned to these genotypes by Young and Hammons (6). All other code numbers in this paper (Figures 2, 3; Tables IV, V) are those assigned by Young and Hammons (6).

The population of varieties portrayed in Figure 1 may be divided into three groups based on oleic acid content. Those in Group 1 (genotypes 1-25) are high in oleic acid, those in Group 2 (genotypes 26-57) are intermediate, while those in Group 3 (genotypes 58-105) are relatively low in oleic acid. Groups 1 and 2 are composed almost exclusively of genotypes that fall into the *A. hypogaea* ssp. *hypogaea* classification, characterized by seed dormancy, while those in Group 3 are predominantly of the *fastigiata* subspecies. Exceptions are genotypes 37, 52, and 57 in Group 2, tentatively classified as nondormant; and genotypes 59, 65, 76, 81, 86, 89, 100 in Group 3, tentatively classified as dormant. Members of the *fastigiata* subspecies are characterized by absence of seed dormancy (10).

Current commercial 'Virginia' and 'Runner' cultivars fall into Group 2, and 'Spanish' and 'Valencia' cultivars are similar in fatty acid composition to the Group 3 genotypes. No commercial US varieties fall within Group 1.

Hybridization resulting from crosses between subspecies of *A. hypogaea* as well as between *A. hypogaea* and other *Arachis* ssp. plays an increasingly important role in the development of new varieties of peanuts, and many of the recently introduced varieties were developed by these methods (11). Thus the subspecies-fatty acid composition relationship apparent in Figure 1 may not continue indefinitely. However, the distribution in Figure 1 could be a useful guide for evaluating progress in progenies of infraspecific and interspecific crosses.

In Group 1 genotypes, oleic and linoleic acid make up ca. 84% of total fatty acids, a value that decreases to ca. 76% for genotypes in Group 3. This decrease is accompanied by an increase in palmitic acid and to a lesser extent by an increase in very long chain (C_{20}-C_{24}) fatty acids. The

TABLE I

Genotype Code Numbers

1[a]	(66)[b]	36	(100)	71	(1)
2	(90)	37	(77)	72	(14)
3	(105)	38	(83)	73	(34)
4	(104)	39	(64)	74	(5)
5	(70)	40	(26)	75	(36)
6	(60)	41	(42)	76	(84)
7	(54)	42	(62)	77	(31)
8	(89)	43	(43)	78	(22)
9	(58)	44	(46)	79	(15)
10	(59)	45	(95)	80	(30)
11	(67)	46	(44)	81	(81)
12	(69)	47	(55)	82	(92)
13	(53)	48	(29)	83	(98)
14	(51)	49	(25)	84	(96)
15	(65)	50	(57)	85	(6)
16	(103)	51	(74)	86	(49)
17	(41)	52	(24)	87	(18)
18	(61)	53	(48)	88	(16)
19	(56)	54	(88)	89	(85)
20	(52)	55	(27)	90	(9)
21	(40)	26	(50)	91	(12)
22	(101)	57	(76)	92	(23)
23	(68)	58	(39)	93	(10)
24	(73)	59	(86)	94	(80)
25	(93)	60	(8)	95	(20)
26	(94)	61	(97)	96	(13)
27	(32)	62	(4)	97	(11)
28	(63)	63	(2)	98	(19)
29	(91)	64	(33)	99	(17)
30	(72)	65	(87)	100	(102)
31	(28)	66	(3)	101	(79)
32	(71)	67	(38)	102	(35)
33	(75)	68	(99)	103	(82)
34	(45)	69	(7)	104	(37)
35	(47)	70	(21)	105	(78)

TABLE II

Fatty Acids of Wild Species of *Arachis*[a]

P.I. Number	Section and series[b] Species	Fatty acids, percent 16:0	18:0	18:1	18:2	18:3	20:0	20:1	22:0	24:0
I. Arachis 1. Annuae										
219823	*A. duranensis*	10.8	2.1	38.5	41.2	<.1	1.5	1.1	3.4	1.5
262133	*Arachis* sp.	8.4	1.8	59.3	24.2	<.1	1.1	1.3	2.4	1.4
I. Arachis 2. Perennes										
338279	*Arachis* sp.[c]	8.9	1.7	40.8	37.1	<.1	1.1	1.9	5.8	2.4
338280	*Arachis* sp.[c]	8.3	1.6	39.0	37.2	<.1	1.2	2.4	6.3	2.7
210554	*A. villosa*	9.6	2.2	51.0	31.8	<.1	1.2	0.9	1.9	1.1
258943	*A. villosa*	9.3	1.7	40.0	41.6	<.1	1.0	1.6	3.2	1.6
210555	*A. correntina*[d]	9.3	2.2	50.9	31.4	<.1	1.2	1.1	2.5	1.3
262808	*A. correntina*[d]	9.5	2.1	59.3	31.9	<.1	1.2	1.2	3.1	1.4
I. Arachis 3. Amphiploides										
219824	*A. monticola*	12.8	3.9	39.4	36.0	<.1	1.8	0.7	3.9	1.5
405933	*A. monticola*	11.4	3.6	43.1	34.7	<.1	1.9	0.9	4.0	1.5
II. Erectoides 2. Tetrafoliolatae										
262842	*A. paraguariensis*	7.6	1.5	45.3	33.7	<.1	1.0	2.4	5.7	2.4
262874	*A. paraguariensis*	8.2	0.9	31.4	48.1	<.1	0.7	2.4	5.5	2.4
III. Rhizomatosae 2. Eurhizomatosae										
243334	*Arachis* sp.[c]	10.4	2.7	43.2	38.9	<.1	0.9	0.8	1.8	1.2
338261	*Arachis* sp.[c]	6.7	1.1	38.8	38.1	.1	1.2	3.4	6.9	3.4
338262	*Arachis* sp.[c]	9.4	2.5	31.1	43.4	.3	1.7	1.9	6.8	2.7
IV. Extranervosae										
263396	*A. villosulicarpa*	11.1	1.4	14.3	50.8	<.1	0.8	1.2	14.0	5.9

[a]Grown at Tifton, GA 1970.

[b]Arranged by R.O. Hammons after Krapovickas (10) and W.C. Gregory (Personal communication).

[c]Species undetermined.

[d]Unpublished (formerly *A. villosa* var. *correntina* Burk.).

TABLE III

Fatty Acids in Four Accessions of *Arachis villosulicarpa*[a]

P.I. No.	Fatty acids, percent							
	16:0	18:0	18:1	18:2	20:0	20:1	22:0	24:0
263396	11.5	2.0	17.1	48.9	1.2	1.3	12.7	4.1
273482	9.5	2.4	15.5	48.0	1.3	1.2	14.5	4.3
336984	9.6	2.6	16.8	49.2	1.4	1.1	13.5	4.3
336985	9.7	2.7	15.4	48.6	1.6	1.0	15.0	4.7
Average	10.1	2.4	16.2	48.7	1.4	1.2	13.9	4.4

[a]Seed furnished by Southern Regional Plant Introduction Station, Experiment, GA.

positive correlation between palmitic and linoleic acid, previously reported by Worthington and Hammons (7), is not as apparent when the examination is limited to genotypes within a group and probably accounts for the failure of other workers to detect this correlation among the fatty acids of *A. hypogaea* (12,13). The total very long chain fatty acid value also appears to be positively correlated with linoleic acid although a great deal of scatter is evident. Individual correlations among these acids have been reported elsewhere (7).

Other species of the genus *Arachis* were also examined for fatty acid distribution patterns. Many of these species are of interest to peanut breeders because of resistance or immunity to insect and disease pests of *A. hypogaea*, and efforts are underway to incorporate this genetic material into commercial cultivars of peanuts. Two current cultivars, Spancross and Tamnut 74, were developed from crosses between *A. hypogaea* L. and *A. monticola* 'Krap. et Rigoni' (14,15).

With one exception (*A. villosulicarpa*) the wild species (Table II) show fatty acid patterns similar to those of Groups 2 and 3 of *A. hypogaea*. This similarity was not totally unexpected since 10 of the 16 accessions are in the same section (*Arachis*) of the genus as is *A. hypogaea* (10). *A. villosulicarpa* 'Hoehne,' PI 263396, is distinctly different from all others, showing a very low level of oleic acid, a high level of linoleic acid, and unusually high levels of behenic (22:0) and lignoceric (24:0) acids. Analysis of seed of this and 3 additional introductions of *A. villosulicarpa* furnished by the Southern Regional Plant Introduction Station (Table III) indicate that this pattern is a species characteristic. *A. villosulicarpa*, the only other species of *Arachis* known to be grown by man for food, is cultivated by Indians in the Mato Grosso of Brazil. It is highly resistant and perhaps immune to Cercospora, a serious leafspot disease of *A. hypogaea*. It is hoped that the genes responsible for leafspot immunity can be transferred or utilized independently of those responsible for the high levels of very long chain saturated fatty acids.

Linolenic acid (18:3) was found in all entries of the wild species (Table II) and at levels of 0.3% and 0.1% in PI 338262 and 338261. These values are the highest found thus far among species of *Arachis* including *A. hypogaea*. All entries of *A. hypogaea* examined in both this and a previous study (9) have contained <0.1% (0.02%-0.08%) linolenic acid.

Of particular interest is the relationship between oil stability and fatty acid patterns. The well established negative correlation between oil stability and linoleic acid content is shown by 3-year average values for *A. hypogaea* genotypes in Figure 1. The yearly r^2 values were .21, .74, and .46 (4). Oil stability values decrease from a high of ca. 18 oven-days for some genotypes low in linoleic acid to values of 12 to 13 days for varieties high in this acid. Although the scatter of stability data indicates varietal differences in this characteristic that are independent of linoleic acid levels, the overall negative correlation between stability and linoleic acid indicates that the stability and shelf life of peanut products could be improved by breeding

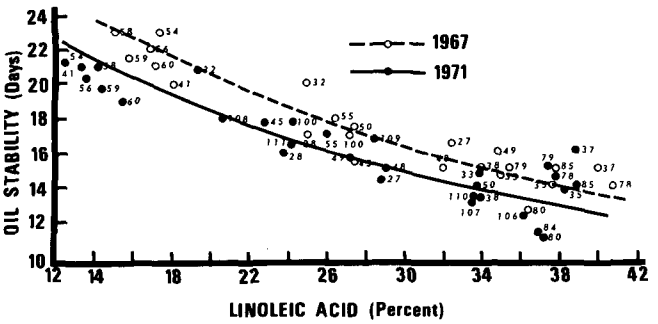

FIG. 2. Relationship between linoleic acid content and stability of peanut oils.

TABLE IV

Comparison of Oil Stability and Linoleic Acid Values for Two Varieties of Peanuts Grown during Five Crop Years

Year	Variety			
	37 White genotype II		80 P.I. 268704	
	18:2 %	Oil[a] stability	18:2 %	Oil[a] stability
1965	38.1	13	35.4	13
1967	40.0	15	40.7	12
1968	40.7	11	39.1	12
1971	38.9	16	36.9	11
1972	39.2	14	37.2	11
Ave.	39.4	13.8	37.9	11.6

[a]Length of autoxidation induction period in days at 60 C.

or selecting for lower levels of linoleic acid and higher levels of oleic acid.

In some crop years the relationship between oil stability and 18:2 content is pronounced (Fig. 2). Examination of data from individual varieties reveals differences in stability that are independent of linoleic acid content. Oil from variety 37 is more stable than that from variety 80, even though 37 has a higher average linoleic acid content. This relationship between these two varieties is rather consistent (Table IV). In four out of five years in which both varieties were grown at the same location, 37 had a higher 18:2 content than 80. In one year (1967) linoleic acid values for the two varieties were not appreciably different; however, the autoxidation induction period of oil from 37 was 25% longer than the oil from 80. In three of the remaining four years, the stability values of oil from 37 were equal to or greater than those from variety 80; in one year the value was slightly lower. The stability curves of three varieties that were grown at a common location, processed in the same manner, and evaluated simultaneously in the same oven are shown in Figure 3. The autoxidation induction period of variety 78 with 40% linoleic acid was ca. 30% longer than variety 80 with 37% linoleic acid. Varieties 37 and 78 have one feature in common—a white testa—but are otherwise unrelated. Growth characteristics are different; 37 was collected in Brazil and 78 in India.

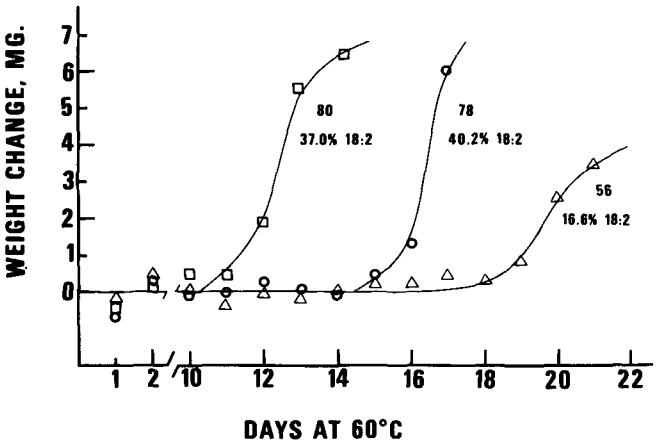

FIG. 3. Stability curves of three varieties of peanuts grown at a common location, similarly processed, and evaluated simultaneously in the same oven.

TABLE V

Levels of Linoleic Acid Content and Stability of Oils and Peanut Butter Prepared from Seven Varieties of Peanuts

Code	Variety	18:2 %	Stability values[a]	
			Oil	Peanut butter
37	White Genotype II	38.8	14.3	24.3
80	PI 268704	37.0	11.0	16.3
109	Florunner	29.0	13.6	26.0
111	PI 290569	25.7	13.6	26.6
28	Va. Bunch 67	24.4	12.0	29.3
32	Fla. 393-7-1	20.3	16.0	35.0
41	Jenkins Jumbo	15.6	17.3	37.0

[a]Length of autoxidation induction period in days at 60 C.

The rate and extent of weight gain during autoxidation is proportional to level of linoleic acid as shown in Figure 3. Maximum weight is attained after 3 to 4 days of autoxidation and decreases gradually thereafter with the evolution of volatile oxidation products.

At the present time we do not know the basis for those differences in oil stability that are unrelated to levels of linoleic acid. Glyceride structure has been reported to have an effect on the stability of oils (16). In addition, Brown et al. (5) reported differences in stabilities of oils that were dependent upon method of oil preparation. Solvent extracted oils were more stable than those prepared by hydraulic press; the most stable oils were obtained by extraction with a chloroform-methanol solvent system. These workers postulated that the increased stability of solvent extracted oil may have been due either to traces of solvent or solvent contaminants remaining in the oil or to a more efficient extraction of seed antioxidants by the more polar solvents. The latter explanation would appear to be the more likely one.

The stability of peanut products is known to be influenced by a number of factors associated with processing, storage, and perhaps climatic conditions during seed development, and in some instances these factors may be the chief determinants of shelf life of peanut butter and other full-fat peanut products. However, the available evidence shows that varietal or genetic factors are of considerable importance in this respect and that stability characteristics of peanut butter, and perhaps other full-fat products, can be inferred from oil stability data. Stability data of oil and peanut butter samples prepared from seven varieties grown in 1972 are shown in Table V. With the exception of peanut butter prepared from variety 37, which is atypical in stability, the stability values of peanut butter samples are inversely related to levels of linoleic acid, and, with the additional exception of oil from variety 28, the oil samples show a similar pattern. In a study of peanut butter samples prepared from 10 peanut genotypes and stored under stress conditions for 4 and 14 months at 38 C, Cecil (8) reported a significant correlation between flavor and aroma and a stability rating composed of 0-25 points each for oil stability values, oleic acid, linoleic acid, and the oleic/linoleic ratio. Thus it appears that these parameters as measured in the laboratory are useful predictors of the stability of full-fat peanut products.

Genetic studies (17,18) have shown peanut oil fatty acids to be quantitatively inherited and under a complex system of genetic control. In hybridization studies, those progenies selected for high yield potential appear to be highest in linoleic acid content, suggesting a genetic relationship between these characteristics. Such a relationship, if it exists, will make difficult the development of new varieties with both improved yield potential and lower levels of linoleic acid.

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