Breeding for improved fatty acid composition in soybean oil

by

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INTRODUCTION

Selection for altered fatty acid composition in many oil crops seems possible considering the available genetic variability (Downy and McGregor (1975). The emphasis in soybeans [Glycine max (L.) Merr.] has been for reducing linolenic acid which causes poor oil quality (Dutton et al., 1951; Evans et al., 1965; Ho et al., 1978; Kalbrener et al., 1974).

Information concerning genetic control of polyunsaturated fatty acid synthesis in soybeans at present indicates that it is under maternal control (Brim et al., 1968; Singh and Hadley, 1968) and it is quantitatively inherited (White et al., 1961). Howell et al. (1972) suggested that the sequential conversion of oleic to linoleic to linolenic acid was under the control of three genes that were additive in their effect.

Wilson et al. (1981) reported that selection for a high ratio of oleic acid to the sum of linoleic and linolenic acids resulted in decreased amounts of linoleic and linolenic acids. Caldwell et al. (1982) indicated that selection for high oleic acid also decreased levels of palmitic acid, but had no effect on stearic acid. One objective of this study was to estimate the relationships among palmitic, stearic, oleic, linoleic and linolenic acids which might give insight into changes due to selection.

Selection for altered fatty acid composition has been confounded by significant genotype x environment interactions, making it difficult to assign exact values for varieties. Caldwell et al. (1982) and Cramer et al. (1981) suggested that the difference in environmental effects is in

magnitude and not in relative ranking. A second objective of this study was to evaluate the possibility of selecting for altered fatty acid composition in Puerto Rico using varieties adapted to Iowa.

A third objective of this study was to evaluate different combinations of resources to be used in a selection program. Eberhart (1970), Eberhart (1972), Fehr (1976), and Fehr (1978) illustrate that the amount of genetic gain per year attained is related to the number of years per cycle of selection. Along with time, the gain per increment of each resource is an important consideration.

LITERATURE REVIEW

Soybean [Glycine max (L.) Merr.] oil constitutes a major portion of the world's edible fats and oils (Munyer, 1979). Improving oil stability and flavor is an important concern. Hydrogenation and winterization are presently used to make soybean oil acceptable for cooking uses. This adds to the costs of the oil and may cause nutritionally undesirable changes (Kummerow, 1975). Eliminating reversion flavors in soybean oil could possible increase its marketability.

Linolenic acid has been identified as the unstable component in soy oil (Dutton et al., 1951; Kalbrener et al., 1974; Okkerse et al., 1967). Evans et al. (1965) suggested that reducing linolenic acid below 5% would achieve an improvement in oil quality. Cowan et al. (1970) indicated that a level of 3% linolenic acid might be acceptable, but 1% would be more desirable.

Research has indicated that linolenic acid synthesis in soybeans and other higher plants occurs by desaturation of oleic to linoleic and then to linolenic acid (Cherif et al., 1975; Simmons and Quakenbush, 1954; Wilson et al., 1981). Selection for low linolenic acid may result in some alteration in the control of this process. Downy and McGregor (1975) suggested that genetic variability existed in soybeans that would permit selection for altered fatty acid composition.

Linolenic acid synthesis is controlled by the maternal parent (Brim et al., 1968; Fehr et al., 1971; Singh and Hadley, 1968). This precludes selection on a single seed basis using F_2 seeds on F_1 plants.

The inheritance of linoleic acid and linolenic acid is believed to be quantitative (White et al., 1961). Howell et al. (1972) suggested that more than three genes acting in an additive manner controlled this trait. De la Roche et al. (1971) indicated that the inheritance of oleic and linoleic acids in maize is controlled by one or two genes and some maternal influence. Poneleit and Alexander (1965) suggested that desaturation of oleic acid in maize was under simple gene control. Oleic acid in safflower seems to be under the control of a single gene with little maternal influence (Knowles, 1968; Yermanos et al., 1967). Linoleic acid in rapeseed seems to be inherited as a quantitative trait and is possibly influenced by the maternal parent (Krzymanski and Downey, 1969).

Heritability estimates for fatty synthesis in soybeans were not available when this study was initiated. Broad-sense heritability estimates calculated in rapeseed for oleic acid ranged from 0.53 to 0.78, for linoleic acid from 0.40 to 0.81, seed for linolenic acid from 0.26 to 0.59 (Kondra and Thomas, 1975).

Selection for low linolenic acid could affect concentrations of the other fatty acids that are precursors of linolenic acid. White et al., (1961) reported positive correlation coefficients of 0.75 to 0.96 between linoleic and linolenic acids in field and greenhouse experiments. Collins and Howell (1957) also found positive correlations between linoleic and linolenic acids. Hammond et al. (1972) and Hammond and Fehr (1975) indicated similar trends between linoleic and linolenic acids, Sekhon et al. (1975) reported a negative correlation between the two fatty acids. Selection for low linolenic acid would probably result in reduction of linoleic acid.

Oleic acid has been shown to be negatively correlated with linoleic and linolenic acids (Hammond et al., 1972; Hammond and Fehr, 1975; Howell et al., 1972; Singh, 1967; Sekhon et al., 1975). This relationship has been used successfully to select for low linolenic acid lines by researchers at North Carolina State University.

Sekhon et al. (1975) reported a significant negative correlation ($r = -0.48$) between stearic and linoleic acid and a significant positive correlation ($r = 0.36$) between stearic and linolenic acid. They indicated nonsignificant correlations between stearic acid and palmitic ($r = -0.11$) and oleic acid ($r = 0.29$). Hammond and Fehr (1975) reported significant negative correlations between stearic acid and palmitic ($r = -0.31$), linoleic ($r = -0.57$) and linolenic ($r = -0.80$) and a significant positive correlation ($r = 0.53$) with oleic acid. The data available would indicate no specific relationship between stearic and linolenic acids.

Hammond and Fehr (1975) indicated nonsignificant negative correlations between palmitic and stearic, oleic, linoleic, and linolenic acids. Sekhon et al. (1975) reported similar correlations with those between palmitic and oleic acids being significant. Selection for stearic, oleic, linoleic or linolenic acid would probably result in no major change in palmitic acid.

Caldwell et al. (1982) reported that after four cycles of selection for high oleic acid, there was a decrease in palmitic, linoleic, and linolenic and little change in stearic acid content. Wilson et al. (1981) reported similar reductions in palmitic, linoleic, and linolenic

acid and an increase in stearic acid. These data agree to a large extent with the correlations referenced earlier.

A significant genotype x environment interaction has been a major problem in assigning discrete values for fatty acid content to individual varieties. A major factor identified by Collins and Howell (1957), Howell and Collins (1957), and Wolf et al. (1982) was the inverse relationship of temperature with the presence of linoleic and linolenic acid in mature soybean seeds. Wolf et al. (1982) reported up to a 40% reduction in linolenic acid content in a hot environment. Appelqvist (1968a,b) reported similar temperature related effects in some Cruciferae cultivars.

Chu and Sheldon (1979), Hammond and Fehr (1975), Howell and Collins (1957), and Kurnik and Jaky (1975) suggested that environment affected the production of unsaturated fatty acids in soybeans. There are no data at present concerning genotype x environment interactions affecting fatty acid synthesis.

Hammond and Fehr (1975) indicated that seed source had little effect on the next generation. Caldwell et al. (1982) and Cramer et al. (1981) indicated that despite variation in fatty acid composition between environments, the relative rankings of lines was essentially the same.

Howell and Collins (1957) indicated that 12 hours of daylight decreased the linolenic acid content, but 16 and 20 hour days made no difference. Chu and Sheldon (1979) and Hammond and Fehr (1975) reported that planting date also affected fatty acid composition in soybeans.

Variability among pods on the same plant and within pods (Collins and Howell, 1957; Cramer et al., 1981) indicates that to adequately test a

plant, a representative sample from the whole plant must be taken. Hammond and Fehr (1975) found that a 10-seed sample was not large enough to eliminate seed-to-seed variation.

A key point to an effective breeding program is to keep the number of years required to complete a cycle as low as possible (Eberhart, 1970; Eberhart, 1972; Fehr, 1976; Fehr, 1978). The ability to select in a winter nursery where the crop is not originally adapted may increase the speed of the program.

MATERIALS AND METHODS

Nineteen cultivars and experimental lines from Maturity Group II of the 1979 Uniform Soybean Tests Northern States and a germplasm line from Iowa State University (Fehr and Bahrenfus, 1980) were evaluated for fatty acid composition in seven environments (Table 1). These twenty lines were grown at the Agronomy and Agricultural Engineering Research Center, Ames, Iowa, and at the Isabela Substation of the University of Puerto Rico in Puerto Rico. The environments were Ames 1979, Ames 1980, Ames 1981, Puerto Rico November, 1980 lighted and nonlighted and Puerto Rico February, 1981 lighted and nonlighted.

The Ames environments were planted on May 9, 1979, May 27, 1980, and May 7, 1981. The Puerto Rico environments were planted on November 1, 1980 and February 15, 1981.

Plots at Ames in 1979 were four rows 6.1 m long with 68 cm between rows. In 1980 and 1981 in Iowa, plots were single rows 1.5 m long with spacing between plots of 68 and 102 cm. At Isabela, Puerto Rico, plots were single rows 0.75 m long with 0.61 m between rows.

Continuous lighting was provided at two of the Puerto Rico environments for 15 days after the time of planting, after which supplemental lighting was reduced to 14.5 hours for about 35 days, and natural daylength thereafter. Plants grown under natural daylength matured in 90 days and those under artificial light matured in 105 days after planting.

The plots at each location were planted in a randomized complete block design. Two replications per location were planted at the rate of 13 seeds per meter, except for Ames 1979 which was planted at the rate

Table 1. Lines evaluated at Ames, Iowa and Isabela, Puerto Rico for fatty acid composition

Line	Originator
Amcor	Illinois Agricultural Experiment Station and USDA-ARS
A2[a]	Iowa Agricultural and Home Economics Experiment Station
A77-211021[b]	Iowa Agricultural and Home Economics Experiment Station
A77-212006[b]	Iowa Agricultural and Home Economics Experiment Station
Beeson	Indiana Agricultural Experiment Station
Beeson 80	Indiana Agricultural Experiment Station
Century	Indiana Agricultural Experiment Station
Corsoy	Iowa Agricultural and Home Economics Experiment Station
Gnome	Ohio Agricultural Experiment Station
Harcor	Agriculture Canada Research Station
H7703[b]	Onio Agricultural Experiment Station
H75-5605[b]	Ohio Agricultural Experiment Station
L73-4673[b]	Illinois Agricultural Experiment Station and USDA-ARS
L75-3674[b]	Illinois Agricultural Experiment Station and USDA-ARS
Nebsoy	Nebraska Agricultural Experiment Station
Pella	Iowa Agricultural and Home Economics Experiment Station
U11239[b]	Nebraska Agricultural Experiment Station
U20235[b]	Nebraska Agricultural Experiment Station
Weber	Iowa Agricultural and Home Economics Experiment Station
Wells II	Indiana Agricultural Experiment Station

[a]Germplasm line.

[b]Experimental line.

of 28 seeds per meter. The plots were not thinned.

Five plants per plot were randomly harvested at maturity at Ames in 1979 and two were selected that had at least 100 seeds per plant. Seed from one of these plants was used to plant the Ames 1980 environment. Two plants with at least 140 seeds were selected for analysis and one of these was used as a seed source for the remaining five environments. Seed from two plants were required for Beeson, Wells II, A77-211021, and Century to obtain sufficient amounts for the five environments. In the remaining environments, five plants per plot were randomly harvested at maturity and two of these plants were randomly selected for analysis.

From each of the two plants selected per plot, two 20-seed samples were taken. After extraction of the oil, two consecutive injections per seed sample were made into the gas chromatograph. Hammond (Department of Food Technology, Iowa State University, Ames, Ia, 1980) reported that the error associated with injections was negligible, thus, the injections were made consecutively to save time and expense.

All of the seed samples were stored at room temperature and the analyses were performed after all of the samples had been accumulated from the seven environments.

Oil extraction was begun by drying the sample in a vacuum oven at 95°C and -1.5 atm. for 15 hours. The samples were crushed with 1055 kg/sq cm in a 30 ml container. Distilled hexane, 3 ml, was added to the crushed samples and allowed to stand for 15 hr.

The extracted oil was converted to methyl esters by putting 0.2 ml of the hexane-oil solution in a 2 ml vial. Next 0.5 ml of 1 N sodium method was added and allowed to react for 2 hr. Then 0.6 ml of distilled

water was added and the esters allowed to separate from the aqueous alcohol phase for 1 hr. A few drops of distilled hexane were added and the top layer, containing approximately 10 µl of ester, was removed and put in a 2 ml vial. The vial was filled with about 1 ml of distilled hexane. About 2 µl of this solution were injected into the gas chromatograph (Beckman GC-5 fitted with hydrogen flame detectors). The column was 6 m x 3.2 mm O.D., packed with EGSSX on Chromsorb w 100/120 mesh and maintained at 185°C. The nitrogen flow was 40 ml/min, hydrogen flow was 50 ml/min and air flow was 400 ml/min. Standard ester mixtures by Nucheck were run on a regular basis for calibration. Peak areas and percentages of palmitic, stearic, oleic, linoleic, and linolenic acid were calculated by a Commodore computer by PET. The fatty acid composition was converted to a percentage of the total fatty acids.

The statistical analysis for the five fatty acids was computed as a randomized complete block design to compare lines. All effects were considered random. The statistical model assumed was:

$$Y_{ijk\ell mn} = \mu + \alpha_i + \beta_{ij} + \gamma_k + \alpha\gamma_{ik} + \varepsilon_{ijk} + \lambda_{ijk\ell} + \phi_{ijk\ell m} + \psi_{ijk\ell mn}$$

where

$Y_{ijk\ell mn}$ = fatty acid percentage for nth injection within the mth seed sample within the ℓth plant of the kth line in the jth replication in the ith environment

μ = population mean

α_i = effect of the ith environment; i = 1 to 7

β_{ij} = effect of the jth replication within the ith environment; j = 1 to 2

γ_k = effect of the kth line; k = 1 to 20

$\alpha\gamma_{ik}$ = interaction of the ith environment with the kth line

ε_{ijk} = whole plot error

$\lambda_{ijk\ell}$ = effect of the ℓth plant within the kth line in the jth replication in the ith environment; ℓ = 1 to 2

$\phi_{ijk\ell m}$ = effect of the mth seed sample within the ℓth plant in the kth line in the jth replication in the ith environment; m = 1 to 2

$\psi_{ijk\ell mn}$ = effect of the nth injection within the mth seed sample in the ℓth plant in the kth line in the jth replication in the ith environment; i = 1 to 2

The analyses of variance and expected mean squares combined over environments in Table 2 were used to obtain variance component estimates. Table 3 shows the analysis of variance of individual environments.

Narrow sense heritabilities were calculated from variance component estimates on a seed sample, plant, plot, and entry mean basis (Hanson et al., 1956).

$$\text{Sample } h^2 = \frac{\sigma^2_G}{\sigma^2_{I/i} + \sigma^2_S + \sigma^2_P + \sigma^2 + \sigma^2_{GE} + \sigma^2_G}$$

$$\text{Plants within plot } h^2 = \frac{\sigma^2_G}{\sigma^2_{I/is} + \sigma^2_{S/s} + \sigma^2_P + \sigma^2_{GE} + \sigma^2_G}$$

$$\text{Plants among plots } h^2 = \frac{\sigma^2_G}{\sigma^2_{I/is} + \sigma^2_{S/s} + \sigma^2_P + \sigma^2 + \sigma^2_{GE} + \sigma^2_G}$$

Table 2. Analysis of variance and expected mean squares for obtaining estimates of variance components for each fatty acid

Source of variation	df		
Environments (E)	E-1	=	6
Replications/E (R/E)	(r-1)E	=	7
Lines (L)	(ℓ-1)	=	19
L x E	(ℓ-1)(E-1)	=	114
L x R/E	(r-1)(ℓ-1)E	=	133
Plants (P)/L x R x E	(p-1)Erℓ	=	280
Samples (S)/P x L x R x E	(s-1)Erℓp	=	560
Injections/S x P x L x R x E	(i-1)Erℓps	=	1120

E = number of environments; E = 7

r = number of replications at an environment; r = 2

ℓ = number of lines; ℓ = 20

p = number of plants per plot; p = 2

s = number of seed samples per plant; s = 2

i = number of injections per seed sample; i = 2

Expected mean squares	Mean square
$\sigma_I^2 + i\sigma_S^2 + is\sigma_P^2 + isp\sigma^2 + ispr\sigma_{LE}^2 + ispr\ell\sigma_E^2$	MS1
$\sigma_I^2 + i\sigma_S^2 + is\sigma_P^2 + isp\sigma^2 + isp\ell\sigma_R^2$	MS2
$\sigma_I^2 + i\sigma_S^2 + is\sigma_P^2 + isp\sigma^2 + ispr\sigma_{LE}^2 + ispre\sigma_L^2$	MS3
$\sigma_I^2 + i\sigma_S^2 + is\sigma_P^2 + isp\sigma^2 + ispr\sigma_{LE}^2$	MS4
$\sigma_I^2 + i\sigma_S^2 + is\sigma_P^2 + isp\sigma^2$	MS5
$\sigma_I^2 + i\sigma_S^2 + is\sigma_P^2$	MS6
$\sigma_I^2 + i\sigma_S^2$	MS7
σ_I^2	MS8

$$\sigma_G^2 = \frac{MS3-MS4}{isprE}$$

$$\sigma_{GE}^2 = \frac{MS4-MS5}{ispr}$$

$$\sigma^2 = \frac{MS5-MS6}{isp}$$

$$\sigma_W^2 = MS6$$

$$\sigma_P^2 = \frac{MS6-MS7}{is}$$

$$\sigma_S^2 = \frac{MS7-MS8}{i}$$

$$\sigma_I^2 = MS8$$

$$\sigma_{PH}^2 = \frac{MS3}{isprE}$$

Table 3. Analysis of variance and expected mean squares for each environment

Source of variation	df			Expected mean squares
Replication (R)	$(r-1)$	=	1	$\sigma_I^2 + i\sigma_S^2 + is\sigma_P^2 + isp\sigma^2 + isp\ell\sigma_R^2$
Lines (L)	$(\ell-1)$	=	19	$\sigma_I^2 + i\sigma_S^2 + is\sigma_P^2 + isp\sigma^2 + ispr\sigma_L^2$
R x L	$(\ell-1)(r-1)$	=	19	$\sigma_I^2 + i\sigma_S^2 + is\sigma_P^2 + isp\sigma^2$
Plant (P)/L x R	$(p-1)r\ell$	=	40	$\sigma_I^2 + i\sigma_S^2 + is\sigma_P^2$
Samples (S)/P x L x R	$(s-1)r\ell p$	=	80	$\sigma_I^2 + i\sigma_S^2$
Injections/S x P x L x R	$(i-1)r\ell ps$	=	160	σ_I^2

r = number of replications; r = 2

ℓ = number of lines; ℓ = 20

p = number of plants per plot; p = 2

s = number of seed samples per plant; s = 2

i = number of injections per seed sample; i = 2

$$\text{Plot } h^2 = \frac{\sigma^2_G}{\sigma^2_{I/isp} + \sigma^2_{S/sp} + \sigma^2_{P/p} + \sigma^2 + \sigma^2_{GE} + \sigma^2_G}$$

$$\text{Entry } h^2 = \frac{\sigma^2_G}{\sigma^2_{I/isprE} + \sigma^2_{S/sprE} + \sigma^2_{P/prE} + \sigma^2/rE + \sigma^2_{GE/E} + \sigma^2_G}$$

where

E = number of environments; E = 7

r = number of replications at an environment; r = 2

p = number of plants; p = 2

s = number of seed samples; s = 2

i = number of injection; i = 2

σ^2_G = genetic variance among lines

σ^2_{GE} = genotype x environment interaction variance

σ^2 = environmental variance among plots

σ^2_P = variance among plants

σ^2_S = variance among seed samples

σ^2_I = variance among injection

Phenotypic correlation coefficients were calculated for all possible combinations of fatty acid percentages with the PROC CORR procedure in SAS (Barr et al., 1979). Rank correlations were calculated according to Snedecor and Cochran (1980).

Genotypic correlations for all possible combinations of fatty acid percentages were calculated on an entry mean basis using analyses of variance and covariance (Service, 1972) and a formula by Wallace et al. (1954).

$$r_g = \frac{C_{g_{ij}} - C_{ge_{ij}}}{\sqrt{M_{gi} - M_{gei}}\ \sqrt{M_{gj} - M_{gej}}} = \frac{\sigma_{gij}}{\sqrt{\sigma^2_{gi}\ \sigma^2_{gj}}}$$

where

C_{gij} = line mean product for two fatty acids

C_{geij} = line x environment mean product for two fatty acids

M_{gi} = line mean square for the first fatty acid

M_{gei} = line x environment mean square for the first fatty acid

M_{gj} = line mean square for the second fatty acid

M_{gej} = line x environment mean square for the second fatty acid

σ_{gij} = covariance between two fatty acids

σ^2_{gi} = genetic variance for the first fatty acid

σ^2_{gj} = genetic variance for the second fatty acid

Predicted gain per cycle and per year were computed for different resource allocations using an equation by Eberhart (1972):

$$\Delta G_y = \frac{k\hat{\sigma}^2_A}{y\sigma_{ph}}$$

where ΔG_y = genetic gain per year, k = selection differential in standard units, $\hat{\sigma}^2_A$ = additive genetic variance estimate, y = number of years per cycle, and σ_{ph} = square root of the phenotypic variance estimate.

The phenotypic variance estimates used in predicting genetic gain were calculated as:

$$\sigma^2_{ph} = \frac{\sigma^2_I}{isprE} + \frac{\sigma^2_S}{sprE} + \frac{\sigma^2_P}{prE} + \frac{\sigma^2}{rE} + \frac{\sigma^2_{GE}}{E} + \sigma^2_G$$

Relative efficiency was computed for each resource allocation arrangement by dividing the estimated genetic gain per cycle by the genetic gain calculated for the resources used in this study then multiplied by 100.

RESULTS

There was significant variation for fatty acid composition among lines across environments and within each environment (Tables 4 to 6). The main effect of environment and the line x environment interaction were significant for each fatty acid (Table 4). The variation associated with plants within lines was significant at the 1% probability level for oleic, linoleic, and linolenic acids and at the 5% level for palmitic acid. The effect of plants within lines was significant in each environment for oleic and linoleic acids.

Variance component estimates for each fatty acid (Table 5) were used to calculate narrow sense heritabilities (Table 8). Heritabilities calculated on an entry mean basis were similar for each fatty acid, with h^2 values ranging from 0.92 to 0.96. Heritability estimates on plot, plant, and seed sample bases for each fatty acid were similar and comparisons among fatty acids on each level were similar.

Palmitic acid had a significant negative correlation with linoleic acid, except on an entry mean basis ($r = 0.16$ to 0.21) and with linolenic acid, except on a plot and entry mean basis (Tables 8 to 12). Correlation coefficients between between palmitic acid and stearic and oleic acids were near zero and significant only on an injection and sample basis.

Stearic acid had a significant ($P < 0.01$) positive correlation with linoleic and linolenic acids, except on an entry mean basis and a significant negative correlation with oleic acid (Tables 9 to 13).

Table 4. Analyses of variance combined over environments for each fatty acid

Source of variation	df	Mean squares for fatty acids				
		Palmitic (16:0)	Stearic (18:0)	Oleic (18:1)	Linoleic (18:2)	Linolenic (18:3)
Environments (E)	6	46.57**	5.31**	999.75**	709.06**	124.40**
Replications/E (R/E)	7	0.41	0.55*	45.42*	35.39**	1.85
Lines (L)	19	22.38**	18.88**	1057.18**	603.19**	61.11**
L x E	114	1.02**	0.71**	83.17**	51.68**	2.66**
L x R/E	133	0.53	0.24	18.33	12.03	0.90
Plants (P)/L x R x E	280	0.56*	0.17	13.49**	8.17**	0.82**
Samples (S)/P x L x R x E	560	0.46**	0.15**	2.34**	2.10**	0.55**
Injections/S x P x L x R x E	1120	0.02	0.01	0.01	0.04	0.02
CV %[a]		9.4	23.5	38.1	13.4	20.6
$\bar{X}$		10.8	3.6	23.9	53.8	7.9

*,**Significant at the 0.05 and 0.01 probability levels.

[a]Line x environment mean squares were used to compute coefficients of variation.

Table 5. Entry means over seven environments for palmitic, stearic, oleic, linoleic, and linolenic acids

Line[a]	Fatty acid				
	Palmitic	Stearic	Oleic	Linoleic	Linolenic
Pella	10.5fghi[b]	4.1abc	25.0bcde	53.6def	6.8h
Harcor	10.8def	3.1jk	27.9a	51.2ghi	7.1gh
L73-4673	10.7defg	3.2ij	25.3abcde	53.7def	7.1gh
Corsoy	10.7defg	3.1ijk	27.3ab	51.7fghi	7.2gh
L75-3674	10.8def	3.1ijk	26.4abc	52.5efgh	7.2gh
A2	10.6efgh	2.9k	26.0abcd	53.1efg	7.4fg
Beeson 80	10.6efgh	3.3hi	25.8abcde	53.0efg	7.4fg
A77-212006	11.6b	3.5gh	27.5ab	50.0i	7.5fg
Nebsoy	10.9de	4.1abc	25.6abcde	51.9fghi	7.5fg
Gnome	10.6efgh	3.8cdef	24.1cde	53.8def	7.7ef
H75-5605	11.9a	3.9cdef	23.3def	52.9efg	8.0de
U11239	10.4ghij	4.2a	24.1cde	53.1efg	8.1de
U20235	11.0d	3.2hij	23.1ef	54.6cde	8.1de
Amcor	10.3hij	3.2hij	27.9a	50.4hi	8.2de
Century	10.7defg	3.6fg	21.3fg	56.1bc	8.3cd
Wells II	10.3ij	3.8cdef	18.6g	58.6a	8.7bc
A77-211021	10.5fghi	3.9bcde	21.2fg	55.6bcd	8.8bc
H7703	11.2c	4.0abcd	19.3g	56.5bc	9.0ab
Beeson	10.2j	3.8cdef	19.7g	57.2a	9.0ab
Weber	11.5b	3.8fg	19.3g	56.2bc	9.3a
$\bar{X}$	10.8	3.6	23.9	53.8	7.9
$S_{\bar{x}}$[c]	0.10	0.09	0.69	0.52	0.16

[a]In order from lowest to highest for linolenic acid.

[b]Means in the same column with the same letter are not significantly different based on Duncan's multiple range test ($P > 0.05$).

[c]Standard error of the mean.

Table 6. Analyses of variance for palmitic, stearic, oleic, linoleic, and linolenic acid in environment

Source of variation	df	Environments						
		Ames 1979[a]	Ames 1980	Ames 1981	PR 1980 Nov L	PR 1980 Nov NL	PR 1981 Feb L	PR 1981 Feb NL
Palmitic acid								
Replications (R)	1	0.11	0.36	0.46	0.14	0.83	0.79	0.21
Lines (L)	19	2.87**	4.56**	3.23**	3.57**	3.12**	6.94**	4.24**
R x L	19	0.37	0.90	0.18	0.29	0.60	0.96	0.43*
Plants (P)/L x R	40	0.37*	0.86	0.13**	0.27	1.20	0.95	0.21
Samples (S)/P x L x R	80	0.17**	0.56**	0.07**	0.27**	0.87**	0.96**	0.30**
Injections/S x P x L x R	160	0.01	0.03	0.01	0.01	0.002	0.04	0.01
CV %[b]		5.9	8.8	4.0	4.9	7.5	8.6	6.0
$\bar{X}$		10.4	10.8	10.6	11.0	10.4	11.4	10.9
Stearic acid								
Replications (R)	1	0.80	0.46	0.14	0.91	0.16	0.01	1.32**
Lines (L)	19	2.85**	1.91**	1.87**	3.40**	4.90**	2.74**	5.45**
R x L	19	0.53*	0.40**	0.07	0.14	0.24	0.22	0.09
Plants (P)/L x R	40	0.28	0.10	0.15**	0.17	0.16	0.19	0.13**
Samples (S)/P x L x R	80	0.31**	0.12**	0.07**	0.15**	0.13**	0.24**	0.06**
Injections/S x P x L x R	160	0.01	0.01	0.001	0.01	0.002	0.02	0.01
CV %		20.1	16.6	7.3	10.7	13.7	13.7	8.7
$\bar{X}$		3.6	3.8	3.7	3.5	3.6	3.4	3.5

Oleic acid

Replication (R)	1	1.35	41.57	49.67**	6.58	1.31	205.12	15.81
Lines	19	70.13**	97.21**	79.27**	218.18**	328.28**	330.10**	433.03**
R x L	19	11.74	17.26**	4.87	16.37	19.80	49.74**	8.53
Plants (P)/L x R	40	19.10**	5.77**	2.83**	13.90**	27.50**	12.23**	13.11**
Samples (S)/P x L x R	80	4.33**	1.92**	0.82**	1.44**	2.06**	2.78**	2.99**
Injections/S x P x L x R	160	0.02	0.02	0.01	0.01	0.01	0.01	0.01
CV %		15.4	17.8	9.8	18.0	18.4	26.3	11.3
$\bar{X}$		22.3	23.4	22.5	22.5	24.2	26.8	25.8

Linoleic acid

Replication (R)	1	1.06	18.16	56.10**	19.73	0.00	137.51**	15.18
Lines (L)	19	41.69**	53.73**	45.52**	123.56**	196.00**	204.95**	247.52**
R x L	19	11.97	8.23*	2.93	11.64	14.80	27.78**	6.85
Plants (P)/L x R	40	8.26**	3.00*	2.19**	9.42**	17.92**	8.07**	8.32**
Samples (S)/P x L x R	80	3.65**	1.82**	0.51**	0.51**	2.03**	2.88**	2.37**
Injections/S x P x L x R	160	0.06	0.04	0.01	0.03	0.01	0.05	0.04
CV %		6.3	5.3	3.1	6.2	7.1	10.3	5.0
$\bar{X}$		55.0	54.2	54.5	54.8	54.5	51.0	52.4

*,**Significant at the 0.05 and 0.01 probability levels.

[a]Nov = November, Feb = February, L = Lighted, NL = Nonlighted, PR = Puerto Rico.

[b]Line x replication mean squares were used to compute coefficients of variation.

Table 6. (Continued)

Source of variation	df	Environments						
		Ames 1979[a]	Ames 1980	Ames 1981	PR 1980 Nov L	PR 1980 Nov NL	PR 1981 Feb L	PR 1981 Feb NL
Linolenic acid								
Replication (R)	1	1.89	5.16	0.54	0.27	0.04	2.67	2.38*
Line (L)	19	5.45**	6.19**	7.59**	13.38**	12.11**	12.51**	19.83**
R x L	19	1.33	1.32	0.40*	0.98	0.87	0.94	0.48**
Plants (P)/L x R	40	3.19	0.48**	0.20	0.56	0.65**	0.39	0.29
Samples (S)/P x L x R	80	2.05**	0.32**	0.08**	0.64**	0.21**	0.20**	0.37**
Injections/S x P x L x R	160	0.04	0.04	0.004	0.01	0.01	0.02	0.006
$\frac{CV}{X}$ %[b]		13.2 8.7	14.9 7.7	7.3 8.7	12.1 8.2	12.7 7.4	13.3 7.3	9.3 7.4

Table 7. Variance components for each fatty acid over seven environments

Variance component		Fatty acid				
		Palmitic	Stearic	Oleic	Linoleic	Linolenic
Genotype	σ^2_G	0.19	0.16	8.70	4.92	0.52
Genotype x Environment	σ^2_{GE}	0.03	0.03	4.05	2.48	0.11
Plot-to-plot	σ^2	-0.004	0.02	0.61	0.48	0.01
Plant	σ^2_P	0.03	0.01	2.79	1.52	0.07
Seed sample	σ^2_S	0.22	0.07	1.17	1.03	0.27
Injection	σ^2_I	0.02	0.01	0.01	0.04	0.02
Phenotypic	σ^2_{PH}	0.20	0.17	9.44	5.39	0.55
Within plot	σ^2_W	0.56	0.17	13.49	8.17	0.82

Table 8. Narrow sense heritabilities on a sample, plant, plot, and entry mean basis for each fatty acid

Basis of h^2	Palmitic	Stearic	Oleic	Linoleic	Linolenic
Entry mean	0.95	0.96	0.92	0.92	0.96
Plot	0.65	0.68	0.58	0.55	0.70
Single plant among plots	0.52	0.62	0.52	0.50	0.61
Single plant within plots	0.52	0.67	0.54	0.52	0.62
Sample	0.40	0.54	0.50	0.47	0.53

Table 9. Phenotypic correlation coefficients among fatty acids on an injection basis

	Fatty acid			
Fatty acid	Stearic	Oleic	Linoleic	Linolenic
Palmitic	0.08**	-0.03	-0.15**	-0.12**
Stearic		-0.44**	0.28**	0.32**
Oleic			-0.95**	-0.74**
Linoleic				0.60**

**Significant at the 0.01 probability level.

Table 10. Phenotypic correlation coefficients among fatty acids on a seed sample basis

	Fatty acid			
Fatty acid	Stearic	Oleic	Linoleic	Linolenic
Palmitic	0.07*	-0.03	-0.15**	-0.12**
Stearic		-0.44**	0.29**	0.32**
Oleic			-0.95**	-0.75**
Linoleic				0.60**

*,**Significant at the 0.05 and 0.01 probability levels.

Table 11. Phenotypic correlation coefficients among fatty acids on a plant basis

	Fatty acid			
Fatty acid	Stearic	Oleic	Linoleic	Linolenic
Palmitic	0.02	-0.02	-0.16**	-0.08*
Stearic		-0.48**	0.35**	0.35**
Oleic			-0.96**	-0.78**
Linoleic				0.65**

*,**Significant at the 0.05 and 0.01 probability levels.

Table 12. Phenotypic correlation coefficients among fatty acids on a plot basis

	Fatty acid			
Fatty acid	Stearic	Oleic	Linoleic	Linolenic
Palmitic	-0.001	-0.04	-0.21**	-0.11
Stearic		-0.50**	0.38**	0.38**
Oleic			-0.96**	-0.80**
Linoleic				0.67**

**Significant at the 0.01 probability level.

Table 13. Phenotypic and genotypic correlation coefficients among fatty acids on an entry mean basis

	Fatty acid							
	Stearic		Oleic		Linoleic		Linolenic	
Fatty acid	P[a]	G	P	G	P	G	P	G
Palmitic	0.05	0.04	-0.001	0.01	-0.21	-0.24	0.05	0.05
Stearic			-0.57**	-0.58	0.43	0.44	0.45*	0.45
Oleic					-0.95**	-0.95	-0.84**	-0.85
Linoleic							0.71**	0.71

*,**Significant at the 0.01 probability level.

[a]Phenotypic (P) and genotypic (G) correlations.

Oleic acid had significant negative correlations with linoleic acid ($r = -0.95$) and linolenic acid ($r = -0.74$ to -0.84). Linoleic acid was significantly correlated with linolenic acid with r values from 0.60 to 0.71 (Tables 9 to 13).

Genotypic correlation coefficients on an entry mean basis were essentially the same as the phenotypic correlation coefficients (Table 13).

A comparison of means across lines for palmitic acid for each environment (Table 14) indicated that the nonlighted Puerto Rico environments on both planting dates produced results similar to Ames. All four Puerto Rico environments produced lower values for stearic acid than the Ames environments.

Both February environments (Table 14) resulted in higher values for oleic acid and lower values for linoleic acid than the other five environments. Three of the Puerto Rico environments had lower mean values for linolenic acid than the Ames environments.

Supplemental lighting in Puerto Rico did not produce significant differences from nonlighted plantings on both dates, except for palmitic acid (Table 14).

Phenotypic correlations among environments (Table 15) for each fatty acid were for the most part highly significant ($r = 0.41$ to 0.96).

Comparison of the rankings of lines in each environment indicated that the four Puerto Rico environments produced similar results for each fatty acid (Tables 16 to 20). Rankings among lines for each fatty acid at the Ames environments were similar (Tables 16 to 20).

Table 14. Mean fatty acid percentages for palmitic, stearic, oleic, linoleic and linolenic acids in each environment

Environment	Fatty acid[a]				
	Palmitic	Stearic	Oleic	Linoleic	Linolenic
Ames 1979	10.4e	3.6bc	22.3c	55.0a	8.7a
Ames 1980	10.8c	3.8a	23.4bc	54.2a	7.7c
Ames 1981	10.6d	3.7b	22.5c	54.5a	8.7a
PR 1980 Nov L[b]	11.0b	3.5cde	22.5c	54.8a	8.2b
PR 1980 Nov NL	10.4e	3.6bcd	24.2b	54.5a	7.4d
PR 1981 Feb L	11.4a	3.4e	26.8a	51.0c	7.3d
PR 1981 Feb NL	10.9bc	3.5de	25.8a	52.4b	7.4d
$\bar{X}$	10.8	3.6	23.9	53.7	7.9
S_x[c]	0.15	0.05	0.67	0.56	0.24

[a]Means in the same column with the same letter are not significantly different based on Duncan's multiple range test.

[b]Nov = November, Feb = February, PR = Puerto Rico, L = Lighted, NL = Nonlighted.

[c]Standard error of the mean.

Table 15. Phenotypic correlation coefficients among environments for each fatty acid

Environments	Ames 1980	Ames 1981	PR 1980 Nov L[a]	PR 1980 Nov NL	PR 1981 Feb L	PR 1981 Feb NL
Palmitic acid						
Ames 1979	0.78**	0.91**	0.71**	0.57**	0.86**	0.77**
Ames 1980		0.90**	0.78**	0.78**	0.77**	0.83**
Ames 1981			0.77**	0.66**	0.80**	0.76**
PR 1980 Nov L				0.81**	0.68**	0.84**
PR 1980 Nov NL					0.58**	0.83**
PR 1981 Feb L						0.74**
Stearic acid						
Ames 1979	0.79**	0.90**	0.75**	0.72**	0.82**	0.77**
Ames 1980		0.79**	0.85**	0.75**	0.83**	0.85**
Ames 1981			0.77**	0.68**	0.79**	0.76**
PR 1980 Nov L				0.93**	0.84**	0.96**
PR 1980 Nov NL					0.76**	0.96**
PR 1981 Feb L						0.81**
Oleic acid						
Ames 1979	0.75**	0.87**	0.67**	0.53*	0.50*	0.52*
Ames 1980		0.82**	0.72**	0.72**	0.66**	0.69**
Ames 1981			0.71**	0.66**	0.53**	0.64**
PR 1980 Nov L				0.77**	0.74**	0.83**
PR 1980 Nov NL					0.65**	0.95**
PR 1981 Feb L						0.69**

Linoleic acid

Ames 1979	0.69**	0.87**	0.73**	0.52*	0.42	0.51*
Ames 1980		0.78**	0.68**	0.72**	0.65**	0.66**
Ames 1981			0.71**	0.62**	0.41	0.57**
PR 1980 Nov L				0.74**	0.76**	0.80**
PR 1980 Nov NL					0.93**	0.93**
PR 1981 Feb L						0.69**

Linolenic acid

Ames 1979	0.72**	0.74**	0.58**	0.56**	0.64**	0.51*
Ames 1980		0.86**	0.85**	0.86**	0.77**	0.83**
Ames 1981			0.80**	0.82**	0.84**	0.77**
PR 1980 Nov L				0.89**	0.88**	0.90**
PR 1980 Nov NL					0.83**	0.95**
PR 1981 Feb L						0.85**

*,**Significant at the 0.05 and 0.01 probability levels.

[a]PR = Puerto Rico, Nov = November, Feb = February, L = Lighted, NL = Nonlighted.

Table 16. Ranking of lines in each environment for palmitic acid

Line	Environments							
	Ames 1979	Ames 1980	Ames 1981	PR 1980 Nov L[a]	PR 1980 Nov NL	PR 1981 Feb L	PR 1981 Feb NL	C[b]
Beeson	1	8	1	2	7	2	2	1
Wells II	2	5	5	3	8	1	1	2
Amcor	7	2	4	1	5	7	3	3
U11239	4	4	3	5	3	5	5	4
A77-211021	3	6	6	12	2	6	4	5
Pella	6	10	11	4	11	3	7	6
Beeson 80	5	7	2	10	10	15	14	7
A2	14	1	7	8	12	9	12	8
Gnome	8	16	12	7	9	4	11	9
Century	11	9	9	9	13	10	9	10
L73-4673	10	3	8	13	14	12	13	11
Corsoy	13	11	16	11	6	8	10	12
L75-3674	16	14	14	6	1	16	6	13
Harcor	15	12	15	14	4	13	8	14
Nebsoy	12	13	10	17	16	11	15	15
U20235	9	15	13	18	17	14	17	16
H7703	17	17	17	15	15	19	18	17
Weber	19	18	18	16	18	18	19	18
A77-212006	20	19	20	19	19	20	16	19
H75-5605	18	20	19	20	20	17	20	20

[a]PR = Puerto Rico, Nov = November, Feb = February, L = Lighted, NL = Nonlighted.

[b]C = Ranking combined over environments; 1 = lowest, 20 = highest percentage of palmitic acid.

Table 17. Ranking of lines in each environment for stearic acid

Line	Environments							
	Ames 1979	Ames 1980	Ames 1981	PR 1980 Nov L[a]	PR 1980 Nov NL	PR 1981 Feb L	PR 1981 Feb NL	C[b]
A2	1	1	1	1	3	1	3	1
Harcor	6	4	2	3	2	4	1	2
L75-3674	5	6	4	4	1	8	2	3
Corsoy	2	11	5	5	4	2	5	4
Amcor	4	5	7	6	6	6	6	5
L73-4673	3	3	3	7	8	5	7	6
U20235	7	2	6	8	7	3	8	7
Beeson 80	16	7	16	2	5	7	4	8
A77-212006	8	13	8	9	9	14	9	9
Century	12	8	11	11	10	9	10	10
Weber	9	12	13	13	12	18	12	11
Gnome	14	16	14	10	11	10	14	12
Beeson	10	9	18	17	15	11	11	13
Wells II	17	10	15	12	16	13	15	14
H75-5605	11	15	10	16	18	16	16	15
A77-211021	15	14	9	14	20	12	17	16
H7703	20	17	17	15	13	20	13	17
Nebsoy	13	19	12	18	19	15	20	18
Pella	19	18	20	19	14	17	18	19
U11239	18	20	19	20	17	19	19	20

[a]PR = Puerto Rico, Nov = November, Feb = February, L = Lighted, NL = Nonlighted.

[b]C = Ranking combined over environments; 1 = lowest, 20 = highest percentage of stearic acid.

Table 18. Ranking of lines in each environment for oleic acid

Line	Environments							
	Ames 1979	Ames 1980	Ames 1981	PR 1980 Nov L[a]	PR 1980 Nov NL	PR 1981 Feb L	PR 1981 Feb NL	C[b]
Wells II	4	3	4	1	1	2	1	1
Weber	5	5	2	2	3	1	2	2
H7703	3	2	1	5	2	3	3	3
Beeson	6	1	6	4	4	4	5	4
A77-211021	8	9	7	3	7	5	7	5
Century	7	6	9	7	5	6	4	6
U20235	17	12	10	6	6	9	6	7
H75-5605	1	4	5	8	14	16	11	8
U11239	10	14	16	9	10	8	8	9
Gnome	9	7	8	10	11	19	9	10
Pella	15	8	11	15	8	18	13	11
L73-4673	14	16	13	12	9	17	10	12
Nebsoy	12	18	18	11	13	14	12	13
Beeson 80	2	11	3	18	16	13	18	14
A2	16	17	19	17	12	10	15	15
L75-3674	11	13	14	16	17	11	16	16
Corsoy	13	15	12	14	19	15	17	17
A77-212006	19	10	20	19	18	7	19	18
Harcor	18	20	15	13	20	12	20	19
Amcor	20	19	17	20	15	20	14	20

[a]PR = Puerto Rico, Nov = November, Feb = February, L = Lighted, NL = Nonlighted.

[b]C = Ranking combined over environments; 1 = lowest, 20 = highest percentage of oleic acid.

Table 19. Ranking of lines in each environment for linoleic acid

Line	Environments							
	Ames 1979	Ames 1980	Ames 1981	PR 1980 Nov L[a]	PR 1980 Nov NL	PR 1981 Feb L	PR 1981 Feb NL	C[b]
A77-212006	1	6	1	2	2	12	1	1
Amcor	2	2	4	1	5	1	7	2
Harcor	6	3	6	12	1	10	2	3
Corsoy	8	5	8	9	3	7	4	4
Nebsoy	7	1	3	6	8	4	8	5
L75-3674	10	9	7	7	6	9	5	6
H75-5605	20	13	15	10	4	2	6	7
Beeson 80	18	10	20	3	7	8	3	8
A2	5	7	5	5	11	13	9	9
U11239	4	4	2	8	9	14	12	10
Pella	9	16	12	4	14	6	10	11
L73-4673	11	11	9	13	13	5	11	12
Gnome	14	15	14	11	10	3	13	13
U20235	3	8	10	14	15	11	15	14
A77-211021	13	14	16	18	12	17	14	15
Century	16	17	13	15	16	15	19	16
Weber	15	12	11	17	17	18	18	17
H7703	12	18	18	16	18	16	16	18
Beeson	17	20	17	19	19	19	17	19
Wells II	19	19	19	20	20	20	20	20

[a]PR = Puerto Rico, Nov = November, Feb = February, L = Lighted, NL = Nonlighted.

[b]C = Ranking combined over environments; 1 = lowest, 20 = highest percentage of linoleic acid.

Table 20. Ranking of lines in each environment for linolenic acid

Line	Environments							
	Ames 1979	Ames 1980	Ames 1981	PR 1980 Nov L[a]	PR 1980 Nov NL	PR 1981 Feb L	PR 1981 Feb NL	C[b]
Pella	1	5	1	2	5	1	6	1
Harcor	2	2	4	8	1	8	2	2
L73-4673	3	1	2	6	8	2	7	3
Corsoy	10	4	5	5	2	5	3	4
L75-3674	7	3	3	4	3	4	5	5
A2	6	6	7	3	7	6	9	6
Beeson 80	17	9	16	1	4	7	1	7
A77-212006	8	8	6	7	6	13	14	8
Nebsoy	4	7	8	9	9	9	8	9
Gnome	5	12	11	10	10	3	11	10
H75-5605	19	16	10	13	11	10	10	11
U11239	16	11	9	12	14	12	13	12
U20235	9	13	13	14	13	11	12	13
Amcor	12	10	15	11	12	14	14	14
Century	13	14	12	15	15	15	15	15
Wells II	11	15	14	16	17	17	19	16
A77-211021	18	17	17	17	16	19	16	17
H7703	20	19	19	18	19	16	18	18
Beeson	14	20	18	19	18	18	17	19
Weber	15	17	20	20	20	20	20	20

[a]PR = Puerto Rico, Nov = November, Feb = February, L = Lighted, NL = Nonlighted.

[b]C = Ranking combined over environments; 1 = lowest, 20 = highest percentage of linolenic acid.

The ranking of lines using means from the three Ames and the four Puerto Rico environments (Table 21) was similar and differed little from the overall ranking using means from seven environments. Phenotypic and rank correlation coefficients among environments (Table 22) were highly significant and exhibited little difference in the ability of either location to establish relative fatty acid composition for the lines used.

Predicted phenotypic variance, genetic gain (ΔG) and relative efficiency were compared to determine an efficient allocation of resources in establishing the relative fatty acid composition of soybean lines. Four assumptions were made when comparing the number of subsamples, replications, and environments: (1) selection intensity among lines was set at 10% ($k = 1.75$), (2) no supplemental lighting would be used in Puerto Rico, thus one environment per planting date, (3) gain per cycle would be calculated for the evaluation of S_1 lines with two seasons of recombination giving four seasons per cycle, and (4) an increase of 5% in relative efficiency would be required when considering an increase in the number of resources used.

Tables 23 to 27 and Figures 1, 6, 11, 16, and 21 provide the results of increasing the number of injections and seed samples for each fatty acid. More than one injection per seed sample had little effect on the rate of genetic gain. Increasing the number of seed samples, using one injection per sample, resulted in estimated genetic gains of 0.07 for palmitic, 0.04 for stearic, 0.06 for oleic, 0.07 for linoleic and linolenic acids. The increases that would be realized for palmitic, stearic, and linolenic acids was greater than the 5% level set for this

Table 21. Ranking of entries in Puerto Rico, Ames, and combined over environments for each fatty acid

Line	Fatty acid					
	Palmitic			Stearic		
	C[a]	PR	Ames	C	PR	Ames
Pella	6	6	9	19	18	20
Harcor	14	11	15	2	2	2
L73-4673	11	14	7	6	8	2
Corsoy	12	8	4	4	3	7
L75-3674	13	10	15	3	5	4
A2	8	9	8	1	1	1
Beeson 80	7	13	3	8	4	4
A77-212006	19	18	19	9	9	8
Nebsoy	15	15	13	18	19	17
Gnome	9	7	11	12	11	16
H75-5605	20	20	20	15	15	11
U11239	4	4	2	20	20	19
U20235	16	16	12	7	7	6
Amcor	3	3	5	5	6	5
Century	10	12	10	10	10	9
Wells II	2	2	4	14	14	15
A77-211021	5	5	6	16	17	12
H7703	17	17	17	17	16	18
Beeson	1	1	1	13	12	13
Weber	18	19	18	11	12	10

[a]C = combined over environments, PR = Puerto Rico, rankings are arranged 1 = lowest 20 = highest percentage of linolenic acid.

Fatty acid								
Oleic			Linoleic			Linolenic		
C	PR	Ames	C	PR	Ames	C	PR	Ames
11	13	10	11	9	11	1	1	1
19	20	18	3	3	5	2	4	3
12	11	15	12	12	10	3	6	2
17	19	14	4	4	7	4	3	6
16	15	13	6	7	9	5	5	4
15	14	19	9	11	6	6	7	5
14	16	6	8	6	17	6	2	14
18	17	17	1	1	1	8	8	7
13	12	16	5	8	3	9	9	8
10	9	9	13	10	13	10	10	9
8	10	3	7	5	16	11	11	16
9	8	12	10	13	4	12	13	11
7	7	11	14	14	8	13	12	10
20	18	20	2	2	2	14	13	12
6	6	7	16	16	15	15	15	12
1	1	4	20	20	20	16	17	15
5	5	8	15	15	14	17	16	17
3	3	1	18	17	18	18	18	20
4	4	2	19	19	19	19	19	18
2	2	5	17	18	12	20	20	19

Table 22. Phenotypic and rank correlation coefficients between the combined Puerto Rico environments and the combined Ames environments

Fatty acid	Correlation	
	Phenotypic	Rank
Palmitic	0.89**	0.74**
Stearic	0.86**	0.82**
Oleic	0.74**	0.74**
Linoleic	0.71**	0.63**
Linolenic	0.84**	0.82**

**Significant at the 0.01 probability level.

Table 23. Estimated phenotypic variance, ΔG, and relative efficiency for palmitic acid using different resource allocations

		σ^2_{ph} a						ΔG						Rel. eff.					
		1	2	3	4	5	6	1	2	3	4	5	6	1	2	3	4	5	6
								No. of injections											
No. of seed samples	1	0.49	0.48	0.48	0.48	0.48	0.48	0.48	0.48	0.48	0.48	0.48	0.48	0.64	0.65	0.65	0.65	0.65	0.65
	2	0.37	0.27	0.36	0.36	0.36	0.36	0.55	0.55	0.55	0.55	0.55	0.55	0.74	0.74	0.75	0.75	0.75	0.75
	3	0.33	0.33	0.33	0.33	0.32	0.32	0.58	0.58	0.58	0.58	0.58	0.58	0.78	0.79	0.79	0.79	0.79	0.79
	4	0.31	0.31	0.31	0.31	0.31	0.31	0.60	0.60	0.60	0.60	0.60	0.60	0.81	0.81	0.81	0.81	0.81	0.81
	5	0.30	0.30	0.30	0.30	0.30	0.30	0.61	0.61	0.61	0.61	0.61	0.61	0.82	0.83	0.83	0.83	0.83	0.83
	6	0.29	0.29	0.29	0.29	0.29	0.29	0.62	0.62	0.62	0.62	0.62	0.62	0.83	0.84	0.84	0.84	0.84	0.84

1 Environment, 1 Replication, 1 Plant, 1-6 Seed samples, 1-6 Injections

								No. of seed samples											
No. of plants	1	0.49	0.37	0.33	0.31	0.30	0.29	0.48	0.55	0.58	0.60	0.61	0.62	0.64	0.74	0.78	0.81	0.82	0.83
	2	0.36	0.30	0.28	0.26	0.26	0.26	0.56	0.61	0.63	0.65	0.66	0.66	0.75	0.83	0.86	0.87	0.88	0.89
	3	0.31	0.27	0.26	0.25	0.24	0.24	0.60	0.64	0.66	0.67	0.67	0.67	0.81	0.86	0.89	0.90	0.91	0.91
	4	0.29	0.26	0.25	0.24	0.24	0.24	0.62	0.66	0.67	0.68	0.68	0.68	0.84	0.89	0.90	0.91	0.92	0.92
	5	0.27	0.25	0.24	0.24	0.24	0.23	0.64	0.67	0.68	0.69	0.69	0.69	0.86	0.90	0.91	0.92	0.93	0.93
	6	0.26	0.25	0.24	0.24	0.23	0.23	0.65	0.67	0.68	0.69	0.69	0.69	0.87	0.91	0.92	0.93	0.93	0.93

1 Environment, 1 Replication, 1-6 Plants, 1-6 Seed samples, 1 Injection

		No. of plants																
No. of replications	1	0.49	0.36	0.31	0.29	0.27	0.26	0.48	0.56	0.60	0.62	0.64	0.65	0.64	0.75	0.81	0.84	0.86	0.87
	2	0.36	0.29	0.26	0.25	0.25	0.24	0.56	0.62	0.65	0.66	0.67	0.68	0.75	0.84	0.87	0.89	0.90	0.91
	3	0.31	0.26	0.25	0.24	0.24	0.24	0.60	0.65	0.67	0.68	0.68	0.69	0.81	0.87	0.90	0.91	0.92	0.93
	4	0.29	0.25	0.24	0.24	0.23	0.23	0.62	0.66	0.68	0.69	0.69	0.69	0.84	0.89	0.91	0.92	0.93	0.93
	5	0.27	0.24	0.24	0.23	0.23	0.23	0.64	0.67	0.68	0.69	0.69	0.69	0.86	0.90	0.92	0.93	0.94	0.94
	6	0.26	0.24	0.24	0.23	0.23	0.23	0.65	0.68	0.69	0.69	0.69	0.70	0.87	0.91	0.93	0.93	0.94	0.94

1 Environment, 1-6 Replications, 1-6 Plants, 1 Seed sample, 1 Injection

		No. of Replications																
No. of environments	1	0.36	0.29	0.26	0.25	0.24	0.24	0.56	0.62	0.65	0.66	0.67	0.68	0.75	0.84	0.87	0.89	0.90	0.91
	2	0.27	0.24	0.23	0.22	0.22	0.22	0.64	0.68	0.70	0.71	0.71	0.72	0.86	0.92	0.94	0.95	0.96	0.97
	3	0.25	0.22	0.22	0.21	0.21	0.21	0.67	0.70	0.72	0.72	0.73	0.73	0.91	0.95	0.97	0.98	0.98	0.99
	4	0.23	0.21	0.21	0.21	0.20	0.20	0.69	0.71	0.73	0.73	0.73	0.74	0.94	0.97	0.98	0.99	0.99	1.00
	5	0.22	0.21	0.21	0.20	0.20	0.20	0.70	0.72	0.73	0.73	0.74	0.74	0.97	0.98	0.99	0.99	1.00	1.00
	6	0.22	0.21	0.20	0.20	0.20	0.20	0.72	0.73	0.73	0.74	0.74	0.74	0.98	0.99	0.99	1.00	1.00	1.00

1-6 Environments, 1-6 Replications, 2 Plants, 1 Seed sample, 1 Injection

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$^{a}\sigma^2_{ph}$ = estimated phenotypic variance, ΔG, = estimated genetic gain, Rel. eff. = relative efficiency.

Table 24. Estimated phenotypic variance, ΔG, and relative efficiency for stearic acid using different resource allocations

		σ^2_{ph} [a]						ΔG						Rel. eff.					
		1	2	3	4	5	6	1	2	3	4	5	6	1	2	3	4	5	6
								No. of injections											
No. of seed samples	1	0.30	0.30	0.29	0.29	0.29	0.29	0.51	0.52	0.52	0.52	0.52	0.52	0.75	0.76	0.76	0.76	0.76	0.76
	2	0.26	0.26	0.26	0.26	0.26	0.26	0.55	0.55	0.55	0.55	0.55	0.55	0.81	0.81	0.81	0.81	0.81	0.81
	3	0.25	0.25	0.24	0.24	0.24	0.24	0.57	0.57	0.57	0.57	0.57	0.57	0.83	0.83	0.83	0.83	0.83	0.83
	4	0.24	0.24	0.24	0.24	0.24	0.24	0.57	0.57	0.57	0.57	0.57	0.57	0.84	0.84	0.84	0.84	0.84	0.84
	5	0.24	0.24	0.23	0.23	0.23	0.23	0.58	0.58	0.58	0.58	0.58	0.58	0.85	0.85	0.85	0.85	0.85	0.85
	6	0.23	0.23	0.23	0.23	0.23	0.23	0.58	0.58	0.58	0.58	0.58	0.58	0.85	0.85	0.85	0.85	0.85	0.85
	1 Environment, 1 Replication, 1 Plant, 1-6 Seed samples, 1-6 Injections																		
								No. of seed samples											
No. of plants	1	0.30	0.26	0.25	0.24	0.24	0.23	0.51	0.55	0.56	0.57	0.58	0.58	0.75	0.81	0.83	0.84	0.85	0.85
	2	0.26	0.24	0.23	0.23	0.22	0.22	0.55	0.58	0.59	0.59	0.59	0.59	0.82	0.85	0.86	0.87	0.87	0.87
	3	0.24	0.23	0.22	0.22	0.22	0.22	0.57	0.59	0.60	0.60	0.60	0.60	0.84	0.86	0.87	0.88	0.88	0.88
	4	0.23	0.22	0.22	0.22	0.22	0.22	0.58	0.59	0.60	0.60	0.60	0.60	0.85	0.87	0.88	0.88	0.88	0.89
	5	0.23	0.22	0.22	0.22	0.22	0.21	0.59	0.60	0.60	0.60	0.60	0.60	0.86	0.88	0.88	0.89	0.89	0.89
	6	0.23	0.22	0.22	0.22	0.21	0.21	0.59	0.60	0.60	0.60	0.60	0.60	0.87	0.88	0.89	0.89	0.89	0.89
	1 Environment, 1 Replication, 1-6 Plants, 1-6 Seed samples, 1 Injection																		

		No. of plants																
No. of replications	1	0.30	0.26	0.24	0.23	0.23	0.23	0.51	0.55	0.57	0.58	0.59	0.59	0.75	0.82	0.84	0.85	0.86	0.87
	2	0.25	0.22	0.22	0.21	0.21	0.21	0.57	0.59	0.60	0.61	0.61	0.61	0.83	0.87	0.89	0.90	0.90	0.90
	3	0.23	0.21	0.21	0.20	0.20	0.20	0.59	0.61	0.62	0.62	0.62	0.62	0.86	0.89	0.91	0.91	0.91	0.92
	4	0.22	0.21	0.20	0.20	0.20	0.20	0.60	0.62	0.62	0.63	0.63	0.63	0.88	0.91	0.92	0.92	0.92	0.92
	5	0.21	0.20	0.20	0.20	0.20	0.20	0.61	0.62	0.63	0.63	0.63	0.63	0.89	0.91	0.92	0.92	0.93	0.93
	6	0.21	0.20	0.20	0.20	0.20	0.20	0.61	0.62	0.63	0.63	0.63	0.63	0.90	0.92	0.92	0.93	0.93	0.93

1 Environment, 1-6 Replications, 1-6 Plants, 1 Seed sample, 1 Injection

		No. of replications																
No. of environments	1	0.26	0.22	0.21	0.21	0.20	0.20	0.55	0.59	0.61	0.62	0.62	0.62	0.82	0.87	0.89	0.91	0.91	0.92
	2	0.21	0.19	0.19	0.18	0.18	0.18	0.61	0.64	0.65	0.65	0.66	0.66	0.90	0.94	0.96	0.96	0.97	0.97
	3	0.19	0.18	0.18	0.18	0.17	0.17	0.64	0.66	0.67	0.67	0.67	0.67	0.94	0.97	0.98	0.98	0.99	0.99
	4	0.18	0.18	0.17	0.17	0.17	0.17	0.65	0.67	0.67	0.68	0.68	0.68	0.96	0.98	0.99	0.99	1.00	1.00
	5	0.18	0.17	0.17	0.17	0.17	0.17	0.66	0.67	0.68	0.68	0.68	0.68	0.97	0.99	1.00	1.00	1.00	1.00
	6	0.18	0.17	0.17	0.17	0.17	0.17	0.67	0.68	0.68	0.68	0.68	0.68	0.98	1.00	1.00	1.00	1.00	1.00

1-6 Environments, 1-6 Replications, 2 Plants, 1 Seed sample, 1 Injection

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[a] σ^2_{ph} = estimated phenotypic variance, ΔG = estimated genetic gain, Rel. eff. = relative efficiency.

Table 25. Estimated phenotypic variance, ΔG, and relative efficiency for oleic acid using different resource allocations

		σ^2_{ph} [a]						ΔG						Rel. eff.					
		1	2	3	4	5	6	1	2	3	4	5	6	1	2	3	4	5	6
								No. of injections											
No. of seed samples	1	17.33	17.33	17.33	17.33	17.33	17.33	3.66	3.66	3.66	3.66	3.66	3.66	0.74	0.74	0.74	0.74	0.74	0.74
	2	16.76	16.76	16.76	16.76	16.76	16.76	3.72	3.72	3.72	3.72	3.72	3.72	0.75	0.75	0.75	0.75	0.75	0.75
	3	16.54	16.54	16.54	16.54	16.54	16.54	3.74	3.74	3.74	3.74	3.74	3.74	0.76	0.76	0.76	0.76	0.76	0.76
	4	16.45	16.45	16.45	16.45	16.45	16.45	3.75	3.75	3.75	3.75	3.75	3.75	0.76	0.76	0.76	0.76	0.76	0.76
	5	16.38	16.38	16.38	16.38	16.38	16.38	3.76	3.76	3.76	3.76	3.76	3.76	0.76	0.76	0.76	0.76	0.76	0.76
	6	16.35	16.35	16.35	16.35	16.35	16.35	3.77	3.77	3.77	3.77	3.77	3.77	0.76	0.76	0.76	0.76	0.76	0.76
		1 Environment, 1 Replication/Environment, 1 plant/Replication, 1-6 Seed samples, 1-6 Injections																	
								No. of seed samples											
No. of plants	1	17.33	16.76	16.54	16.45	16.38	16.35	3.66	3.72	3.74	3.75	3.76	3.77	0.74	0.75	0.76	0.76	0.76	0.76
	2	15.34	15.05	14.97	14.92	14.87	14.87	3.89	3.92	3.94	3.94	3.95	3.95	0.79	0.79	0.80	0.80	0.80	0.80
	3	14.70	14.50	14.43	14.40	14.38	14.36	3.97	4.00	4.01	4.01	4.02	4.02	0.80	0.81	0.81	0.81	0.81	0.81
	4	14.36	14.20	14.17	14.12	14.12	14.12	4.02	4.04	4.05	4.05	4.05	4.05	0.81	0.82	0.82	0.82	0.82	0.82
	5	14.17	14.03	14.00	13.98	13.96	13.96	4.05	4.06	4.07	4.07	4.07	4.08	0.82	0.82	0.82	0.82	0.82	0.82
	6	14.03	13.94	13.90	13.88	13.88	13.88	4.07	4.08	4.09	4.09	4.09	4.09	0.82	0.82	0.83	0.83	0.83	0.83
		1 Environment, 1 Replication/Environment, 1-6 Plants, 1-6 Seed samples, 1 Injection																	

No. of plants

No. of replications																		
	1	17.33	15.34	14.70	14.36	14.17	14.03	3.66	3.89	3.97	4.02	4.05	4.07	0.74	0.79	0.83	0.81	0.82	0.82
	2	15.05	14.05	13.72	13.55	13.47	14.41	3.93	4.06	4.11	4.14	4.15	4.16	0.79	0.82	0.83	0.84	0.84	0.84
	3	14.29	13.62	13.41	13.30	13.22	13.18	4.03	4.13	4.16	4.18	4.19	4.19	0.81	0.83	0.84	0.84	0.85	0.85
	4	13.90	13.41	13.24	13.16	13.10	13.08	4.08	4.16	4.19	4.20	4.21	4.21	0.83	0.84	0.85	0.85	0.85	0.85
	5	13.68	13.28	13.14	13.08	13.04	13.00	4.12	4.18	4.20	4.21	4.22	4.22	0.83	0.84	0.85	0.85	0.85	0.85
	6	13.53	13.18	13.08	13.02	12.99	12.97	4.14	4.19	4.21	4.22	4.22	4.23	0.84	0.85	0.85	0.85	0.85	0.85

1 Environment, 1-6 Replications, 1-6 Plants, 1 Seed sample, 1 Injection

No. of replications

No. of environments																		
	1	15.34	14.05	13.62	13.41	13.28	13.18	3.89	4.06	4.13	4.16	4.18	4.19	0.79	0.82	0.83	0.84	0.84	0.85
	2	12.03	11.39	11.17	11.05	10.98	10.94	4.39	4.51	4.56	4.58	4.59	4.60	0.89	0.91	0.92	0.93	0.93	0.93
	3	10.92	10.48	10.34	10.27	10.22	10.20	4.61	4.70	4.74	4.75	4.76	4.77	0.93	0.95	0.96	0.96	0.96	0.96
	4	10.37	10.05	9.93	9.88	9.85	9.83	4.73	4.81	4.83	4.85	4.85	4.86	0.96	0.97	0.98	0.98	0.98	0.98
	5	10.03	9.77	9.68	9.64	9.61	9.60	4.81	4.87	4.89	4.89	4.91	4.91	0.97	0.98	0.99	0.99	0.99	0.99
	6	9.94	9.73	9.65	9.62	9.60	9.58	4.83	4.88	4.90	4.91	4.91	4.92	0.98	0.99	0.99	0.99	0.99	0.99

1-6 Environments, 1-6 Replications, 2 Plants, 1 Seed sample, 1 Injection

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$^{a}\sigma^2_{ph}$ = estimated phenotypic variance, ΔG = estimated genetic gain, Rel. eff. = relative efficiency.

Table 26. Estimated phenotypic variance, ΔG, and relative efficiency for linoleic acid using different resource allocations

		σ^2_{ph} [a]						ΔG						Rel. eff.					
		1	2	3	4	5	6	1	2	3	4	5	6	1	2	3	4	5	6
								No. of injections											
No. of seed samples	1	10.46	10.46	10.46	10.46	10.46	10.46	2.66	2.66	2.66	2.66	2.67	2.67	0.72	0.72	0.72	0.72	0.72	0.72
	2	9.84	9.84	9.84	9.84	9.84	9.84	2.73	2.73	2.73	2.73	2.73	2.73	0.74	0.74	0.74	0.74	0.74	0.74
	3	9.84	9.84	9.84	9.84	9.84	9.84	2.76	2.76	2.76	2.76	2.76	2.76	0.74	0.74	0.74	0.74	0.74	0.74
	4	9.65	9.65	9.65	9.65	9.65	9.65	2.77	2.77	2.77	2.77	2.77	2.77	0.75	0.75	0.75	0.75	0.75	0.75
	5	9.65	9.65	9.65	9.65	9.65	9.65	2.78	2.78	2.78	2.78	2.78	2.78	0.75	0.75	0.75	0.75	0.75	0.75
	6	9.65	9.65	9.65	9.65	9.65	9.65	2.78	2.78	2.78	2.78	2.78	2.78	0.75	0.75	0.75	0.75	0.75	0.75
	1 Environment, 1 Replication, 1 Plant, 1-6 Seed samples, 1-6 Injections																		
								No. of seed samples											
No. of plants	1	10.46	9.84	9.84	9.65	9.65	9.65	2.66	2.73	2.76	2.77	2.78	2.78	0.72	0.74	0.74	0.75	0.75	0.75
	2	9.11	8.95	8.79	8.79	8.79	8.79	2.84	2.88	2.90	2.91	2.91	2.91	0.77	0.78	0.78	0.78	0.78	0.79
	3	8.79	8.63	8.48	8.48	8.48	8.48	2.91	2.94	2.95	2.96	2.96	2.96	0.78	0.79	0.80	0.80	0.80	0.80
	4	8.48	8.34	8.34	8.34	8.34	8.34	2.95	2.97	2.98	2.98	2.99	2.99	0.79	0.80	0.81	0.80	0.80	0.81
	5	8.34	8.34	8.20	8.20	8.20	8.20	2.97	2.99	3.00	3.00	3.00	3.00	0.80	0.81	0.81	0.81	0.81	0.81
	6	8.34	8.20	8.20	8.20	8.20	8.20	2.99	3.00	3.00	3.00	3.00	3.00	0.80	0.81	0.81	0.81	0.81	0.81
	1 Environment, 1 Replication, 1-6 Plants, 1-6 Seed samples, 1 Injection																		

No. of plants

No. of replications	1	10.46	9.11	8.79	8.48	8.34	8.34	2.66	2.84	2.91	2.95	2.97	2.99	0.72	0.77	0.78	0.79	0.80	0.80
	2	8.95	8.34	8.07	7.94	7.94	7.81	2.88	2.99	3.03	3.05	3.06	3.07	0.78	0.81	0.82	0.82	0.83	0.83
	3	8.48	7.94	7.81	7.69	7.69	7.69	2.97	3.05	3.07	3.09	3.10	3.10	0.80	0.82	0.83	0.83	0.83	0.84
	4	8.20	7.81	7.69	7.69	7.69	7.57	3.01	3.07	3.10	3.11	3.11	3.12	0.81	0.83	0.83	0.84	0.84	0.84
	5	8.07	7.81	7.69	7.57	7.57	7.57	3.04	3.09	3.11	3.12	3.12	3.13	0.82	0.83	0.84	0.84	0.84	0.84
	6	7.94	7.69	7.57	7.57	7.57	7.57	3.06	3.10	3.12	3.13	3.13	3.13	0.83	0.84	0.84	0.84	0.84	0.84

1 Environment, 1-6 Replications, 1-6 Plants, 1 Seed sample, 1 Injection

No. of replications

No. of environments	1	9.11	8.34	7.94	7.81	7.81	7.69	2.84	2.99	3.05	3.07	3.09	3.10	0.77	0.81	0.82	0.83	0.83	0.84
	2	7.04	6.60	6.46	6.39	6.34	6.31	3.24	3.35	3.39	3.41	3.42	3.43	0.87	0.90	0.91	0.92	0.92	0.92
	3	6.34	6.07	5.94	5.90	5.86	5.85	3.42	3.50	3.53	3.55	3.56	3.56	0.92	0.94	0.95	0.96	0.96	0.96
	4	5.99	5.76	5.69	5.66	5.63	5.62	3.52	3.59	3.61	3.62	3.63	3.63	0.95	0.97	0.97	0.98	0.98	0.98
	5	5.77	5.59	5.53	5.51	5.50	5.49	3.58	3.64	3.66	3.67	3.67	3.68	0.97	0.98	0.99	0.99	0.99	0.99
	6	5.63	5.48	5.43	5.41	5.39	5.38	3.63	3.68	3.70	3.70	3.71	3.71	0.98	0.99	1.00	1.00	1.00	1.00

1-6 Environments, 1-6 Replications, 2 Plants, 1 Seed sample, 1 Injection

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[a] σ^2_{ph} = estimated phenotypic variance, ΔG = estimated genetic gain, Rel. eff. = relative efficiency.

Table 27. Estimated phenotypic variance, ΔG, and relative efficiency for linolenic acid using different resource allocations

		$\sigma^{2\,a}_{ph}$						ΔG						Rel. eff.					
		1	2	3	4	5	6	1	2	3	4	5	6	1	2	3	4	5	6
								No. of injections											
No. of seed samples	1	0.99	0.98	0.98	0.98	0.98	0.98	0.92	0.92	0.92	0.92	0.92	0.92	0.74	0.74	0.74	0.74	0.74	0.74
	2	0.86	0.85	0.85	0.85	0.85	0.85	0.99	0.99	0.99	0.99	0.99	0.99	0.80	0.80	0.80	0.80	0.80	0.80
	3	0.81	0.80	0.80	0.80	0.80	0.80	1.02	1.02	1.02	1.02	1.02	1.02	0.82	0.82	0.82	0.82	0.82	0.82
	4	0.78	0.78	0.78	0.78	0.78	0.78	1.03	1.03	1.03	1.03	1.04	1.04	0.83	0.83	0.83	0.84	0.84	0.84
	5	0.77	0.77	0.77	0.77	0.77	0.77	1.04	1.04	1.04	1.04	1.04	1.04	0.84	0.84	0.84	0.84	0.84	0.84
	6	0.76	0.76	0.76	0.76	0.76	0.76	1.05	1.05	1.05	1.05	1.05	1.05	0.85	0.85	0.85	0.85	0.85	0.85
	1 Environment, 1 Replication, 1 Plant, 1-6 Seed samples, 1-6 Injections																		
								No. of seed samples											
No. of plants	1	0.99	0.86	0.81	0.78	0.77	0.76	0.92	0.99	1.02	1.03	1.04	1.05	0.74	0.80	0.82	0.83	0.84	0.85
	2	0.82	0.75	0.73	0.71	0.71	0.70	1.01	1.06	1.07	1.08	1.09	1.09	0.81	0.85	0.87	0.87	0.88	0.88
	3	0.76	0.72	0.70	0.69	0.68	0.68	1.05	1.08	1.09	1.10	1.10	1.11	0.85	0.87	0.88	0.89	0.89	0.89
	4	0.73	0.70	0.68	0.68	0.67	0.67	1.07	1.10	1.11	1.11	1.11	1.12	0.86	0.88	0.89	0.90	0.90	0.90
	5	0.71	0.68	0.68	0.67	0.67	0.67	1.08	1.10	1.11	1.12	1.12	1.12	0.87	0.89	0.90	0.90	0.90	0.90
	6	0.70	0.68	0.67	0.67	0.66	0.66	1.09	1.11	1.12	1.12	1.12	1.12	0.88	0.90	0.90	0.90	0.90	0.91
	1 Environment, 1 Replication, 1-6 Plants, 1-6 Seed samples, 1 Injection																		

No. of plants

No. of replications	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18
1	0.99	0.82	0.76	0.73	0.71	0.70	0.92	1.01	1.05	1.07	1.08	1.09	0.74	0.81	0.85	0.86	0.87	0.88
2	0.81	0.73	0.70	0.68	0.67	0.67	1.01	1.07	1.10	1.11	1.11	1.12	0.82	0.87	0.88	0.89	0.90	0.90
3	0.75	0.69	0.67	0.66	0.66	0.65	1.05	1.10	1.11	1.12	1.13	1.13	0.85	0.88	0.90	0.90	0.91	0.91
4	0.72	0.68	0.66	0.66	0.65	0.65	1.07	1.11	1.12	1.13	1.13	1.13	0.87	0.89	0.90	0.91	0.91	0.91
5	0.70	0.67	0.66	0.65	0.65	0.65	1.09	1.12	1.13	1.13	1.13	1.13	0.88	0.90	0.91	0.91	0.92	0.92
6	0.69	0.66	0.65	0.65	0.65	0.64	1.10	1.12	1.13	1.13	1.14	1.14	0.89	0.90	0.91	0.91	0.92	0.92

1 Environment, 1-6 Replications, 1-6 Plants, 1 Seed sample, 1 Injection

No. of replications

No. of environments	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18
1	0.82	0.73	0.69	0.68	0.67	0.66	1.01	1.07	1.10	1.11	1.12	1.12	0.81	0.87	0.88	0.89	0.90	0.90
2	0.67	0.62	0.61	0.60	0.60	0.59	1.11	1.16	1.17	1.18	1.18	1.19	0.90	0.93	0.94	0.95	0.95	0.96
3	0.62	0.59	0.58	0.57	0.57	0.57	1.16	1.19	1.20	1.21	1.21	1.21	0.94	0.96	0.97	0.97	0.97	0.98
4	0.60	0.57	0.56	0.56	0.56	0.56	1.18	1.21	1.22	1.22	1.22	1.22	0.95	0.97	0.98	0.98	0.99	0.99
5	0.58	0.56	0.55	0.55	0.55	0.55	1.20	1.22	1.23	1.23	1.23	1.23	0.97	0.98	0.99	0.99	0.99	0.99
6	0.57	0.56	0.55	0.55	0.55	0.55	1.21	1.22	1.23	1.23	1.23	1.23	0.97	0.99	0.99	0.99	0.99	0.99

1-6 Environments, 1-6 Replications, 2 Plants, 1 Seed sample, 1 Injection

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$^{a}\sigma^2_{ph}$ = estimated phenotypic variance, ΔG = estimated genetic gain, Rel. eff. = relative efficiency.

comparison (Figures 2, 7, and 22); however, in each case the use of two plants and one seed sample per plant would give slightly higher gains. Estimated gains for linoleic and oleic acid were increased substantially by using two plants with one seed sample each.

The combination of using two environments with two replications, two plants per replication, one seed sample per plant, and one injection per sample provided an increase in genetic gain for palmitic acid from 0.56 to 0.68 (Table 23), 0.55 to 0.64 for stearic acid (Table 24) and 1.01 to 1.16 for linolenic acid (Table 27). The gain per year for each of these fatty acids (Figures 5, 10, and 25) was less when two environments are used.

The combination of two environments, one replication per environment, two plants per replication, one seed sample per plant, and one injection per seed sample provided estimated gains for oleic and linoleic acid that were greater than using one environment and one or two replications. The gains per year when using two environments were substantially less than when one environment was used (Figures 14, 15, 19, and 20).

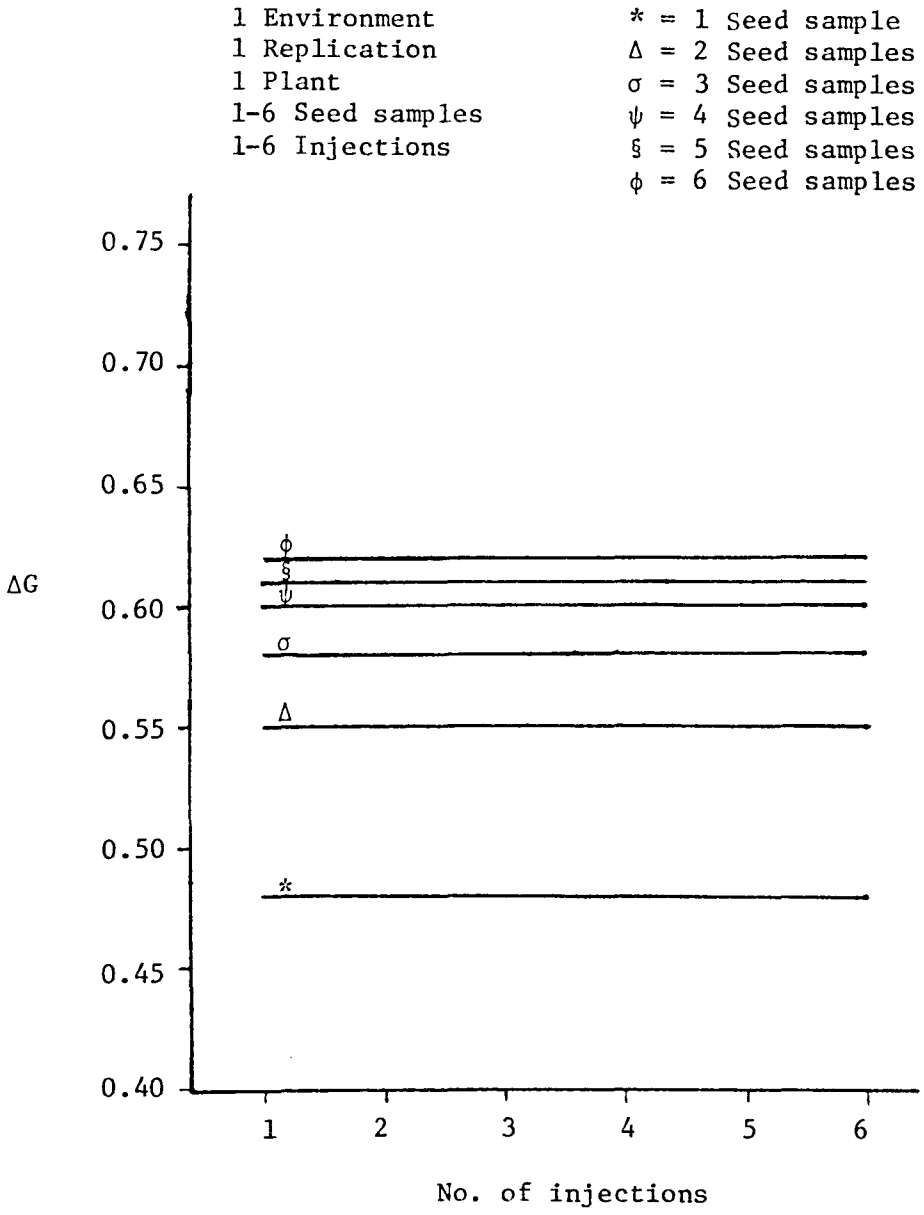

Figure 1. Relationship of expected gain to numbers of seed samples and injections per seed sample for palmitic acid

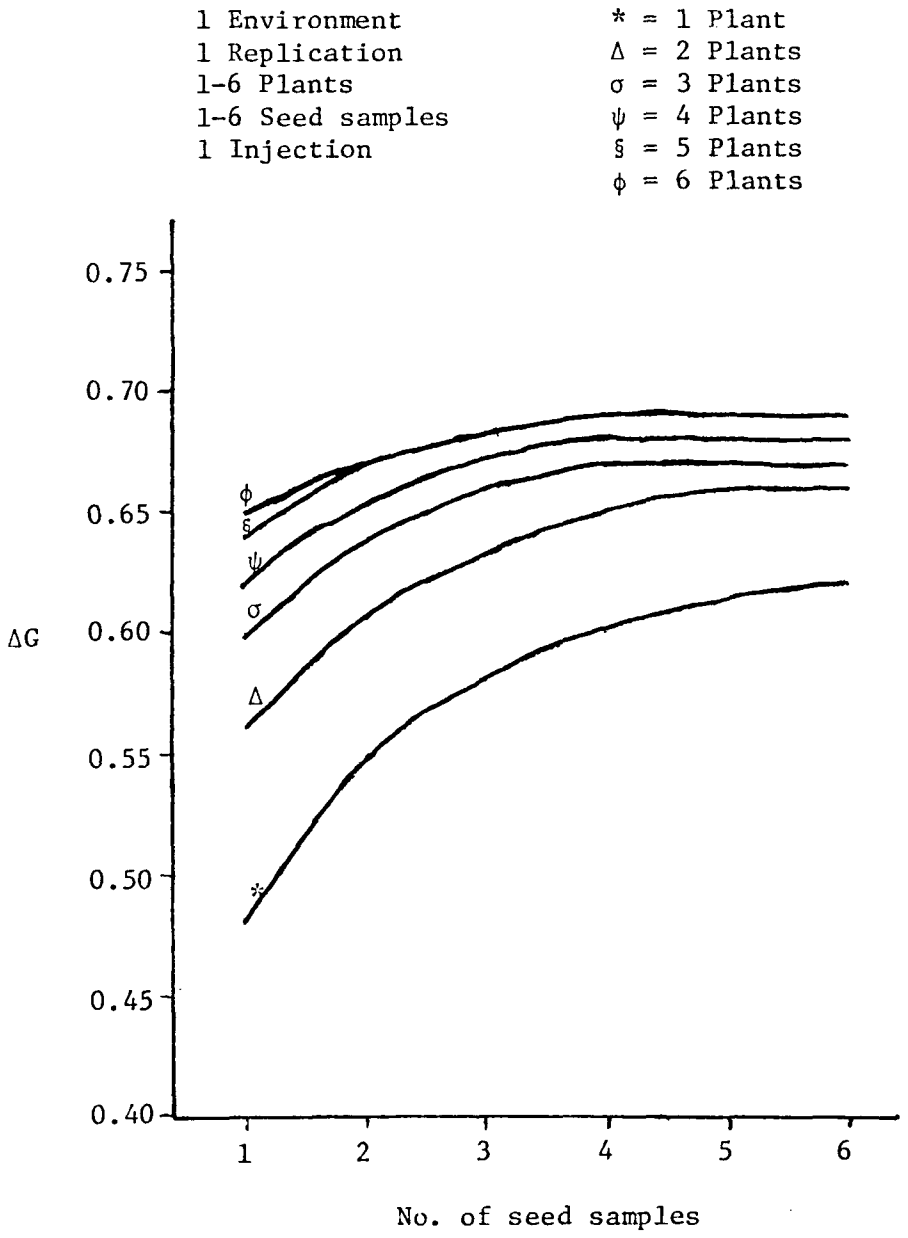

Figure 2. Relationship of expected gain to numbers of plants and seed samples per plant for palmitic acid

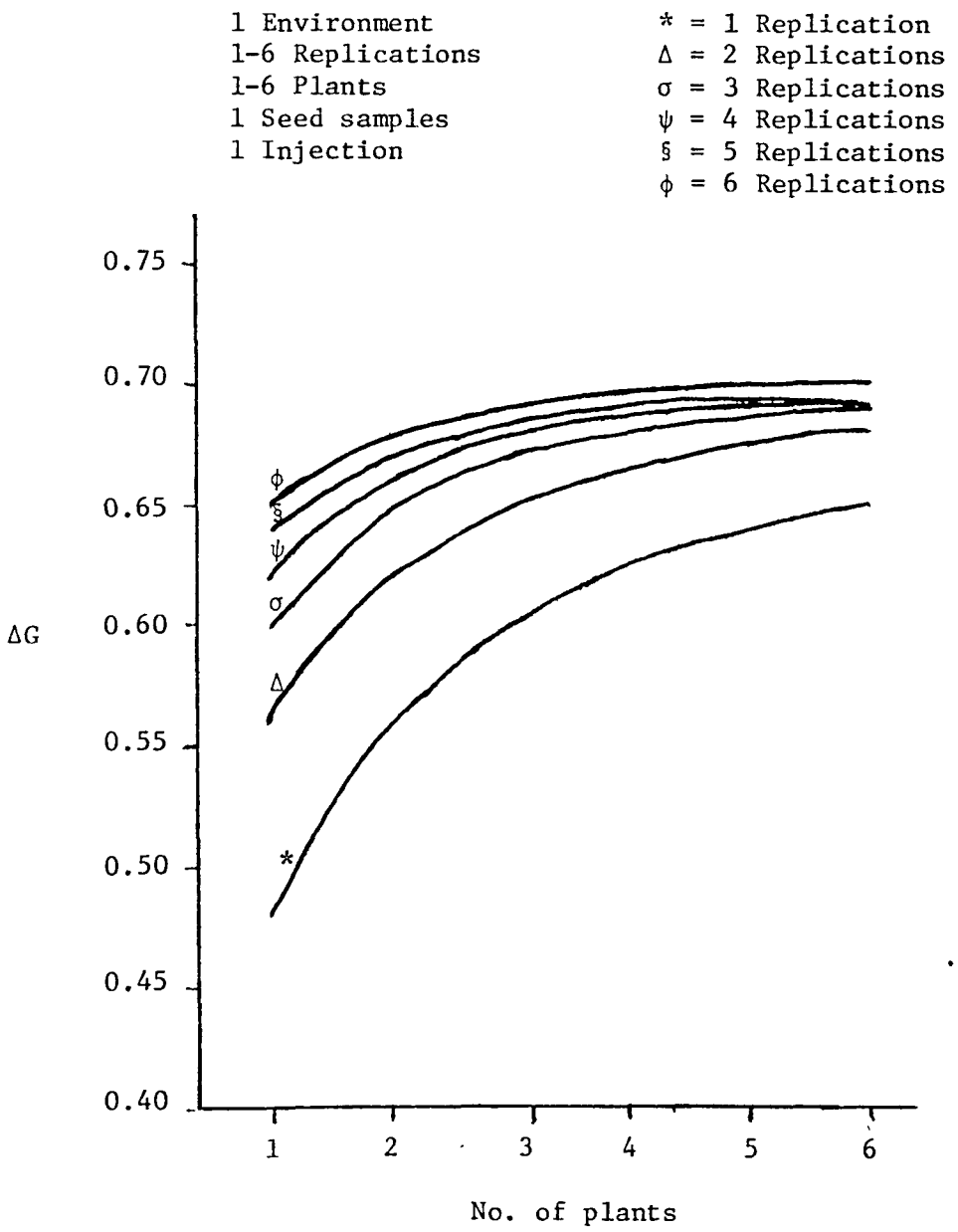

Figure 3. Relationship of genetic gain to numbers of replications and plants per replication for palmitic acid

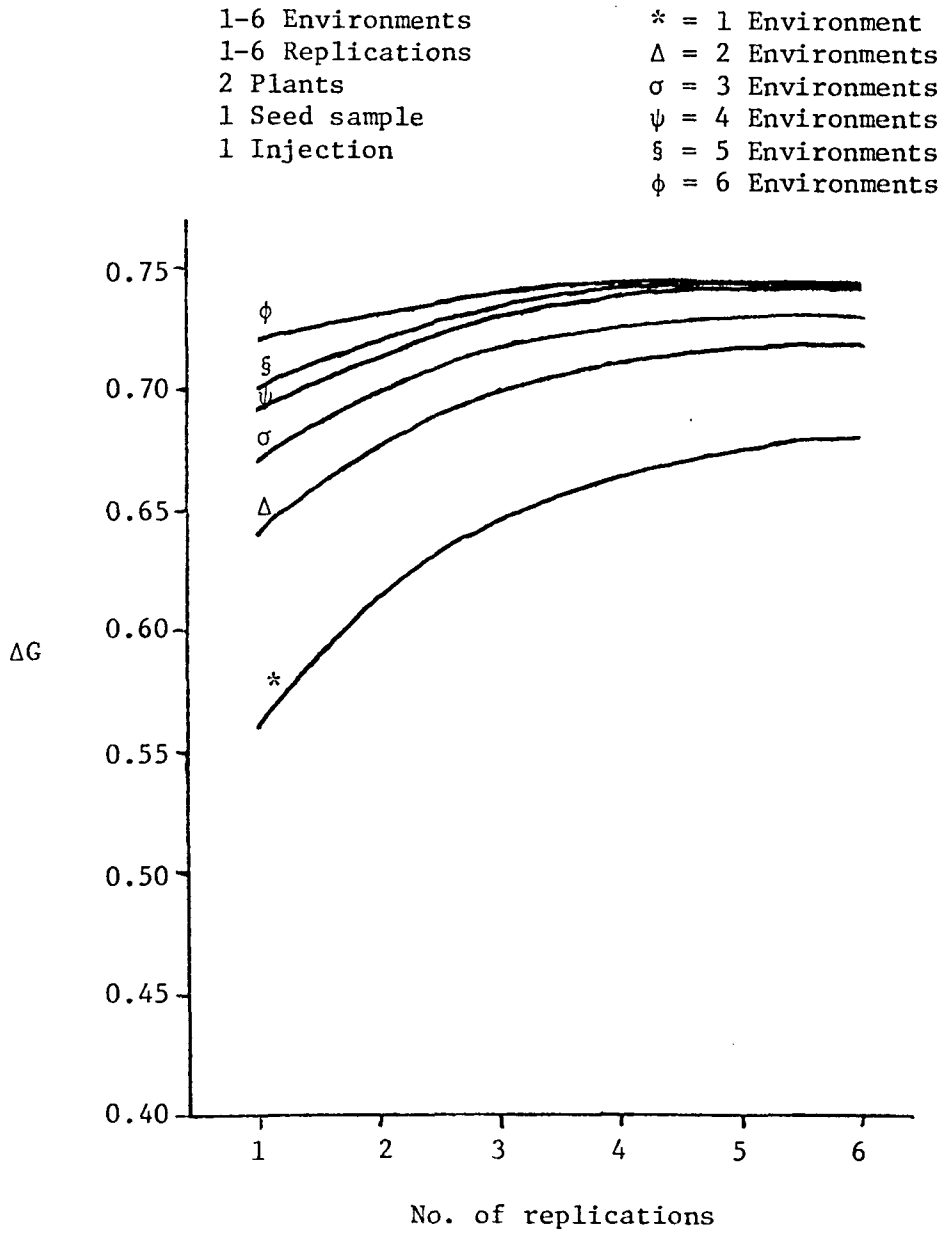

Figure 4. Relationship of genetic gain to numbers of environments and replications per environment for palmitic acid

1-6 Environments
1-6 Replications
2 Plants
2 Seed samples
1 Injection

* = 1.3 yr/cycle 1 Environment
Δ = 1.6 yr/cycle 2 Environments
σ = 2.0 yr/cycle 3 Environments
ψ = 2.3 yr/cycle 4 Environments
§ = 2.6 yr/cycle 5 Environments
ϕ = 3.0 yr/cycle 6 Environments

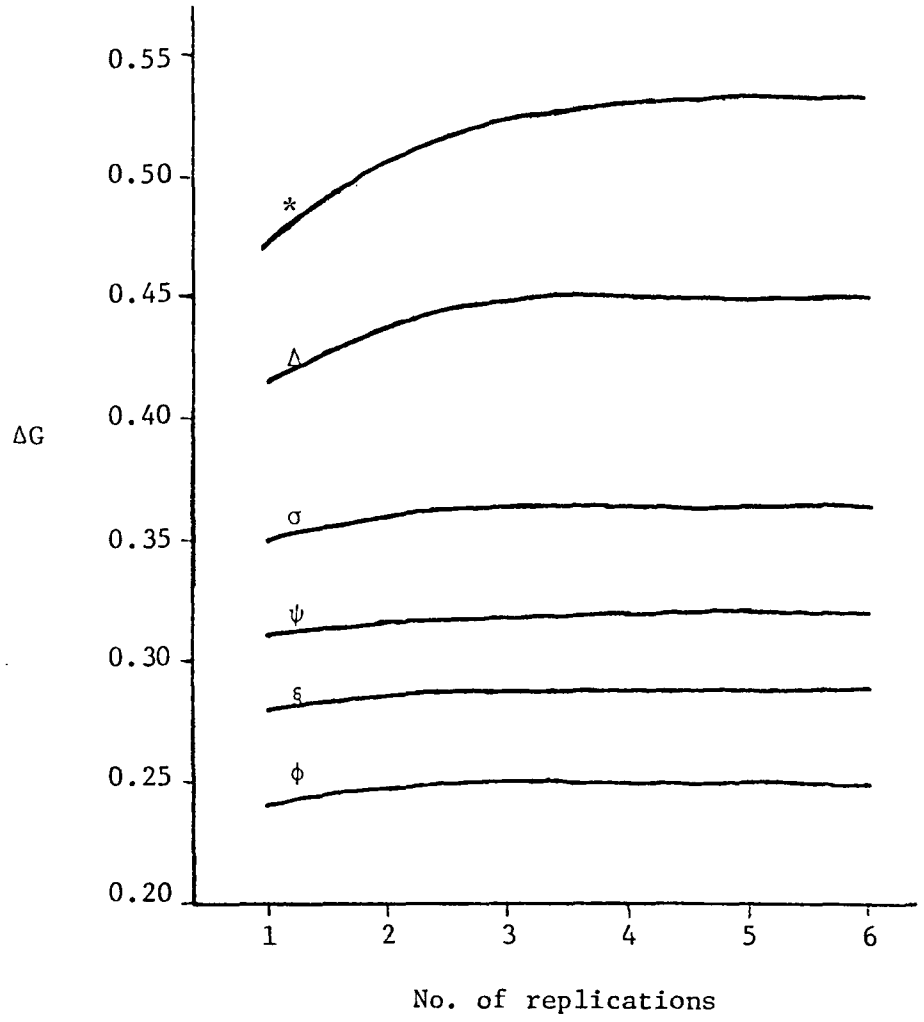

Figure 5. Relationship of expected gain per cycle to numbers of environments and replications per environment for palmitic acid

1 Environment
1 Replication
1 Plant
1-6 Seed samples
1-6 Injections

* = 1 Seed sample
Δ = 2 Seed samples
σ = 3, 4 Seed samples
ψ = 5, 6 Seed samples

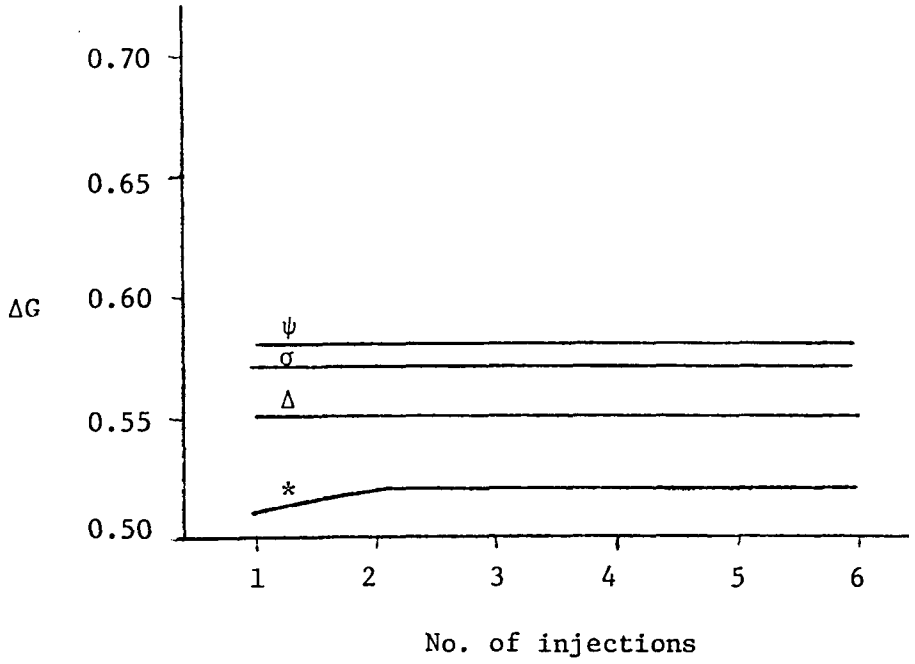

Figure 6. Relationship of expected gain to numbers of seed samples and injections per seed sample for stearic acid

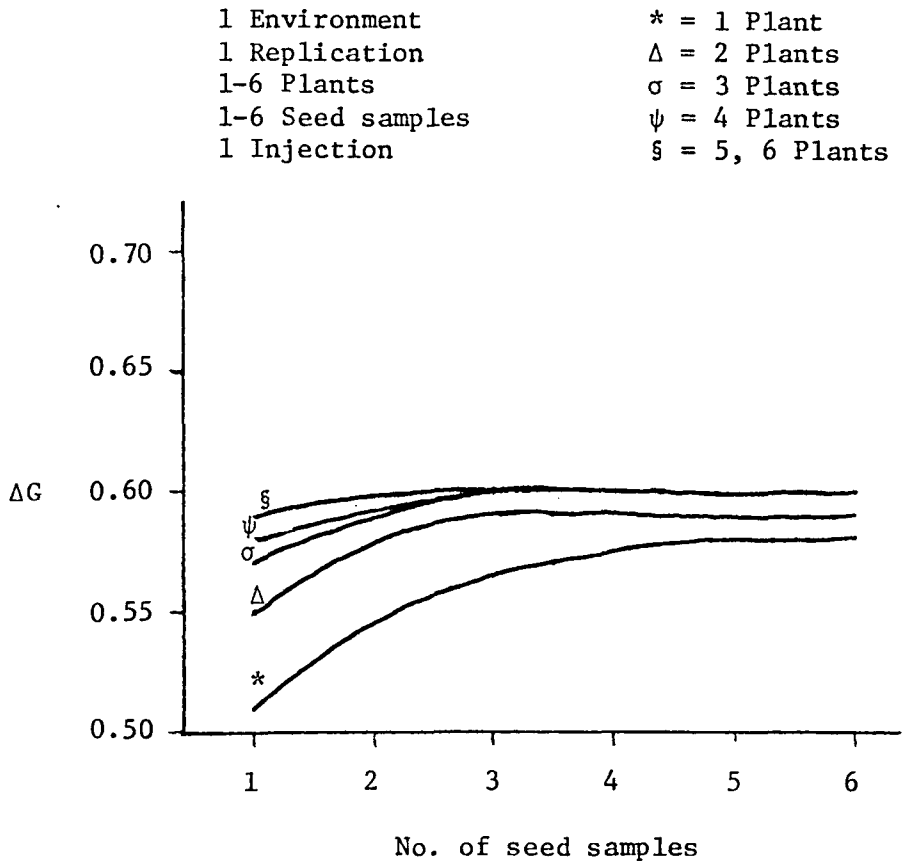

Figure 7. Relationship of expected gain to numbers of plants and seed samples per plant for stearic acid

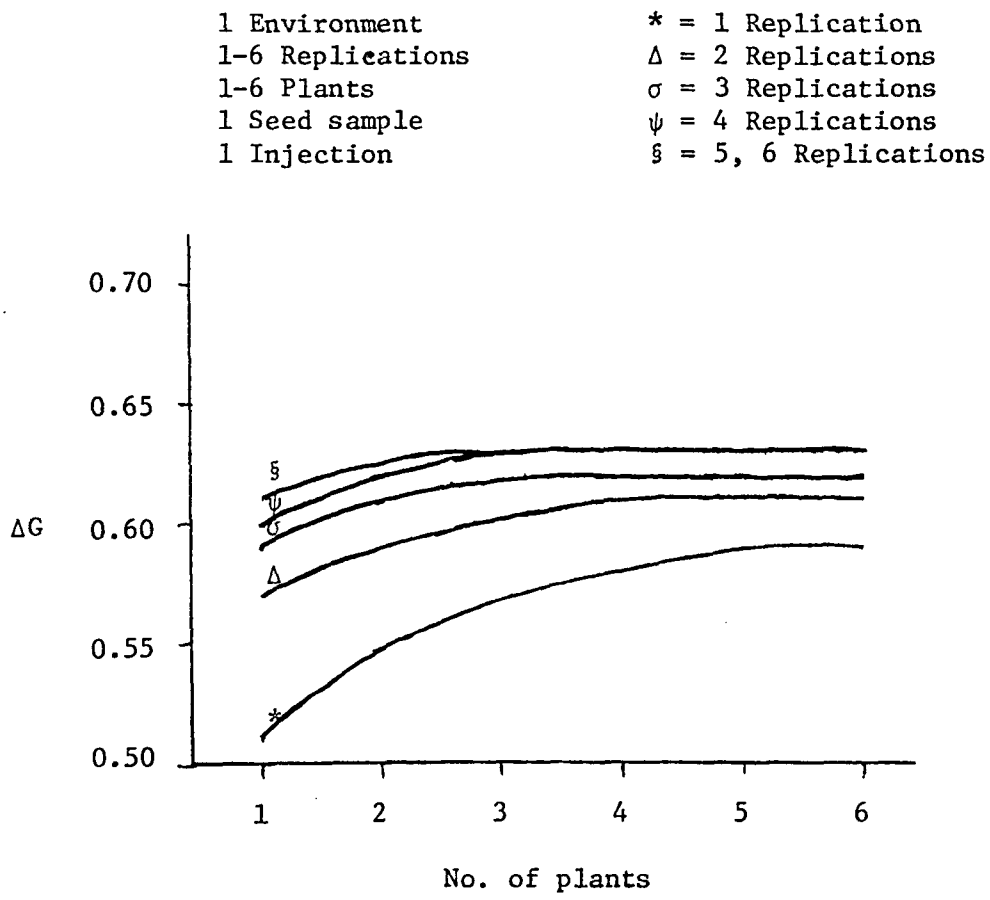

Figure 8. Relationship of expected gain to numbers of replications and plants per replication for stearic acid

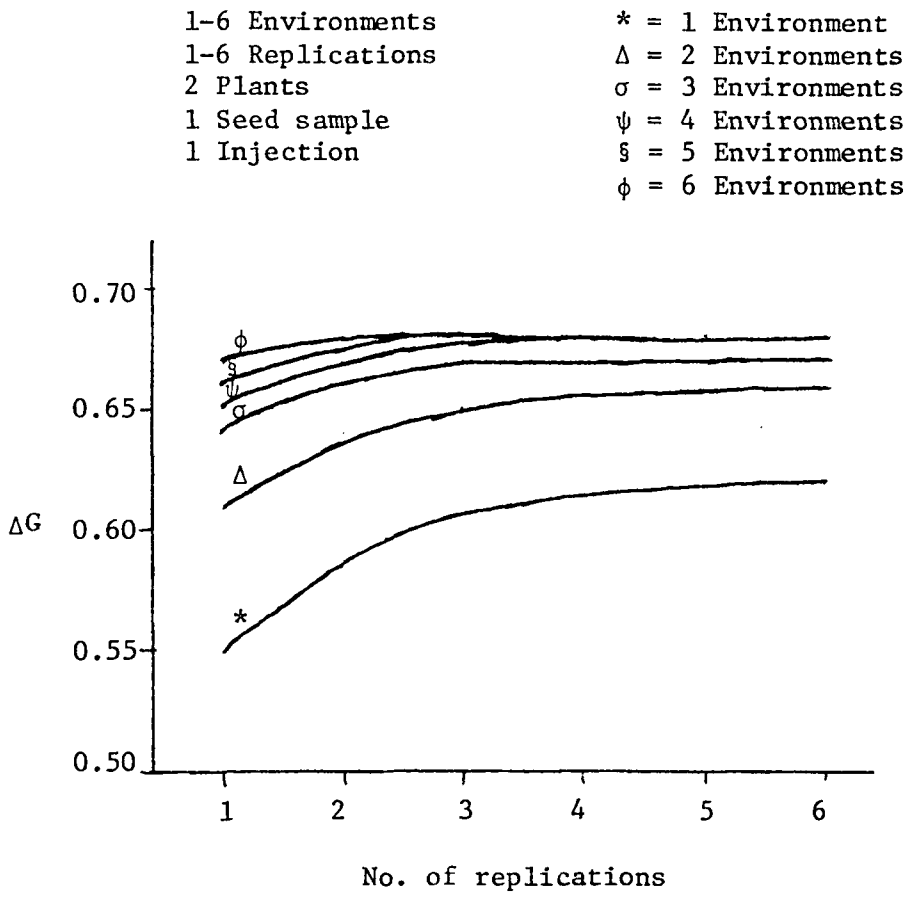

Figure 9. Relationship of expected gain to numbers of environments and replications per environments for stearic acid

1-6 Environments
1-6 Replications
2 Plants
1 Seed sample
1 Injection

* = 1.3 yr/cycle 1 Environment
Δ = 1.6 yr/cycle 2 Environments
σ = 2.0 yr/cycle 3 Environments
ψ = 2.3 yr/cycle 4 Environments
§ = 2.6 yr/cycle 5 Environments
ϕ = 3.0 yr/cycle 6 Environments

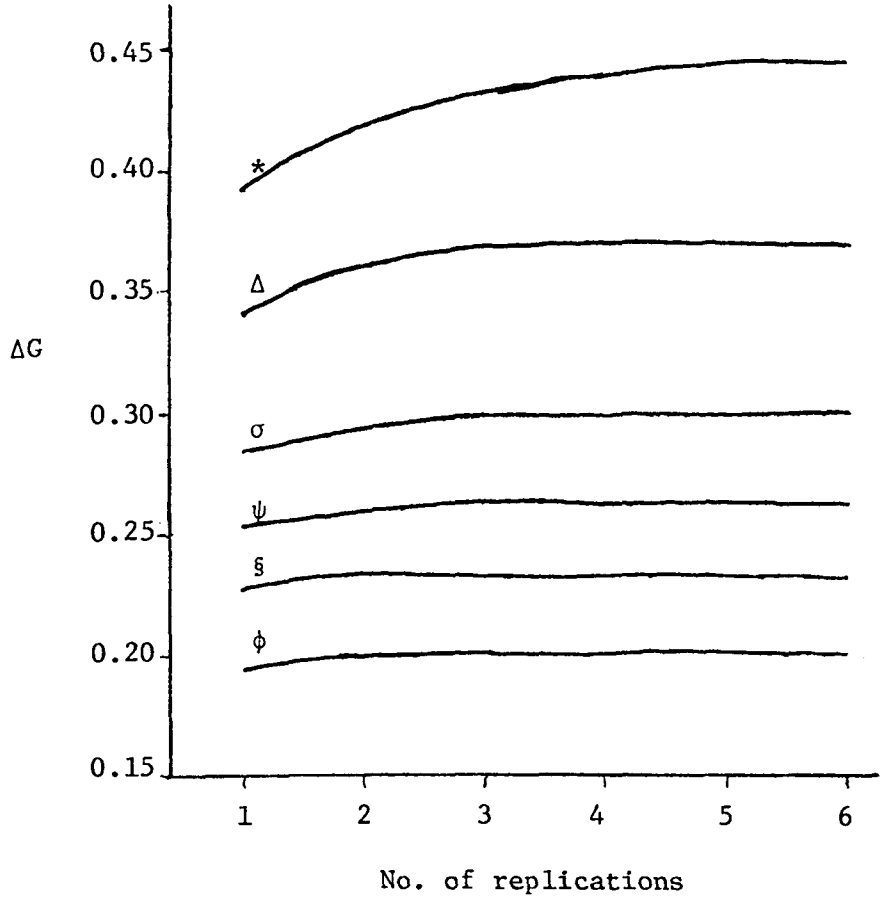

Figure 10. Relationship of expected gain per cycle to numbers of environments and replications per environments for stearic acid

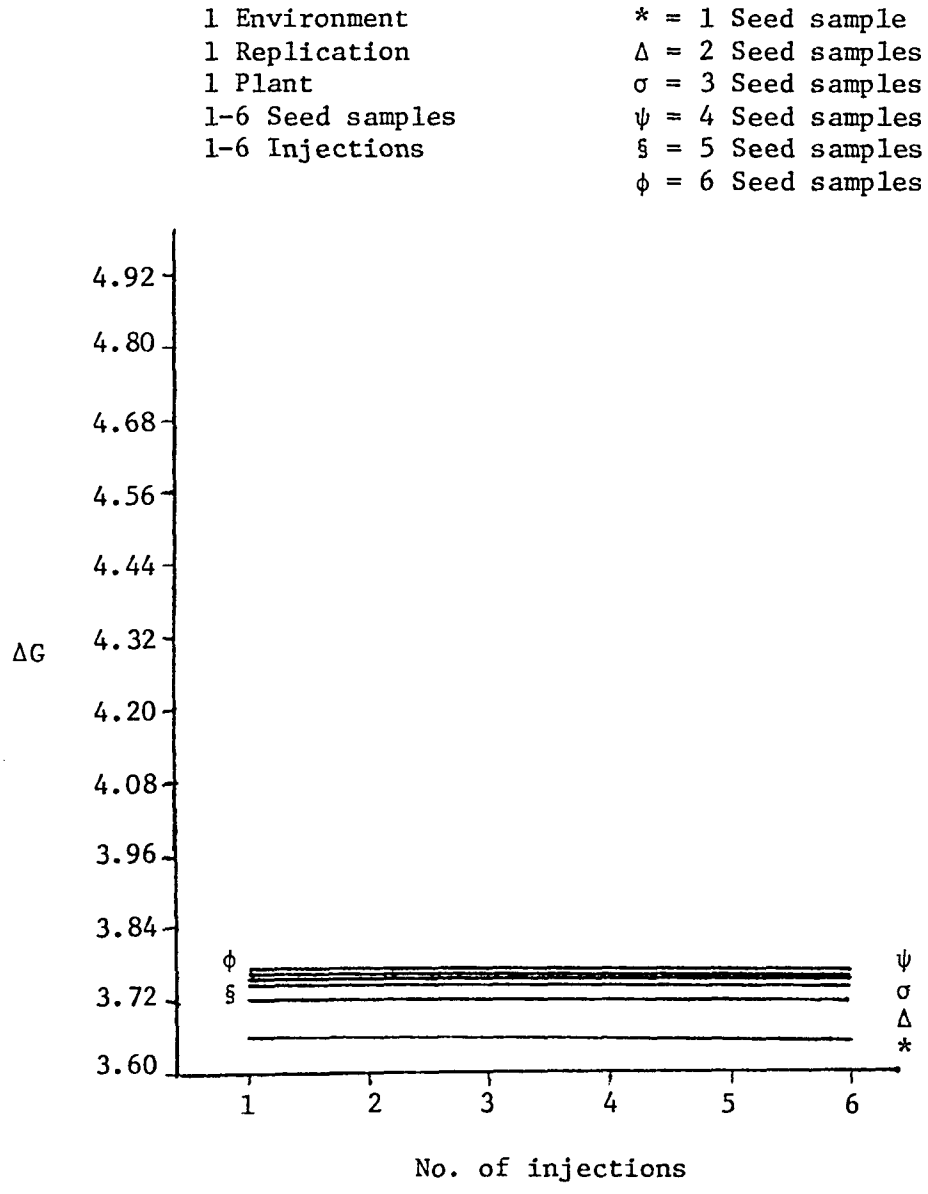

Figure 11. Relationship of expected gain to numbers of seed samples and injections per seed samples for oleic acid

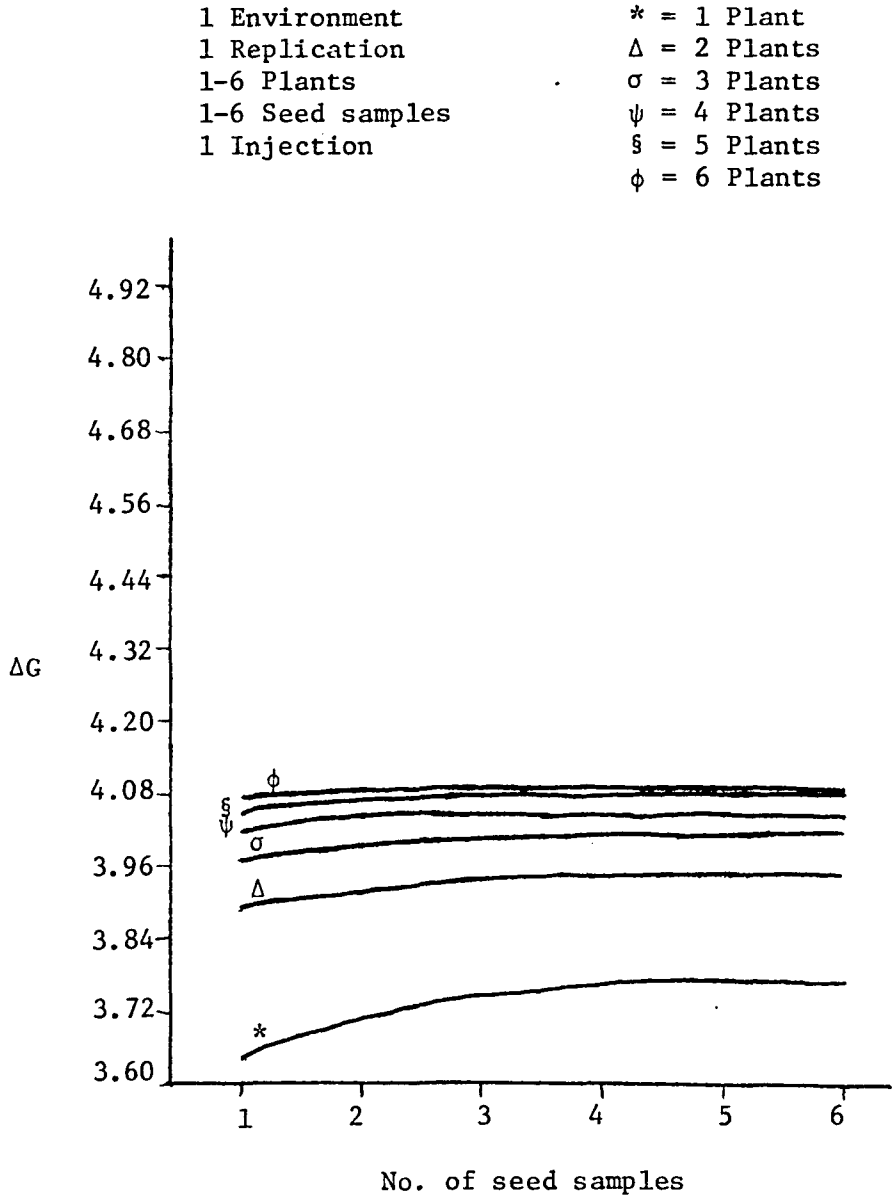

Figure 12. Relationship of expected gain to numbers of plants and seed samples per plant for oleic acid

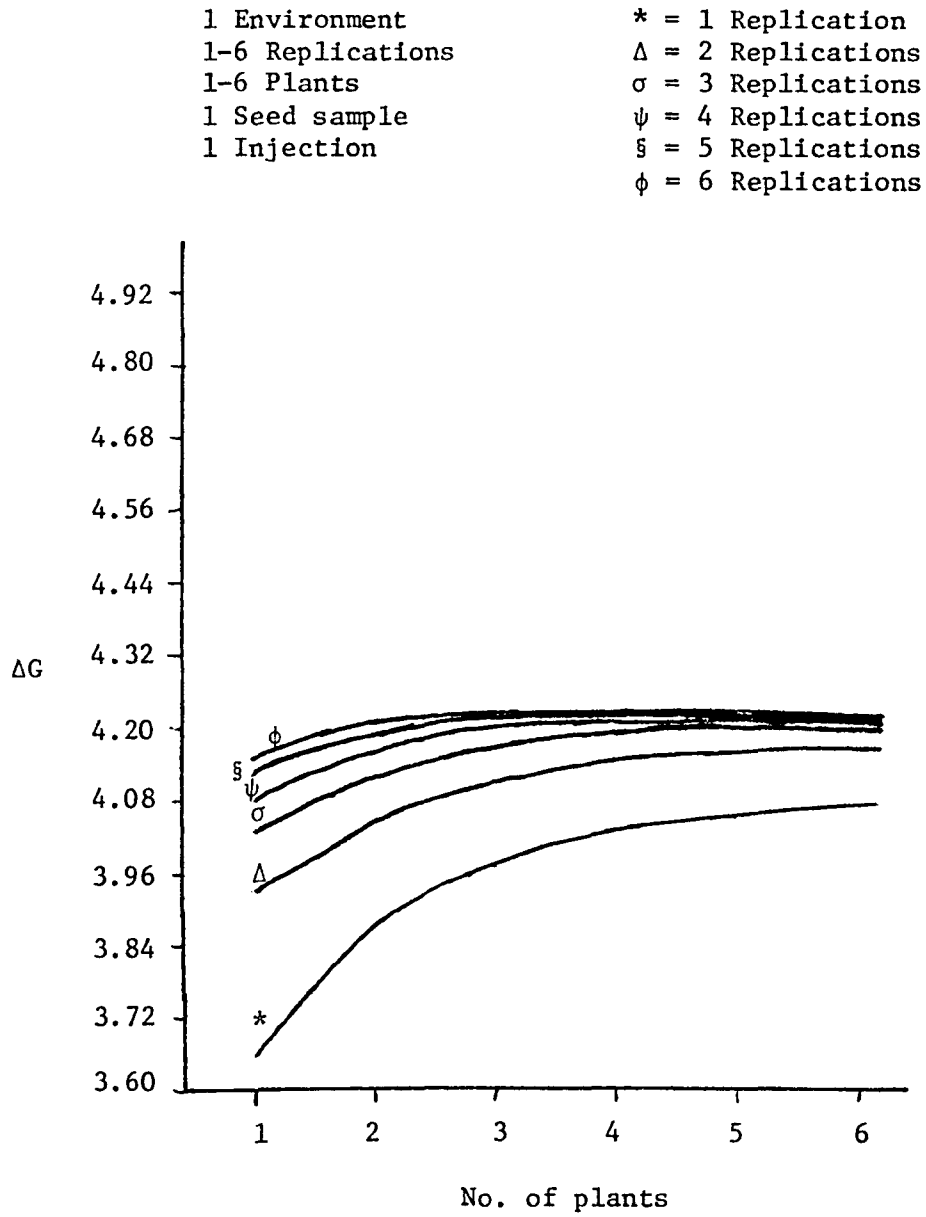

Figure 13. Relationship of expected gain to numbers of replications and plants per replication for oleic acid

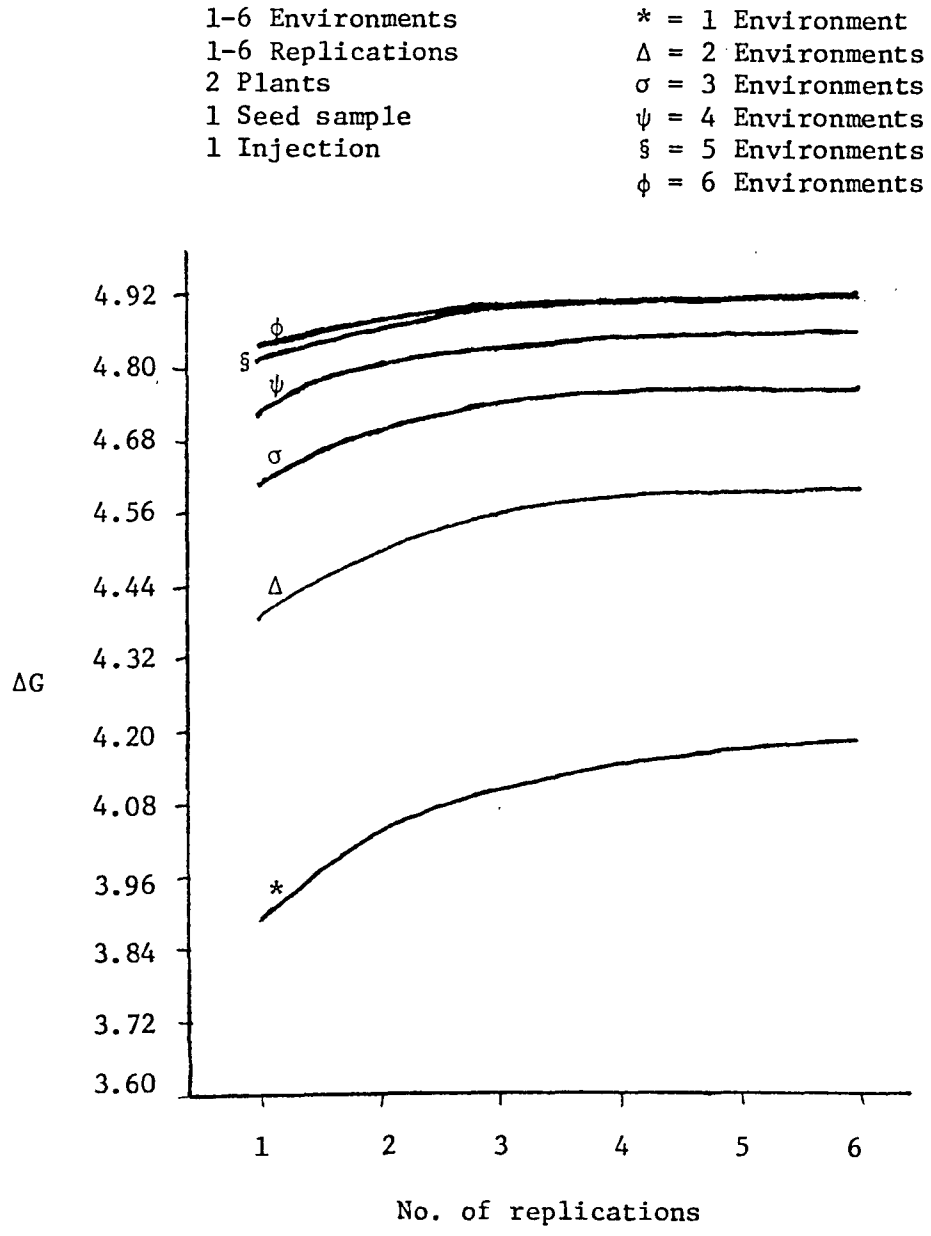

Figure 14. Relationship of expected gain to numbers of environments and replications per environment for oleic acid

1-6 Environments
1-6 Replications
2 Plants
1 Seed sample
1 Injection

* = 1.3 yr/cycle 1 Environment
Δ = 1.6 yr/cycle 2 Environments
σ = 2.0 yr/cycle 3 Environments
ψ = 2.3 yr/cycle 4 Environments
§ = 2.6 yr/cycle 5 Environments
ϕ = 3.0 yr/cycle 6 Environments

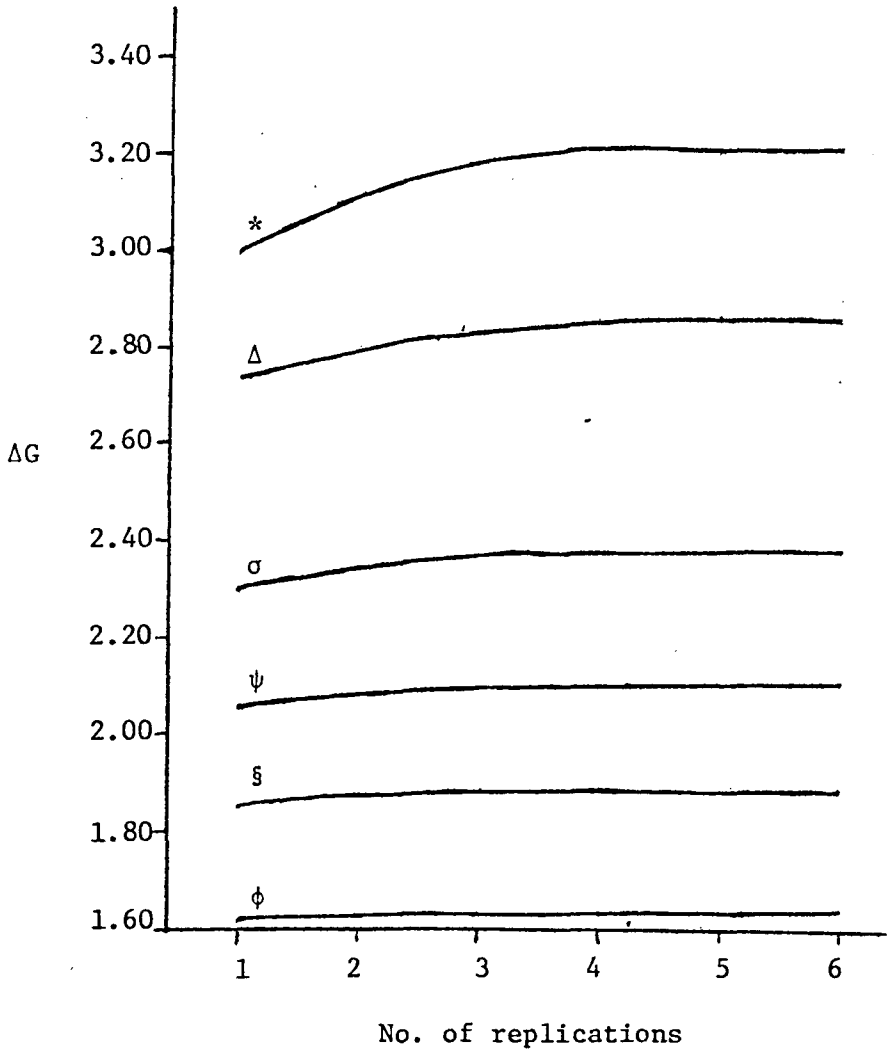

Figure 15. Relationship of expected gain per cycle to numbers of environments and replications per environment for oleic acid

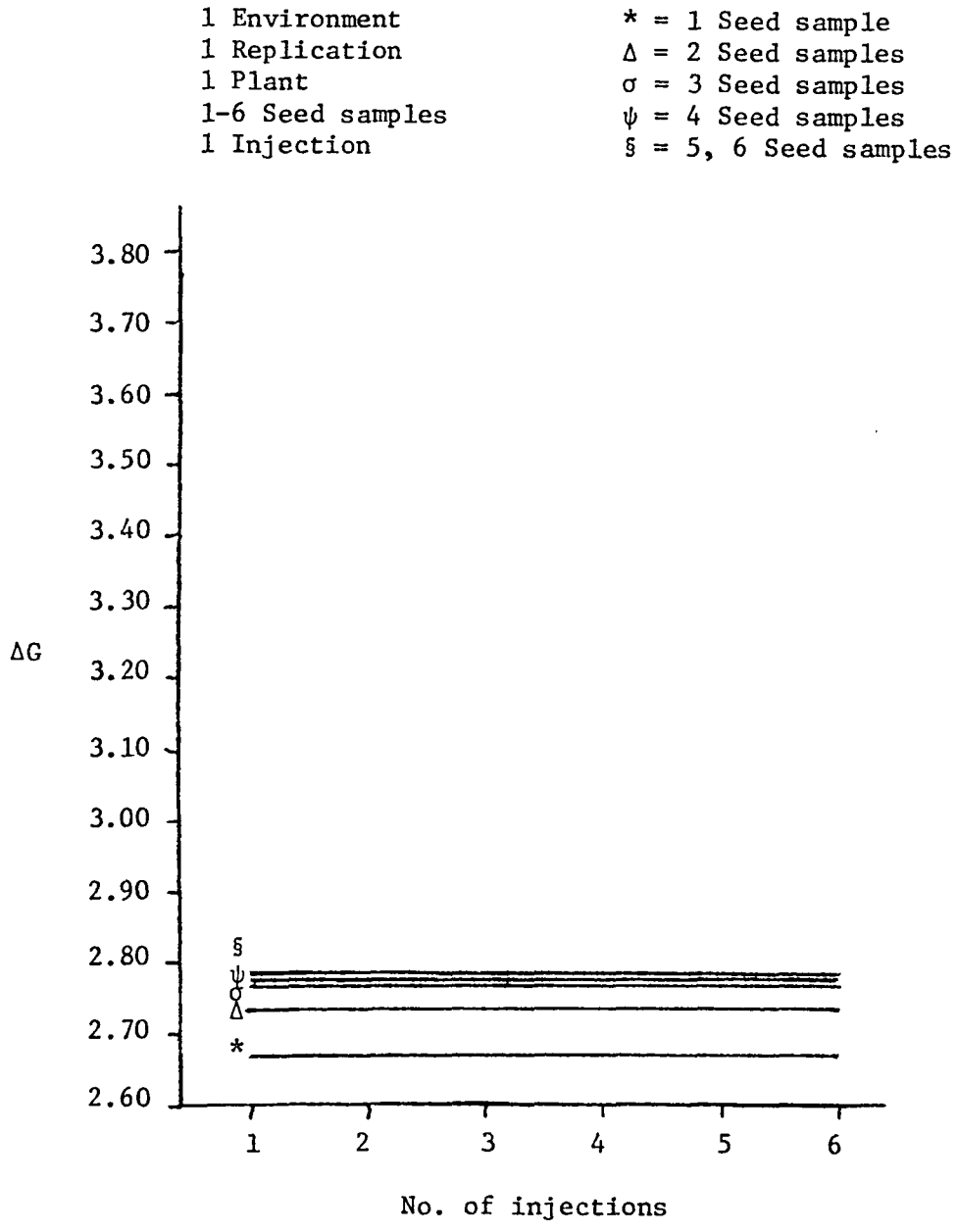

Figure 16. Relationship of expected gain to numbers of seed samples and injections per seed sample for linoleic acid

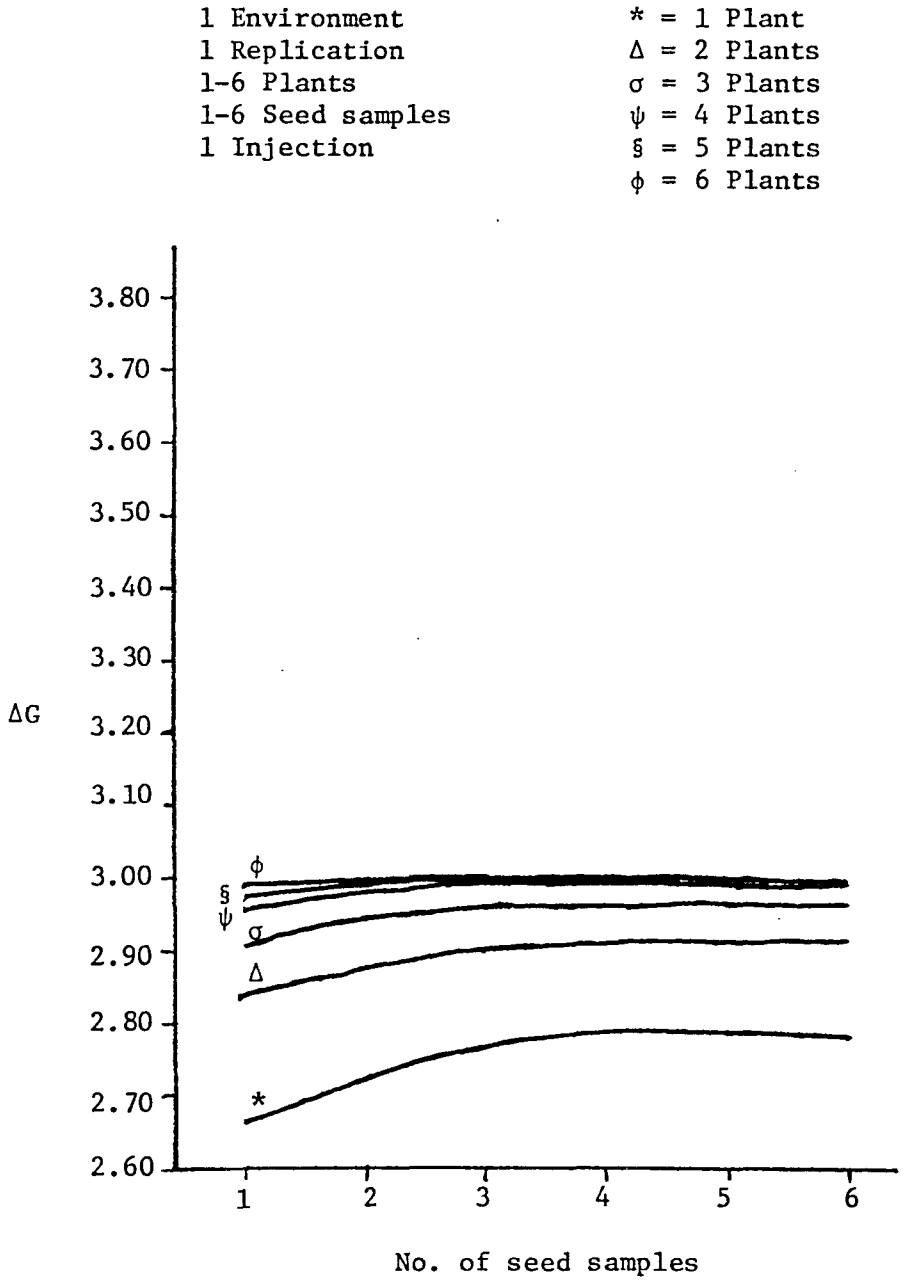

Figure 17. Relationship of expected gain to numbers of plants and seed samples per plant for linoleic acid

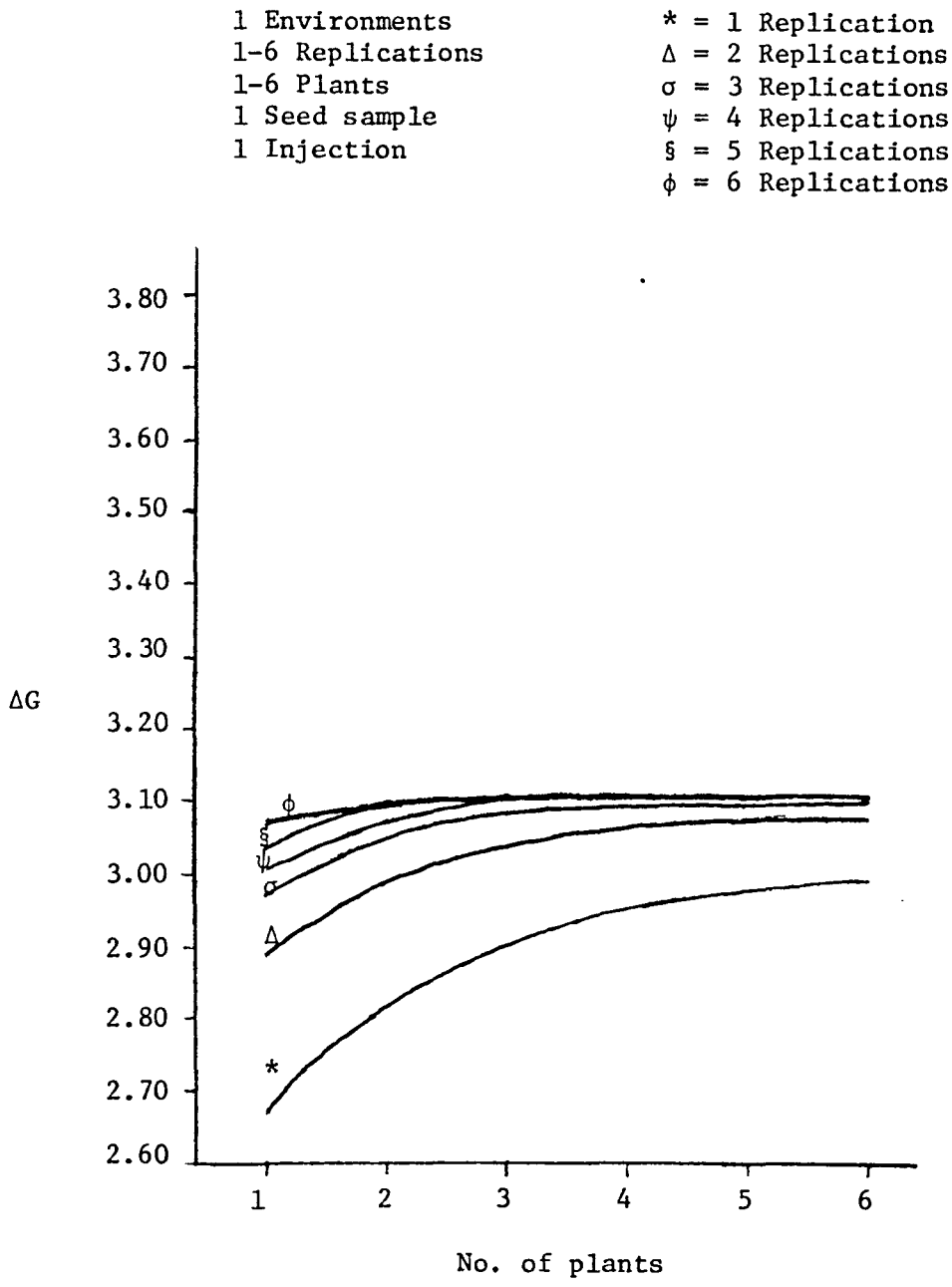

Figure 18. Relationship of expected gain to numbers of replications and plants per replication for linoleic acid

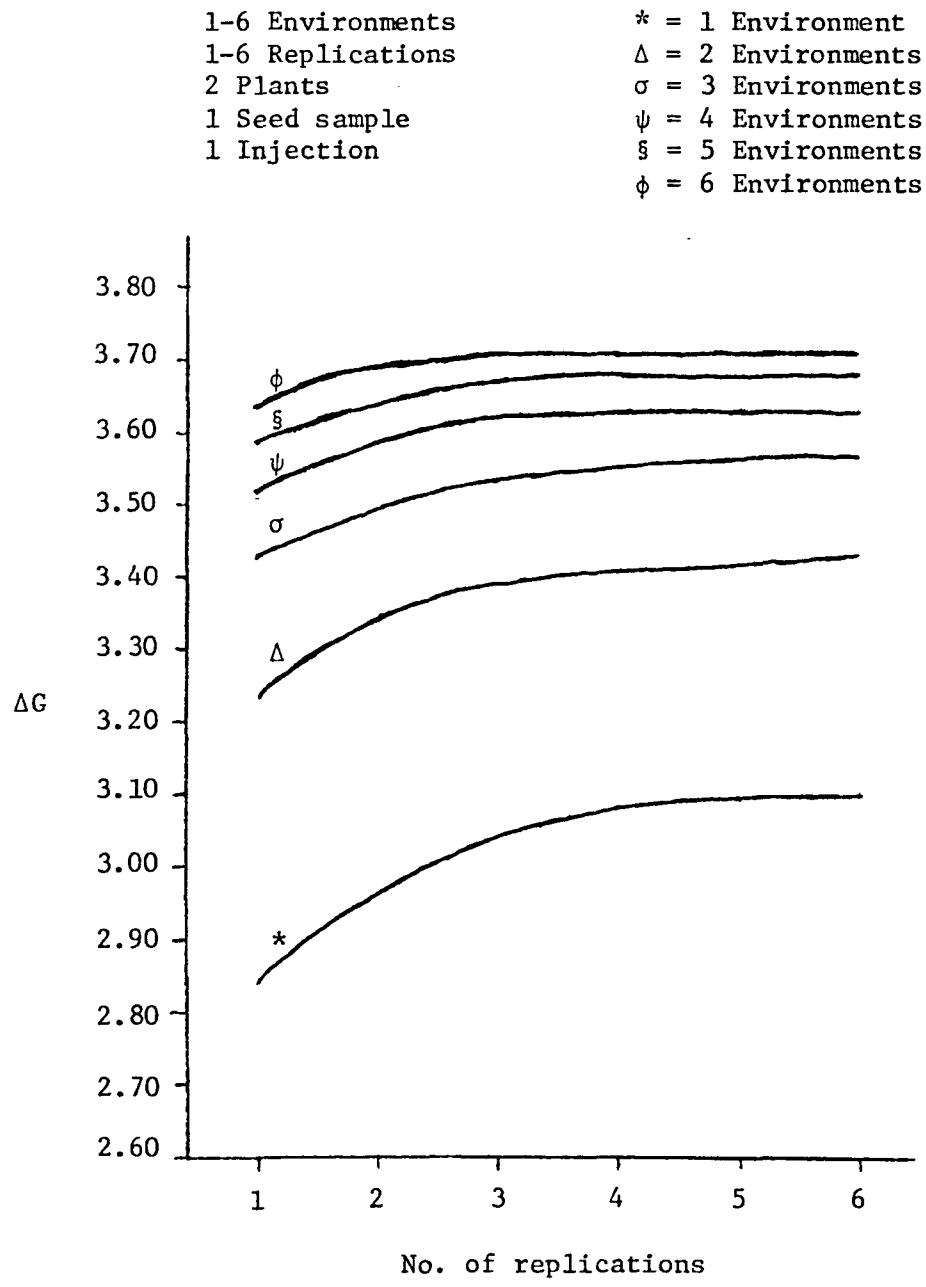

Figure 19. Relationship of expected gain to numbers of environments and replications per environment for linoleic acid

1-6 Environments
1-6 Replications
2 Plants
1 Seed sample
1 Injection

* = 1.3 yr/cycle 1 Environment
Δ = 1.6 yr/cycle 2 Environments
σ = 2.0 yr/cycle 3 Environments
ψ = 2.3 yr/cycle 4 Environments
§ = 2.6 yr/cycle 5 Environments
ϕ = 3.0 yr/cycle 6 Environments

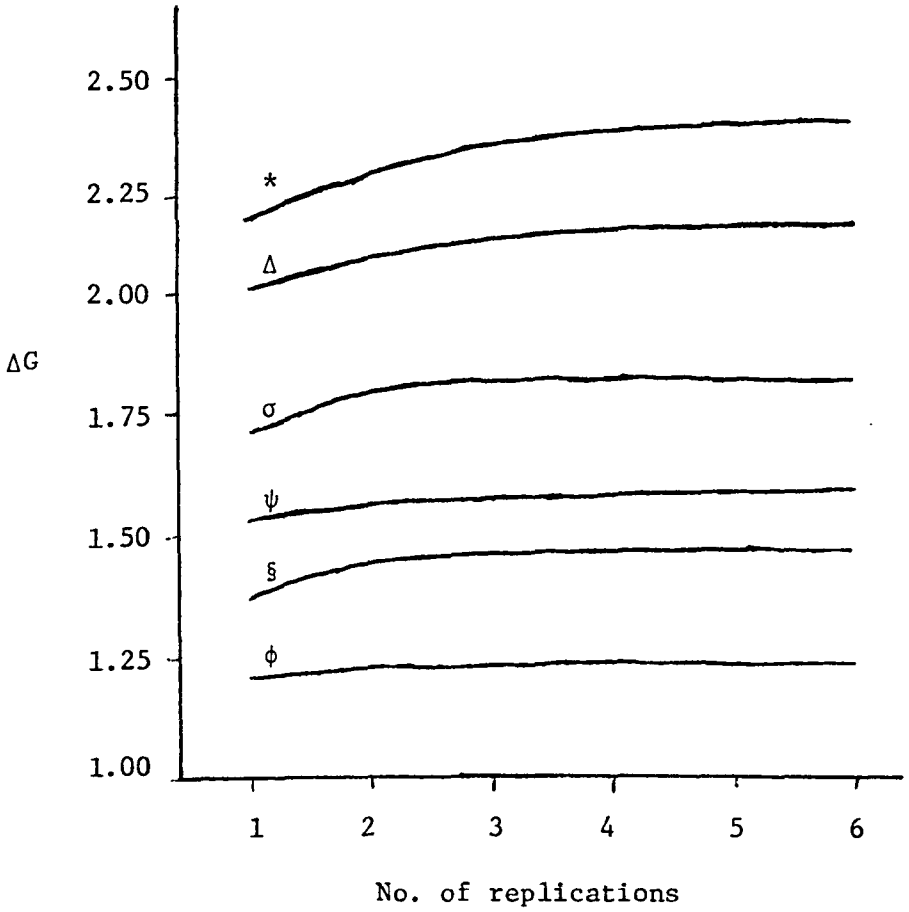

Figure 20. Relationship of expected gain per cycle to numbers of environments and replications per environment for linoleic acid

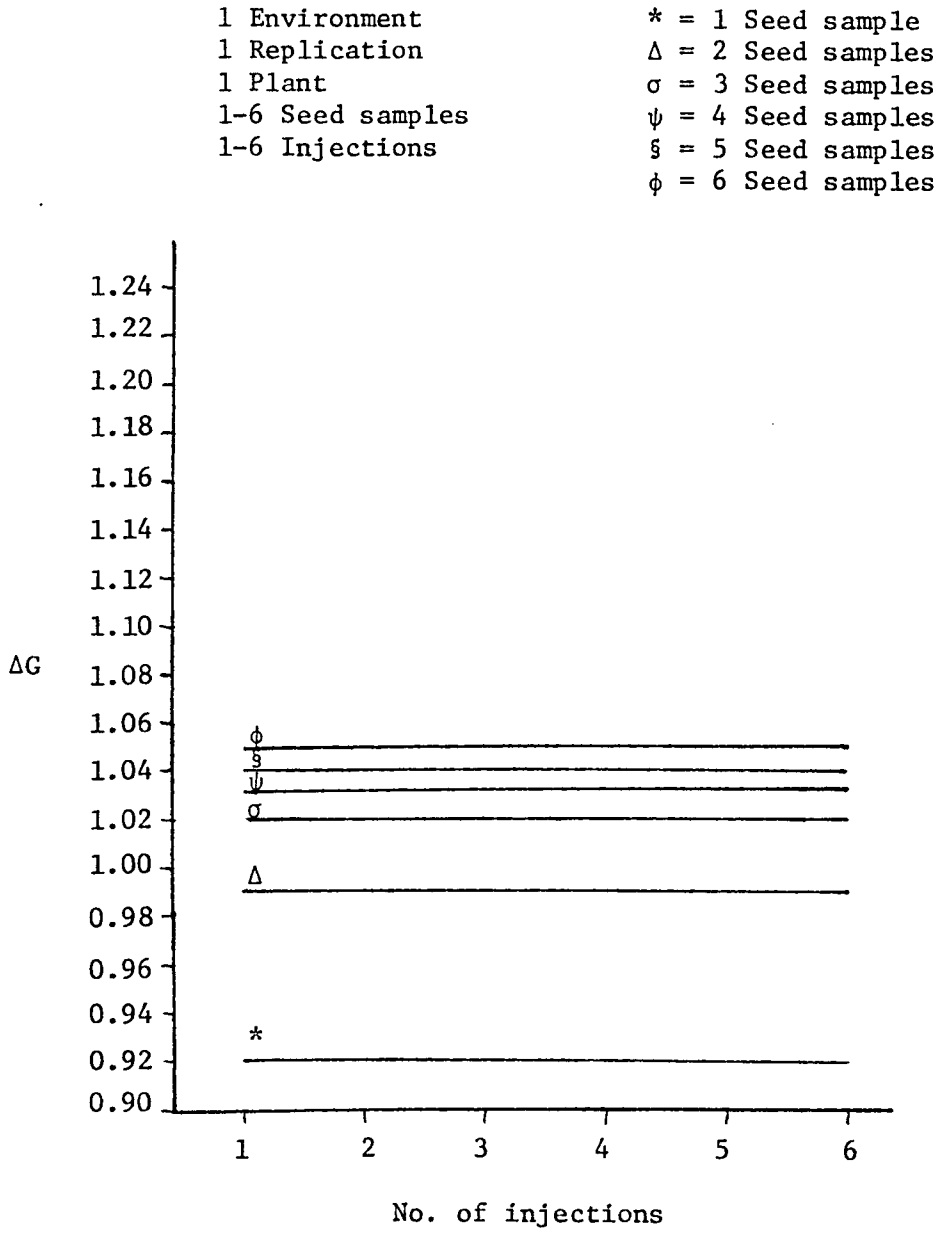

Figure 21. Relationship of expected gain to numbers of seed samples and injections per seed sample for linolenic acid

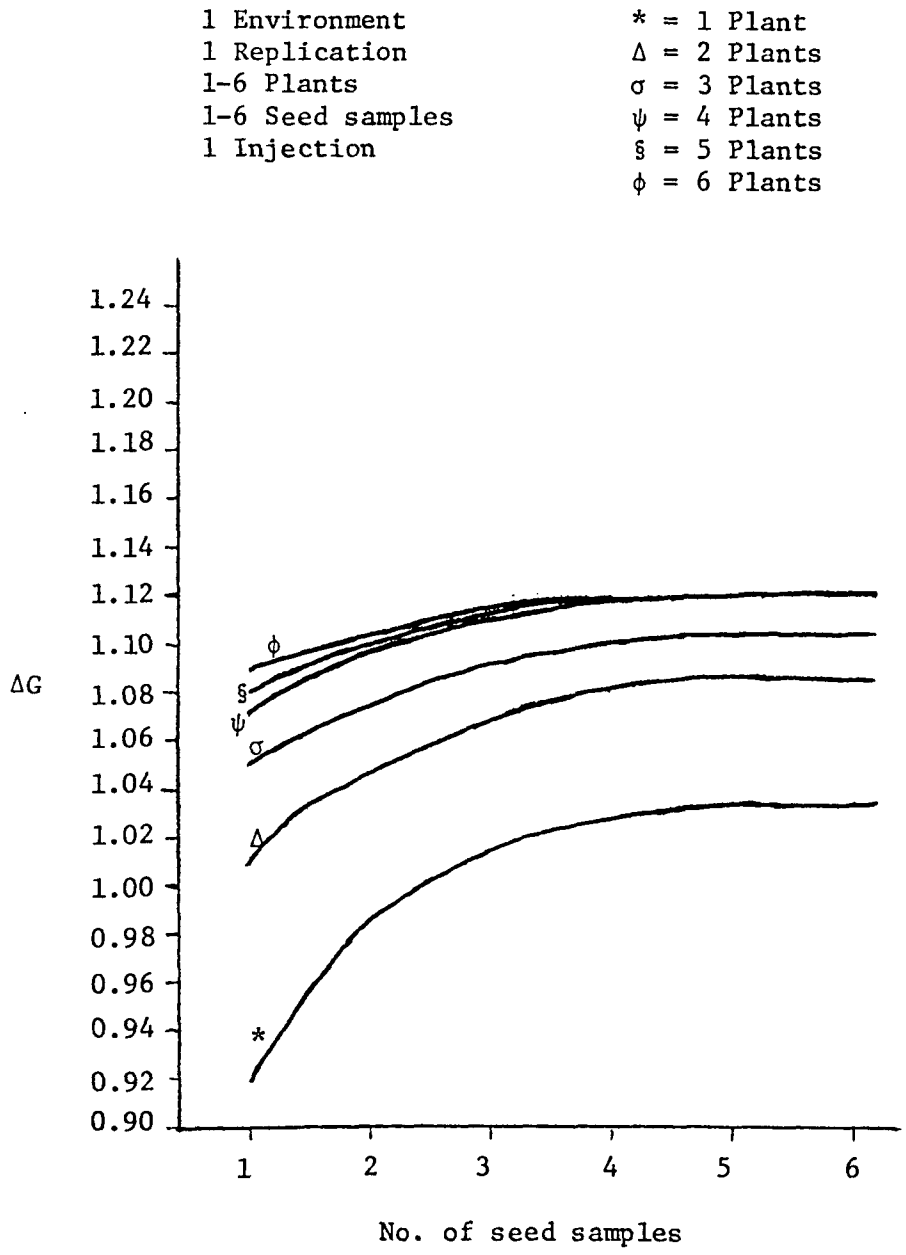

Figure 22. Relationship of expected gain to numbers of plants and seed samples per plant for linolenic acid

1 Environment
1-6 Replications
1-6 Plants
1 Seed sample
1 Injection

* = 1 Replication
Δ = 2 Replications
σ = 3 Replications
ψ = 4 Replications
§ = 5 Replications
ϕ = 6 Replications

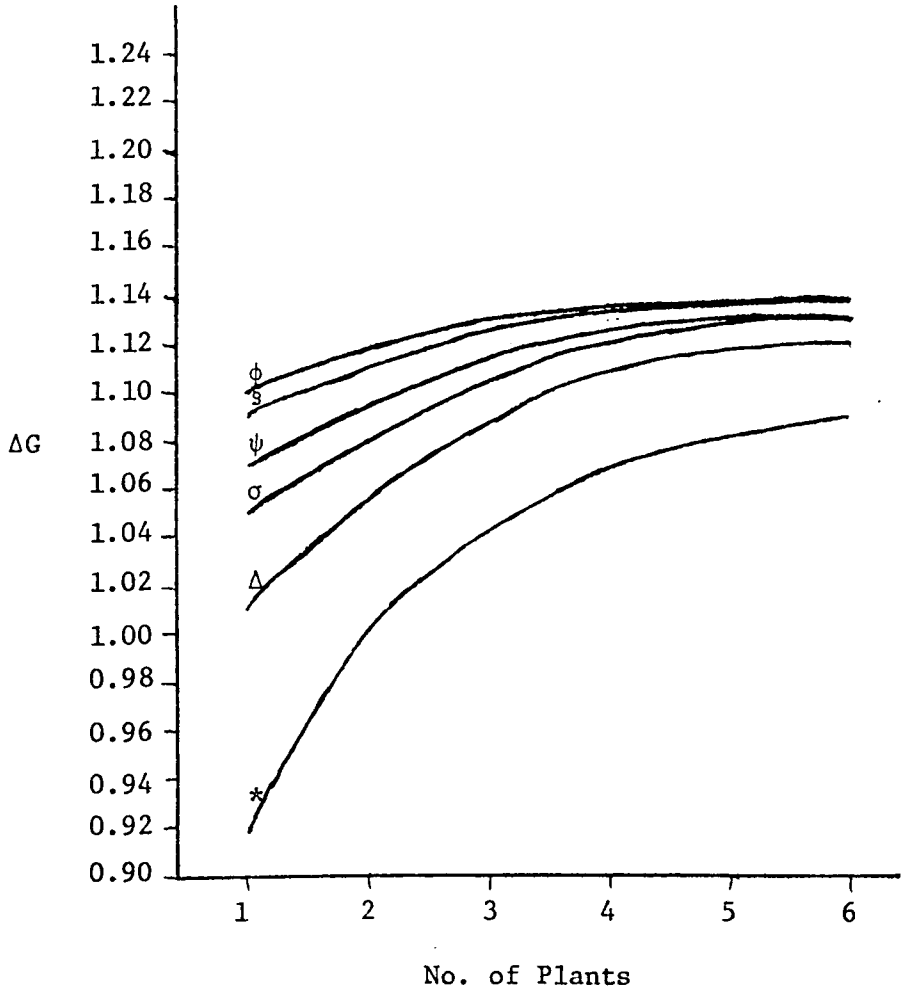

Figure 23. Relationship of expected gain to number of replications and plants per replication for linolenic acid

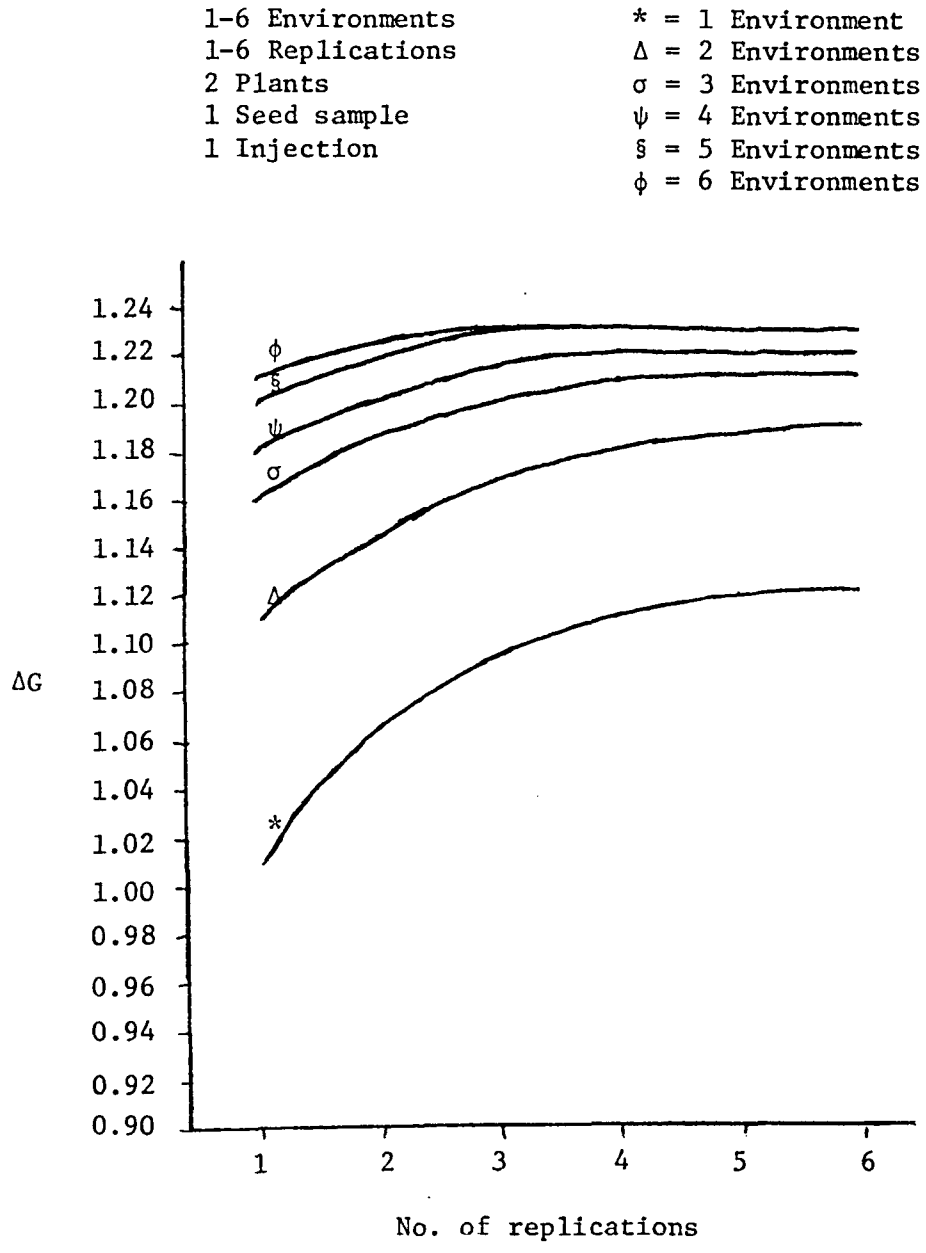

Figure 24. Relationship of expected gain to numbers of environments and replications per environment for linolenic acid

1-6 Environments
1-6 Replications
2 Plants
1 Seed sample
1 Injection

* = 1.3 yr/cycle 1 Environment
Δ = 1.6 yr/cycle 2 Environments
σ = 2.0 yr/cycle 3 Environments
ψ = 2.3 yr/cycle 4 Environments
§ = 2.6 yr/cycle 5 Environments
ϕ = 3.0 yr/cycle 6 Environments

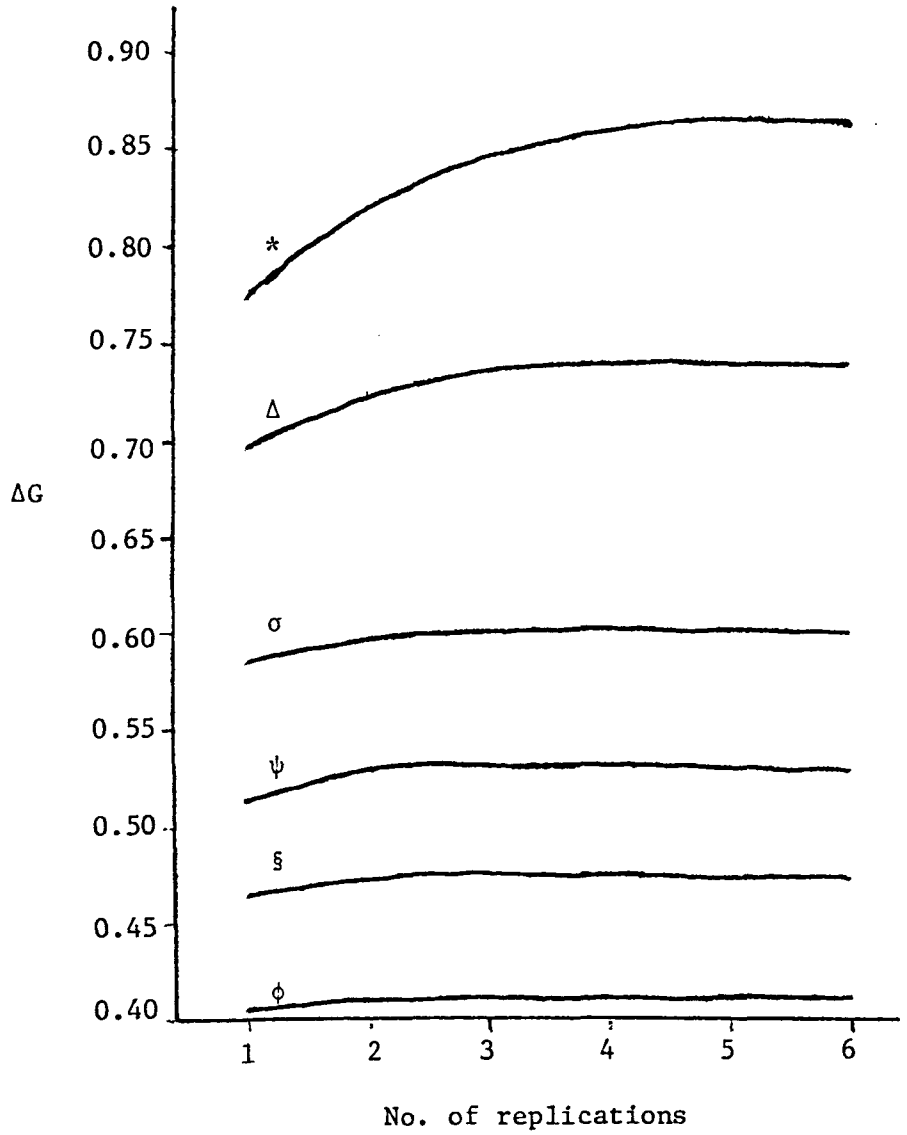

Figure 25. Relationship of expected gain per cycle to numbers of environments and replications per environment for linolenic acid

DISCUSSION

The first objective of this study was to estimate the relationship between palmitic, stearic, oleic, linoleic, and linolenic acids in soybean oil. Correlation coefficients would indicate a slight change in palmitic acid content if selection were practiced on stearic, oleic, or linolenic acids. There is a slight negative correlation with linoleic acid that could affect palmitic acid. Stearic acid exhibited a significant positive correlation with linoleic and linolenic acids that may lead to slight reductions in stearic acid in a breeding program for low linolenic acid. Significant negative correlations between oleic acid stearic, linoleic, and linolenic acids would suggest the reduction in the latter three oil components if selection for high oleic acid were practiced.

Burton et al. (1981) and Caldwell et al. (1982) reported decreases in palmitic, linoleic, and linolenic acids after selection for high oleic acid content. Howell et al. (1972) and Wilson et al. (1981) also indicated reductions in linoleic and linolenic acids after selecting for high oleic acid.

A second objective was to determine the importance of the line x environment interaction. The interaction was statistically significant. Phenotypic correlations among environments were large, and the correlation between Puerto Rico and Ames were also significant. Ranking of lines and rank correlations between Puerto Rico and Ames environments indicated that both locations were able to establish the same relative relationship among lines. Selection for fatty acid composition among

lines grown in different environments can be confounded unless parent lines or some other common entry of known performance is included at both locations. Cramer et al. (1981) found a significant genotype x environment interaction for the soybean lines they used. Caldwell et al. (1982) and Cramer et al. (1981) found significant differences in magnitude induced by environments, but not in relative ranking among lines.

The third objective was to evaluate different resource allocations for an effective selection program. Using more than one injection per seed sample provided little or no increase in genetic gain. The variance associated with injections was small and when it was used to test seed samples within plants, it produced significant results for each fatty acid. The increased genetic gain for each fatty acid using multiple seed samples per plant is about the same as testing two or more plants per plot. Collins and Cartter (1956) and Cramer et al. (1981) reported variation among pods on the same plant which could be reduced by adequately sampling each portion of the plant. By limiting seed samples to one per plant, more individual plants per line could be sampled, an important consideration in a segregating population.

Based on the assumptions made and the variance terms obtained, the allocation of resources for oleic and linoleic acids suggested is two environments, one replication, two plants per replication, one seed sample per plant, and one injection per seed sample. Recommended resources for palmitic, stearic, and linolenic acids are two

environments, two replications, two plants per replication, one seed sample per plant, and one injection per seed sample.

Another alternative would be to reduce the variance within plots. It would be less expensive to grow six or twelve plants per plot than using multiple environments. As previous stated, the gains from using multiple plants per plot with one seed sample per plant was as effective as using one plant with multiple seed samples. One representative seed sample from six or twelve plants could give adequate gains.

Using the variance components for linolenic acid (Table 7) in the genetic gain equation, estimates were obtained for six or twelve plants per plot. The phenotypic variance was computed for these estimates using the equation:

$$\sigma^2_{ph} = \frac{\frac{\sigma^2_W}{n} + \sigma^2}{r} + \frac{\sigma^2_{GE}}{E} + \sigma^2_G$$

where

σ^2_{PH} = phenotypic variance

σ^2_W = within plot variance

σ^2 = plot to plot variance

σ^2_{GE} = genotype x environment variance

σ^2_G = genotypic variance

n = number of plants per plot; n = 6, 12

r = number of replications per environment; r = 1 to 2

E = number of environments; E = 1 to 2

The estimates (Table 28) indicate that using plots of twelve plants with one representative seed sample would be as effective as using two environments and two plants per plot.

Another consideration is the number of samples each method can handle. For example, if each environment were limited to 5,000 seed samples, the number of lines that can be handled using six or twelve plants is twice that of the other methods. Using two environments to evaluate a group of lines would not allow evaluation of as many lines each year if only one environment were used.

Table 28. Estimated genetic gains for two alternative plot arrangements

No. of environments		6 plants		12 plants		2 plants w/individual seed samples	
		ΔG	No. of lines handled	ΔG	No. of lines handled	ΔG	No. of lines handled
1	1 rep	1.03	5,000	1.08	5,000	1.01	2,500
	2 reps	1.09	2,500	1.12	2,500	1.07	1,250
2	1 rep	1.13	5,000	1.13	5,000	1.11	2,500
	2 reps	1.15	2,500	1.16	2,500	1.16	1,250

CONCLUSIONS

Palmitic acid is not highly correlated with stearic, oleic, or linolenic acid, but it is negatively correlated with linoleic acid. Oleic acid is negatively correlated with stearic, linoleic, and linolenic acids. Stearic, linoleic, and linolenic acids are positively correlated with one another. Selection for low linolenic acid would probably result in an increase in oleic acid and a decrease in linoleic acid.

The use of Puerto Rico as a selection environment is not restricted by a genotype x environment interaction. Selection among lines adapted to Iowa for low linolenic acid would probably be successful in Puerto Rico without supplemental lighting.

An effective allocation of resources for selection for low linolenic acid would be to use one environment, two replications per environment, six to twelve plants per replication with one representative seed sample of twenty or more seeds and one injection per seed sample.

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