

Agronomic Management System and Precipitation Effects on Soybean Oil and Fatty Acid Profiles

Juan Gao,^{*} Xinmei Hao, Kurt D. Thelen, and G. Phillip Robertson

ABSTRACT

This study investigated long-term agronomic management systems and precipitation level effects on soybean [*Glycine max* (L.) Merr.] total oil content and fatty acid composition. Management systems evaluated included conventional (CT), no-till (NT), low chemical input (LI), and zero chemical input (ORG). Total oil content and major fatty acids profiles were analyzed by accelerated solvent extractor (ASE 200) and gas chromatography with flame ionization detector (FID). The results showed these four management systems have limited influence on soybean grain total oil content and oleic acid (O) and linoleic acid (L) compositions. The NT management system significantly improved soybean oil yield on a land-area basis as a result of higher annual grain yields. Soybeans grown under the NT management system had as high or higher palmitic acid (P) composition than the other three management systems; similarly, the CT treatments had as low or lower linolenic acid (LN) composition in soybean when compared with the other three management systems. The levels of stearic acid (S), O, L, and LN had a significant quadratic relationship ($R^2 = 0.64\text{--}0.75$) with total (July–September) precipitation. The oil quality ratio of O/(L + LN) had a quadratic relation with precipitation.

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Abbreviations: CT, tilled conventional chemical-input system; den df, denominator degree of freedom; FAME, fatty acid methyl ester; FID, flame ionization detector; KBS, Kellogg Biological Station; L, linoleic acid; LI, tilled low chemical-input system; LN, linolenic acid; LTER, long-term ecological research site; NT, nontilled conventional chemical-input system; num df, numerator degree of freedom; O, oleic acid; ORG, tilled no-chemical-input system; P, palmitic acid; S, stearic acid.

ORIGINATING IN EAST ASIA, soybean [*Glycine max* (L.) Merr.] has been grown as an essential dietary component because of its high grain protein (25.5–58.9%) and oil (12.0–23.0%) content. It is grown widely throughout the world but primarily in the United States (32%), Brazil (28%), Argentina (21%), China (7%), and India (4%) (soystat, 2008). The oil extracted from soybean is widely used for frying oil, as a component of margarine and salad dressing, and in the production of inks, paints, and cosmetics (Sheaffer and Moncada, 2008). Soybean oil is also the principal oil used for biodiesel in the United States, and it was reported that 68.6 million gallons of soybean oil was converted to biodiesel (91.5%) in 2005 (Urbanchuk, 2006). The esters of soybean oil have similar physical properties, such as viscosity, cetane number, and energy content to No. 2 diesel fuel, while reducing carbon, sulfur, and nitrogen exhaust emissions (Knothe and Dunn, 2001).

Five major fatty acids, palmitic acid (P, C16:0), stearic acid (S, C18:0), oleic acid (O, C18:1), linoleic acid (L, C18:2), and

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linolenic acid (LN, C18:3), commonly occur in vegetable oil and compose over 98% of the fatty acids in soybean oil (Canakci and Gerpen, 2001; Dubois et al., 2007). Among these five major fatty acids, P and S have saturated structures, which are stable during storage and have higher melting points (62 and 69.3°C). The saturated fatty acids are very useful in improving food taste and making candles, soaps, plastics, oil pastes, and cosmetics. However, in biodiesel, saturated fatty acids contribute to the cold flow issues associated with the fuel. The unsaturated fatty acids, such as O (13.4°C), L (−5°C), and LN (−11.3°C), have lower melting points and unstable properties. Polyunsaturated fatty acids, L and LN, are essential fatty acids for human dietary needs and serve as primers in the synthesis of longer and more desaturated fatty acids. In the food and biodiesel industry, O is a more ideal fatty acid, having a relatively lower melting point than the saturated fatty acids and longer shelf life than polyunsaturated fatty acids. Total oil content and fatty acid composition of soybean varies with different genotypes and environmental conditions (Cherry et al., 1985; Dardanelli et al., 2006; Piper and Boote, 1999; Primomo et al., 2002; Wilcox and Cavins, 1992). Temperature and precipitation are the two major environmental factors (Dornbos and Mullen, 1992). In former reports, seed oil content initially increased with temperature, reaching a plateau at around 22°C (Sato and Ikeda, 1979), or 25 to 28°C (Wolf et al., 1982), and decreased when temperatures exceeded these levels (Gibson and Mullen, 1996a; Gibson and Mullen, 1996b). Higher ambient air temperatures during the seed-filling period increased O content and decreased L and LN content; P and S are relatively stable to environmental change (Dornbos and Mullen, 1992). Increased drought stress could decrease seed oil content (Dornbos and Mullen, 1992). Other factors, such as planting date (Wilcox and Cavins, 1992), availability of mineral nutrient (except nitrogen) (Leffel and Hanson, 1961), cultural practices, soil type, and weed, disease, and insect pressure may also affect seed oil content of soybean (Primomo et al., 2002).

Management practices, including tillage, fertilizer input, and plant rotation, have been reported to have significant effects on agricultural systems, including weed community (Swanton et al., 2006), soil structure (Grandy et al., 2006b; Wright and Hons, 2004), soil fertility (Kelley and Sweeney, 1998), crop yield, and spatial variability (Grandy et al., 2006a; Harder et al., 2007; Howard et al., 1998; Hussain et al., 1999; Keim et al., 1999; Kravchenko et al., 2005; Smith et al., 2007).

Although the effect of environment and genotype (Hou et al., 2006; Oliva et al., 2006; Piper and Boote, 1999; Primomo et al., 2002) has been studied, very little has been reported on agronomic management system effects on soybean oil content and fatty acid composition. In this study, four agronomic management systems were

evaluated in a 17-yr, long-term crop research site. The objective of this study was to investigate management systems and precipitation effects on soybean seed oil content and fatty acid profile.

MATERIALS AND METHODS

Site Description and Data Collection

The study was conducted at the Michigan State University, Kellogg Biological Station (KBS), a long-term ecological research site (LTER). Four agronomic management systems comprised the treatments: tilled with conventional chemical inputs (CT); no-till with conventional chemical inputs (NT); tilled with low chemical inputs (LI), and tilled with no chemical inputs (ORG). The dominant soil types were well-drained Kalamazoo silt loam (Typic Hapludalfs) and Oshtemo sand loam (Typic Hapludalfs) (Davis et al., 2005). The site was established in 1988. The experiment was designed as a randomized complete block and each treatment had six replicates (more information can be found at <http://lter.msu.edu/data>). From 1993 to 2006, a rotation of corn–soybean–winter wheat was grown at the site. Among those years, soybean was grown in 1994, 1997, 2000, 2003, and 2006 (the information of soybean varieties and planting and harvest dates can be found in Table 1). A more detailed description of these four management systems and site history can be found in Smith et al. (2007).

A total of 120 soybean grain samples were collected from six reps with four treatments over five soybean rotational years: 1994, 1997, 2000, 2003, and 2006. Soybean grain was collected at harvest, ground, and stored at 4°C. Average growing season daily (maximum, mean, minimum) temperature (Table 2) and total precipitation (Fig. 1) was obtained from the KBS LTER Meteorological Station.

Total Oil Content

Total oil content was measured following the method of Matthäus and Brühl (2001). One gram of powdered soybean grain was weighted and oil was extracted using an accelerated solvent extractor ASE 200 (Dionex Corporation, Sunnyvale, CA) equipped with 11-mL stainless-steel extractor cells. The dead volume of the extractor cell was filled with Ottawa sand (Fisher Scientific, Rochester, NY). The following conditions were set on the ASE system: preheat for 6 min, heat for 6 min, oven temperature at 105°C, static time for 10 min, flush volume 70%, purge time for 60 s, two static cycles, and extraction pressure at 1000 psi. After processing, the extraction solvent hexane was

Table 1. Soybean [*Glycine max* (L.)] varieties, planting, and harvest dates in these rotation years.

Year	Variety	Relative maturity	Planting date (May)	Harvest date (Oct.)
1994	Pioneer 9202	2.0	17–20	06–11
1997	Pioneer 9172	2.0	21–23	17–21
2000	Pioneer 9172	2.0	25(CT), 26(NT) [†]	11–19
2003	NK S20-F8	2.0	26–29	06–07
2006	NK S20-F8	2.0	26	26

[†]Seeds with LI and ORG were planted 7 June 2000. CT, tilled conventional chemical input; NT, nontilled conventional chemical input; LI, tilled low chemical input; ORG, tilled no chemical input.

evaporated by purging oxygen-free compressed nitrogen (AGA Gas Inc., Cleveland, OH) above the surface. The residue hexane was removed in an oven at 100°C for about 70 min. The total oil content (*C*) was calculated using AOCS Official Method AM 2-93 (AOCS, 2000) as follows:

$$\%C = 100 \times \frac{Q_w}{[W \times (1 - \text{moisture}\%)]} \quad [1]$$

in which Q_w (g) is the total oil extracted, W (g) is the weight of the ground sample, and *moisture%* is the moisture percentage of the ground sample as measured using a Moisture Analyzer A&D MF-50 (A&D Company Ltd., San Jose, CA)

Fatty Acid Methyl Ester Reaction

The in situ fatty acid methyl ester (FAME) reactions were conducted in basic conditions following the method of Hammond et al. (Hammond, 1991). Soybean powder (10 mg) was soaked in 1 mL hexane (GC grade, EMD Chemicals Inc., Gibbstown, NJ) for a minimum 2-h extraction, then 0.5 mL 1 N sodium methoxide (Sigma-Aldrich Inc., Allentown, PA) was added to the mixture. The reaction mixture was shaken using a Labnet Vortex Mixer (Labnet International, Inc., Woodbridge, NJ) for half an hour to 1 h, after which the reaction was stopped by adding 2 mL distilled water. When the reaction mixture separated into two clear layers, the upper layer containing the FAME was removed and stored at 4°C until analysis. The transesterification rate was >98%, and results were not significantly different from those obtained from powdered samples in an acid condition (Liu et al., 1995a; Liu et al., 1995b).

Gas Chromatography with Flame Ionization Detector

After FAME reaction, samples were analyzed using a 6890 Agilent GC-FID (gas chromatography with flame ionization detector) equipped with a 30 m × 0.53 mm × 0.5 μm Supelcowax 10 capillary column (SUPELCO Inc., Bellefonte, PA) and HP 7673 autosampler (Agilent Technologies, Santa Clara, CA). Gas chromatograph running conditions were as follows: oven temperature increased from 195 to 240°C at 4°C/min and held at 240°C for 2 min, injection volume was 1 μL with split ratio 20:1; inlet temperature was 220°C; and detector temperature was 290°C. Ultra-high-purity (99.999%) helium (AGA Gas Inc., Cleveland, OH) was used as carrier gas at a flow of 3.3 mL/min. Concentrations of the five major FAMEs (P, S, O, L, and LN methyl ester) were corrected with external standard (Supeclo, Bellefonte, PA) calibration curves (0.05, 0.1, 0.2, 0.5, 1, 2, 5 mg mL⁻¹). The data were expressed as a percentage of the total five major fatty acids. Triplicate analyses were conducted for all samples.

Statistical Analysis

Analyses of variance (ANOVA) was employed to determine the effect of the four management systems on the total oil content and fatty acid composition. A linear mixed statistical model, similar to the one proposed by Kravchenko et al. (2005), was used:

Table 2. Average daily maximum, mean, and minimum temperatures in July, August, and September of rotation years. Max., average daily maximum temperature (°C); Mean, average daily mean temperature (°C); Min., average daily minimum temperature (°C).

Year	July			August			September		
	Max.	Mean	Min.	Max.	Mean	Min.	Max.	Mean	Min.
1994	26.62	20.87	15.43	24.38	18.45	12.65	23.36	16.59	10.63
1997	27.25	21.05	15.25	24.09	18.48	13.59	21.44	15.79	10.58
2000	25.52	19.75	14.28	25.65	20.01	15.06	21.75	15.73	10.54
2003	28.15	21.23	14.56	28.46	21.27	15.25	22.81	16.01	9.83
2006	29.29	23.05	16.92	27.53	21.13	14.96	19.74	15.02	9.71

$$y_{ijk} = \mu + \text{block}_j + \text{tr}_i + \text{plot}_{ij} + \text{year}_k + \text{year}_k \times \text{tr}_i + e_{ijk} \quad [2]$$

where y is total oil content or percentage of the major fatty acids in oil, μ is the grand mean of total oil content or percentage of the major fatty acid, block is the random effect of blocks (replications), plot was the random effects of a plot, tr is the fixed effect of treatment, year was the fixed effect of year, year × tr was the interaction effect of year and treatment, and e is the residual. The analysis was conducted by PROC MIXED in SAS 9.0 (SAS Institute, Cary, NC).

Temperature and precipitation were the two major environmental effects evaluated. Because average daily temperatures during these 5 yr were fairly constant (Table 2), temperature was not included in the analysis. A quadratic model was used to describe the relationship of total precipitation and total oil content or fatty acid components. To test whether the four treatments had the common intercepts and common slopes, general linear test approach, as described in Kutner et al. (2004), was conducted using PROC GLM in SAS 9.0 (SAS Institute, Cary, NC). This approach was often used to test whether a reduced or restricted model was significantly different with a full or unrestricted model, when fitted with the same data, by comparing the two error sums of squares obtained from the two models. For the full model in the study, the intercepts and slopes were assumed to be different for the four treatments, while the four

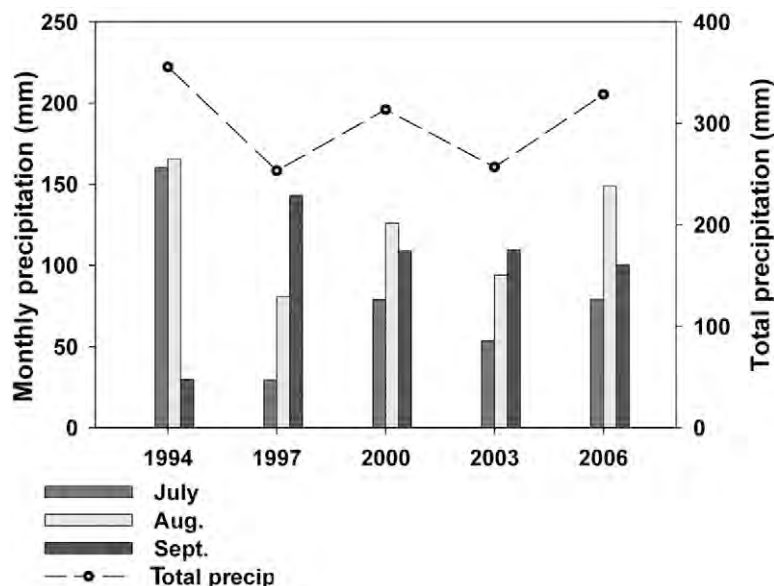


Figure 1. Total precipitation during July to September in research years.

treatments presumably had the common intercepts and slopes for the reduced model. When the reduced model was found significantly different with the full model, further analyses were conducted to compare the four treatments pairwise. To that end, three indicator or dummy variables were created as X1 (= 1 if tr = NT; = 0 otherwise), X2 (= 1 if tr = LI; = 0 otherwise), and X3 (= 1 if tr = ORG; = 0 otherwise). The overall regression model was

$$\begin{aligned}
 y_{ijk} = & \beta_0 + \beta_{01}X1 + \beta_{02}X2 + \beta_{03}X3 + \beta_{11}\text{precip}_k \\
 & + \beta_{11}X1\text{precip}_k + \beta_{12}X2\text{precip}_k + \beta_{13}X3\text{precip}_k \\
 & + \beta_{21}\text{precip}_k^2 + \beta_{21}X1\text{precip}_k^2 + \beta_{22}X2\text{precip}_k^2 \\
 & + \beta_{23}X3\text{precip}_k^2 + e_{ijk} \quad [3]
 \end{aligned}$$

The significances of estimated coefficients (β_{11} , β_{12} , β_{13} , β_{21} , β_{22} , and β_{23}) indicate treatments respond differently, relative to CT, with the change of precipitation. The difference between NT and LI was determined by testing whether $\beta_{11} = \beta_{12}$ and $\beta_{21} = \beta_{22}$ simultaneously using CONTRAST statement in the PROC GLM. Similar tests were performed for NT vs. ORG and LI vs. ORG. The regression analysis was conducted separately for total oil content or the fatty acid composition. Unless specified, an α level of 0.05 was used to determine the significance of effects and comparisons.

RESULTS AND DISCUSSION

The range of soybean total oil content (Table 3) was 18.61 to 25.14% of seed dry mass, which was consistent with the range reported by Dardanelli et al. (2006). The five major fatty acids compositions were: P, 10.26 to 12.29%; S, 3.67 to 6.10%; O, 18.72 to 28.23%; L, 48.23 to 57.15%; and LN, 5.38 to 9.68%. These levels were in the range reported by Cherry et al. (1985). Because the treatments \times year interaction was significant for total oil content ($p < 0.001$) and fatty acids composition ($p < 0.001$), the effects of treatments were evaluated within each year and the effects of years were evaluated within the same treatment. Table 4 shows the estimated least square means from the model and significance of comparisons across treatments within years (numerator) or across years within the same treatment (denominator).

Total Oil Content

Total oil content was not significantly different among the four management systems in 1994, 1997, and 2006. However, treatment differences were found in 2000 and 2003. The LI management system resulted in significantly lower soybean total oil content than the other three management systems in 2000, while soybean grown under the CT and NT management systems had lower total oil content than the LI and ORG management systems in 2003. The results did not show a consistent trend of management system effects on total oil content. In general, the alternative agriculture management systems of NT, LI, and ORG were not found to reduce total oil content in soybean seed.

Table 3. Means and range of soybean [*Glycine max* (L.)] oil content (% of dry mass) and five major fatty acids (% of oil content) from five rotation years (1994, 1997, 2000, 2003, and 2006). Tr, treatment; CT, tilled conventional chemical input; NT, nontilled conventional chemical input; LI, tilled low chemical input; ORG, tilled no chemical input; P, palmitic acid; S, stearic acid; O, oleic acid; L, linoleic acid; LN, linolenic acid.

Tr	Oil	Mean	Range	
CT	oil content (%)	21.96 \pm 1.57	19.64–25.14	
	P	10.98 \pm 0.35	10.26–11.65	
	S	5.04 \pm 0.58	3.82–5.88	
	O	23.75 \pm 2.44	19.54–27.98	
	L	52.60 \pm 2.29	48.23–56.34	
NT	LN	7.64 \pm 1.14	5.38–9.36	
	oil content (%)	21.96 \pm 1.49	19.47–24.66	
	P	11.02 \pm 0.48	10.07–12.29	
	S	4.92 \pm 0.77	3.67–5.96	
	O	23.14 \pm 2.31	18.72–26.58	
LI	L	52.94 \pm 2.23	50.09–57.02	
	LN	7.98 \pm 1.11	6.28–9.68	
	oil content (%)	21.65 \pm 1.30	18.61–23.85	
	P	10.91 \pm 0.37	10.07–11.58	
	S	4.90 \pm 0.71	3.72–5.94	
ORG	O	23.45 \pm 2.40	19.34–27.80	
	L	52.87 \pm 2.38	48.91–57.15	
	LN	7.88 \pm 1.02	6.14–9.43	
	oil content (%)	22.01 \pm 1.09	19.89–24.19	
	P	10.93 \pm 0.28	10.36–11.44	
Difference of means between treatments	S	4.88 \pm 0.73	3.74–6.10	
	O	23.45 \pm 2.39	19.79–28.23	
	L	52.88 \pm 2.43	48.88–55.80	
	LN	7.86 \pm 1.05	6.09–9.25	
		CT – NT	CT – LI	CT – ORG
	oil content (%)	0.002	0.316	–0.045
	P	–0.041	0.074	0.047
	S	0.111	0.14	0.157
	O	0.611	0.303	0.305
	L	–0.339	–0.278	–0.287
	LN	–0.342	–0.239	–0.222

Total oil content was found to be significantly different across the five soybean rotation years for the study, as indicated in Table 4. Total oil content in 2006 was significantly higher than other years for all treatments, probably because of the weather conditions in 2006, with slightly higher daily temperatures (20.00°C) from July to September. The temporal variation of total oil content between seasons can be affected by many factors, including but not limited to seed genotype, weather conditions, and soil properties. With so many variables involved, it became difficult to evaluate the extent to which the four management systems might contribute to the temporal variability in total soybean oil content observed in this study.

Although soybean oil content as a percentage of seed weight was found to be similar among the four management systems, oil yield on a land-area basis (Table 5) was significantly different across treatments because of differences in soybean grain yield (KBS, website). In 1997, soybean grain yield under the NT management system was significantly higher than yields under the CT management system. Similarly, in 2003, plots in the NT management system had significantly higher grain yield than those in the LI and ORG systems, and in 2006, soybean yield with NT was significantly greater than CT and ORG. Therefore, given relatively consistent oil concentration levels, total soybean oil produced on a land-area basis would be expected to be greater for the higher-yielding NT management system. Long-term NT management has been reported to improve soil physical structure (Grandy et al., 2006b) and accumulate more surface soil C (Grandy and Robertson, 2007). This may explain the relatively higher soybean grain yields associated with the NT system on the relatively coarse textured KBS soils.

Fatty Acids Composition

In each year studied (Table 4), there was no significant treatment effect on O and L; in 2000 and 2006, treatment effects were not significant on P composition; in 1994, 2000, and 2003, there was no significant treatment effect on LN composition. In 1994, P composition in soybean grown with NT management was significantly greater than those with CT and ORG treatments, and S composition with NT treatments was significantly greater than CT, LI, and ORG treatments. In 1997, P levels from soybean in the NT treatments were greater than the LI treatment, S levels from the CT treatment were greater than LI and ORG treatments, and LN levels from soybean grown in CT treatments were lower than the NT, LI, and ORG treatments. In 2003, P with CT was greater than LI and ORG, and S with CT was less than NT, LI, and ORG treatments. In 2006, S with CT was less than that with NT, LI, and ORG, and LN with CT was less than NT, LI, and ORG treatments. From the results above, soybean oil from NT management systems was as high or higher in P composition than those grown under tillage management systems (CT, LI, ORG). Similarly, soybean oil from

Table 4. Least square means of total oil content (% of dry mass) and five major fatty acid compositions (% of total oil) from ANOVA MIXED Model. Tr, treatment; P, palmitic acid; S, stearic acid; O, oleic acid; L, linoleic acid; Ln, linolenic acid; CT, tilled conventional chemical input; NT, nontilled conventional chemical input; LI, tilled low chemical input; ORG, tilled no chemical input.

Tr	Total oil content	P	S	O	L	LN	O/(L + LN)
%							
1994							
CT	21.00 a/c [†]	11.29 b/a	5.44 b/a	24.91 a/ab	49.84 a/d	8.52 a/a	0.427 a/a
NT	20.61 a/b	11.76 a/a	5.81 a/a	23.54 a/b	50.51 a/d	8.38 a/b	0.400 a/bc
LI	20.96 a/bc	11.43 ab/a	5.46 b/b	25.11 a/ab	49.70 a/c	8.29 a/b	0.433 a/ab
ORG	20.70 a/b	11.26 b/a	5.41 b/b	25.30 a/ab	49.57 a/c	8.45 a/a	0.436 a/ab
1997							
CT	22.71 a/ab	10.82 ab/bc	5.27 a/ab	25.57 a/a	52.07 a/c	6.26 b/c	0.440 a/a
NT	23.18 a/a	11.04 a/b	4.98 ab/b	24.29 a/ab	52.59 a/bc	7.10 a/c	0.408 a/ab
LI	21.36 a/ab	10.57 b/c	4.70 b/c	23.73 a/bc	53.71 a/b	7.29 a/c	0.391 a/bc
ORG	21.83 a/ab	10.86 ab/bc	4.73 b/c	23.64 a/b	53.64 a/b	7.12 a/b	0.391 a/b
2000							
CT	21.75 b/bc	11.08 a/ab	3.99 a/c	19.99 a/c	55.86 a/a	9.07 a/a	0.308 a/c
NT	22.27 a/a	11.06 a/b	3.80 a/d	19.61 a/c	56.37 a/a	9.16 a/a	0.299 a/d
LI	19.75 c/c	11.13 a/ab	3.87 a/d	20.50 a/d	55.43 a/a	9.07 a/a	0.318 a/d
ORG	21.70 ab/ab	11.18 a/ab	3.80 a/d	20.89 a/c	55.22 a/a	8.91 a/a	0.326 a/c
2003							
CT	20.25 b/c	11.18 a/a	5.35 b/ab	25.43 a/a	51.37 a/cd	6.67 a/c	0.438 a/a
NT	20.41 b/b	10.80 ab/bc	5.66 ab/a	25.93 a/a	51.21 a/cd	6.40 a/d	0.450 a/a
LI	22.56 a/ab	10.73 b/bc	5.85 a/a	26.09 a/a	50.97 a/c	6.36 a/d	0.455 a/a
ORG	22.68 a/a	10.70 b/c	5.87 a/a	26.16 a/a	51.01 a/c	6.27 a/c	0.457 a/a
2006							
CT	24.26 a/a	10.53 a/c	5.13 a/b	22.85 a/b	53.83 a/b	7.66 b/b	0.372 a/b
NT	23.32 a/a	10.44 a/c	4.37 b/c	22.34 a/b	53.99 a/b	8.86 a/ab	0.356 a/c
LI	22.95 a/a	10.67 a/c	4.60 b/c	21.80 a/cd	54.59 a/ab	8.35 a/b	0.347 a/cd
ORG	23.12 a/a	10.66 a/c	4.57 b/c	21.25 a/c	54.98 a/ab	8.54 a/a	0.335 a/c

[†]Different letters in nominator indicate means are significantly different between treatments in a single year ($p < 0.05$); different letters in denominator indicate that means are significantly different between years within one treatment ($p < 0.05$).

the CT treatment had as low or lower LN composition than the other three treatments, which may have relevance to growers of low-linolenic-acid specialty soybeans. Oleic and L fatty acid levels were not affected by these four management systems during the five studied rotation cycles.

Table 5. Treatments and annual yields (Mg ha⁻¹) of soybean [*Glycine max* (L.)] from 1994 to 2006. CT, tilled conventional chemical input; NT, nontilled conventional chemical input; LT, tilled low chemical input; ORG, tilled no chemical input.

Year	Management system			
	CT	NT	LT	ORG
1994	3.01	2.86	3.09	3.19
1997	1.51 b [†]	2.04 a	1.81 a	1.65 a
2000	2.66	2.92	2.87	2.90
2003	1.62 ab	1.87 a	1.20 b	1.01 b
2006	2.87 b	3.60 a	3.21 b	2.97 b

[†]Means followed by a different letter in a row are significantly different from each other ($p < 0.05$).

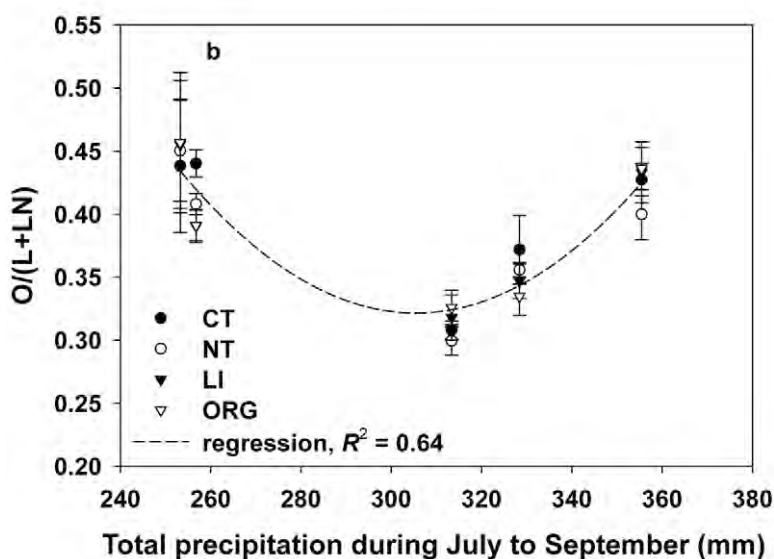
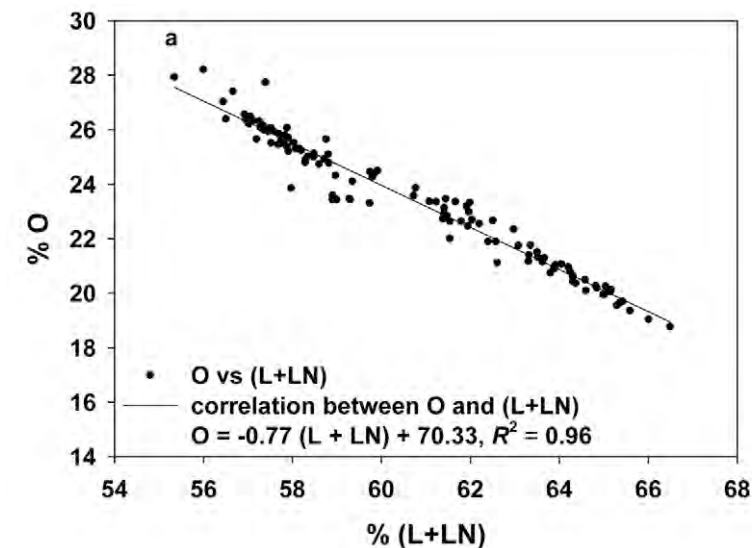


Figure 2. (a) Correlation of O and (L + LN) compositions in soybean [*Glycine max* (L.)] oil; (b) regression between the ratio of O/(L + LN) and total precipitation. O, oleic acid; L, linoleic acid; LN, linolenic acid.

Across years (Table 4), soybean oil in 2000 had lower S and O composition and higher L and LN composition than oil in the other years. Conversely, soybean oil in 2003 had higher S and O and lower L and LN levels. The LI and ORG management systems had similar soybean oil fatty acid composition trends across the rotational years of this study. The differences in fatty acids composition across years was well correlated with total precipitation.

In comparing these five major fatty acid compositions, it was found that S, O, L, and LN had strong correlations. For example, O and S had a positive correlation ($R = 0.84$, $p < 0.0001$), O and L had a negative correlation ($R = -0.90$, $p < 0.0001$), and O and (L + LN) had a strong negative correlation ($R = -0.98$, $p < 0.0001$, Fig. 2a). Similarly, Oliva et al. (2006) reported that O and L had a strong negative correlation ($R = -0.93$, $p < 0.0001$). Increasing temperature has been reported to decrease L

and LN compositions while increasing O composition (Dornbos and Mullen, 1992). Increasing O and decreasing L and LN in soybean oil profile are beneficial to the food and biodiesel industry. Therefore, the ratio O/(L + LN) could be used as a general indicator of oil quality. The average ratio in this study was 0.389 ± 0.057 . The results (Table 4) showed no management system effect on this oil quality ratio ($p > 0.05$). For the CT and NT management systems the ratio was significantly lower in 2000 than in other years; for the LI and ORG management systems the ratio was significantly lower in 2000 and 2006 than in 1994 and 2003.

Regression Model

Temperature and precipitation during the grain-fill period are considered critical to soybean total oil content and oil fatty-acid profiles (Piper and Boote, 1999). Because temperatures during these 5 yr were relatively constant (Table 2), daily temperatures (maximum, mean, minimum) from July to September did not correlate with soybean total oil content and oil fatty-acid profiles in this study. Only precipitation and its quadratic component were included in the regression model (Eq. [3]). With the full regression model, the total precipitation during seed filling explained only 0.63% variation in total oil content, indicating the total precipitation had little effect on soybean total oil content. The regression model was significant for fatty acid composition differences across years (Table 6). For S, O, L, and LN, there was no significant difference between the reduced model and the full model ($p > 0.05$), suggesting that the studied management systems had little effect on the relationship between precipitation levels and S, O, L and LN. Therefore, a common model was used to describe the relationships of S, O, L, and LN to precipitation levels for the four management systems. For P, the reduced model was found significantly different with the full model ($p = 0.014$), indicating the relationships of P to the precipitation level were different in the four management systems. Further analysis using Eq. [3] demonstrated that the relationship of P to precipitation in NT was different from those in the other three management systems, while the relationship was similar between CT, LI, and ORG. Fatty acid P in soybean grain from the NT management system was more dependent of precipitation than that from the other three management systems.

Apparently, L and O had greater slopes, thus were more sensitive to precipitation than the other three fatty acids (Table 6). Linoleic and LN had an opposite response to precipitation compared with P, S, and O. When total precipitation was in the midrange (302 to 305 mm) during July through September, the percentages of S or O

were at their minimum observed values (4.1 and 20.7%) (Fig. 3b, 3c). Conversely, in the precipitation mid-range, L and LN levels were at their maximum (55.9 and 8.7%) (Fig. 3d, 3e). The range of total precipitation from July through September in these 5 yr was 253.24 to 355.47 mm (KBS, website).

The ratio of O/(L + LN) was considered a good indicator of oil quality. Hence the regression analysis was also conducted between the ratio of O/(L + LN) and total precipitation, but the results showed little effect from the studied management systems on the regression. When total precipitation fell below 305 mm, the ratio decreased as precipitation increased, arriving at a minimum value of 0.322; when total precipitation continued to increase, the ratio also increased (Fig. 2b). When total precipitation was 265 or 345 mm, the ratio was at the mean value of 0.389, while for total oil yield (Fig. 3f, $R^2 = 0.77$), values increased as precipitation increased, arriving at a maximum value of 674 mg ha⁻¹ when total precipitation was 335 mm and then decreasing as total precipitation continued to increase. Thus, for maximum oil yield and maximum oil quality (O/(L + LN)), the optimum total precipitation in KBS area was 345 to 355 mm.

CONCLUSIONS

This research provides useful information about long-term agronomic management system effects and precipitation influence on soybean oil quality for food or biodiesel applications. The results showed total oil content was not significantly different among management systems, and the difference from year to year could not be explained by precipitation levels. However, for management systems on a land-area basis, the NT management system produced more total oil because of higher soybean grain yields. The agronomic management systems did not affect O and L fatty acid levels but did impact P, S, and LN levels. Soybeans grown under the NT management system had as high as or higher P composition than the other three management systems; similarly, the CT treatments had as low or lower LN composition in soybean when compared with the other three management systems. Oleic and L were significantly influenced by seasonal precipitation levels, while P, S, and LN were relatively stable. The oil quality ratio of O/(L + LN) had a quadratic relation with precipitation.

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Table 6. Estimated coefficients of precipitation for five fatty acids, quality ratio, and soybean [*Glycine max* (L.)] oil yield by Eq. [3]. P, palmitic acid; S, stearic acid; O, oleic acid; L, linoleic acid; LN, linolenic acid.

Variable		β_0	β_1	$\beta_2 (10^{-3})$	R^2
P	CT, LI, ORG	21.4 ±2.5	-0.074 ±0.017	0.13 ±0.03	0.28
	NT	34.2 ±5.5	-0.16 ±0.04	0.28 ±0.06	0.50
S		58.1 ±3.3	-0.32 ±0.02	0.54 ±0.04	0.64
O		176 ±11	-1.0 ±0.1	1.7 ±0.13	0.64
L	CT, NT, LI, ORG	-115 ±10	1.1 ±0.1	-1.9 ±0.1	0.72
LN		-35.7 ±4.2	0.27 ±0.03	-0.42 ±0.05	0.75
O/(L + LN)		4.18 ±0.27	-0.025 ±0.002	0.041 ±0.003	0.65
Oil yield		5030 ±680	34 ±5	-51 ±8	0.77

References

- AOCS. 2000. AM 2-93: Determination of oil content in oilseeds. *In* Official methods and recommended practices of the American Oil Chemists' Society. AOCS, Champaign, IL.
- Canakci, M., and J.V. Gerpen. 2001. Biodiesel production from oils and fats with high free fatty acids. *Trans. ASAE* 44:1429-1436.
- Cherry, J.H., L. Bishop, P.M. Hasegawa, and H.R. Leffert. 1985. Differences in the fatty acid composition of soybean seed produced in northern and southern areas of the U.S.A. *Phytochemistry* 24:237-241.
- Dardanelli, J.L., M. Balzarini, M.J. Martinez, M. Cuniberti, S. Resnik, S.F. Ramunda, R. Herrero, and H. Baigorri. 2006. Soybean maturity groups, environments, and their interaction define mega-environments for seed composition in Argentina. *Crop Sci.* 46:1939-1947.
- Davis, A.S., K.A. Renner, and K.L. Gross. 2005. Weed seedbank and community shifts in a long-term cropping systems experiment. *Weed Sci.* 53:296-306.
- Dornbos, D.L., and R.E. Mullen. 1992. Soybean seed protein and oil contents and fatty-acid composition adjustments by drought and temperature. *J. Am. Oil Chem. Soc.* 69:228-231.
- Dubois, V., S. Breton, M. Linder, J. Fanni, and M. Parmentier. 2007. Fatty acid profiles of 80 vegetable oils with regard to their nutritional potential. *Eur. J. Lipid Sci. Technol.* 109:710-732.
- Gibson, L.R., and R.E. Mullen. 1996a. Soybean seed composition under high day and night growth temperatures. *J. Am. Oil Chem. Soc.* 73:733-737.
- Gibson, L.R., and R.E. Mullen. 1996b. Soybean seed quality reductions by high day and night temperature. *Crop Sci.* 36:1615-1619.
- Grandy, A.S., T.D. Loecke, S. Parr, and G.P. Robertson. 2006a. Long-term trends in nitrous oxide emissions, soil nitrogen, and crop yields of till and no-till cropping systems. *J. Environ. Qual.* 35:1487-1495.
- Grandy, A.S., and G.P. Robertson. 2007. Land-use intensity effects on soil organic carbon accumulation rates and mechanisms. *Ecosystems* (NY, Print) 10:58-73.
- Grandy, A.S., G.P. Robertson, and K.D. Thelen. 2006b. Do productivity and environmental trade-offs justify periodically cultivating no-till cropping systems? *Agron. J.* 98:1377-1383.
- Hammond, E.G. 1991. Organization of rapid analysis of lipids in many individual plants. Springer, Berlin.
- Harder, D.B., C.L. Sprague, and K.A. Renner. 2007. Effect of soybean row width and population on weeds, crop yield, and economic return. *Weed Technol.* 21:744-752.

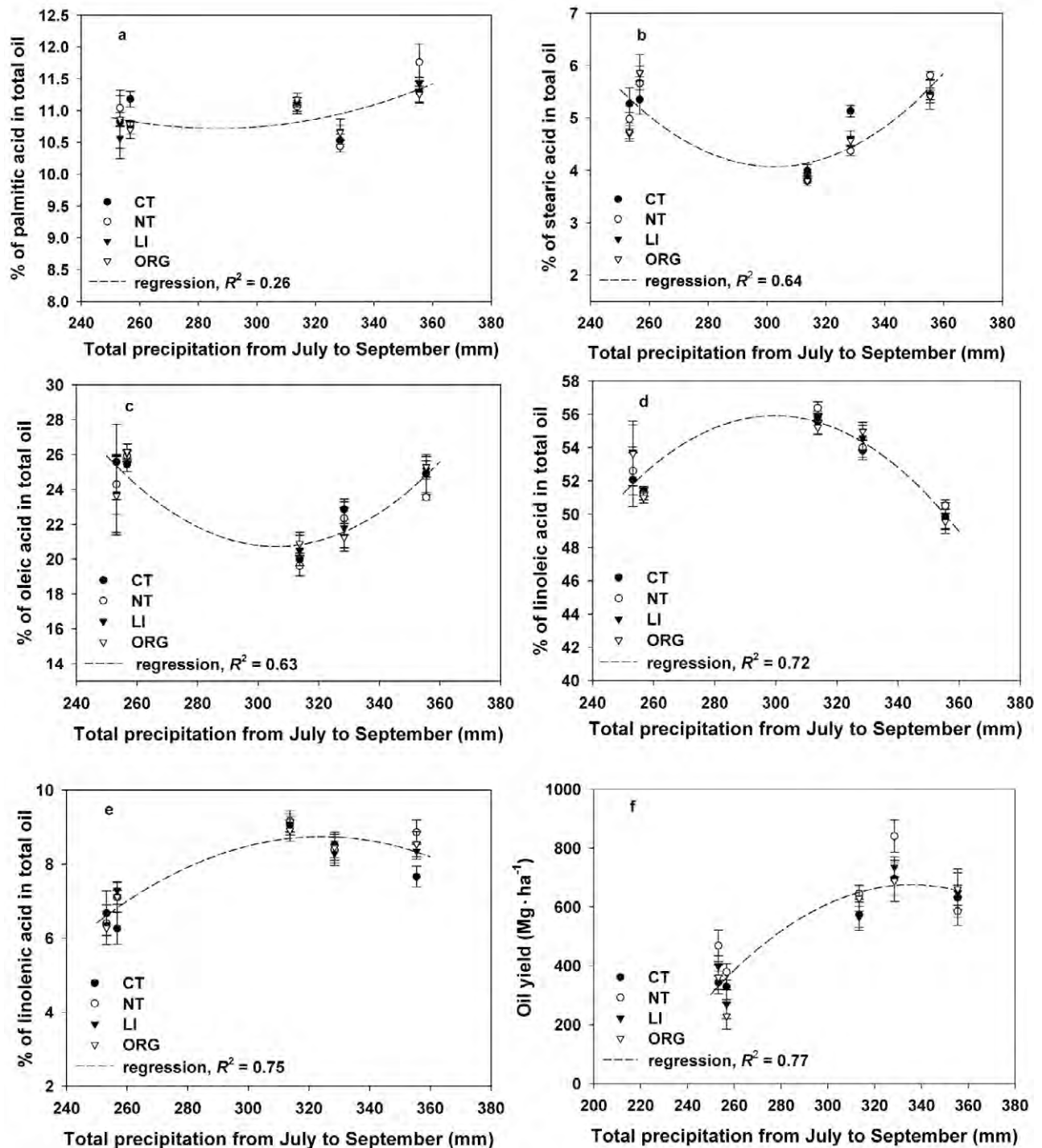


Figure 3. Five fatty acids in oil profiles and oil yield at different treatments in five rotation years with regression of precipitation: (a) palmitic acid, P; (b) stearic acid, S; (c) oleic acid, O; (d) linoleic acid, L; (e) linolenic acid, LN; (f) oil yield. CT, tilled conventional chemical input; NT, nontilled conventional chemical input; LI, tilled low chemical input; ORG, tilled no chemical input.

Hou, G., G. Ablett, K. Pauls, and I. Rajcan. 2006. Environmental effects on fatty acid levels in soybean seed oil. *J. Am. Oil Chem. Soc.* 83:759–763.

Howard, D.D., A.Y. Chambers, and G.M. Lessman. 1998. Rotation and fertilization effects on corn and soybean yields and soybean cyst nematode populations in a no-tillage system. *Agron. J.* 90:518–522.

Hussain, I., K.R. Olson, and S.A. Ebelhar. 1999. Impacts of tillage and no-till on production of maize and soybean on an eroded Illinois silt loam soil. *Soil Tillage Res.* 52:37–49.

KBS. Website. The KBS LTER Sites [Online] <http://www.lter.kbs.msu.edu/>.

Keim, K.R., A. Eren, and L.H. Edwards. 1999. Long-term study of planting date effects on seed yield in soybean. *J. Prod. Agric.* 12:288–292.

Kelley, K.W., and D.W. Sweeney. 1998. Effects of wheat-residue management on double-cropped soybean and subsequent crops. *J. Prod. Agric.* 11:452–456.

Knothe, G., and R.O. Dunn. 2001. Biofuels derived from vegetable oils and fats. *In* F.D. Gunstone and R.J. Hamilton (ed.)

- Oleochemical manufacture and applications. Sheffield Academic Press, Sheffield, UK.
- Kravchenko, A.R., G.P. Robertson, K.D. Thelen, and R.R. Harwood. 2005. Management, topographical, and weather effects of spatial variability of crop grain yields. *Agron. J.* 97:514–523.
- Kutner, M.H., C.J. Nachtsheim, J. Neter, and W. Li. 2004. *Applied linear statistical models*. 5th ed. McGraw-Hill, New York.
- Leffel, R.C., and W.D. Hanson. 1961. Early generation testing of diallel crosses of soybeans. *Crop Sci.* 1:169–174.
- Liu, K.S., E.A. Brown, and F. Orthoefer. 1995a. Fatty-acid composition within each structural part and section of a soybean seed. *J. Agric. Food Chem.* 43:381–383.
- Liu, K.S., F. Orthoefer, and E.A. Brown. 1995b. Association of seed size with genotypic variation in the chemical-constituents of soybeans. *J. Am. Oil Chem. Soc.* 72:189–192.
- Matthäus, B., and L. Brühl. 2001. Comparison of different methods for the determination of the oil content in oilseeds. *J. Am. Oil Chem. Soc.* 78:95–102.
- Oliva, M.L., J.G. Shannon, D.A. Sleper, M.R. Ellersieck, A.J. Cardinal, R.L. Paris, and J.D. Lee. 2006. Stability of fatty acid profile in soybean genotypes with modified seed oil composition. *Crop Sci.* 46:2069–2075.
- Piper, E.L., and K.J. Boote. 1999. Temperature and cultivar effects on soybean seed oil and protein concentrations. *J. Am. Oil Chem. Soc.* 76:1233–1241.
- Primomo, V.S., D.E. Falk, G.R. Ablett, J.W. Tanner, and I. Rajcan. 2002. Genotype \times environment interactions, stability, and agronomic performance of soybean with altered fatty acid profiles. *Crop Sci.* 42:37–44.
- Sato, K., and T. Ikeda. 1979. Growth-responses of soybean plant to photoperiod and temperature. 4. Effect of temperature during the ripening period on the yield and characters of seeds. *Jpn. J. Crop. Sci.* 48:283–290.
- Sheaffer, C.C., and K.M. Moncada. 2008. *Introduction to agronomy, food, crops, and environment*. CENGAGE Delmar Learning, Clifton Park, NY.
- Smith, R.G., F.D. Menalled, and G.P. Robertson. 2007. Temporal yield variability under conventional and alternative management systems. *Agron. J.* 99:1629–1634.
- soystat. 2008. World soybean production 2007. Available at <http://www.soystats.com/2008/Default-frames.htm>. United Soybean Board.
- Swanton, C.J., B.D. Booth, K. Chandler, D.R. Clements, and A. Shrestha. 2006. Management in a modified no-tillage corn-soybean-wheat rotation influences weed population and community dynamics. *Weed Sci.* 54:47–58.
- Urbanchuk, J.M. 2006. Contribution of the biodiesel industry to the economy of the United States. Available at http://www.biodiesel.org/resources/reportsdatabase/reports/gen/20060930_gen373.pdf.
- Wilcox, J.R., and J.F. Cavins. 1992. Normal and low linolenic acid soybean strains: Response to planting date. *Crop Sci.* 32:1248–1251.
- Wolf, R.B., J.F. Cavins, R. Kleiman, and L.T. Black. 1982. Effect of temperature on soybean seed constituents: Oil, protein, moisture, fatty-acids, amino-acids, and sugars. *J. Am. Oil Chem. Soc.* 59:230–232.
- Wright, A.L., and F.M. Hons. 2004. Soil aggregation and carbon and nitrogen storage under soybean cropping sequences. *Soil Sci. Soc. Am. J.* 68:507–513.