Peanut Science (1977) 4, 67-70

Internal Pericarp Color as a Subjective Maturity Index for Peanut Breeding[1]

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ABSTRACT

Fruit of 10 peanut (*Arachis hypogaea* L.) genotypes differing in botanical type and geographical source were evaluated to establish parameters for making reliable maturity determinations on the basis of internal pericarp color (IPC), and to compare the IPC, kernel density (KD) and arginine maturity index (AMI) methods of estimating peanut maturity.

Kernels from fruits with detectable, non-disease related internal pericarp darkening were significantly lower in density than kernels from fruits without internal pericarp darkening. No difference in density was detected between kernels from fruits differing in intensity of internal pericarp darkening. In general, two-seeded fruits with detectable non-disease related darkening in at least one orbital of the pericarp were mature, whereas fruits with no internal pericarp darkening were immature.

Mature fruit percentages were determined on sample sizes of 25, 50, 75, and 100 fruits. Although standard errors were consistently high for 25-fruit samples, means and standard errors were similar for sample sizes of 50, 75, and 100 fruits, indicating that estimates based on 50-fruit samples were reliable.

Post-harvest stability of IPC was evaluated from determinations made on five dates at 30-day intervals. IPC did not change sufficiently during the 120 day period to affect the maturity estimates.

Kernel samples classified as mature by the IPC method were significantly lower in density and free arginine content than kernels classified as immature. Correlations among maturity estimates using the IPC, KD and AMI methods were highly significant, with all coefficients exceeding 0.95.

The results indicate that peanut samples can be evaluated reliably for maturity by subjective classification of IPC. Maturity estimates on the basis of IPC were as effective as those determined using either the KD or AMI methods. The IPC method requires no sophisticated equipment and it is simple, rapid and nonsacrificial which makes it particularly useful in breeding programs involving large numbers of maturity determinations.

Additional index words: *Arachis hypogaea* **L., kernel density, arginine maturity index.**

Considerable interest has been expressed in the development of peanut (*Arachis hypogaea* L.) cultivars that produce high percentages of harvestable mature fruits. Recent results (4) have shown that peanut genotypes grown under common environments differ in their ability to produce high proportions of mature fruits, indicating that genotypes producing high percentages of mature fruits at harvest might be achieved through breeding. Programs designed to identify and utilize genes conditioning this character will involve maturity evaluations of large numbers of genotypes during segregating generations, and reproduction from select fruits in variety development programs. Maturity evaluations used in breeding for this character, therefore, must be reliable, rapid, and nonsacrificial.

Several methods have been developed for determining maturity in peanuts. Sharon (16), Emery et al. (3), and others (1,5) reported that optical density of oil extracted from peanut kernels is a potential index of peanut maturity. Free arginine content of peanut kernels was reported to be a reliable index for maturity in peanuts by Newell (12), Mason et al. (10), and Young and coworkers (17, 19, 20). Results obtained by Aristizabal et al. (1), Kramer et al. (9), and Miller and Burns (11) indicated that kernel density can be used for estimating maturity in peanuts. These indices are accurate and objective, but are both time consuming and expensive. They require relatively large numbers of fruit for reliable maturity determinations, and some require destruction of the fruit.

The most common method of determining maturity in peanuts is based on color of the internal surface of the pericarp. Darkening of the internal surface of the pericarp has been shown (11,13,15) to be directly related with kernel maturity. This method can be used to classify individual pods for maturity, and when used subjectively, is inexpensive, rapid, and nonsacrificial. Subjective methods of determining maturity in peanuts, however, have been criticized (2) because of inconsistencies that might occur when the methods are used by different people on different cultivars and in different seasons. This study was conducted to establish parameters which would maximize the effectiveness of maturity determinations made subjectively on the basis of internal pericarp color (IPC), and to compare results obtained using these techniques with both the kernel density (KD) and arginine maturity index (AMI) methods for estimating peanut maturity.

Materials and Methods

Fruit of three peanut (*Arachis hypogaea* L.) cultivars, 'Starr', 'New Mexico Valencia A', and 'Florunner', of the Spanish, Valencia, and Virginia botanical types, respectively, and seven Spanish-type plant introductions (139919, 149639, 248759, 259611, 268750, 288021, and 341885) of diverse geographical sources were used in the studies. The fruit were produced on Patilo-type soil near College Station, Texas. Recommended insect, disease, and weed control practices were followed during each production season. Fertilizer and gypsum were applied according to soil test recommendations and water was applied as needed by sprinkler irrigation.

[1]Contribution from Dep. of Soil and Crop Sci., Texas A&M Univ., College Station, TX 77843. Part of a dissertation submitted by the senior author in partial fulfillment of the requirements for the Ph.D. degree. Research supported in part by a Tom Slick Graduate Research Fellowship. Approved as T.A. no. 13395 of the Texas Agric. Exp. Stn., College Station, TX 77843.

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Harvests were made at several dates which encompassed the fruit maturation period of each genotype to obtain fruit representing a range in maturity. All fruit were dried artificially to 13% moisture immediately after harvest and stored at room temperature until used. Fruit used in each comparison were produced during the preceeding growing season.

Peanut fruits having discoloration on the internal surface of the pericarp commonly are considered mature, whereas fruits without discoloration are considered immature. Maturity classification problems occur, however, with fruits which exhibit slight discoloration in only one orbital of the pericarp. To quantity the relationship between IPC and kernel maturity, fruits from a composite sample comprised of approximately 250 multiseeded fruits of each genotype were classified individually on the basis of internal pericarp color and segregated into one of three kernel classes. Fruits with no detectable darkening on the internal surface of the pericarp were considered immature; fruits with detectable non-disease related darkening in one orbital of the pericarp were considered intermediate; and fruits with distinct non-disease related darkening in at least one orbital of the pericarp were considered mature. Sixteen 20 g kernel samples were selected at random from each kernel class and analyzed for density, using a Beckman Air Comparison Pycnometer.

In a separate study, fruit of Starr, Florunner, and PI 248759 were evaluated to determine the minimum sample size necessary for reliable maturity evaluations. Four subsamples of 25 two-seeded fruits selected at random from samples of each genotype were analyzed for percentage of mature fruits on the basis of IPC. Fruits with detectable non-disease related internal pericarp darkening were considered mature whereas fruits without detectable darkening were considered immature. After each subsample was classified, maturity estimates were combined allowing comparisons between estimates based on 25, 50, 75, and 100 fruits. A total of 56 samples were analyzed for each genotype.

To determine if IPC changes sufficiently after harvest to alter maturity estimates, 100 two-seeded fruits of each of three genotypes, Starr, Florunner, and PI 268750, were evaluated for percentage of mature fruits at five dates after harvest. Evaluations were made on the basis of IPC at 30 day intervals beginning as soon after harvest as possible. Four replications were conducted.

The reliability of maturity estimates determined on the basis of IPC was evaluated by comparing results obtained using this method with both the KD (1) and AMI (8, 18) methods. Two samples of approximately 500 multiseeded fruits were selected at random from each of the ten genotypes and analyzed for maturity on the basis of IPC. Kernels from fruits with detectable non-disease related darkening on the internal surface of the pericarp were considered mature, whereas fruits without darkening were considered immature. Two 25 g subsamples of kernels were selected at random from both the mature and immature kernel classes of each genotype. Each subsample was then evaluated for kernel density using a Beckman Air Comparison Pycnometer, and for free arginine content using the Sakaguchi reaction, as modified by Izumi (6, 7) and Young and coworkers (18, 20). Correlations were calculated between each on the three methods using subsample estimates. Analyses of variance were computed on data from all experiments and means were compared using Duncan's New Multiple Range Test.

Results and Discussion

Peanut kernels segregated on the basis of IPC had highly significant differences in density (Table 1). Kernels from fruits exhibiting darkening on the internal surface of the pericarp (mature and intermediate maturity classes) had lower densities than kernels from fruits without internal pericarp darkening (immature maturity class). An inverse relationship between kernel density and fruit maturity also has been reported in peanuts by others (1,11).

No significant difference in density was detected between kernels from the intermediate and mature maturity classes (Table 1). Distinction between kernels in these two classes was based on IPC intensity, with kernels in the intermediate maturity class arising from fruits with only slight darkening in one orbital of the pericarp. The results indicate that peanut fruit exhibiting detectable non-disease related darkening in at least one orbital of the pericarp are mature. Several instances were observed, however, where fruit of New Mexico Valencia A, which produces a high frequency of 4- and 5-seeded pods, had detectable darkening in the basal orbitals of the pod, but noticably immature seed in orbitals at the apical end of the pod. Inclusion of these immature seeds into the intermediate maturity class undoubtedly resulted in the slightly higher density obtained for the intermediate maturity class than for the mature maturity class (Table 1). Detectable non-disease related darkening in more than one orbital of the pericarp is needed for reliable maturity determinations of fruit that contain more than two seeds.

Table 1. Density of peanut kernels separated for maturity on basis of internal pericarp color.

Maturity class	Density (g/cc)
Mature	1.089 a[1/]
Intermediate	1.091 a
Immature	1.149 b

[1/] Means followed by the same letter are not significantly different at the 0.01 probability level (Duncan's New Multiple Range).

Maturity estimates based on sample sizes of 25, 50, 75, and 100 fruits did not differ significantly for any of the genotypes evaluated (Table 2). As an average over genotypes, the percentage of mature fruit estimates for the four sample sizes differed by only 0.5 percentage points. Standard errors calculated for each genotype and over genotypes, however, were consistently larger for maturity estimates based on 25 fruits than for estimates based on the larger sample sizes (Table 2). Means and standard errors were comparable for maturity estimates based on 50, 75, and 100 fruits, indicating that 50 fruits are adequate for reliable maturity determinations in peanuts. Comparisons among maturity estimates based on IPC, KD, and free arginine content on samples of less than 50 fruits demonstrated that

Table 2. Effect of sample size on percentage of mature fruit estimates of 3 peanut genotypes.

	Fruit Number			
Genotype	25	50	75	100
Starr	82.2 ± 3.5[1/]	81.4 ± 2.3	82.2 ± 1.4	81.6 ± 2.0
Florunner	56.4 ± 4.9	56.6 ± 2.3	56.8 ± 2.6	56.8 ± 3.4
PI 248759	74.5 ± 5.2	74.8 ± 2.5	75.1 ± 2.7	74.5 ± 3.0
Mean	71.0 ± 4.6 a[2/]	70.9 ± 2.3 a	71.4 ± 2.3 a	71.0 ± 2.8 a

[1/] Mean and standard error of 56 samples.

[2/] Means followed by the same letter are not significantly different at the 0.05 probability level (Duncan's New Multiple Range).

estimates based on IPC were as reliable as those made using either of the other two methods. Variations in maturity estimates on sample sizes of

less than 50 fruits were due primarily to sampling error, with little variation attributable to method of determination.

Delaying the time after harvest at which maturity determinations were made had no effect on the maturity estimates (Table 3). Mature fruit percentages were similar at five evaluation dates after harvest for each genotype. These results indicate that IPC intensity does not change sufficiently after harvest to affect maturity evaluations.

Differences in IPC intensity were detected among genotypes. Mature fruits of both Florunner and the white-seeded genotype PI 268750 appeared to have slightly less intense darkening than did mature fruits of the other genotypes evaluated. Results of Schenk (14) also indicate that peanut genotypes may differ in IPC intensity, and it has been suggested (2) that these differences may complicate maturity determinations based on IPC. The variation in IPC intensity observed among the genotypes we studied, however, was very slight and was not sufficient to reduce the reliability of mature determinations.

Table 3. Percentage of mature fruits of 3 genotypes at 5 dates after harvest.

	Days after harvest				
Genotype	0	30	60	90	120
Starr	87.8[1/]	88.0	87.0	89.3	86.3
Florunner	67.8	65.3	63.0	63.5	62.8
PI 268750	64.0	63.8	66.5	64.5	62.8
Mean	71.5 a[2/]	72.3 a	72.2 a	72.4 a	70.6 a

1/ Estimates based on 400 fruit.

2/ Means followed by the same letter are not significantly different at the 0.05 probability level (Duncan's New Multiple Range).

Density and free arginine content data from peanut kernels separated on the basis of IPC are included in Table 4. Kernels of all genotypes that were classified as mature on the basis of IPC were significantly lower in density and had significantly lower AMI values than kernels classified as mature. When averaged over genotypes, densities of 1.083 and 1.143 were obtained for the mature and immature kernel classes, respectively, which compares favorably with the results of others (1, 11).

AMI values, determined by multiplying the optical density reading of the filtrate containing free arginine at 520 nm by 100 (8), averaged 16.7 for the mature kernels and 66.8 for the immature kernels (Table 4). Young and coworkers (17, 18) stated that peanut samples having AMI values below 30 are mature, whereas samples having values higher than 35 are immature. AMI values obtained in our study were not above 21.2 for mature kernels or below 51.2 for immature kernels, indicating that the maturity classifications based on IPC were reliable for all genotypes.

Correlations among the three methods of determining maturity were highly significant. The coefficient between the IPC and AMI methods was 0.96, between the IPC and KD methods was 0.98, and between the AMI and KD methods was 0.95. There was no instance of maturity misclassification in the samples analyzed.

Table 4. Density and AMI values of peanut kernels of 10 genotypes separated for maturity on the basis of internal pericarp color.

	Density (g/cc)		AMI	
Genotype	Mature	Immature	Mature	Immature
Starr	1.085	1.139	15.0	63.2
New Mex. Val. A.	1.090	1.148	20.3	66.0
Florunner	1.078	1.136	18.8	74.0
PI 139919	1.085	1.155	15.1	81.0
PI 149635	1.090	1.140	15.6	51.2
PI 248759	1.094	1.144	18.3	61.5
PI 259611	1.085	1.147	12.9	59.6
PI 268750	1.077	1.147	21.2	82.8
PI 288921	1.081	1.145	13.8	62.0
PI 341885	1.070	1.136	16.4	66.5
Mean	1.083 a[1/]	1.143 b	16.7 A	66.8 B

1/ Means followed by the same letter are not significantly different at the 0.01 probability level (Duncan's New Multiple Range).

The reliability of maturity estimates depends not only on the method of determination but also on the size of the fruit sample analyzed. Our results indicate that reliable maturity estimates can be obtained from 50 peanut fruits regardless of method used. Maturity determinations made subjectively on the basis of IPC were as reliable as other methods for samples that contain less than 50 fruits. Maturity evaluations based on IPC are simple, can be made in the field on individual plants, and require no sophisticated equipment. The method has the added features of being rapid and nonsacrificial which makes it a particularly useful tool in breeding programs.

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