1% with the absorbance values obtained at 450 mμ by use of magnesium oxide–Celite columns.

Concerning the selection of wave length for absorbance measurement, other wave lengths and appropriate coefficients may be used to estimate the carotene with greater stoichiometric accuracy since the cis isomers have lower coefficients than the all-trans isomer at 450 mμ. Results calculated at this wave length as all-*trans*-β-carotene are low. Biological activities of the cis isomers relative to absorbancies are even lower than for the all-trans form (*3*, *5*). Hence, calculation as suggested at 450 mμ may reflect the biological potency with less error than calculations at other wave lengths which indicate higher carotene content.

Literature Cited

(1) Assoc. Offic. Agr. Chemists," Methods of Analysis," 7th ed., 1950.
(2) Bickoff, E. M., White, L. M., Bevenue, A., Williams, K. T., *J. Assoc. Offic. Agr. Chemists* **31**, 634 (1948).
(3) Deuel, H. J., Jr., Sumner, E., Johnston, C., Polgar, A., Zechmeister, L., *Arch. Biochem.* **6,** 157 (1945).
(4) Johnson, R. M., Baumann, C. A., *J. Biol. Chem.* **169,** 85 (1947).
(5) Kemmerer, A. R., Fraps, G. S., *Ibid.*, **161,** 305 (1945).
(6) Moore, L. A., and Ely, Ray, *Ind. Eng. Chem., Anal. Ed.* **13,** 600 (1941).
(7) Pepkowitz, L. P., *J. Biol. Chem.* **155,** 219 (1944).
(8) Wiseman, H. G., Stone, S. S., Savage, H. L., Moore, L. A., *Anal. Chem.* **24,** 681 (1952).
(9) Zscheile, F. P., Whitmore, R. A., *Ibid.*, **19,** 170 (1947).

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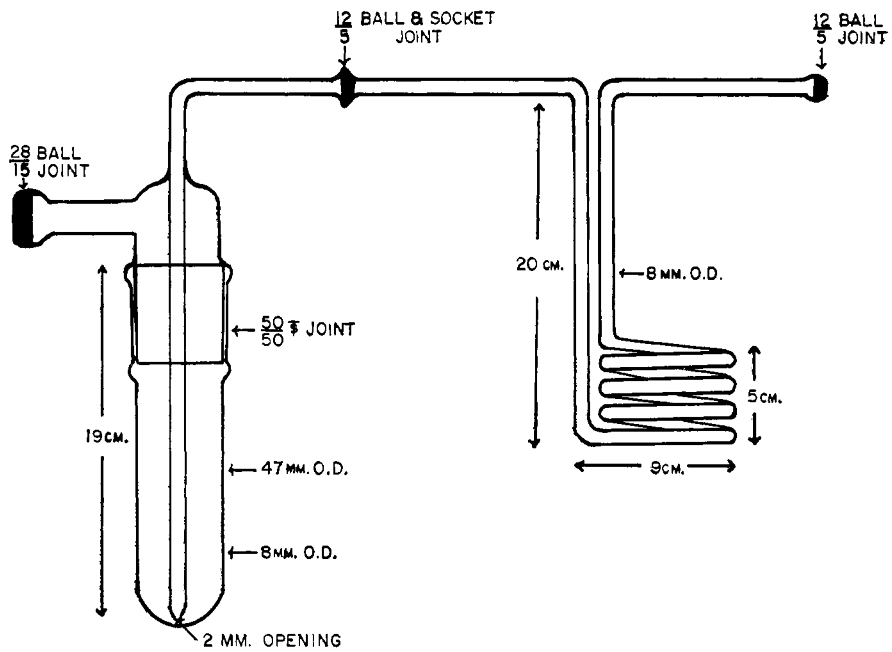
Determination of Antioxidants in Edible Fats—Correction

Figure 4 was omitted from the article on Determination of Antioxidants in Edible Fats [Constance Anglin, J. H. Mahon, and R. A. Chapman, J. AGR. FOOD CHEM. **4,** 1018 (1956)]. It is printed below.

▼ Figure 4. Distilling flask and superheater coil



NUTRITIVE VALUE OF BEANS

Nutrients in Central American Beans

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Because of the special nutritional importance of beans in Central America and Panama, the factors influencing protein, methionine, lysine, and tryptophan content of 25 varieties were studied. Niacin, thiamine, and riboflavin were also determined. Over-all differences in nitrogen and tryptophan content among varieties and between localities were highly significant. The fertility of the land significantly alters the yield and riboflavin content of the kidney bean, but the content of nitrogen, methionine, lysine, tryptophan, niacin, and thiamine is not detectably affected by fertility differences.

THE KIDNEY BEAN, *Phaseolus vulgaris*, is second only to maize in importance in the diets of the people throughout most of Central America. It is not unusual for these beans to account for 20 to 30% of the protein in the diet (*3*, *5*, *20*). Under these circumstances the quality of their protein becomes of crucial importance. However, data on the variations in nutritive value of the kidney bean, especially in essential amino acids, are surprisingly limited. The most complete report is that of Jaffé (*16*), who described the chemical composition, digestibility, protein efficiency, and limiting essential amino acids in a group of bean samples from Venezuela.

Large differences in thamine and riboflavin content of beans grown at different locations have been reported by Eheart and associates (*4*). Guyer, Kramer, and Ide (*10*) observed that as harvest was delayed the yield increased but the content of ascorbic acid and of yellow pigment decreased. The relation between environment and yield was studied by Post (*18*), who concluded that weather has a profound influence. Similarly, Casseres and Thompson (*2*) on the basis of a standard test of six commercial varieties of snap bean found that winter yields were significantly greater than summer yields.

Because of the special nutritional importance of beans in Central America and Panama, a study was undertaken of the factors influencing protein, methionine, lysine, and tryptophan content of 25 varieties grown in two localities in Guatemala for both the early and late planting of 1951. Determinations were also made of niacin, thiamine, and riboflavin. As improved varieties can be introduced only on the basis of better agronomic characteristics, the content of these nutrients in the various samples was in each case related to yield.

Plan of Experiment

The known agronomic characteristics of the 25 varieties of beans selected for the trial are summarized in Table I. They were planted in 1951 at two locations (Alameda and Bárcenas) during May 15 to 17 and again on September 5 and 6. Although four completely randomized blocks were used at each location, one of these in each planting was damaged by animals, so that the corresponding yield data could not be used. In each replication the varieties were assigned at random to plots consisting of two 10-foot rows $2^1/_2$ feet apart. Thirty-five seeds were spaced approximately evenly over each length of the 10 feet and covered with about 2 inches of soil. The blocks of 25 plots were separated from each other by an alley of 3 feet.

A 5-foot-wide border row, separated from the experimental area by an alley of 3 feet, was planted on all four sides of the experimental area. No thinning and only one or two weedings were done. The early planting was harvested between August 5 and 20 and the late planting was harvested between November 22 and December 12. Varieties 5089 and 5091 were not included in the early planting.

The seed samples for biochemical analyses were obtained from the late crop and thoroughly mixed before a sample of 50 grams was drawn. Four samples were taken for each variety from the planting in Bárcenas, one from each of the four replicates.

Chemical Methods

The determinations of moisture and the digestions of nitrogen were done by the Association of Official Agricultural Chemists official method (*1*). The distillations and titrations were completed as recommended by Hamilton and Simpson (*11*). Riboflavin was measured fluorometrically by the method of Hodson and Norris (*13*) and thiamine assays were carried out by the thiochrome method of Hennessey and Cerecedo (*12*). Niacin was determined by the chemical method of Friedemann and Frazier (*6*). However, the color of the reaction between nicotinic acid $CNBr_2$ and *p*-aminophenol (metol) and the color of the sample were measured with three tubes and their respective blanks according to the method of Melnick (*17*). The concentration of the color of the reaction was read with 420-mμ filter in an Evelyn colorimeter.

Tryptophan was estimated by the microbiological method of Wooley and Sebrell (*27*), with the modification that prior to the enzymatic hydrolysis the material was autoclaved at 15 pounds pressure with 75 ml. of water for 30 minutes and the assay media were prepared as suggested by Greene and Black (*8*). Methionine and lysine were determined by the microbiological methods of Horn, Brasse, and Blum (*14*, *15*). Each sample was assayed in duplicate at four concentrations and the entire procedure of hydrolysis and analysis was repeated if the results were not consistent.

Table I. Known Agronomic Characteristics of Bean Varieties Included in Guatemalan Trial of 1951

Variety No.	Origin[a]	Color of Seeds	Days to Maturity	Weight/100 Seeds, Grams	Anthracnose	Rust	Isariopsis
2226B	Sta. Cruz del Quiché	Red	85	21.3	MR	MR	MR
2228	Sta. Cruz del Quiché	Black	90	23.8	MR	MR	MR
2449	Mexico HGO. 14-A3	Pinto	90	25.3	MS	MR	MS
2450	Mexico HGO. 33-A1	Black	100	25.1	MR	MR	MS
2458	Mexico AGC. 9-3	Violet	90	23.9	R	MR	MS
2464	Mexico Chis 84-3	Black	90	20.8	S	MR	MR
2465	Mexico Chis 88-1	Mixed	90	24.1	MS	MR	MS
T2468A	Mexico 22 A-2	Yellow	90	24.9	R	MR	MS
2472	Mexico GTO. 3-A2	Black	100	19.4	MR	MR	MS
2473	Mexico GTO. 10-A5	Black	100	20.8	R	MR	MS
2476	Mexico MOR. C-A1	Pinto	95	30.2	R	R	MR
2503	Sta. Rosa Bárcenas	Black	105	24.2	MR	MR	MS
2504	Rabinal, B. Verapaz	White	90	20.6	MS	MS	MR
2524	Parramos, Chimaltenango	Black	105	21.7	MR	MR	MR
2525	Parramos, Chimaltenango	White	90	22.3	MR	MR	MS
2526	Parramos, Chimaltenango	Red	100	21.5	MR	R	MR
2798	Panajachel	Black	90	23.6	MR	MR	MR
2804	Chichicastenango	Black	90	20.4	MS	MR	MR
2806	Chichicastenango	Black	95	21.8	MR	MR	MR
2808	Tecpán	Red	85	22.9	MR	R	MS
2809	Tecpán	Black	90	24.2	MR	MR	MS
2824	Ipala, Chiquimula	Black	90	18.1	S	MS	MR
2829	Chiquimula	Black	85	21.7	MR	S	MS
5089	Salcajá, Quezaltenango	Black	90	25.1	MS	MS	MR
5091	La Alameda, Chimaltenango	Black	100	23.7	R	MR	MS

[a] Varieties not marked as originating in Mexico originated in Guatemala.

S. Susceptible MS. Moderately susceptible MR. Moderately resistant R. Resistant

Table II. Analysis of Variance of Yield of Bean per Plot

Source of Variation	D.F.	Sum of Squares	Mean Square	F
Varieties	22	519,854.19	2,369.74	
Plantings	1	11,134.08	11,134.08	
Variety × planting	22	582,676.68	26,485.30	4.915[a]
Location	1	41,564.38	41,564.38	
Variety × location	22	421,702.71	19,168.30	3.557[a]
Planting × location	1	182,079.43	182,079.43	
Variety × location × planting	22	118,561.39	5,389.15	1.861[b]
Replicates within planting × location	8	310,274.82	38,784.35	13.392[a]
Experimental error	176	509,728.50	2,896.18	

[a] *P* less than 0.01.
[b] *P* less than 0.05.

Results

Yield The analysis of variance of yield in grams per plot is given in Table II. The ranking of the varieties was different in the two plantings and in the two locations, as shown by the significant variety × planting and variety × location interactions. Moreover, the differences in rank of the varieties due to plantings changed from one location to the other, as indicated by the significant variety × planting × location interaction. Highly significant differences were also found among replicates at each location in each planting.

The F values were not significant at the $P = 0.05$ level for the average differences between varieties, plantings, or locations. As the planting × location interaction was also not significant, it can be concluded that the effect of plantings on the average yield of the entire group of varieties was similar in the locations.

In order to evaluate the relative importance of the factors and their interactions, the component of variance corresponding to each one of these was computed. Table III shows that only 22% of the total variation is attributable to unknown random causes and 78% of the total variation observed is attributable to factors that were included in the present study. The interactions involving varietal differences account for 50% of the total variation.

The average yields per acre of the varieties in this study are included in Table IV. The yields ranged from 9.66 to 20.19 bushels per acre.

Nitrogen and Protein The average nitrogen content of each variety is given in Table IV and indicates that protein content (N × 6.25) ranged from 20.1 to 27.9% with an average for all samples of 24.1%

Table III. Estimates of Components of Variance of Some Factors Affecting Yield of Beans

Component	Estimated Magnitude	Relative Magnitude, % of Total
Variety × planting	3,516.02	27
Random causes	2,896.18	22
Variety × location	2,296.52	17
Planting × location	2,076.74	16
Replicates within planting × location	1,560.36	12
Variety × planting × location	830.99	6
Total	13,176.81	100

Table IV. Average Yield and Nutrient Content of Bean Varieties Included in Guatemalan Trial of 1951[a]

Variety No.	Av. Prod., Bu./Acre	Nitrogen, %	Methionine		Lysine		Tryptophan		Niacin, Mg./100 G.	Thiamine, Mg./100 G.	Riboflavin, Mg./100 G.
			%	G./16 g. N	%	G./16 g. N	%	G./16 g. N			
2226B	19.90	3.79	0.33	1.39	1.96	8.27	0.17	0.72	2.46	1.17	0.17
2228	15.81	4.00	0.20	0.80	2.05	8.19	0.14	0.56	2.08	0.91	0.18
2449	13.50	3.36	0.19	0.90	1.86	8.86	0.14	0.67	2.02	0.77	0.16
2450	16.19	3.21	0.23	1.15	1.78	8.86	0.19	0.94	1.97	0.82	0.17
2458	17.80	3.86	0.29	1.20	1.74	7.22	0.15	0.62	1.90	0.90	0.17
2464	18.08	4.32	0.23	0.85	2.04	7.55	0.15	0.56	2.26	1.18	0.17
2465	16.91	3.92	0.22	0.90	1.83	7.47	0.20	0.82	2.28	0.93	0.18
T2468A	19.82	3.39	0.20	0.94	1.85	8.74	0.14	0.66	1.68	0.85	0.16
2472	17.88	3.35	0.21	1.01	1.86	8.88	0.14	0.67	2.16	0.91	0.17
2473	19.43	3.59	0.21	0.93	1.69	7.54	0.15	0.67	1.85	0.78	0.18
2476	17.59	3.69	0.27	1.17	1.91	8.29	0.19	0.82	2.21	1.04	0.18
2503	10.95	4.20	0.31	1.18	2.29	8.72	0.19	0.72	2.45	1.16	0.18
2504	15.95	3.83	0.17	0.70	2.20	9.18	0.21	0.88	2.41	0.88	0.16
2524	9.66	4.07	0.27	1.06	1.92	7.55	0.21	0.83	2.31	1.13	0.18
2525	15.01	4.01	0.24	0.96	2.02	8.06	0.22	0.88	2.31	1.02	0.17
2526	17.64	4.02	0.24	0.96	1.84	7.33	0.17	0.67	2.30	1.22	0.18
2798	11.83	4.02	0.26	1.04	2.14	8.51	0.15	0.59	2.29	1.03	0.17
2804	18.23	3.93	0.24	0.98	1.90	7.73	0.17	0.69	2.41	0.97	0.18
2806	19.74	4.02	0.26	1.04	2.20	8.75	0.16	0.64	2.60	0.94	0.17
2808	17.88	3.76	0.21	0.90	2.01	8.54	0.16	0.67	2.15	1.20	0.18
2809	15.23	4.07	0.30	1.18	1.92	7.55	0.15	0.59	2.34	1.09	0.17
2824	18.55	3.98	0.27	1.09	2.10	8.45	0.20	0.80	2.33	1.00	0.18
2829	20.19	3.85	0.28	1.17	2.01	8.35	0.16	0.67	2.03	1.00	0.18
5089	18.02[b]	3.81	0.26	1.09	2.08	8.74	0.16	0.67	2.01	1.14	0.16
5091	16.26[b]	4.46	0.29	1.04	2.42	8.69	0.17	0.61	2.95	1.34	0.23
Pooled standard error	2.99	0.12	0.03	...	0.12	...	0.01	...	0.15	0.07	0.01
Maximum	20.19	4.46	0.33	1.39	2.42	9.18	0.22	0.94	2.95	1.34	0.23
Minimum	9.66	3.21	0.17	0.80	1.69	7.22	0.14	0.56	1.68	0.77	0.16
Over-all mean	16.82	3.81	0.25	1.03	1.98	8.24	0.17	0.70	2.22	1.01	0.18

[a] All values are corrected to 10% moisture.
[b] Yields based on fall planting only.

Table V. Analysis of Variance of Nutrient Content of Bean Varieties

Source of Variation	Degrees of Freedom	Mean Square for the Nutrients						
		Nitrogen	Methionine	Lysine	Tryptophan	Niacin	Thiamine	Riboflavin
Varieties	24	0.5602[a]	0.0094	0.1927[b]	0.0033[a]	0.4013[a]	0.1379[a]	0.0007
Localities	1	2.8694[a]	0.1895[a]	0.9976[a]	0.0406[a]	0.2700	0.0002	0.0005
Var. × loc.	24	0.0902[a]	0.0062[a]	0.0914[a]	0.0012[a]	0.1321	0.0268[b]	0.0005[a]
Reps. within loc.	4	0.0163	0.0002	0.0215	0.0002	0.0220	0.0151	0.0007
Exptl. error	96	0.0244	0.0011	0.0189	0.0002	0.1029	0.0146	0.0002

[a] P less than 0.01.
[b] P less than 0.05.

Table VI. Values of Components of Variance in Nutrients in Bean Varieties

Nutrients	Total Magnitude	Components of Variance as % of Total: Varieties	Var. × loc.	Localities	Error	Coefficient of Variation, %
Nitrogen	3.5605	47	13	25	15	11
Methionine	0.2064	45	28	8	19	31
Lysine	1.3221	23	33	18	26	14
Tryptophan	0.0455	27	20	40	13	23
Niacin	0.9283	28	6	1	65	18
Thiamine	0.1946	50	11	0	39	19
Riboflavin	0.0026	9	29	0	57	10

and a pooled standard error of 0.8%. The analysis of variance of nitrogen content is given in the first column of mean squares in Table V.

The over-all differences in nitrogen content among varieties and between localities were highly significant. The relative nitrogen content of the varieties was also significantly different for the two locations, as shown by the highly significant variety × location interaction. However, replicates at the same location did not differ significantly in nitrogen content. The magnitude of variation due to each of the principal components is given in Table VI, together with the coefficient of variation. Under the environmental conditions to which these varieties were exposed in the second trial, 47% of the variation was due to differences in genotype, 25% to the effect of locations, 13% to interaction between genotype and the locations, and 15% to other causes.

In Table IV, amino acid content is expressed in terms of both grams per 100 grams of bean at 10% moisture and grams per 16 grams of nitrogen (grams per 100 grams of protein).

Methionine The average methionine content of the varieties, given in Table IV, ranged from 0.17 to 0.33%, with an average methionine content for all samples of 0.25% and a pooled standard error of 0.03%. The analysis of variance of methionine content is given in the second column of mean squares in Table V.

The varietal differences were negligibly small. However, the difference between localities was highly significant, as were also the differences in the rank of the varieties in methionine content for the two locations. Replicates at the same location did not differ significantly in methionine content. Although the magnitude of the varietal differences was small, Table VI shows that they account for 45% of the total variation. Only 8% of the total variation is explained by the differences between localities, 28% by the interaction between locality and varieties, and 19% by other causes.

Lysine The average lysine content of each variety given in Table IV ranged from 1.69 to 2.42%, with an average for all samples of 1.98% and a pooled standard error of 0.12%. The analysis of variance of lysine content is given in the third column of mean squares in Table V.

The over-all differences among varieties in lysine content were significant and that between localities was highly significant. The rank of the varieties in lysine content was also significantly different for the two locations, as shown by the highly significant variety × location interaction. Replicates at the same location did not differ significantly in lysine content.

Table VI shows that under the conditions of the second trial, 23% of the variation in lysine content was due to differences in genotype, 18% to the effect of location, 33% to interaction between genotype and location, and 26% to other causes.

Tryptophan The average tryptophan content of the varieties, given in Table IV, ranged from 0.14 to 0.22%, with an average for all samples of 0.17% and a pooled standard error of 0.01%. The analysis of variance of tryptophan content is given in the fourth column of mean squares in Table V.

The over-all differences in tryptophan content among varieties and between localities were highly significant. Moreover, the relative tryptophan content of the varieties was significantly different for the two locations, as shown by the highly significant variety × location interaction. Replicates at the same location did not show significant differences in tryptophan content.

Table VI shows that, under the conditions of the second trial, 27% of the variation in tryptophan was due to differences in genotype, 40% to the effect of location, 20% to interaction between genotype and location, and 13% to other causes.

Niacin The average niacin content, given in Table IV, ranged from 1.68 to 2.95 mg. per 100 grams, with an average for all samples of 2.22 mg. per 100 grams and a pooled standard error of 0.15 mg. per 100 grams.

The analysis of variance of niacin content given in the fifth column of mean squares in Table V showed highly significant over-all differences in niacin content among varieties. However, the difference between localities was negligibly small, as were also the differences in the rank of the varieties in niacin content at the two locations. Replicates at the same location did not differ significantly in niacin content.

Table VI shows that under the conditions of the second trial 28% of the variation in niacin content was due to differences in genotype, 1% to the effect of location, 6% to interaction between genotype and location, and 65% to other causes.

Thiamine The average thiamine content given in Table IV ranged from 0.77 to 1.34 mg. per 100 grams with an average for all samples of 1.01 mg. per 100 grams and a pooled standard error of 0.07 mg. per 100 grams.

The analysis of variance of thiamine content, given in the sixth column of mean squares in Table V, showed not only the over-all differences among varieties but also the differences in the rank of the varieties for the two locations to be highly significant. Nevertheless, the difference between localities was negligibly small and replicates at the same location did not differ significantly in thiamine content.

Table VI shows that under the conditions of the second trial 50% of the variation in thiamine content was due to differences in genotype, 11% to interaction between genotype and location, and 39% to other causes.

Riboflavin The average riboflavin content given in Table IV ranged from 0.16 to 0.23 mg. per 100 grams, with an average for all samples of 0.18 mg. per 100 grams and a pooled standard error of 0.01 mg. per 100 grams.

The analysis of variance of riboflavin content, given in the seventh column of mean squares in Table V, showed the over-all differences in riboflavin content among varieties and between localities to be negligibly small. However, the rank of the varieties in riboflavin content was very different for the two locations, as shown by the highly significant variety × location interaction. Replicates at the same location also differed highly significantly in riboflavin content.

Table VI shows that under the conditions of the second trial 9% of the variation in riboflavin was due to differences in genotype, 29% to interaction between genotype and location, and 57% to other causes.

Relationship between Yield and Nutrients

Three series of linear correlation coefficients were computed from the covariance analyses of each of the nutrients paired with yield. All intervariety coefficients of correlation between yield and the different nutrients were negative in sign. The very small positive coefficient between yield and riboflavin was the only exception. The coefficients between yield and nitrogen (−0.635), lysine (−0.507), niacin (−0.408), and thiamine (−0.574) were significant. Among the variety × locality coefficients, none was significant at the 5% level with the exception of that between yield and thiamine (−0.468). None of the intrareplicate coefficients between yield and nutrients were significant.

Interrelationship among Nutrients Analyzed

Covariance analyses were done for all possible pairs of nutrients. With certain exceptions, linear regression corrections for one nutrient at a time failed to change the relative importance of the factors that affected each of the other nutrients. The exceptions were: (1) The varietal differences in niacin became negligibly small when lysine was held constant, as did the varietal differences for lysine when any one of the other nutrients was held constant. (2) The locality to locality difference in lysine disappeared when nitrogen or methionine was held constant.

In these studies the intrareplicate coefficients computed from covariance analyses measure the extent to which the variation in one nutrient is reduced by considering the content of the other nutrient constant among plants in a replicate. Out of 21 intrareplicate coefficients only three were statistically significant at the 1% level. These were between nitrogen and tryptophan, lysine and niacin, and methionine and lysine. Although the intrareplicate coefficients of correlation were negative between either riboflavin or methionine and nitrogen, tryptophan, and thiamine, they were very small and could be attributed to sampling error.

The variety × locality coefficients indicate the extent to which the genetic-environmental interactions determine the reduction in the variation in one nutrient when the content of another is held constant. Only three of these were statistically significant at the 5% level, those between nitrogen and lysine (0.541), nitrogen and niacin (0.383), and niacin and thiamine (0.446).

The intervariety coefficients which indicate the genetic part of the relationship between the content of the nutrients were all positive and are shown in Table VII. They were statistically significant between nitrogen, niacin, or riboflavin as one of the variables and each of the other nutrients except tryptophan. For the latter the only significant intervariety coefficient of correlation was that involving niacin.

Table VII. Intervariety Coefficients of Linear Correlation between Nutrients in Bean Varieties Included in Guatemalan Trial of 1951

	Methionine	*Lysine*	*Tryptophan*	*Niacin*	*Thiamine*	*Riboflavin*
Nitrogen	0.443[a]	0.670[a]	0.268	0.693[b]	0.726[b]	0.451[a]
Methionine		0.258	0.138	0.399[a]	0.541[b]	0.326
Lysine			0.275	0.721[b]	0.550[b]	0.407[a]
Tryptophan				0.421[a]	0.224	0.183
Niacin					0.616[b]	0.541[b]
Thiamine						0.561[b]

[a] P less than 0.05.
[b] P less than 0.01.

Discussion

The results were analyzed in terms of the content of amino acid per 100 grams of beans, because the purpose of the study was to provide data which might help in the selection of varieties whose amino acid composition would better complement the Central American diet. Expressing the data in terms of grams of amino acid per 100 grams of protein would not accomplish this purpose. Moreover, the results of the covariance analysis show that the relative importance of the factors affecting amino acid content would not be altered by holding nitrogen constant.

The data show that the fertility of the land significantly alters both the yield and the riboflavin content of the kidney bean. However, the content of nitrogen, methionine, lysine, tryptophan, niacin, and thiamine is not detectably affected by the fertility differences, as the differences between replicates at each locality were not significant. This agrees with the findings of Scharrer and Schreiber (*19*) that different fertilizer levels increase the over-all yield and the total amount of protein in beans without materially influencing the relative percentages of crude protein, pure protein, and digestible crude protein.

The high-yielding varieties in the early planting were not the same that gave high yields in the late one. Similarly, varieties that were high yielding at Alameda were relatively low in yield at Bárcenas. The following tabulation of the four highest yielding varieties at each locality during each planting illustrates this.

Alameda		*Bárcenas*	
Early	*Late*	*Early*	*Late*
2458	2450	2829	2465
2464	2476	2808	2473
2829	2425	2809	T2468A
2806	2798	2824	2806

In this listing all the entries are different except 2829 and 2806, which appear twice. Therefore, if maximum yield is desired, it will be necessary to develop different varieties of bean for each type of locality during each of the planting seasons.

With the exception of the effect of replication on the yield, all the statistically significant interactions involve variety as one of the factors. This indicates the existence of a very large interaction between the genetic composition and environment in the yield of beans.

Therefore, a high-yielding variety in one environment may be a low-yielding one in another environment. As a consequence, the difference in yield among the varieties was negligible when averaged over the two plantings and the two locations. In this group of 25 varieties of kidney beans not a single variety was found which will yield equally well under the conditions of the two localities or both planting seasons.

Nevertheless, when data from the two localities were combined, highly significant varietal differences were detected in the contents of nitrogen, lysine, tryptophan, niacin, and thiamine. As the varietal differences are basically genetic, the gene composition controlling the content of each one of these nutrients must be different among the varieties studied. The conclusion that genetic differences are of major importance is further supported by the highly significant variety × locality interactions found for most of the nutrients. These results are in agreement with those of Gough and Lantzel (*7*), who in eight bean varieties grown at three locations in New Mexico in three different years observed significant differences in thiamine and niacin due to varieties, localities, and years; and in riboflavin due to varieties and localities only.

For nitrogen, methionine, tryptophan, niacin, and thiamine contents, the varietal differences (basically genetic) are more important than variety × locality (genetic × environmental) interactions. Hence, the most effective way to increase the amount of these nutrients in beans is selection and distribution of the varieties which are richer in these nutrients. Because the intervariety coefficients of correlation between pairs of these nutrients were all positive, selection among varieties for any one of these nutrients will not result in losses in the other nutrients.

Interaction between varieties and localities is a particularly important source of variation in lysine. Furthermore, the effect of localities—i.e., the crop-climate—is a significant factor in determining the content of lysine. Consequently, in order to increase the lysine supplied by beans, selected varieties should be grown in regions where their genetic make-up has the best chance of resulting in maximum nutrient value as well as yield. This would require both selecting among the varieties and choosing appropriate regions to grow them.

Locality to locality difference in the content of nitrogen, methionine, and tryptophan was large. Hence, growing bean varieties in suitable regions will increase the average content of these three nutrients.

In general, the content of each of the nutrients determined was higher for beans planted at Bárcenas than in Alameda. As sufficient information is not available, it is impossible to attribute all this difference to the dissimilarity of the "crop-climate" at the two localities. It is probable that the piece of land used in Bárcenas was more fertile than that in Alameda. As 39% or more of the total variance in the content of niacin, thiamine, and riboflavin was associated with unknown random causes, more work is needed to explore the factors that affect the content of these nutrients. The coefficients of variation for the nutrients analyzed ranged from 10 to 31%, the largest being for methionine and tryptophan.

The intervariety coefficients of correlation between pairs of these nutrients were all positive and a majority of them were highly significant. This implies one of the two following hypotheses: First, the presence of genes that cause the content of one nutrient to increase affects the content of other nutrients, causing the latter to increase also. In other words, the same set of genes affects several nutrients in the same direction. Second, the sets of genes affecting the different nutrients are linked very closely in the coupling phase. The data available in this study are not adequate for testing either of the above-mentioned hypotheses. Furthermore, even though most of the coefficients of correlation are significantly different from zero, they are also significantly different from unity.

The highly significant negative intervariety coefficients of correlation between yield and the nutrients nitrogen, thiamine, niacin, and methionine indicate that among the varieties included in this trial, the high-yielding ones tended to be low in these nutrients and vice versa. Furthermore, this relationship between yield and the nutrients is primarily genetic in nature and the variety × locality and intrareplicate coefficients are almost all negligibly small. Greenwood (*9*) concluded that in pinto beans under normal conditions—i.e., where the crops were not heavily infested with beetles or were not affected by hail storms—a negative correlation existed between yield and the percentage of protein. The present results are in agreement with her findings.

If it is necessary to select varieties from among those studied, for use in both localities, those numbered 2824, 2465, 2829, and 2806 seem to be most promising on the basis of yield and the general content of the nutrients.

Summary

The content of nitrogen, methionine, lysine, tryptophan, niacin, thiamine, and riboflavin was determined in 25 bean varieties grown in two different locations in Guatemala. Yield data on these varieties in the two locations were obtained for two different plantings. Yield ranged from 9.66 to 20.19 bushels per acre (average, 16.82 bushels per acre). No variety was found to yield equally well under the conditions of the two locations or the two plantings. The protein content ranged from 20.1 to 27.9% (average 24.1%) and was influenced significantly by both variety and location.

Methionine varied from 0.17 to 0.33% (average 0.25%), lysine from 1.69 to 2.42% (average 1.98%), and tryptophan from 0.14 to 0.22% (average 0.17%). This variation was due to both varietal and location factors, except in the case of methionine where varietal differences were not significant.

Niacin varied from 1.68 to 2.95 mg. per 100 grams (average 2.22 mg. per 100 grams), thiamine from 0.77 to 1.34 mg. per 100 grams (average 1.01 mg. per 100 grams), and riboflavin from 0.16 to 0.23 mg. per 100 grams (average 0.18 mg. per 100 grams). The differences in niacin and thiamine were due largely to varietal factors. The riboflavin values did not differ significantly among varieties or between locations.

All intervariety coefficients of correlation between yield and the different nutrients were negative in sign, except that between yield and riboflavin which was negligibly small. Since the intervariety coefficients of correlations between pairs of the nutrients were all positive, selection among varieties for any one of these nutrients will not result in losses in the others.

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Literature Cited

(1) Assoc. Offic. Agr. Chemists, "Official Methods of Analysis," 7th ed., 1950.

(2) Casseres, E. H., Thompson, H. C., *Turrialba* **1,** 44 (1951).

(3) Castillo, A. S., Flores, M., "Estudios dietéticos en El Salvador. II. Cantón Platanillos, Municipio de Quezaltepeque, Departamento de La Libertad," Suplemento No. 2 Boletín de la Oficina Sanitaria Panamericana, "Publicaciones Científicas del Instituto de Nutricion de Centro América y Panamá," p. 54 (1955).

(4) Eheart, J. F., *et al.*, "Vitamin Studies on Lima Beans," Georgia, Miss., S. Carolina, Virginia Agr. Expt. Stas., Southern Coop. Ser. Bull. **5** (1946).

(5) Flores, M., Reh, E., "Estudios de hábitos dietéticos en poblaciones de Guatemala. I. Magdalena Milpas Altas," Suplemento No. 2 Boletín de la Oficina Sanitaria Panamericana, "Publicaciones Científicas del Instituto de Nutrición de Centro América y Panamá," p. 90 (1955).

(6) Friedemann, T. E., Frazier, E. I., *Arch. Biochem.* **26,** 361 (1950).

(7) Gough, H. W., Lantzel, E. M., *Food Research* **15,** 308 (1950).

(8) Greene, R. D., Black, A., *J. Biol. Chem.* **155,** 1 (1944).

(9) Greenwood, M. L., *New Mexico Agr. Expt. Sta. Bull.* **213** (1933).

(10) Guyer, R. B., Kramer, A., Ide, L. E., *Proc. Am. Soc. Hort.* **56,** 303 (1950).

(11) Hamilton, L. F., Simpson, S. G., "Talbot's Quantitative Chemical Analysis," 9th ed., Macmillan, New York, (1946).

(12) Hennessey, D. J., Cerecedo, L. R., *J. Am. Chem. Soc.* **61,** 179 (1939).

(13) Hodson, A. Z., Norris, L. C., *J. Biol. Chem.* **131,** 621 (1939).

(14) Horn, M. J., Brasse, J. D., Blum, A. E., *Ibid.*, **166,** 321 (1946).

(15) *Ibid.*, **169,** 71 (1947).

(16) Jaffé, W. G., *Arch. Venezolanos Nutric* **1,** 107 (1950).

(17) Melnick, D., *Cereal Chem.* **19,** 53 (1942).

(18) Post, J. J., *Mededel Direct. Tuinbouw.* **13,** 23 (1950).

(19) Scharrer, K., Schreiber, R., *Bodenk. u. Pflazenernähr.* **30,** 360 (1943).

(20) Sogandares, L., Galindo, A. P. de, Mejía, H. P., "Estudios dietéticos de grupos urbanos y rurales de la República de El Salvador," Suplemento No. 1 Boletín de la Oficina Sanitaria Panamericana, "Publicaciones Científicas del Instituto de Nutrición de Centro América y Panamá," p. 27 (1953).

(21) Wooley, G. L., Sebrell, Jr., W. H., *J. Biol. Chem.* **157,** 141 (1945).

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