Mineral Composition of Cottonseed is Affected by Fertilization Management Practices

Zhongqi He,* Mark Shankle, Hailin Zhang, Thomas R. Way, Haile Tewolde, and Minori Uchimiya

ABSTRACT

Cottonseed and its products can be used as human food, animal feed, and industrial raw material. Chemical composition is one of the critical parameters to evaluate cottonseed's quality and potential end use. Whereas there are reports on the improvement of cotton (*Gossypium hirsutum* L.) lint yield by poultry litter (PL) fertilization, there are no data about the impact of PL application on mineral composition of cottonseed. In this study, we collected seed samples in 2 yr from cotton fertilized with PL and chemical inorganic fertilizer (CIF) with or without winter wheat (*Triticum aestivum* L.) cover crop (CC) at the Mississippi Agricultural and Forest Experiment Station. Contents of 18 elements (Al, As, B, C, Ca, Cd, Cu, Fe, K, Mg, Mn, N, Na, Ni, P, Pb, S, and Zn) and ash in these cottonseed samples were determined by inductively coupled plasma-based or dry combustion methods. Cover crop did not significantly impact the chemical composition of cottonseed. Compared to no fertilizer control, both CIF and PL altered the levels of Ca, Mg, Cu, Fe, Mg, Mn, P (possibly Na and K), and ash contents but not C, N, Zn, Ni, Cd, Pb, Al, S, B and As. The impact of subsurface band vs. broadcast application methods or fall vs. spring application timing on mineral composition was not obvious. Cottonseed K, Mg, and P contents were higher with PL fertilization but lower with CIF than those of the control cottonseed (no fertilizer). The data of the three elements were highly correlated ($\alpha < 0.01$), indicating the three elements were co-accumulated in cottonseed.

Cotton is an essential source of natural fiber. For every 100 kg of lint fiber ginned from cotton, 150 kg of cottonseed is produced (Yu et al., 2012). Whole cottonseed and its products can be used as human food, animal feed, and industrial raw materials. For example, cottonseed oil is used as edible oil in food processing and restaurants (O'Brien and Wakelyn, 2005), and has the potential to be used as biofuel (Meneghetti et al., 2007). Whole seed and cottonseed meal are valuable dairy feedstuff (Arieli, 1998; Bertrand et al., 2005). Although not widely used, cottonseed meal can be used as an adhesive for wood-gluing applications (Hogan and Arthur, 1951; Lambuth, 2003). The chemical composition is an important parameter in evaluating cottonseed quality for different applications. Genetic, environmental, and agronomic approaches have been used to alter cottonseed composition (Cherry et al., 1978). Cottonseed oil and protein contents can vary from 17 to 27% and 12 to 32%, respectively, among genetic variations (Dowd et al., 2010; Kohel et al., 1985; Yu et al., 2012). Elmore et al. (1979) found that N fertilization increased N levels in cottonseed with altered amino acid concentration. Pettigrew and Dowd (2011) reported that varying planting dates or irrigation regimes altered cottonseed composition in terms of protein, oil, gossypol, and soluble carbohydrates.

Poultry litter (a mixture of manure and other external materials, such as bedding materials) is a byproduct of the poultry industry and contains valuable plant nutrients. Managed appropriately, land application of PL as a fertilizer is an efficient and environmentally-acceptable method of recycling nutrients and organic matter. Long-term application (up to 20 yr) of PL into pasture soil continuously increased the hay yield in the Sand Mountain region of Alabama (He et al., 2008). Similarly, fertilizing cotton plant with PL often results in a yield increase (Endale et al., 2002; Reddy et al., 2007; Tewolde et al., 2009a). However, the information on the effect of PL application on mineral composition of plants is very limited even though PL contains many mineral elements (Schroder et al., 2011). Citak and Sonmez (2010) evaluated the effects of the types of fertilizers (farmyard manure, chicken manure, blood meal, and chemical fertilizer) on nutrient content of the edible part of cabbage (*Brassica oleracea*) plants during two consecutive seasons. They found that the mineral contents of cabbage receiving organic applications tended to be higher than cabbage receiving chemical fertilizers, and on the whole, cabbage responded the best to farmyard and chicken manure as a mixture or separately. Demir et al. (2010) measured 17 elements (N, P, K, Ca, Mg, S, Fe, Zn, Cu, Mn, Mo, Cl, Si, Br, Rb, Sr, and Ba) in tomato (*Lycopersicon esculentum* Mill.) leaves and fruits grown in soil fertilized with poultry manure. These researchers concluded that the increased fruit yield, the increased concentration of

Z. He, and M. Uchimiya, USDA-ARS, Southern Regional Research Center, 1100 Robert E Lee Blvd., New Orleans, LA 70124; H. Tewolde, USDA-ARS, Crop Science Research Laboratory, Mississippi State, MS 39762; H. Zhang, Dep. of Plant and Soil Sciences, Oklahoma State University, Stillwater, OK 74078; T.R.Way, USDA-ARS, National Soil Dynamics Laboratory, Auburn, AL 36832; M. Shankle, Pontotoc Ridge-Flatwoods Branch Experiment Station, Mississippi State Univ., Pontotoc, MS 38863. Received 13 Sept. 2012. *Corresponding author (zhongqi.he@ars.usda.gov).

Published in Agron. J. 105:341–350 (2013)
doi:10.2134/agronj2012.0351

Copyright © 2013 by the American Society of Agronomy, 5585 Guilford Road, Madison, WI 53711. All rights reserved. No part of this periodical may be reproduced or transmitted in any form or by any means, electronic or mechanical, including photocopying, recording, or any information storage and retrieval system, without permission in writing from the publisher.

Abbreviations: CC, cover crop; −CC, without cover crop; +CC, with cover crop; CIF, chemical inorganic fertilizer; PL, poultry litter.

Table 1. pH and concentration of selected nutrients in poultry litter and background soil taken to a depth of 15 cm on 5 Dec. 2007 and analyzed by the Mehlich 3 procedure (Mehlich, 1984).

Year	pH	Moisture	Total N	Total C	Ca	K	Mg	Na	P	B	Cu	Fe	Mn	Zn
		g kg^{-1}								mg kg^{-1}				
Soil	6.46		0.877	9.909	1.175	0.081	0.024		0.050		0.29	254	71.8	1.26
Litter														
2008	7.16	323	28.7	254.2	19.6	26.8	5.84	7.80	12.2	40.0	296	455	519	411
2009	7.12	362	27.3	224.6	20.6	27.8	6.14	8.46	14.2	48.0	291	542	531	402
2010	7.68	239	23.8		17.4	26.6	5.87	6.12	12.8	35.9	285	703	544	413

Table 2. Monthly maximum and minimum air temperature and total precipitation at the experimental site in 2009 and 2010.

Month	2009			2010		
	Maximum temperature	Minimum temperature	Total precipitation	Maximum temperature	Minimum temperature	Total precipitation
	°C		mm	°C		mm
January	10.3	–1.0	83	7.5	–3.0	137
February	15.2	0.9	102	7.9	–2.5	77
March	17.7	6.1	157	15.4	3.6	98
April	22.3	9.0	71	24.5	10.1	55
May	25.5	16.0	285	27.1	16.2	220
June	31.9	19.9	55	32.5	21.5	54
July	30.2	19.7	200	33.2	22.4	68
August	30.3	19.6	169	34.0	22.8	48
September	27.5	18.2	195	31.3	17.0	43
October	20.1	9.9	324	25.9	9.0	71
November	17.5	5.1	55	18.0	4.9	139
December	9.1	–0.3	96	9.4	–1.7	50
Avg./sum	21.5	10.3	1791	22.2	10.0	1059

Zn (an element required in the human diet) and the lower concentration of potentially harmful Br in the fruit make poultry manure a valuable soil amendment for tomato production. Tewolde et al. (2011) comparatively evaluated mineral nutrients of mid-season cotton plants fertilized with PL and ammonium nitrate. Their data suggested that the better yield performance of cotton fertilized with PL compared to ammonium nitrate fertilization was due to a more ideal soil pH, favorable tissue Mn concentration, and improved K and B nutrition. They did not report effects of PL application on the mineral composition of cottonseed. Therefore, we collected cottonseed samples in two successive years from cotton fertilized with PL and chemical fertilizer with or without CC, and determined the ash and 18 elemental contents of these samples. The objective of this study was to evaluate the mineral profiles of cottonseed as affected by CIF and PL fertilization.

MATERIALS AND METHODS

Field Study

Cotton (cultivar DP143B2RF and DP0924B2RF for 2009 and 2010, respectively) seed was collected after harvest in 2009 and 2010 from research conducted in 2008 to 2010 at the Mississippi Agricultural and Forest Experiment Station near Pontotoc, in a Falkner silt loam soil (fine-silty, siliceous, active, thermic Aquic Paleudalfs). Initially, the soil had a pH of 6.4 (1:1 soil/water), and total N and organic C of 0.88 and 9.9 g kg^{-1}, respectively (Table 1). The experiment consisted of a factorial combination of three management factors. The factors included PL application methods, application timing, and winter CC. Poultry litter (5.6 Mg ha^{-1} was subsurface banded in the spring (LSS) or fall (LSF) or surface broadcast in the spring (LBS) or fall (LBF) with or without a winter CC. These factors were compared against a standard commercial fertilization (CIF) and an unfertilized control (Con) in a randomized complete block design with a split-plot arrangement of the treatments replicated four times. Winter CC was applied to the main plot and a factorial combination of the application method and timing to subplots. Each subplot consisted of four 1.02-m wide rows with 21.3-m length. The field was managed as a no-till system. For consistent observations, each plot received the same treatment over the experimental years. Monthly air temperatures and total precipitation at the experimental site in 2009 and 2010 are listed in Table 2.

Poultry litter was obtained from a local broiler chicken operation in the fall before each season and the amount for each subplot weighed after passing the PL through a device that broke large caked-manure pieces to pass a 12-mm screen and homogenizing the litter (Table 1). The litter for the fall application timing was immediately applied and that for the spring application was placed in large plastic bags and stored in covered plastic tubs under shade during the winter until application in the spring. The fall PL applications were made on 11 Feb. 2008 (delayed due to wet Fall), 6 Nov. 2008, and 24 Nov. 2009 and the spring applications on 7 May 2008, 19 May 2009, and 22 Apr. 2010. The litter for the surface broadcast was applied by hand. The litter for the subsurface band was applied with a four-row patented prototype implement that opens a 5 cm-wide trench and places the litter about 5 cm below the soil surface (Pote et al., 2011; Way et al., 2010). The litter bands were spaced 1.02 m apart which was the same spacing as the

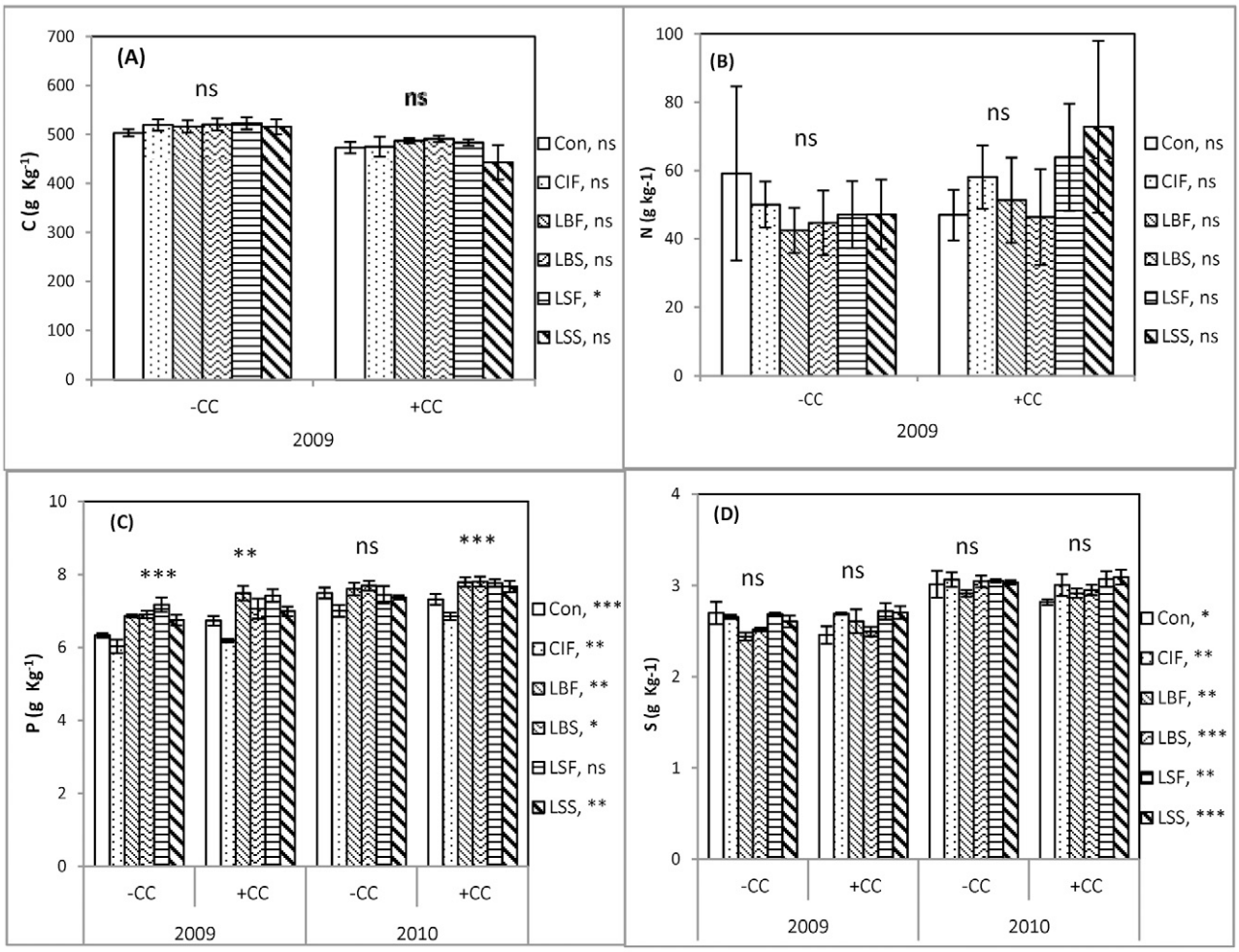

Fig. 1. Contents of nonmetal C, N, P, and S in seed harvested from cotton grown in 2009 and 2010 with (+CC) or without (–CC) winter cover crop management. See the Field Study description for fertilization treatment abbreviations. Data are averages of field triplicates with standard error bars. Symbols *, **, *, and ns are for statistically significant impact at α = 0.05, 0.01, 0.001 and not significant at α = 0.05, respectively, by the fertilization (symbols at the top of above the data bars) or cover crop treatments under each fertilization (symbols besides the fertilization legends).**

cotton rows. The CIF treatment received 101 kg ha^{-1} N as urea-ammonium nitrate solution as a sidedress around the first square stage each season on 10 July 2008, 10 July 2009, and 9 July 2010. The CIF treatment also received 84 kg K ha^{-1} as KCl in 2008 and 121 kg K ha^{-1} in 2009 and 2010 based on soil test recommendations from Mississippi State University. No P fertilization was necessary according to the recommendation.

Each season, wheat CC was planted shortly before or after applying the litter in the fall and killed 2 to 3 wk before planting cotton on 19 May 2008, 21 May 2009, and 26 May 2010 with a four-row no-till planter. The cotton was planted 15 cm away from the litter band at the same 1.02-m row spacing as the litter bands. Cotton from the middle two rows was harvested on 22 Oct. 2008, 12 Nov. 2009, and 29 Sept. 2010 with a two-row spindle picker.

Seed Sample Preparation

About 1.0-kg subsample was taken from the cotton harvested in 2009 and 2010, ginned on a 10-saw tabletop gin, and the cottonseed delinted with concentrated H_2SO_4 and dried in a forced-air oven at 65°C. Approximately 120 g dried seeds from each sample were then ground in a stainless steel coffee grinder and sieved to pass a 0.5 mm stainless steel sieve (no. 35 U.S. Standard Test Sieve). Part of the sample that did not pass through this sieve was further ground in the coffee grinder and sieved again. The part of samples that was too persistent for the coffee grinder and did not pass this sieve was ground in a stainless steel Wiley Mill to pass a 1-mm screen (no. 20) and thoroughly blended with the remainder of the sieved sample. The portion that had to be ground by the Wiley Mill was mostly hull pieces and was only a small fraction of the total.

Ash and Elemental Analysis

Seed ash content was determined after 1.0 g of each ground sample was ashed in a muffle furnace at 550°C. The mineral content of the seed was analyzed following the digestion (Jones and Case, 1990) in which 0.50 g of ground cottonseed sample was digested in 10.0 mL of concentrated trace metal grade HNO_3 for 1 h in the HotBlock Environmental Express block digester. The sample was heated to 115°C for 2 h and 15 min, then cooled and brought to 50mL with deionized (DI) water. The concentrations of 13 elements (i.e., Al, B, Ca, Cu, Fe, K, Mg, Mn, Na, Ni, P, S, and Zn) in these digests were determined by a Spectro CirOs ICP spectrometer (Mahwah, NJ). The concentrations of As, Pb, and Cd in these digests were determined separately using the same instrument to have better detection

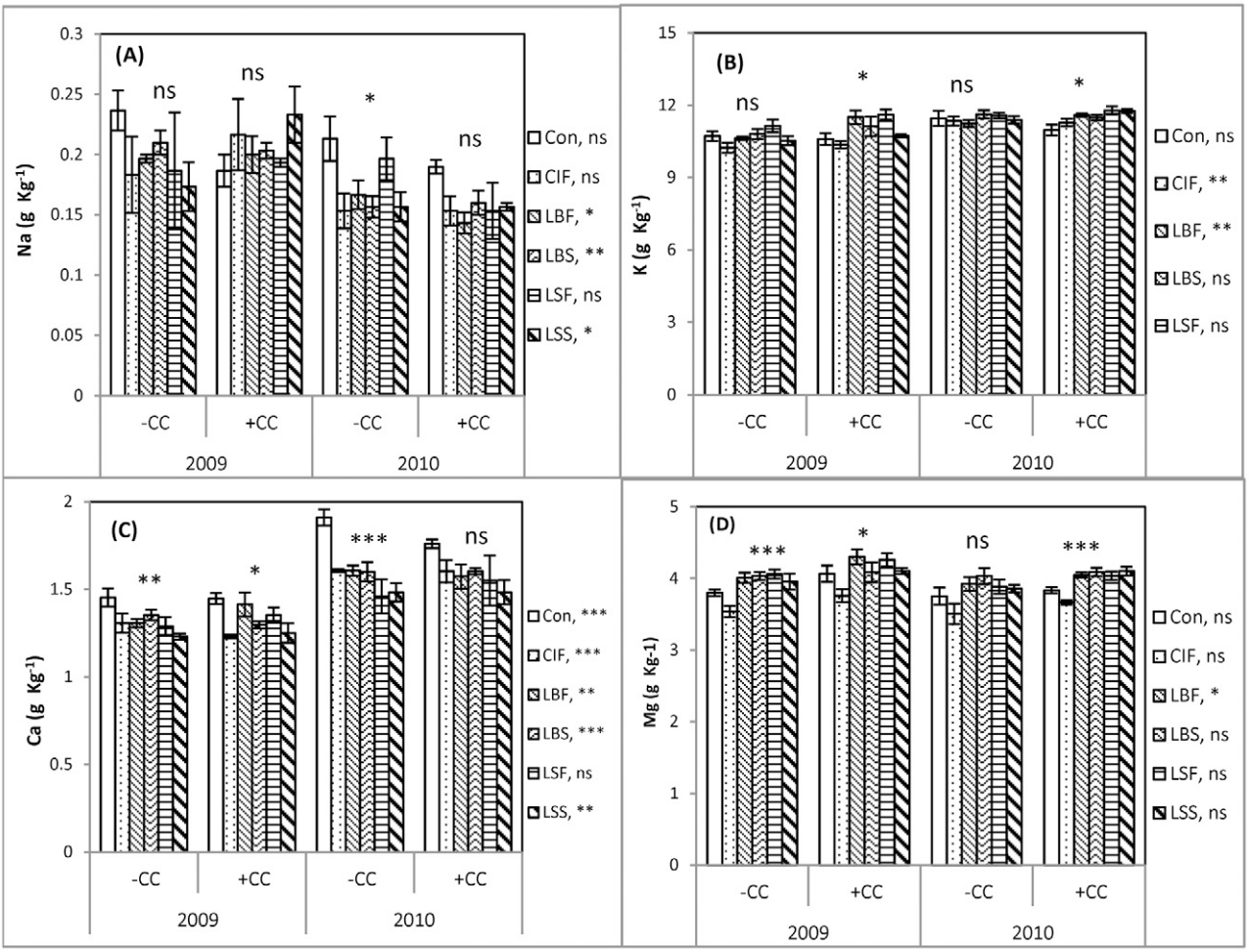

Fig. 2. Contents of macro metal K, Na, Ca, and Mg in seed harvested from cotton grown in 2009 and 2010 with (+CC) or without (−CC) winter cover crop management. See the Field Study description for fertilization treatment abbreviations. Symbols *, **, *, and ns are for statistically significant impact at α = 0.05, 0.01, 0.001 and not significant at α = 0.05, respectively, by the fertilization (symbols at the top of the data bars) or cover crop treatments under each fertilization (symbols besides the fertilization legends).**

limits. The concentrations of C and N were determined by dry combustion using PerkinElmer 2400 Series II CHNS/O analyzer (PerkinElmer, Shelton, CT).

Statistical Analysis

Although there were four field replicates for each year, the laboratory elemental analysis of cottonseed samples was performed only on three field replicates. The data analysis package in Microsoft Excel 2007 was used for statistical analysis. The Descriptive Statistics Tool Data was used to calculate averages and standard errors. "Single-factor" and "two-factor with replicates" ANOVA were used to evaluate the effects of management treatments on changes of element and ash contents of cottonseed. The Correlation Analysis Tool was used to analyze correlation coefficients between element and ash contents.

RESULTS

Nonmetal Elements (Carbon, Nitrogen, Phosphorus, Sulfur, Boron, and Arsenic)

Based on the 2009 data, the contents of C and N were 495.8 ± 23.4 and 52.5 ± 8.4 g kg^{-1}, respectively. The management practices did not significantly ($\alpha = 0.05$) impact either C or N content in cottonseed (Fig. 1A and 1B). Seed P content varied from 6.0 to 7.8 g kg^{-1} and S content varied from 2.4 to 3.1 g kg^{-1} (Fig. 1C and 1D). The P content of the cottonseed samples with the same fertilization, except that of PL applied subsurface band in Fall (labeled as LSF), was significantly different ($\alpha \leq 0.05$) among the four sets of data (Fig. 1C). Our data indicated that the CC management increased the P content of cotton seed slightly; however, there were major differences within growing season/year. For example, the averages of the P content was 6.68, 6.95, 7.44 and 7.53 mg kg^{-1}, for 2009−CC, 2009+CC, 2010−CC, and 2010+CC, respectively (2009 and 2010 treatments without [−CC] and with [+CC] cover crop management). Compared to the no-fertilizer control, CIF fertilization decreased the P content, but PL fertilization increased the P content. No apparent differences between the PL application methods were observed. On average, CIF decreased the P content of cottonseed by 6.5% and PL increased the P content by 5.6% relative to no fertilization. The difference in the P content between CIF and PL samples was 13.0%. The cottonseed S content was not influenced by the treatments but the 2 yr differed substantially (Fig. 1D). The average of the S content was 2.60, 2.61, 3.02, and 2.98 mg kg^{-1}, for 2009−CC, 2009+CC, 2010−CC, and 2010+CC,

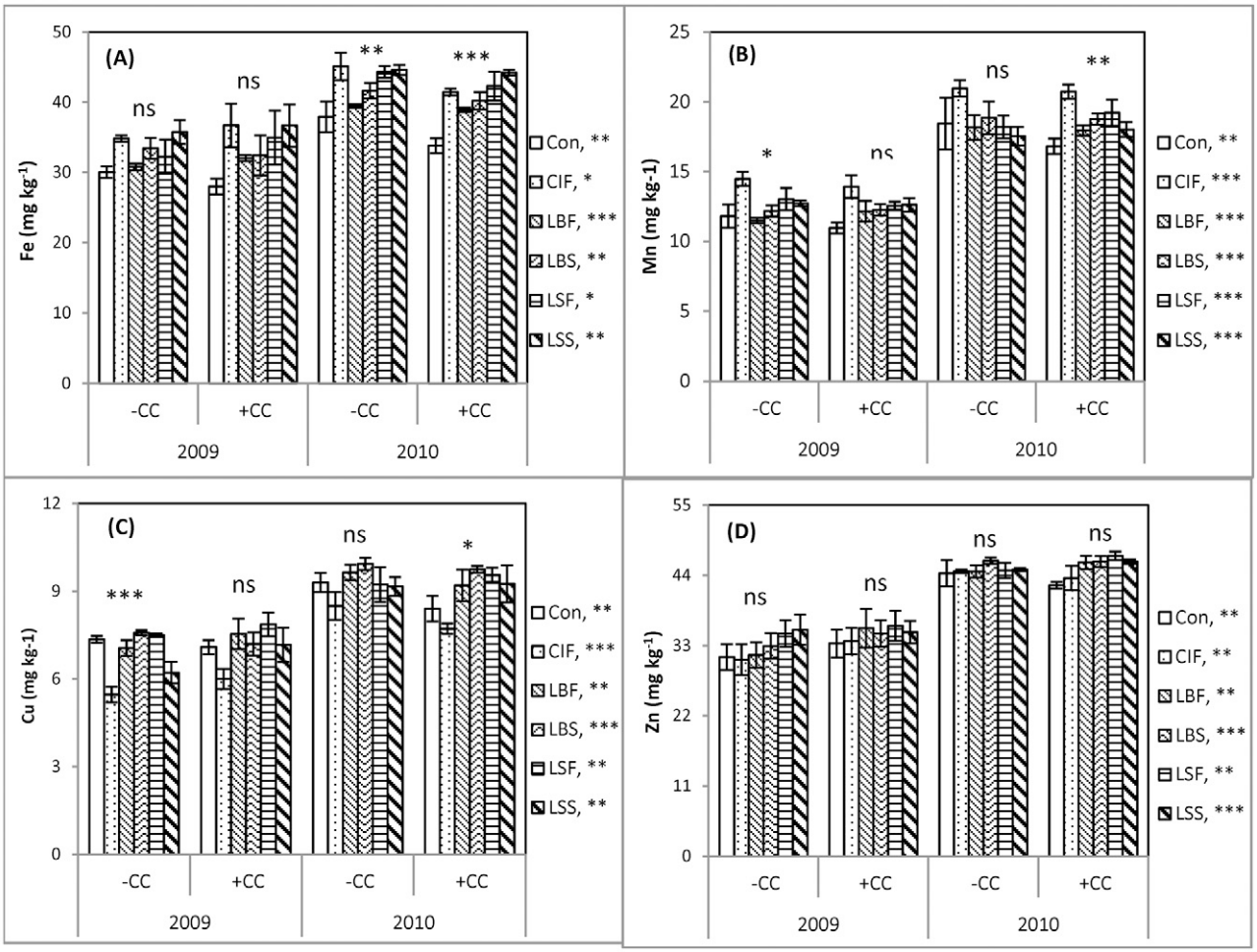

Fig. 3. Contents of micrometal Fe, Mn, Cu, and Zn in seed harvested from cotton grown in 2009 and 2010 with (+CC) or without (–CC) winter cover crop managements. See the Field Study description for fertilization treatment abbreviations. Symbols *, **, *, and ns are for statistically significant impact at α = 0.05, 0.01, 0.001 and not significant at α = 0.05, respectively, by the fertilization (symbols at the top of the data bars) or CC treatments under each fertilization (symbols besides the fertilization legends).**

respectively. Similar to P, S content was also affected by growing years, but not by the CC or fertilization.

The contents of micro nonmetal elements B and As in those cottonseed samples varied from 16.8 to 20.5 mg kg^{--1} and from 0.004 to 0.011 mg kg^{-1}, respectively (Fig. 1C and 1D). However, fertilization and CC management did not significantly ($\alpha \leq 0.05$) affect these micro nonmetal elements.

Macrometal Elements (Potassium, Sodium, Calcium, and Magnesium)

On average, the Na content was 0.18 g kg^{-1} of cottonseed (Fig. 2A). Compared to the unfertilized control, fertilization lowered Na content in most samples. However, the variances of the field replicates were so large that three of the four groups of data were not statistically significant at $\alpha \leq 0.05$. The average of K content of cottonseed was 11.1 g kg^{-1} or 50-fold higher than the total Na content (Fig. 2B). The K content in these cottonseed samples without the cover crop management (–CC) was basically unchanged over the fertilization treatments. On the other hand, both years' data showed fertilization treatments with the CC management significantly impacted the K content in cottonseed. Relative to the unfertilized control, PL application with CC management increased the K content in cottonseed by an average of 6.8% in 2009 and 6.3% in 2010.

Higher Ca content was detected in 2010 than 2009 samples (Fig. 2C). There were no consistent effects of CC management on the Ca content. The Ca content in the four control cottonseed samples was 1.45, 1.45, 1.91, and 1.76 g kg^{-1}, respectively. The averages across the five PL- and chemical-fertilized treatments for 2009–CC, 2009+CC, 2010–CC, and 2010+CC were 1.31, 1.31, 1.55, and 1.56 g kg^{-1}, respectively. This suggests that fertilization lowered the Ca content in cottonseed. The effects of fertilizer type and application method, however, were not obvious. On average, fertilization reduced the Ca content by 10.7, 9.5, 18.5, and 11.1%, respectively, for 2009 and 2010 data without or with CC managements. Compared to the Ca data, the Mg content varied less with the year and CC factors when the fertilization treatments were the same (Fig. 2D). Similar to the Ca content, the Mg content was lower in the cottonseed with chemical fertilizer, compared to the control. However, unlike the Ca content data, the Mg content was higher in the cottonseed samples with PL than the corresponding Control samples. The Mg content in the four fertilizer controls of –CC and +CC samples of 2009 and 2010 ranged from 3.75 to 4.07 g kg^{-1}. The CIF decreased the Mg content by 6.8, 7.6, 6.4, and 4.4%, in the same order. The PL increased the Mg content by 5.8, 2.9, 4.8 and 6.0% under the same managements. Combining the four sets of data, the average decrease

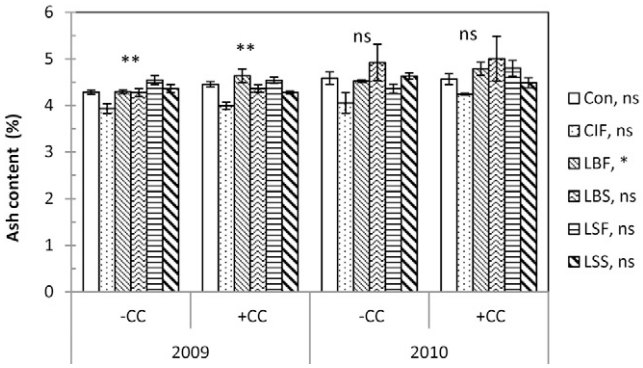

Fig. 4. Ash content of seed harvested from cotton grown in 2009 and 2010 with (+CC) or without (–CC) winter cover crop management. See the Field Study description for fertilization treatment abbreviations. Symbols *, **, and ns are for statistically significant impact at α = 0.05, 0.01, 0.001 and not significant at α = 0.05, respectively, by the fertilization (symbols at the top of the data bars) or CC treatments under each fertilization (symbols besides the fertilization legends).

by CIF was 6.3%; the average increase by PL was 4.9%. The difference in the Mg content between CIF and PL samples was 12.2%, comparable to that change of P content in these cottonseed samples.

Micrometal Elements (Iron, Manganese, Copper, Zinc, Nickel, Cadmium, Lead, and Aluminum)

Within each fertilization treatment, CC significantly affected the four metal Fe, Mn, Cu, and Zn contents in the cottonseeds ($\alpha \leq 0.05$) (Fig. 3). Additionally, the four elements were higher in all 2010 samples than in the corresponding 2009 samples without or with the CC managements. For example, the Fe content was 32.9, 33.5, 42.2, and 40.2 mg kg^{-1}, for 2009–CC, 2009+CC, 2010–CC, and 2010+CC, respectively. The 2010 cottonseed samples had greater micrometals content than the 2009 samples and the differences depended on the presence or absence of CC. For example, the Fe, Mn, Cu, and Zn content of the 2010 cottonseed samples exceeded those of the 2009 samples by 28.4, 48.1, 35.4, and 36.9% with CC and 20.0, 49.6, 25.8, and 30.0% without CC, respectively.

Fertilization affected the contents of the four metals in cottonseed in different patterns. The impacts of fertilization treatments on the Fe content in cottonseed were similar in the four sets of data although only the 2010 data were statistically significantly affected (Fig. 3A). Compared to the corresponding controls, chemical fertilization increased the Fe content in cottonseeds by 16.0, 31.1, 19.0 and 22.7%,for 2009–CC, 2009+CC, 2010–CC, and 2010+CC, respectively. This represents an average increase of 22.2%. Poultry litter application also increased the Fe content but to a small extent. The general impact order of PL application methods on the Fe content was LBF $\leq$ LBS $\leq$ LSF $\leq$ LSS as shown in Fig. 3A. The average increases in the two seasons without or with CC managements were 9.0, 14.0, 18.6, and 24.6% with the four PL applications compared to the control. These observations implied that PL applied in Spring and/or with subsurface band application would exert greater impacts on the Fe content in cottonseed. The levels of Mn in cottonseeds ranged from 11 to 21 mg kg^{-1}, less than a half of that of Fe (Fig. 3B). Similar to the Fe content, chemical inorganic fertilizer increased the Mn content with an average of 21.7%, compared to that of control. The PL application had less impacts and the differences between the PL application methods were not distinct. The Mn content in the cottonseed with PL was only 6.6% higher than the control. On average, the Fe content in 2010 samples was 28.4 and 20.1% higher than those in 2009 samples, respectively, without and with the CC. The corresponding Mn content in 2010 samples was 48.5 and 49.7% higher than those in 2009 samples.

The Cu levels in these cottonseed samples were not >10 mg kg^{-1} (Fig. 3C). In contrast to Fe and Mn, the Cu content in cottonseed with chemical fertilizer was lower than the corresponding controls by 25.7, 15.3, 8.6 and 8.2% for 2009–CC, 2009+CC, 2010–CC, and 2010+CC, respectively. On the other hand, no consistent effects of PL application and the application methods on the Cu content were observed. By the averages of LBF, LBS, LSF and LSS, PL application changed the Cu content by –3.5, 5.0, 2.1, and 12.3%, compared to the corresponding controls in 2009 and 2010 seasons without or with CC managements. The Zn content in cottonseed seemed not impacted by fertilization managements (Fig. 3D).

Among other four (i.e., Ni, Cd, Pb, and Al) elements, the Ni levels in all cottonseed digests including those with PL fertilization was below the digest blank (0.3 mg kg^{-1}), indicating the very low Ni content in all these cottonseed samples. The contents of two non-nutrient elements, Cd and Pb, were detectable, but extremely low in these cottonseed samples. For example, Cd content ranged from 0.000 to 0.003 mg kg^{-1} whereas Pb content ranged from 0.001 to 0.017 mg kg^{-1}. Another non-nutrient element Al was relatively higher in the cottonseed samples, ranged from 33 to 42 mg kg^{-1}. None of the four element (Ni, Ca, Pb, and Al) contents in the cottonseeds were significantly ($\alpha = 0.05$) changed by any of the cropping management factors we tested.

Ash Content

The ash content was 4.28, 4.45, 4.58, and 4.56% of dry matter, for 2009–CC, 2009+CC, 2010–CC, and 2010+CC, respectively. Among the six sets of data with the same fertilization treatments, only one set (LBF) was significantly different at $\alpha = 0.01$(Fig. 4), indicating little change of the ash content in cottonseed over growing year and CC managements. The changes of ash content impacted by the fertilization treatments was statistically significant ($\alpha = 0.01$) for 2009 data, but not for 2010 data even though the impact patterns of the 2-yr data were similar. In all four sets of data, the ash content of cottonseed with chemical fertilizer, 3.93, 3.99, 4.05, and 4.24% of dry matter, was lower than the corresponding control samples. Comparison of the four groups of data led us to conclude that the methods of PL application showed inconsistent, little effects on the ash content of these cottonseed samples. Thus, we calculated the averages of ash content of the four PL applications (LBF, LBS, LSF, and LSS) as 4.37, 4.45, 4.61, and 4.77% of dry matter, for 2009–CC, 2009+CC, 2010–CC, and 2010+CC, respectively. These data indicate that the ash content of cottonseed with PL fertilization was higher than that of cottonseed with chemical fertilizer. The differences were 0.44, 0.46, 0.56 and 0.53% of dry matter between CIF- and PL-applied samples for 2009–CC, 2009+CC, 2010–CC, and 2010+CC. On average, the ash content in PL-applied

Table 3. Effects of fertilization (Fer) and cover crop (CC) treatments on selected elements and ash contents in cottonseed based on the averages of 2-yr data. (+CC and –CC: with or without winter cover crop management).

Treatment	P		S	K	Mg	Ca	Fe	Mn	Cu	Ash
ANOVA	$P > F$									
CC	0.006		0.566	0.067	0.626	0.892	0.261	0.676	0.906	0.078
Fer	<0.001		0.012	0.002	0.001	<0.001	<0.001	0.002	<0.001	<0.001
Fer × CC	0.566		0.067	0.120	0.937	0.590	0.544	0.838	0.243	0.522
Content	g kg^{-1}						mg kg^{-1}			%
	–CC	+CC								
Con†	6.91ab‡	7.03b	2.75abc	10.9ab	3.81ab	1.64c	32.4a	14.5a	8.03b	4.47b
CIF	6.52a	6.52a	2.86bc	10.8a	3.59a	1.44ab	39.5d	17.5b	6.92a	4.05a
LBF	7.24bcd	7.64c	2.72a	11.2bc	4.04c	1.48b	35.3b	14.9a	8.36bc	4.56b
LBS	7.30d	7.43bc	2.75abc	11.3bc	4.02c	1.46b	36.9c	15.5a	8.61c	4.64b
LSF	7.31cd	7.59c	2.88bc	11.5c	4.03c	1.41ab	38.5cd	15.8a	8.54bc	4.56b
LSS	7.06bc	7.33bc	2.86b	11.1b	3.98bc	1.36a	40.3d	15.2a	7.95b	4.44b

† Con, control; CIF, chemical inorganic fertilizer; LBF, surface broadcast in the fall; LBS, surface broadcast in the spring; LSF, subsurface banded in the fall; LSS, subsurface banded in the spring.

‡ Content values followed by different letters within the same column are significantly different at $\alpha \leq 0.05$.

cottonseed was 0.5% of dry matter higher than that in inorganic fertilizer-applied cottonseed, representing 12.3% change of ash content.

DISCUSSION

Among the 18 elements we tested, 13 (C, N, P, K, Ca, Mg, S, B, Cu, Fe, Mn, Ni, and Zn) are essential elements for plant growth. Copper, Mn, Ni, and Zn are also potentially toxic to plants when concentrations are too high. Aluminum, As, Cd, and Pb are nonessential elements whose presence are of concern to both human and ecosystem health (Fageria et al., 2002; Bolan et al., 2004; Schroder et al., 2011). Sodium is a non-essential element for plants although there are reports that some crops benefit from Na in the soil (Dias et al., 2009; Helmke and Sparks, 1996). Modern chemical inorganic fertilizer applications supply P, K, and several other minerals to soil to promote plant growth. Poultry litter can provide more minerals to soil as those minerals have been added into poultry feed for various purposes and excreted into PL (Schroder et al., 2011). Due to the different mobility, those minerals accumulate in soils with different rates (He et al., 2008, 2009; Richards et al., 2011; Tewolde et al., 2011). Tewolde et al. (2010, 2011) demonstrated in an earlier study that the better lint yield performance of PL-fertilized cotton than for CIF-fertilized cotton was not due to better N nutrition of the PL-fertilized cotton, rather it was caused by other mineral nutrients in PL and other benefits associated with PL addition. In another study, Tewolde et al. (2005a, 2005b) found that cotton grown in media with nutrients only from PL had normal growth, showing that the PL provided all essential nutrients for normal plant growth. In the present study, we tried to understand the effects of CIF and PL fertilization with different application methods on cottonseed mineral composition harvested in two successive years. Data presented in this study indicated that neither the CC management nor PL application times were critical factors in determining the mineral contents in cottonseed. However, the levels of many minerals in cottonseeds were affected by the growing year factor. The annual differences could be due to differences in weather conditions between the 2 yr. It is reported that weather factors, such as rainfall and temperature, affect ash and chemical contents of cottonseeds (Bertrand et al., 2005). For example, higher rainfall is reported to result in higher ash content (Bertrand et al., 2005). In contrast, we observed lower ash content in the wetter 2009, indicating the complicated effects of weather factors. In addition, several element contents were lower in 2009 than 2010, indicating possible cumulative effects of repeated treatments on the same plots. To eliminate the year effect, we further analyzed the effects of fertilization and CC treatments on the elemental and ash contents in cottonseed based on the averages of 2-yr data. The results showed that the contents of eight elements and ash were significantly ($\alpha \leq 0.05$) impacted by fertilization (Table 3). Cover crop management impacted only the P content ($\alpha \leq 0.05$). There was no significant ($\alpha \leq 0.05$) impact by the interactions of the CC and fertilization treatments. The CC management seems a long-term factor that is worth further evaluation when samples from long-term experimental fields become available.

Historical data indicated a decreasing trend of ash and several mineral contents of cottonseed. For example, the ash content of whole cottonseed reported by the National Research Council had decreased from 4.8% in 1969 to 1989 to 4.2% (n = 193, SD = 2.1%) in 2001 (Bertrand et al., 2005; NRC, 2001). Bertrand et al. (2005) reported ash content of eight cottonseed samples from 3.7 to 4.2% with an average of 4.0%. Whereas it is true that this trend accompanied by reduced seed sizes is related to increased selection for lint quality (Bertrand et al., 2005), our data suggest that fertilization and possibly other growing conditions affect the ash content. Inadequate N fertilization seems to lead to larger ash content in the seed than adequate N fertilization, an observation that may be an indication of reduced photoassimilate accumulation in the seed and not necessarily an indication of increased mineral accumulation. The lower ash content of the CIF treatment than the unfertilized control is likely an indication of greater photoassimilate accumulation and larger seed size, not necessarily reduced mineral levels in the seed of the CIF treatment. Cotton that was fertilized with PL had adequate N fertilization and therefore must have the same level of photoassimilate accumulation and seed size as the cotton fertilized with the

Table 4. Correlation coefficients among seven elements and ash contents in cottonseed based on the year 2009 and 2010 data and the averages of the 2-yr data.

Elements	P	Ca	K	Mg	Fe	Cu	Mn
A. 2009 data (*n* = 12)							
Ca	ns†						
K	0.893***	ns					
Mg	0.964***	ns	0.840***				
Fe	ns	–0.787**	ns	ns			
Cu	0.795**	ns	0.805*	0.818**	ns		
Mn	ns	–0.608*	ns	ns	0.749**	–0.665*	
Ash	0.899***	ns	0.831***	0.890***	ns	0.800**	–0.607*
B. 2010 data (*n* = 12)							
Ca	ns						
K	0.583*	ns					
Mg	0.906***	ns	0.567*				
Fe	ns	–0.736**	ns	ns			
Cu	ns	ns	ns	ns	ns		
Mn	ns	ns	ns	ns	ns	ns	
Ash	0.834***	ns	ns	0.810**	ns	ns	ns
C. Averages of 2-yr/cover crop data (*n* = 6)							
Ca	ns						
K	0.898*	ns					
Mg	0.989***	ns	0.856*				
Fe	ns	–0.938**	ns	ns			
Cu	0.950**	ns	0.839*	0.923**	ns		
Mn	ns	ns	ns	ns	ns	ns	
Ash	0.932**	ns	ns	0.918**	ns	0.990***	ns

* Indicates the coefficients are statistically significant at $\alpha = 0.05$.
** Indicates the coefficients are statistically significant at $\alpha = 0.01$.
*** Indicates the coefficients are statistically significant at $\alpha = 0.001$.
† ns, indicates no statistically significant ($\alpha = 0.05$) coefficients observed.

CIF treatment (Tewolde et al., 2010; 2011). However, cotton fertilized with PL had higher seed ash content than the CIF treatment probably because PL supplied other mineral nutrients (i.e., P, K, Ca, Mg, Fe, B, etc.) and resulted in greater seed mineral content than the CIF treatment which received only N and K fertilizers. Relative to inorganic fertilizers, PL application has been known to increase concentration of P, K, Mg, and other elements in leaves and other plant parts (Tewolde et al., 2011; 2009b) and may lead to greater accumulation of these elements in the seed and to higher seed ash content.

In our experiment, the fertilizer type (i.e., CIF or PL) was the single most important factor in influencing the mineral contents of cottonseed. Compared to the no-fertilizer control, fertilization impacted the mineral contents in three ways: (i) little or no changes; (ii) decrease; and (iii) increase in the content of a mineral in cottonseed. Ten elements (C, N, Zn, Ni, Cd, Pb, Al, S, B, and As) belong to the first category. Calcium belongs to the second category, and Fe and Mn belong to the third. Magnesium, Cu, and P are the mixing scenarios of categories 2 and 3 as the contents of the elements decreased with CIF fertilization, but increased with PL fertilization. Fertilization increased the content of K, and decreasing the content of Na in cottonseed; however, more data are needed to draw a definitive conclusion. Interestingly, the ash content in these cottonseed samples also changed in a way similar to that of Mg, Cu, and P. Correlation coefficient analysis showed the ash content positively correlated with the contents of P, K, and Cu, but negatively correlated with that of Mn in the 2009 data, but positively correlated with only P and Mg contents for the 2010 data (Table 4). These relationships imply that the increase of ash content by PL fertilization was mainly caused by P and Mg accumulation. Both the 2009 and 2010 data also showed positive correlations between ash content and P, Mg, and K, implying the co-existence of the three elements in cottonseed in the form of Mg and K phosphate. Based on 58 samples of eight cotton varieties grown at 13 locations, Stansbury et al. (1953) reported that 95.7% of the total P increase in cottonseed was attributed to phytate P. Whereas phytate can contain other elements, such as Ca and Fe in plant seeds (Lott and West, 2001), the positive correlations we observed between ash content and P, Mg, and K prompted us to conclude that PL fertilization increased the Mg/K phytate in these cottonseed samples. It should be pointed out that although the majority of P in PL is in the form of phytate (Cade-Menun, 2011; He et al., 2006), plants take up mainly inorganic P for their growth (He et al., 2003; Seeling and Jungk, 1996). The organic P moiety of phytate in PL applied to soil should be first hydrolyzed to inorganic P, then taken up by the cotton plant. However, the Mg/K phytate forms in PL from poultry grain feedstuff might have provided the appropriate ratios for the cotton to take up synergistically Mg, K, and P to favor the accumulation of Mg/K phytate in cottonseed. The CIF fertilization did not provide

this kind of advantage. Based on the P data, the difference in phytate content between the CIF- and PL-fertilized cottonseed samples was 13%. The negatively charged phosphate in phytate strongly binds to metallic cations of Ca, Fe, K, Mg, Mn, and Zn making them insoluble and thus unavailable as nutritional factors (Bohn et al., 2008). Thus, the impact of the elevating phytate level in cottonseed by PL application is worthy of further research.

An interesting observation of the correlation coefficient analysis is the negative correlation of Ca with Fe, and possibly Mn. Our data showed that both CIF and PL fertilization decreased Ca content in cottonseed even though PL contained a significant amount of Ca and extractable soil Ca increased with PL application into the cotton field (Tewolde et al., 2011). Demir et al. (2010) found that fruit Ca content of tomato (*Lycopersicon esculentum* Mill.) was significantly reduced by increased rate of applied poultry manure. They explained their observation by (i) drawing-away of Ca from the fruits due to the enhanced growth of the plant with application of poultry manure and (ii) Ca dilution due to increased fruit yields. These explanations are also applicable to our case. Based on the correlation results, an additional explanation would be repelling (competing) effects of Fe and Mn on storage of Ca in cottonseed as Ca, Fe, and Mn are all localized in globoid crystals of the plant seed (Lott, 1981).

CONCLUSION

This research demonstrated the importance to determine the contents of several elements (Al, As, B, C, Ca, Cd, Cu, Fe, K, Mg, Mn, N, Na, Ni, P, Pb, S, and Zn) and ash in cottonseed samples. Our study with cotton grown in a Falkner silt loam soil in northeastern Mississippi showed that the contents of the elements determined in cottonseed samples were not affected by the presence of wheat CC or the PL application timing (spring vs. fall) and method (broadcast vs. subsurface band). The contents of six elements (Ca, Fe, Cu, Mn, Zn, P, and S) were higher in 2010 cottonseed samples than the corresponding 2009 samples. The fertilization type (CIF vs. PL) showed outstanding but not necessarily the same impacts on the contents of six elements (Ca, Fe, Mn, P, Cu, and Mg) and ash. Fertilization decreased Ca, but increased Fe and Mn contents. The CIF fertilization decreased, but PL fertilization increased P, Mg, Cu, and ash contents. Correlation coefficient analysis suggested that P, Mg, and K contributed to the increase of ash content, probably in the form of the mixture of K/Mg phytate compounds.

ACKNOWLEDGMENTS

Mention of trade names or commercial products in this publication is solely for the purpose of providing specific information and does not imply recommendation or endorsement by the U.S. Department of Agriculture. USDA is an equal opportunity provider and employer.

REFERENCES

Arieli, A. 1998. Whole cottonseed in dairy cattle feeding: A review. Anim. Feed Sci. Technol. 72:97–110. doi:10.1016/S0377-8401(97)00169-7

Bertrand, J.A., T.Q. Sudduth, A. Condon, T.C. Jenkins, and M.C. Calhoun. 2005. Nutrient content of whole cottonseed. J. Dairy Sci. 88:1470–1477. doi:10.3168/jds.S0022-0302(05)72815-0

Bohn, L., A.S. Meyer, and S.K. Rasmussen. 2008. Phytate: Impact on environment and human nutrition. A challenge for molecular breeding. J. Zhejiang Univ. Sci. B 9:165–191. doi:10.1631/jzus.B0710640

Bolan, N.S., D.C. Adriano, and S. Mahimairaja. 2004. Distribution and bioavailability of trace elements in livestock and poultry manure by-products. Crit. Rev. Environ. Sci. Technol. 34:291–338. doi:10.1080/10643380490434128

Cade-Menun, B.J. 2011. Characterizing phosphorus in animal waste with solution ^{31}P NMR spectroscopy. In: Z. He, editor, Environmental chemistry of animal manure. Nova Science Publ., New York. p. 275–299.

Cherry, J.P., J.G. Simmons, and R.J. Kohel. 1978. Potential for improving cottonseed quality by genetic and agronomic practices. In: M. Friedman, editor, Nutritional improvement of food and feed proteins. Plenum Press, New York. p. 343–364.

Citak, S., and S. Sonmez. 2010. Influence of organic and conventional growing conditions on the nutrient contents of white head cabbage (*Brassica oleracea* var. capitata) during two successive seasons. J. Agric. Food Chem. 58:1788–1793. doi:10.1021/jf903416a

Demir, K., O. Sahin, Y.K. Kadioglu, D.J. Pilbeam, and A. Gunes. 2010. Essential and non-essential element composition of tomato plants fertilized with poultry manure. Sci. Hortic. (Amsterdam) 127:16–22. doi:10.1016/j.scienta.2010.08.009

Dias, A.S., A.L. Fernando, and F.C. Lidon. 2009. III.Heat stress in Triticum: Kinetics of Na, K and P accumulation. Braz. J. Plant Physiol. 21:143–152.

Dowd, M.K., D.L. Boykin, W.R. Meredith, B.T. Campbell, F.M. Bourland, J.R. Gannaway et al. 2010. Fatty acid profiles of cottonseed genotypes from the national cotton variety trials. J. Cotton Sci. 14:64–73.

Elmore, C.D., W.I. Spurgeon, and W.Q. Thom. 1979. Nitrogen fertilization increases N and alter amino acid concentration of cottonseed. Agron. J. 71:713–716. doi:10.2134/agronj1979.00021962007100050005x

Endale, D.M., M.L. Cabrera, J.L. Steiner, D.E. Radcliffe, W.K. Vencill, H.H. Schomberg, and L. Lohr. 2002. Impact of conservation tillage and nutrient management on soil water and yield of cotton fertilized with poultry litter or ammonium nitrate in the Georgia Piedmont. Soil Tillage Res. 66:55–86. doi:10.1016/S0167-1987(02)00013-2

Fageria, N.K., C. Baligar, and R.B. Clark. 2002. Micronutrients in crop production. Adv. Agron. 77:185–268. doi:10.1016/S0065-2113(02)77015-6

He, Z., D.M. Endale, H.H. Schomberg, and M.B. Jenkins. 2009. Total phosphorus, zinc, copper, and manganese concentrations in Cecil soil through ten years of poultry litter application. Soil Sci. 174:687–695. doi:10.1097/SS.0b013e3181c30821

He, Z., C.W. Honeycutt, and T.S. Griffin. 2003. Enzymatic hydrolysis of organic phosphorus in extracts and resuspensions of swine manure and cattle manure. Biol. Fertil. Soils 38:78–83. doi:10.1007/s00374-003-0634-1

He, Z., Z.N. Senwo, R.N. Mankolo, and C.W. Honeycutt. 2006. Phosphorus fractions in poultry litter characterized by sequential fractionation coupled with phosphatase hydrolysis. J. Food Agric. Environ. 4:304–312.

He, Z., I.A. Tazisong, Z.N. Senwo, and D. Zhang. 2008. Soil properties and macro cations status impacted by long-term applied poultry litter. Commun. Soil Sci. Plant Anal. 39:858–872. doi:10.1080/00103620701880925

Helmke, P.A., and D.L. Sparks. 1996. Lithium, sodium, potassium, rubidium, and cesium. In: D.L. Sparks, editors, Methods of soil analysis: Part 3. Chemcial methods. SSSA, Madison, WI. p. 551–574.

Hogan, J.T., and J.C. Arthur, Jr. 1951. Preparation and utilization of cottonseed meal glue for plywood. J. Am. Oil Chem. Soc. 28:20–23. doi:10.1007/BF02639743

Jones, J.B., and V.W. Case. 1990. Sampling, handling, and analyzing plant tissue samples. In: R.L. Westerman, editor, Soil testing and plant analysis. 3rd ed. SSSA, Madison, WI. p. 404–410.

Kohel, R.J., J. Glueck, and L.W. Rooney. 1985. Comparison of cotton germplasm collections for seed-protein content. Crop Sci. 25:961–963. doi:10.2135/cropsci1985.0011183X002500060015x

Lambuth, A.L. 2003. Protein adhesives for wood. In: A. Pizzi and K. L. Mittal, editors, Handbook of adhesive technology. 2nd ed. Marcel Dekker, New York. p. 457–478.

Lott, J.N.A. 1981. Protein bodies in seed. Nord. J. Bot. 1:421–432. doi:10.1111/j.1756-1051.1981.tb00708.x

Lott, J.N.A., and M.M. West. 2001. Elements present in mineral nutrient reserves in dry *Arabidopsis thaliana* seeds of wild type and *pho*1, *pho*2, and *man* 1 mutants. Can. J. Bot. 79:1292–1296.

Mehlich, A. 1984. Mehlich 3 soil extractant: A modification of Mehlich 2 extractant. Commun. Soil Sci. Plant Anal. 15:1409–1416. doi:10.1080/00103628409367568

Meneghetti, S.M.P., M.R. Meneghetti, T.M. Serra, D.C. Barbosa, and C.R. Wolf. 2007. Biodiesel production from vegetable oil mixtures: Cottonseed, soybean, and castor soils. Energy Fuels 21:3746–3747. doi:10.1021/ef070039q

NRC. 2001. Nutrient requirements of dairy cattle. 7th rev. ed. Natl. Academy Press, Washington, DC.

O'Brien, R.D., and P.J. Wakelyn. 2005. Cottonseed oil: An oil for trans-free options. Food Technol. 16:677–679.

Pettigrew, W.T., and M.K. Dowd. 2011. Varying planting dates or irrigation regimes alters cottonseed composition. Crop Sci. 51:2155–2164. doi:10.2135/cropsci2011.02.0085

Pote, D.H., T.R. Way, P.J.R. Kleinman, P.A. Moore, J.J. Meisinger, K.R. Sistani et al. 2011. Subsurface application of poultry litter in pasture and no-till soils. J. Environ. Qual. 40:402–411. doi:10.2134/jeq2010.0352

Reddy, K.C., R.K. Malik, S.S. Reddy, and E.Z. Nyakatawa. 2007. Cotton growth and yield response to nitrogen applied through fresh and composted poultry litter. J. Cotton Sci. 11:26–34.

Richards, J., H. Zhang, J.L. Schroder, J.A. Hattey, W.R. Raun, and M.E. Payton. 2011. Micronutrient availability as affected by the long-term application of phosphorus fertilizer and organic amendments. Soil Sci. Soc. Am. J. 75:927–939. doi:10.2136/sssaj2010.0269

Schroder, J.L., H. Zhang, J.R. Richards, and Z. He. 2011. Sources and contents of heavy metals and other trace elements in animal manures. In: Z. He, editor, Environmental chemistry of animal manure. Nova Science Publ., New York. p. 385–414.

Seeling, B., and A. Jungk. 1996. Utilization of organic phosphorus in calcium chloride extracts of soil by barley plants and hydrolysis by acid and alkaline phosphatases. Plant Soil 178:179–184. doi:10.1007/BF00011581

Stansbury, M.F., W.A. Pons, and C.L. Hoffpauir. 1953. Phosphorus compounds in cottonseed kernels; influence of variety of cottonseed and environment. J. Agric. Food Chem. 1:75–78. doi:10.1021/jf60001a011

Tewolde, H., A. Adeli, K.R. Sistani, and D.E. Rowe. 2011. Mineral nutrition of cotton fertilized with poultry litter or ammonium nitrate. Agron. J. 103:1704–1711. doi:10.2134/agronj2011.0174

Tewolde, H., N. Buehring, A. Adeli, K.R. Sistani, D.E. Rowe, and R.G. Pratt. 2009a. Cotton response to chicken litter in rotation with corn in clayey soil. Agron. J. 101:626–634. doi:10.2134/agronj2008.0154x

Tewolde, H., N. Buehring, A. Adeli, K.R. Sistani, D.E. Rowe, and R.G. Pratt. 2010. Equivalency of broiler litter to ammonium nitrate as a cotton fertilizer in an upland soil. Agron. J. 102:251–257. doi:10.2134/agronj2009.0244

Tewolde, H., M.W. Shankle, A. Adeli, K.R. Sistani, and D.E. Rowe. 2009b. Macronutrient concentration in plant parts of cotton fertilized with broiler litter in a marginal upland soil. Soil Tillage Res. 105:1–11. doi:10.1016/j.still.2009.04.007

Tewolde, H., K.R. Sistani, and D.E. Rowe. 2005a. Broiler litter as a sole nutrient source for cotton: Nitrogen, phosphorus, potassium, calcium, and magnesium concentrations in plant parts. J. Plant Nutr. 28:605–619. doi:10.1081/PLN-200052633

Tewolde, H., K.R. Sistani, and D.E. Rowe. 2005b. Broiler litter as a micronutrient source for cotton: Concentrations in plant parts. J. Environ. Qual. 34:1697–1706. doi:10.2134/jeq2005.0009

Way, T.R., M.R. Siegford, and D.E. Rowe. 2010. Applicator system and method for the agricultural distribution of biodegradable and non-biodegradable materials. U.S. Patent 7721,662. Date issued: 25 May 2010.

Yu, J., S. Yu, S. Fan, M. Song, H. Zhai, X. Li, and J. Zhang. 2012. Mapping quantitative trait loci for cottonseed oil, protein and gossypol content in a *Gossypium hirsutum* x *Gossypium barbadense* backcross inbred line population. Euphytica 187:191–201. doi:10.1007/s10681-012-0630-3