

Boron and Manganese Effects on Protein, Oil, and Fatty Acid

Composition of Oil in Soybeans

Fred C. Boswell and R. E. Worthington*

Studies were conducted on three soil types to determine whether foliar applied manganese and boron are beneficial or detrimental with respect to protein, oil, and fatty acid composition of soybean seed. Mn and B treatments did not greatly affect percent total oil content as determined by nmr. Percent oil ranged from 20.7 on the Etowah soil when 1.22 kg/ha Mn was applied the initial year to a high of 23.8 on the Norfolk soil the second year when the

same Mn rate was applied. Although slight protein increases were noted when B and Mn treatments were applied, values from the treated plots were not significant when compared to the check plots. Soil sites and years influenced total oil and protein more than micronutrient treatments. Further, Mn and B had no effect on the various fatty acid components.

The soybean [*Glycine max.* (L) Merrill] is becoming a leading crop in the Southeast, and with increased economic importance of the crop, more effort is being directed towards increasing crop yields and quality (Boswell and Anderson, 1969; Parker and Harris, 1962). With larger quantities of soybeans and soybean products being channeled into the world's food supply, the effects of cultural practices on seed quality become increasingly important.

Since the acid soils of the Southeast vary greatly in micronutrient content, the effects of these elements on seed composition, quality, and yield are receiving considerable attention. The effects of these nutrients are of added interest since many of the presently used high analysis fertilizers are practically devoid of micronutrients.

Numerous fatty acid studies with soybean oil (Alderks, 1949; Howell and Cartter, 1953; Howell and Collins, 1957; Johnson and Jellum, 1969; Simmons and Quackenbush, 1954) have been conducted. However, information on the effects of applied micronutrients on protein and oil content, and fatty acid composition of oil in soybeans is sparse.

With increased use of micronutrients, the effects of these materials on soybean quality should be investigated and documented. These studies were conducted to determine whether foliar applications of manganese and boron (Mn and B) affect protein and oil content of seed or fatty acid composition of oil.

MATERIALS AND METHODS

Field experiments, utilizing Bragg variety of soybeans, were established on Norfolk, Davidson, and Etowah soils, rep-

resentative of the Coastal Plain, Piedmont, and Limestone Valley regions, respectively. Prior to initiation, dolomitic limestone was applied at rates sufficient to adjust the soil reaction near pH 6.2. Eight treatments were applied in a latin square design. The treatments were: Check, 0.112 kg/ha B (as Solubor, a commercial product containing 20.5% B); 0.280 kg/ha B; 0.560 kg/ha B; 1.22 kg/ha Mn (as $MnSO_4 \cdot H_2O$); 2.44 kg/ha Mn; 4.07 kg/ha Mn; and 0.280 kg/ha B + 2.44 kg/ha Mn. These materials were dissolved in 250 ml of deionized water and foliar-applied to 3.65 m row length when soybean plants began to bloom. At harvest, samples were taken for protein, oil, and fatty acid determinations. The studies were conducted, in place, for the 3-year study period. Samples from individual plots were not analyzed, but equal samples were combined from eight replications to form composite samples for each treatment. Duplicate samples of each treatment were analyzed for total protein and oil and fatty acid composition of oil. In order to statistically evaluate treatment and year effects on protein and oil content and fatty acid composition, locations were used as replications.

Fatty acid methyl esters were prepared by treating sample material with a 3% solution of sulfuric acid in methanol, as described previously (Jellum and Worthington, 1966), and held at 20°C under nitrogen until analyzed.

Analyses were made either on an F&M Model 700, or MicroTek Model 220 gas chromatograph, equipped with flame ionization detectors and an Infotronics CRS 100 electronic integrator. The individual fatty acid esters were identified and estimated according to previously developed procedures (Worthington and Holley, 1967), except quantitative measurements were made with columns packed with 70/80 mesh Chromosorb W (AW) (DMCS) coated with 10% (W/W) stabilized diethylene glycol succinate (Analabs, Inc.).

*Departments of Agronomy and Food Science, University of Georgia, College of Agricultural Experiment Stations, Georgia Station, Experiment, Ga. 30212

Table I. Protein Content (%) of Soybeans as Affected by B and Mn on Three Soils for 3 Years

Treatment and Rate, kg/ha	Norfolk soil			Davidson soil			Etowah soil			Treatment Mean ^a
	1967	1968	1969	1967	1968	1969	1967	1968	1969	
Check	40.9	41.8	41.1	39.6	41.3	40.1	38.5	37.4	36.3	39.7
B - 0.11	40.4	42.3	42.0	39.6	40.2	39.5	39.0	37.5	38.4	39.9
B - 0.28	41.3	42.5	42.5	40.0	41.4	38.6	39.6	37.9	36.6	40.0
B - 0.56	41.1	43.1	42.5	40.3	41.4	40.8	38.5	37.9	37.6	40.4
Mn - 1.22	41.5	42.7	41.7	40.0	42.1	40.5	39.0	37.7	37.2	40.3
Mn - 2.44	41.5	41.9	41.9	40.0	42.0	40.0	38.7	37.7	36.0	40.0
Mn - 4.07	41.4	42.2	40.7	39.9	42.8	39.6	38.3	37.6	36.5	39.9
B + Mn - 0.28 + 2.44	41.3	41.7	40.5	39.5	41.6	40.2	38.7	37.1	37.0	39.7
Mean	41.2	42.3	41.6	39.9	41.6	39.9	38.8	37.6	37.0	40.0
σ	0.35	0.45	0.72	0.25	0.71	0.64	0.38	0.25	0.73	0.23

Year	Protein Percent	Soil sites	Protein Percent
1967	39.9 ab ^b	Norfolk	41.9 a
1968	40.5 a	Davidson	40.5 b
1969	39.5 b	Etowah	37.7 c

^a ANOV for treatment—N.S. ^b Values followed by the same letter are not significantly different.

Table II. Total Oil Content (%) of Soybeans as Related to Temperature for 3 Years at Three Soil Sites^a

Year	Total Oil Percent	Mean Temper- ature, ^b °C	Soil Site	Total Oil Percent	Mean Temper- ature, °C
1967	21.3 a ^c	22.3	Norfolk	21.9 a	24.3
1968	22.5 b	24.3	David- son	22.0 ab	22.9
1969	22.5 b	23.2	Etowah	22.3 b	22.4
Stat.	0.01		Stat.	0.05	
Sig.			Sig.		

^a Mean of all treatments. ^b August and September. ^c Values followed by the same letter are not significantly different.

RESULTS AND DISCUSSION

The water extractable B content of the soils, prior to the establishment of the studies, was 0.06, 0.18, and 0.18 ppm, while the N ammonium acetate extractable Mn was 1.20, 59.3, and 114.1 ppm for the Norfolk, Davidson, and Etowah soils, respectively. Organic matter ranged from 1.53% on the Norfolk to 2.49% on the Davidson soil. Cation exchange capacities for the soils were 4.16, 9.06, and 11.12 mequiv/100 g for the Norfolk, Davidson, and Etowah soils, respectively.

Percent protein of the soybeans, as influenced by treatments, ranged from 39.7 on the check treatment to 40.4 when 0.56 kg/ha of B was applied (Table I). There was a tendency for

protein to increase as the rate of B increased. Conversely, as the rate of Mn increased, protein tended to decrease. These differences in treatment averages were not significant (P = 0.05). When comparing the percent protein of the soybeans grown at various soil sites, significant differences were noted. Soybeans grown in the Coastal Plains (Norfolk site) were significantly higher in protein (P = 0.01) than those grown in the Piedmont (Davidson site), which were significantly higher than those grown in the Limestone Valley (Etowah site). Protein was also inversely related to total oil content of soybeans grown at the respective sites. As expected, significant differences in protein occurred from year to year but were not always related to total oil.

Differences in yearly averages of total oil were statistically significant (P = 0.01) with the oil content in 1967 being significantly less than in 1968 or 1969; no difference in the latter 2 years was noted (Table II).

Total oil content of the soybeans grown on the Norfolk soil site was significantly less (P = 0.05) than that of those grown on the Etowah soil site, but not less than that of the Davidson soil (Table II). Some workers (Howell and Cartter, 1953; Simmons and Quackenbush, 1954) have shown a positive correlation between environmental temperature during certain stages of pod filling and percent oil in mature beans. Similar effects were observed at two of the three locations (Table II). Temperature at the Norfolk site was

Table III. Total Oil Content (%) of Soybeans as Affected by B and Mn on Three Soils for 3 Years^a

Treatment and Rate kg/ha	Norfolk soil			Davidson soil			Etowah soil			Treatment Mean ^b
	1967	1968	1969	1967	1968	1969	1967	1968	1969	
Check	21.0	22.7	21.9	21.8	21.8	22.7	21.0	23.3	22.5	22.1
B - 0.11	21.2	22.0	21.9	21.5	22.1	23.2	21.4	23.1	22.2	22.1
B - 0.28	21.3	21.8	21.9	21.5	22.4	22.8	21.0	23.1	22.8	22.1
B - 0.56	21.6	21.8	21.9	21.2	21.8	22.7	21.2	22.7	22.8	22.0
Mn - 1.22	21.5	23.8	21.6	21.2	22.0	23.1	20.7	23.4	22.8	22.2
Mn - 2.44	21.5	22.7	21.6	21.5	21.7	23.1	21.2	22.8	22.8	22.1
Mn - 4.07	21.2	22.7	21.6	21.0	21.8	23.1	21.4	23.1	22.8	22.1
B + Mn - 0.28 + 2.44	21.8	22.4	21.6	21.0	21.8	23.2	21.2	22.7	22.8	22.1
Treatment and Soil Mean	21.4	22.5	21.8	21.3	21.9	23.0	21.1	23.0	22.7	22.1
σ	0.24	0.62	0.15	0.26	0.22	0.20	0.22	0.25	0.21	0.05

^a Nmr. ^b ANOV for treatment—N.S.

higher than for the Davidson soil site which was, in turn, higher than the Etowah site during pod filling (August and September), thus resulting in an inverse relationship. The highest temperatures for August and September were recorded in 1968, followed by 1969 and then 1967, which is directly related to the total oil content values.

Percent oil and temperature relationship cannot be fully evaluated for soils, since all soils were not represented at a given site.

Utilizing locations as replications, treatment effects on total oil were evaluated statistically. Treatment effects were nonsignificant ($P = 0.05$) and varied only 0.2% (Table III). The coefficient of variation was 2.51%. Percent total oil content of the soybeans ranged from a low of 20.7 on the Etowah soil, when 1.22 kg/ha Mn was applied the initial year, to a high of 23.8 on the Norfolk soil the second year, when these same rates of Mn were applied.

Fatty acid compositions of soybean oil from the various soils in 1969 are presented in Table IV as representative values for the 3-year period. Rates of B or Mn had no effect on fatty acid components. Palmitic acid (16:0) was slightly less at the Norfolk soil site, when compared to the Davidson and Etowah soil sites. At the Etowah soil site oleic acid (18:1) values were somewhat less than values for soybeans grown on the Norfolk or Davidson sites but not significantly different.

A summary of the fatty acid composition of the oil from soybeans grown on the three soil sites for the 3 years is presented in Table V. Since treatments were not different, the values are the eight treatment averages. Palmitic acid (16:0) was not different for soil sites or years. Stearic (18:0), oleic (18:1), arachidic (20:0) and behenic (22:0) values were significantly less for the Etowah site, when compared to the Davidson and Norfolk sites. These fatty acid values were less in 1969 as compared to 1967, although not always

Table IV. Fatty Acid Composition of Soybean Oil as Affected by B and Mn on Three Soils in 1969

Soil and Rate of Mn - B, kg/ha	Fatty acid ^a						
	16:0	18:0	18:1	18:2	20:0	18:3	22:0
	Percent						
Norfolk							
Check	10.3	4.4	21.1	55.6	0.5	7.9	0.7
B - 0.11	10.4	4.5	21.3	54.7	0.4	7.8	0.8
B - 0.28	10.6	4.4	21.2	54.7	0.4	8.0	0.7
B - 0.56	10.5	4.5	21.1	55.0	0.4	7.9	0.7
Mn - 1.22	10.9	4.4	21.4	55.0	0.4	8.0	0.7
Mn - 2.44	10.5	4.4	20.9	55.0	0.4	8.1	0.7
Mn - 4.07	10.6	4.4	20.2	55.3	0.4	8.3	0.8
Mn + B - 2.44 + 0.28	10.7	4.5	21.0	54.8	0.3	8.2	0.5
Davidson							
Check	11.3	4.4	21.2	54.2	0.4	7.3	0.8
B - 0.11	11.1	4.5	21.4	54.2	0.5	7.2	0.8
B - 0.28	11.1	4.5	21.7	54.2	0.5	7.2	0.8
B - 0.56	10.9	4.6	22.0	53.9	0.4	7.1	0.8
Mn - 1.22	11.1	4.4	21.1	54.7	0.4	7.4	0.9
Mn - 2.44	11.3	4.6	21.3	54.3	0.5	7.2	0.8
Mn - 4.07	11.2	4.5	21.7	53.9	0.5	7.2	0.8
Mn + B - 2.44 + 0.28	10.7	4.5	21.7	54.1	0.5	7.6	0.8
Etowah							
Check	11.3	3.9	19.1	54.5	0.4	9.6	0.7
B - 0.11	11.4	3.9	18.9	54.5	0.4	9.4	0.7
B - 0.28	11.1	3.9	19.2	55.0	0.4	9.4	0.7
B - 0.56	11.1	4.0	19.3	55.2	0.4	9.5	0.7
Mn - 1.22	11.2	4.0	19.4	54.8	0.4	9.3	0.7
Mn - 2.44	11.2	4.2	19.7	54.4	0.4	9.2	0.7
Mn - 4.07	11.0	4.1	19.4	54.8	0.4	9.3	0.7
Mn + B - 2.44 + 0.28	10.9	4.2	19.2	55.0	0.4	9.3	0.8

^a No significant difference due to Mn or B.

Table V. Fatty Acid Composition of Soybean Oil from Three Soils for 3 Years

Soil and year	Fatty acid						
	16:0	18:0	18:1	18:2	20:0	18:3	22:0
	Percent						
Norfolk							
1967	10.8	4.8	20.9	53.8	0.7	8.2	0.5
1968	10.6	4.4	21.0	55.0	0.4	8.0	0.7
1969	11.4	3.8	19.4	54.7	0.3	8.5	0.3
Soil mean	10.9	4.4 a	20.5 a	54.5 a	0.5 a	8.2 a	0.5 a ^a
Davidson							
1967	11.2	4.4	19.6	55.0	0.6	8.8	0.4
1968	11.1	4.5	21.5	54.2	0.5	7.3	0.8
1969	11.1	3.8	18.0	56.7	0.4	8.8	0.3
Soil mean	11.1	4.2 a	19.7 a	55.3 ab	0.5 a	8.3 a	0.5 a
Etowah							
1967	10.7	4.1	19.1	55.7	0.5	9.4	0.3
1968	11.1	4.0	19.3	54.8	0.4	9.4	0.7
1969	10.9	3.6	17.3	56.3	0.3	10.0	0.3
Soil mean	10.9	3.9 b	18.6 b	55.6 b	0.4 b	9.6 b	0.4 b
Soil mean Sig.	N.S.	0.01	0.01	0.01	0.01	0.01	0.05
Year Means							
1967 average	10.9	4.4 a	19.9 ab	54.9 a	0.6 a	8.8 ab	0.4 a
1968 average	10.9	4.3 a	20.6 b	54.7 a	0.4 b	8.2 a	0.7 b
1969 average	11.1	3.7 b	18.2 a	55.9 b	0.3 b	9.1 b	0.3 a
Year mean Sig.	N.S.	0.01	0.01	0.01	0.01	0.01	0.01

^a Values followed by the same letter are not significantly different.

significant. A reciprocal relationship existed for linoleic (18:2) and linolenic (18:3) fatty acids for soils and years.

Applications of B and Mn did not alter the fatty acid distribution or total oil content. Significant yield effects were not obtained (data to be reported elsewhere), although the heavier application rates of B and Mn resulted in visual toxicity symptoms.

These data indicate that normal climatic fluctuations and soil variation may be expected to have a greater effect on oil quantity and composition than foliar applications of the micronutrients B or Mn, even when these elements are added in sufficient quantity to cause visual foliar toxicity.

ACKNOWLEDGMENT

Appreciation is expressed to personnel of the U.S. Regional

Soybean Laboratory, Urbana, Ill., for total oil analyses and U.S. Borax for supplying Solubor.

LITERATURE CITED

- Alderks, O. H., *J. Amer. Oil Chem. Soc.* **26**, 126 (1949).
Boswell, Fred C., Anderson, O. E., *Agron. J.* **61**, 58 (1969).
Howell, R. W., Cartter, J. L., *Agron. J.* **45**, 526 (1953).
Howell, R. W., Collins, F. I., *Agron. J.* **49**, 593 (1957).
Jellum, M. D., Worthington, R. E., *Crop Sci.* **6**, 251 (1966).
Johnson, B. J., Jellum, M. D., *Agron. J.* **61**, 379 (1969).
Parker, M. B., Harris, H. B., *Agron. J.* **54**, 480 (1962).
Simmons, R. O., Quackenbush, F. W., *J. Amer. Oil Chem. Soc.* **31**, 601 (1954).
Worthington, R. E., Holley, K. T., *J. Amer. Oil Chem. Soc.* **44**, 515 (1967).

*Received for review November 5, 1970. Accepted February 4, 1971.
Approved as Journal Series Paper No. 903, University of Georgia,
College of Agricultural Experiment Stations, Georgia Station, Ex-
periment, Ga. 30212*

Correction

COTTONSEED PROTEIN PRODUCTS—COMPOSITION AND FUNCTIONALITY

In this article by Wilda H. Martinez, Leah C. Berardi, and Leo A. Goldblatt [*J. AGR. FOOD CHEM.* **18**(6), 961 (1970)], on page 964, the last sentence of the first paragraph in column two should read: "Grinding to a narrow range or single particle size would make nitrogen concentration by air separation impossible."