

**Comparison of Poultry Litter and Commercial Fertilizer Rate and Application Timing
on Environmental Nitrogen Loss, Corn Grain Yield, and Mineral Composition**

by

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Abstract

For efficient use of poultry litter (PL) as a nutrient source, it is critical to balance the amount of nitrogen (N) needed to maximize corn (*Zea Mays* L.) grain yield while reducing N loss to the environment. Besides, there are limited reports on whether the application of PL enrich corn ear (including grain, cob and husk) with some of the mineral elements it supplies such as nitrogen (N), phosphorus (P), potassium (K), calcium (Ca), and magnesium (Mg). The mineral composition of corn ear greatly impacts its nutritional value and potential use as human food and animal feed. This 2-year study (2018–19) investigated the effects of N source [PL and conventional fertilizer (CF) such as urea], application rate [0 (control), 168 (low rate) and 336 (high rate) kg total N ha⁻¹], and time (single and split application) on corn grain yield, environmental N loss (ENL), and ear mineral composition studied at three locations [E.V. Smith Research Center (EVS), Wiregrass Research and Extension Center (WREC), and Tennessee Valley Research and Extension Center (TVREC)]. In a single application, the target N rate was applied pre-plant whereas in a split application, one-fourth of the target N rate was applied pre-plant and the remaining three-fourth side dressed at the V6 growth stage. A field-scale partial N budget was used to quantify environmental N loss (ENL). Nitrogen inputs included N contribution from the fertilizer treatments, and the background N (soil N mineralization, crop seed, biological N fixation, and atmospheric N deposition) while N outputs included plant N uptake, residual soil inorganic N (NH₄-N + NO₃-N) and total N lost to the environment i.e. ENL. Plant and soil samples (0-15 and 15-30 cm depths) were collected at harvest each year for measuring aerial dry matter and residual inorganic N content, respectively. Ear samples were also harvested at physiological maturity each year and analyzed for the contents of 11 mineral elements (N, P, K, Ca, Mg, S, B, Zn, Mn, Fe, and Cu). Relative to CF, the application of PL increased grain yield at EVS whereas no significant difference in yield was found at the other two locations (WREC and TVREC). No response to application

timing for grain yield was observed at WREC but the split application of PL reduced grain yield at other two locations (EVS and TVREC). Increased application of PL resulted in greater grain yield at WREC and TVREC compared to a low rate. Averaged across EVS and WREC, drier growing conditions of 2019 lowered grain yield by about 29% than in 2018. Aerial plant dry biomass followed similar patterns as grain yield at EVS and WREC whereas the highest whole plant N concentration was attained from urea application relative to PL, regardless of the study site, application rate, and timing. We found no consistent results for ENL among N sources with significantly greater ENL reported from PL at WREC but no differences at TVREC. At EVS, urea had greater ENL than PL at a single application whereas the opposite was true at a split application. Application timing did not influence ENL at WREC and TVREC. However, ENL increased with increasing N rate ranging from 32 to 52% of the total N input. Poultry litter, regardless of application rate, time, and location, did not increase the concentration of selected mineral elements in the ear. Corn fertilized with CF had the highest ear concentrations of N, P, K, Ca, Mg, Zn, Mn, Fe, and Cu at all locations which increased with increasing N rate. Ear N concentration was dependent on the level of its plant-availability in the soil. However, ear levels of P, K, and other mineral elements were not in proportion to their soil levels rather dependent on ear N concentration. These results indicated that optimal levels of plant-available N (PAN) in the soil, irrespective of whether derived from PL or CF, ensures maximum accumulation of ear N along with P, K, and other elements.

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List of Abbreviations

BMPs	Best management practices
CF	Conventional fertilizer
ENL	Environmental nitrogen loss
EVS	E.V. Smith Research Center
PAN	Plant-available N
PL	Poultry litter
WREC	Wiregrass Research and Extension Center
TVREC	Tennessee Valley Research and Extension Center

I. Literature Review

Introduction

The United States has the largest broiler (*Gallus gallus domesticus*) industry in the world marketing about 8.9 billion birds annually (USDA-NASS, 2019). Consequently, over 13 million tons of poultry litter (PL) is generated (1.5 kg litter/broiler; Mitchell and Tu, 2005) which consists of manure (bird excreta) mixed with spilled feed, feathers, and bedding material such as wood shavings of pine (*Pinus palustris* P. Miller or *Pinus elliottii* Englem.), hulls of rice (*Oryza sativa* L.) or peanut (*Arachis hypogaea* L.). The industry has witnessed rapid growth over the last decade especially in the south-eastern United States (MacDonald, 2008) with a majority of the broiler production (about two-thirds) concentrated in five southeast states (North Carolina, Georgia, Alabama, Mississippi, and Arkansas). Currently, Alabama ranks second nationally in broiler production behind Georgia with 1.12 billion birds produced in 2018, generating roughly 1.68 million tons of PL (USDA-NASS, 2019).

Poultry litter is regarded as a valuable source of plant nutrients particularly nitrogen (N) (Poffenbarger et al., 2015) with an average fertilizer equivalent grade of 3-3-2 (N-P₂O₅-K₂O) (Stephenson et al., 1990; Mitchell and Donald, 1999). Historically, much of the PL generated was primarily applied to hayfields and pasture lands (Sistani et al., 2008; Mitchell and Tu, 2005). According to Moore et al. (1995), 90% or more of PL generated is being land-applied as a source of plant nutrients. The addition of PL to the soil also positively influences many soil physical, chemical, and microbiological properties (Entry et al., 1997), thereby improving overall soil health (Kingery et al., 1994; Mitchell and Tu, 2005; Schomberg et al., 2009). Like any other organic manure, PL application increases soil organic matter (SOM) (Tejada and Gonzales, 2008; Tejada et al., 2008; Adeli et al., 2011). Higher SOM favors soil aggregation increases soil permeability to air and water, soil nutrient and water storage capacities, and decreases soil bulk density (Johnson et al., 2005; Celik et al., 2004; Leroy et al., 2008). Both

the population and diversity of various soil microorganisms such as bacteria, fungi, and actinomycetes are influenced by litter application (Pratt and Tewolde, 2009; Acea and Carballas, 1996) with a positive impact on soil enzyme activity (Acosta-Martinez et al., 2006; Chu et al., 2007). Enhanced microbial activity promotes nutrient cycling, formation of soil humus, and decomposition of several resistant compounds (Zak et al., 1994; Wu et al., 2011).

Land application of PL is often based on crop N requirement (Pote et al., 1996). Although N is present in abundance (about 78%) in the air as a dinitrogen (N_2) gas, it is the most limiting nutrient for crop production (Tafteh and Sepaskhah, 2012). This is because crop uptake of N from the soil occurs primarily in two inorganic forms: nitrate (NO_3) and ammonium (NH_4). Nitrogen input accounts for about 40% of the total production cost; therefore, PL can be used as a relatively cheap alternative N source (Hollis, 2013). The majority of the litter N (up to 75%) occur in the form of ammonium and uric acid (Schefferle, 1965). The uric acid-N gets readily transformed into NH_4 -N in most soils which get further converted into NO_3 -N via nitrification (Sims and Wolf, 1994).

It has been estimated that only 50% of the fertilizer N applied is utilized by the growing crop (Krupnik et al., 2004; Smil, 1999). The main cause of this low crop N-use efficiency (NUE) is the loss of N to the environment via several pathways such as denitrification, ammonia (NH_3) volatilization, nitrate leaching, runoff, and soil erosion (Bouwman et al., 2002; IFA-FAO, 2001), resulting in air and water pollution (Erisman et al., 2007).

Nitrogen losses in Agricultural systems

Agriculture is a major contributor to the emissions of reactive N [NO_3 , ammonia (NH_3), NH_4 , nitrous oxide (N_2O)] into the environment. Of the total emissions in the United States, half of NO_3 (Smith et al., 1997), three-fourth of N_2O (USEPA, 2010), and 84% of NH_3 (USEPA, 2010) emissions are contributed by agriculture. Recent decades have witnessed a

significant increase in the use of N fertilizers with application expected to reach 186 million Mg N yr⁻¹ by mid-21st century (Muschiatti-Piana et al., 2018). Enhanced use of N fertilizers along with improved crop genetics and agronomic practices, have consistently increased crop yields over the last century (Cassman et al., 2002); however, excessive use may negatively impact crop yields, reduce fertilizer NUE, (Eickhout et al., 2006; Janzen et al. 2003; Smil, 1999; Varvel and Peterson, 1990) and degrade the environment through the release of NH₃, nitrogen oxides (NO_x), nitrous oxide (N₂O) gases (Franzluebbbers 2007; Herrero et al. 2010) and NO₃ leaching; thus N application mandates careful handling in modern agriculture (Ju and Christie, 2011).

Nitrate leaching

Nitrate leaching is the primary cause of groundwater contamination in agroecosystems that are dominated by sandy soils (Akbariyeh et al., 2018; Mitsch and Day, 2006) and have heavy rainfalls or irrigation conditions. Any N present beyond the rooting zone has the potential to leach down to the aquifer (Delgado et al., 2005). In some cases, the groundwater NO₃ content may exceed the maximum contaminant level (MCL) of 10 mg L⁻¹ for potable water set by the USEPA (United States Environmental Protection Agency). High NO₃ content (> 10 mg L⁻¹) in drinking water is detrimental to human health especially for pregnant women, and infants less than 6 months old (Spalding and Exner, 1993). Enrichment of the surface water bodies, such as lakes and rivers, with NO₃, may result in eutrophication and hypoxia (Rabalais et al., 1996).

On average, up to 30% of the total N input can be lost from different cropping systems via NO₃ leaching (Raun and Schepers, 2008) and increases with increasing N input (Jn-Baptiste et al., 2012). Among cereals, greater NO₃ leaching losses have been reported for corn (*Zea Mays L.*) (St. Luce et al., 2011).

Ammonia volatilization

Ammonia volatilization, another important pathway in the nitrogen cycle, is the release of NH_3 gas from the agricultural system (Harrison and Webb, 2001). Synthetic N fertilizers are the largest source of NH_3 emissions globally (Yan et al., 2003) accounting for about 80–90% of the total anthropogenic NH_3 emissions (Battye et al., 2003). Based on 148 different studies which measured NH_3 volatilization, Bouwman et al. (2002) reported mean NH_3 loss globally to be 10-19%, and 19-29% from the applied commercial N fertilizers and animal manure, respectively.

Ammonia is a potential pollutant in the environment (Galloway et al., 2003). Once into the atmosphere, NH_3 can readily form N_2O , greenhouse gas (Ferm, 1998). Emitted NH_3 eventually make its way back to the land surface via atmospheric deposition and induces soil acidification (van Breemen et al., 1982). Higher amounts of H^+ and Al^{3+} ions in the soil were found to be toxic to plant roots (Van Den Berg et al., 2005; Poschenrieder et al., 2008), soil organisms (Kuperman and Edwards 1997), reduce soil fertility (Kochian, 1995), impacts soil microbial community (Rousk et al., 2010) which in turn affects several other processes including N mineralization (Freckman, 1988; Ferris et al., 1998).

Fertilizer or manure N addition is expected to increase the potential for NH_3 loss by elevating soil $\text{NH}_4\text{-N}$ levels (Ma et al., 2010; Rochette et al., 2013). However, the effect of N rate on NH_3 volatilization is not consistent with studies indicating positive (Black et al., 1985), negative (Thompon et al., 1990; Tian et al., 2001) or no response (Dhyani and Mishra, 1992; Saravanan et al., 1987) to the N application rate. Sharpe et al. (2004) reported an NH_3 loss of 5.4% and 24% from the surface application of PL during the summer of 2001 and 2000, respectively. However, maximum volatilization occurred within 48 h of litter application in both years. Other studies have also reported rapid NH_3 volatilization rates immediately following litter application (Cabrera et al., 1993; Marshall et al., 1998). Marshall et al. (1998)

in an open-field study on three Major Land Resource Areas (MLRAs) of the southeast US including Coastal Plain (Alabama) found that between 1.70 and 6.40% of total N applied was lost as NH₃ within 14 d of litter application. Variable NH₃ losses in the studies may be related to the climatological differences such as air temperature and wind speed following litter application (Harper and Sharpe, 1995). Greater volatilization during summer 2000 in the Sharpe et al. (2004) study was attributed to higher air temperature (about 5°C) and wind speed (about twice) than in 2001. In general, dry conditions such as low soil moisture and relative humidity favor NH₃ volatilization (Sommer et al., 1991; Nathan and Malzer, 1994). Incorporation of PL into the soil greatly reduces volatilization losses (Webb et al., 2005) as NH₄ present at the soil surface is more prone to volatilization loss (Terman, 1979).

Denitrification

With the limited supply of oxygen in the soil, the soil-inhabiting nitrifying bacteria mainly *Bacillus*, *Pseudomonas*, *Thiobacillus* reduce nitrate to nitric oxide (NO) and N₂O (Firestone, 1982). These bacteria are facultative anaerobes, capable of using NO₃ as an electron acceptor during respiration under limited oxygen or anaerobic conditions. The stepwise reduction of NO₃ to N₂O and nitric oxide (NO) is facilitated by nitrification (aerobic) and denitrification (anaerobic) biochemical processes (Davidson, 1991; Conrad, 1996). These processes are controlled by several factors, including soil moisture, pH, temperature, oxygen levels, and microbial activity (Firestone, 1982; Tiedje, 1988; Aulakh et al., 1992).

The agriculture sector is responsible for 68% of the total anthropogenic United States N₂O emissions with synthetic fertilizers contributing about one-fourth of it (USEPA, 2013). Nitrous oxide emissions ranged from 0.2 to 6.3% of applied N in the US Midwest corn belt (Flynn and Smith, 2010, Linquist et al., 2012). Nitrous oxide is a 300 times stronger greenhouse gas, when compared to carbon dioxide (CO₂), with 120 years of lifetime (Robertson and Grace,

2004; IPCC, 1996). The N₂O gets oxidized in the atmosphere to form NO and other nitrous oxides (NO_x) forms which deplete stratospheric ozone (Bliefert, 1994; Duxbury, 1994).

Bouwman et al. (2002) proposed a loss of 0.009 kg N₂O-N per kg of N applied which increases exponentially with increasing N input (Bouwman et al., 2002; Kim et al., 2012; Millar et al., 2010; Van Groenigen et al, 2011). The use of organic manures such as PL in crop production can elevate N₂O and NO emissions from the field (Akiyama and Tsuruta, 2003a, b; Veltof et al., 2003; Jones et al., 2007). In addition to available N, organic fertilizers also supply organic C, which may stimulate soil microbial activity. The resultant anaerobic conditions in the soil due to enhanced O₂ consumption by the microbes promote denitrification (Granli and Bøckman, 1994). Sistani et al. (2011) found that the daily N₂O fluxes and cumulative N₂O emission were significantly higher from PL than from commercial fertilizers (such as urea, urea ammonium nitrate, ammonium nitrate) in both years (2009 and 2010) of a no-till corn study. As a percentage of total N applied, the 2-yr mean N₂O loss was 4.5% for PL, and 1.5% for dry granular urea. Hayakawa et al. (2009) also reported larger N₂O emission rates from PL than chemical fertilizer when incorporated into the soil to a depth of 15 cm.

Estimating nitrogen losses via nitrogen budget

The soil N cycle is a complicated process especially in today's intensive agricultural systems (Kim et al., 2012). To avoid the negative environmental impact of N loss from agricultural fields and evaluate fertilizer NUE, we need to better understand and determine major pathways (as discussed above) via which N is lost from the system (Gao et al., 2014). Quantification of individual N loss pathways is complex and involves time, additional equipment, and labor. For instance, it is difficult to accurately measure N leaching and involves costly lysimeter field trails (Uhlen, 1994). However, one of the cost-effective, simple, and robust way of measuring total N loss to the environment is using a N budget. A N budget is defined as the record of N inputs and outputs at the field level (Onenema et al., 2003). In other

words, it is a summary of N imports and exports for a given system (Shober et al., 2011). Nitrogen budgets have been used since the late-19th century (Lawes et al., 1882) and provide a framework to identify dominant N flows and assess agroecosystem performance and environmental safety (Watson and Atkinson, 1999; Ross et al., 2008).

Nitrogen budgets are based on the principle of mass conservation where the difference in total N inputs and N outputs into an agricultural system must equal the change in the system's total N (Legg and Meisinger, 1982; Meisinger and Randall, 1991). A N budget can be defined as:

$$\Sigma N \text{ inputs} - \Sigma N \text{ outputs} = N \text{ stored within, or lost from, the agroecosystem}$$

The N inputs may include fertilization rate (N_a), biological N fixation (N_b), soil N mineralization (N_c), atmospheric N deposition (N_d), crop seed N (N_e), and non-symbiotic N fixation (N_f). Therefore, the total N input (N_{ti}) can be calculated as a sum of all inputs:

$$N_{ti} = N_a + N_b + N_c + N_d + N_e + N_f$$

The N outputs can be crop N removal (N_g), volatilization loss (N_h), denitrification loss (N_i), leaching loss (N_j), and runoff loss (N_k). Total N output (N_{to}) was calculated as:

$$N_{to} = N_g + N_h + N_i + N_j + N_k$$

A positive difference between N inputs and outputs represents N surplus while a negative difference indicates N loss. The calculated N surplus indicates potential N loss from a farming system, which includes all possible pathways such as leaching, ammonia volatilization, denitrification, and runoff/erosion (Halberg et al., 1995).

Several studies have been conducted on N budgeting in different agroecosystems (Barry et al., 1993; Frissel, 1978; Ross et al., 2008; Pieri et al., 2011; Prasad and Hochmuth, 2016). However, it is challenging to compare these studies since they are based on different conceptual

models with varied study goals, system boundaries, and level of detail (Watson and Atkinson, 1999). For instance, the goals for the N balance study may vary from identifying major N flow pathways to estimating excess- or unaccounted-N in the system. The reliability of the budget depends upon the detail and accuracy of the available data. Some N inputs such as crop yield are relatively easy to measure compared to other soil processes such as mineralization, symbiotic N fixation, etc. (Meisinger and Randall, 1991; Smaling and Fresco, 1993). Fertilizer N input into a system can be defined easily with high accuracy but errors in estimating manure N input may range from 30% to 50% (Meisinger and Randall, 1991) adding uncertainty to the budget. Lower N balance values indicate higher accounting of N inputs and outputs in an agroecosystem (Sainju et al., 2016).

This mass balance tool was successfully used to estimate both total N loss to the environment and potential N leaching after 19 years of fertilization including urea and pig manure (Huang et al., 2017). The total N loss was estimated as 24 to 48% of total N input from incorporated fertilizer treatments. Duan et al. (2016) also employed the concept of mass balance to quantify total environmental N loss (ENL) at three sites under 8 different fertilization regimes and found that N loss ranged between 20% and 63% of total N input. Korsæth and Eltun (2000) predicted N runoff from different arable and forage cropping systems with cattle slurry fertilization using a mass balance approach and concluded that the N balance model was well suited to predict N runoff especially for arable crops. Several other studies have also used agricultural N balance as an indicator of the risk of N loss from specific farm units (Lord et al., 2002; Constantin et al., 2010; Liu et al., 2003).

Despite extensive research conducted, there may arise an imbalance in the N budget with inputs exceeding outputs (Schlesinger, 2009). Also, this approach is challenged by the slow changes in the soil N status for short-term experiments. For sustainable production, a farm's N budget should be balanced. If there is net N loss from the system, the soil will become

N deficient over time. On the other hand, if there is an N surplus in the system, there is potential for leaching and surface runoff losses. The best management practices (BMPs) should target achieving the maximum crop yield with the lowest ENL. Therefore, examining a N balance can assist in developing efficient nutrient management strategies.

Influence of PL on crop production

Due to its nutrient value and organic matter content, PL is widely used for crop production especially in poultry-production areas (Mitchell and Mask, 1992; Mitchell and Donald, 1999; Huang and Lu, 2000; Stevenson et al., 1990). Numerous studies have evaluated PL as a nutrient source against conventional fertilizers on cotton (*Gossypium hirsutum* L.) (Tewolde et al., 2010a; Tewolde et al., 2011), soybean (*Glycine max* L.) (Adeli et al., 2005; Adeli et al., 2015), and corn (*Zea mays* L.) (Adeli et al., 2012; Tewolde et al., 2013; Endale et al., 2008; Watts and Torbert, 2011; Liebhardt, 1976; Perkins et al., 1964, Sims 1987; Nyakatawa and Reddy, 2002) and shown positive yield response in corn and soybean (Sistani et al., 2008; Adeli et al., 2005; Watts and Torbert, 2011). In northeast Alabama on a fine sandy loam, grain yield was consistently increased by long-term addition of PL from 1991 to 2001 (Watts and Torbert, 2011). Endale et al. (2008) reported 18% higher corn grain yield from PL compared to commercial fertilizers (ammonium nitrate or sulfate) when applied at 168 kg plant-available N ha⁻¹.

Applying PL to row crops has been reported to increase the content of mineral elements in the soil (Adeli et al., 2010; Tewolde et al., 2011). These increased soil nutrient levels may further enrich plant tissues such as leaves and stem (He et al., 2013; Tewolde et al., 2005b, 2007, 2010a). Fertilizing cotton with PL can increase seed phosphorus (P), potassium (K), magnesium (Mg), and copper (Cu) concentrations compared to inorganic fertilizer (He et al., 2013). In a study with soybeans grown on the soil where sewage sludge and metal salts were applied, Ham and Dowdy (1978) attributed the enrichment of soybean seeds with mineral

elements to enhanced plant nutrient availability and uptake. Farmaha et al. (2011) also observed a positive correlation between harvested soybean seeds and leaf concentrations of P and K to varied application rates of P and K fertilizers. Benefits of PL application have also been reported in horticultural crops. For example, in cabbage (*Brassica oleracea*), the application of organic manures including PL increased mineral contents of the edible part relative to chemical fertilizers (Citak and Sonmez, 2010). In tomato (*Lycopersicon esculentum* Mill.), PL elevated levels of zinc (Zn; critical element for human nutrition) and lowered bromine (Br; potentially harmful) concentration in the fruit highlighting its value in tomato production (Demir et al., 2010).

Corn grain is used both as animal feed and for human consumption. As such, the mineral composition of corn grain impacts its nutritional value and potential use. When used as livestock feed, inadequate grain mineral content adversely affects animal physiology and reduce growth (Schutte, 1964; Underwood and Somers, 1969). According to Olson and Frey (1987), corn grain fails to meet dietary mineral requirements of P, calcium (Ca), sodium (Na), manganese (Mn), Zn, and Cu in pigs and poultry and require mineral supplements from external sources. Sauberlich et al. (1953) found that increasing corn grain N content improved its nutritive quality.

Nitrogen mineralization from PL

The effects on crop growth and yield from PL application depends upon the nutrient availability to plants especially N. Total litter N can be highly variable ranging from 2 to 6% (Nicholson et al., 1996; Stephenson et al., 1990) and changes based on bird type (layer or broiler), feed, and housing conditions (Gordillo & Cabrera, 1997; Mowrer et al., 2014; Nahm, 2005; Stephenson et al., 1990). Since organic N is the dominant litter N form with 20 to 40% of the total N present in the inorganic form (mostly ammonia) (Sims, 1986, 1987; Westerman et al., 1987), estimation of rate at which organic N mineralizes under in-situ conditions is

critical to calculating its fertilizer value. The amount of N released from litter is influenced by environmental factors such as soil (pH, nutrient status, biological activity), weather (temperature and humidity), and litter quality (Paul and Clark, 1996).

Several studies have investigated the kinetics of N mineralization rate from different animal manures (Castellanos and Pratt, 1981; Chae and Tabatabai, 1986; Bonde and Lindberg, 1988; Cabrera et al., 1993). In a laboratory incubation study with 107 manure samples, N mineralization ranged from 0% to more than 50% with an average of 10-20% (Van Kessel and Reeves, 2002). Gordillo and Cabrera (1997) reported 27-91% of the total N as net N mineralized in an incubation study with 15 PL samples for 112 days. A recent incubation study by Cassity-Duffey et al. (2020) found that 10-55% of the total N in 15 PL samples got mineralized over 99 days. However, dynamic field conditions make these results less relevant (Flowers and Arnold, 1983).

Under field conditions, it is commonly assumed that about 50% of the total litter N becomes plant available in a single growing season (Rasnake, 2002; Sistani et al., 2008; Vest et al., 1994; CAES 2007; Schomberg et al., 2011). However, some studies have demonstrated that this assumption may not accurately depict actual N availability from PL (Tewolde et al., 2009a, 2013). Eghball et al. (2002) reported 55% of the litter N becomes plant available in the first year of application with the remaining 45% in the succeeding years. On the other hand, Ruiz and Sawyer (2008) have shown that only 35% of the litter N applied to corn in the spring was available for plant uptake in the first year. Warren et al. (2006) overestimated the litter N availability factor (assuming 60%) and reported a 32% reduction in corn grain yield. Sims (1986) reported 25 to 40% organic N mineralization at 25° C and 17 to 64% at 40° C.

Conservation tillage

Conservation tillage systems such as no-till have been widely adopted by growers since the 1960s (Phillips and Young, 1973; CTIC, 2009) primarily to lower soil erosion, fuel usage, and increase soil water retention (Larson, 1981; Campbell et al., 1984). The use of no-till retains most of the crop residues at the soil surface leading to increased SOM, improved aeration, and water infiltration (Radcliffe et al., 1988). However, achieving these benefits can be challenging in the south-eastern United States due to the hot and humid climate which facilitates faster decomposition of surface residues (Watts and Torbert, 2011). The region's soils are typically sandy with low organic matter content and water holding capacities; thus, increasing the risk of soil compaction by heavy agricultural machinery (Carreker et al., 1977). Such soil conditions may limit root growth, lowering crop productivity (Watts and Torbert, 2011).

Application of PL combined with no-till may stimulate positive effects on crop productivity from added organic matter (Watts et al., 2010; Edwards et al., 1992; Bauer and Black, 1994). Over 5-yr (2001–2005), Endale et al. (2008) reported 31% greater corn grain yield from the combined effect of no-till and PL application relative to conventional tillage and fertilizer. However, Watts and Torbert (2011) reported 9-yr mean continuous corn grain yield from conventional tillage with litter application as 7507 kg ha⁻¹ compared to 6282 kg ha⁻¹ from no-till with litter application but provided no specific information.

Poultry litter is usually surface applied with or without incorporation into the soil. However, the implementation of no-till mandates surface application of PL, which may increase the risk of N losses. Compared to incorporated-PL, surface-broadcasted litter has been reported to have higher NH₃ volatilization (Sharpe et al., 2004; Webb et al., 2005; Sommer and Hutchings, 2001), and NO₃ leaching losses (Nyakatawa et al., 2001); thereby reducing fertilizer efficacy of PL. When stored uncovered, about one-tenth of the NH₄-N in the PL can be lost

within 2 months (Sims, unpublished data). In addition, due to the continuous supply of oxygen at the soil surface, litter $\text{NH}_4\text{-N}$ may get transformed into $\text{NO}_3\text{-N}$ (via nitrification); thus, increasing the risk of nitrate leaching.

Research has shown that PL incorporated at the rate of 135 kg plant-available N ha^{-1} increased corn grain yield by 12% (590 kg ha^{-1}) compared with non-incorporated PL (Jn-Baptiste et al., 2013). Also, in cotton, subsurface banding of PL at 6.7 Mg ha^{-1} increased lint yield by 68 kg ha^{-1} (1052 vs. 984 kg ha^{-1}) relative to surface broadcasting (Tewolde et al., 2009b). In Mississippi, on an upland soil with low organic matter, Adeli et al. (2008) found that mixing PL in the top 5 cm of the soil retained more nutrients over no incorporation, thereby preventing losses of nutrients and building soil fertility. However, Sistani et al. (2008) reported no significant interaction between tillage (no-till and conventional tillage) and PL rates (11 and 22 Mg ha^{-1}) for corn grain yield in a 4-year experiment. According to Jn-Baptiste et al. (2013), when nonincorporated, application of PL at a high rate (270 kg plant-available N ha^{-1}) may compensate for N losses by supplying excess N and cause similar grain yield compared to PL incorporation.

Application timing of poultry litter

To synchronize N release from PL with crop needs, application timing is one of the critical factors to be considered. Substantial research has been conducted to optimize the application timing of N fertilizer (Aldrich, 1984; Randall et al., 2003; Randall and Vetsch, 2005). Nitrogen application coinciding with major crop uptake has shown to maximize corn grain yields (Aldrich, 1984; Fox et al., 1986; Mitsch et al., 2001).

Ruiz Diaz and Sawyer (2008) evaluated three application times of PL (late fall, winter, and spring pre-plant) in Iowa and reported similar corn grain yield although soil $\text{NO}_3\text{-N}$ levels in early June during the growing season were higher with the spring application. They

suggested that the fall application of PL may benefit growers logistically and economically, particularly when there is time constrain during spring planting. However, fall and winter PL applications may have greater N losses due to increased exposure to the environment (Beckwith et al., 1998; Smith et al., 2002; Hansen et al., 2004; van Es et al., 2004). Significant N loss via leaching or denitrification may occur with N applied too early in the season (Blackmer et al., 1989; Magdoff, 1991). In regions with a warm fall and mild winter such as Alabama, Jn-Baptiste et al. (2013) reported lower corn grain yield from PL applied in the fall than spring application, regardless of the rate (68, 135, and 270 kg plant-available N ha⁻¹). Similar results were also reported in Mississippi (Tewolde and Sistani, 2010; Adeli et al., 2011).

Application rate of poultry litter

Nitrogen fertilizer is a costly input and often needed in large amounts, thus necessitating high application rates. Sistani et al. (2010) reported increasing bermudagrass hay production with increasing rates of surface-broadcasted PL. Balkcom et al. (2003) found similar peanut yield between PL applied at three rates (1.9, 3.8, and 7.6 Mg ha⁻¹) and the control on 13 farm fields. Tewolde et al. (2010a), on the other hand, documented the effect of six different rates (2.2 to 13.5 Mg ha⁻¹) of surface-broadcasted PL on cotton and found the highest lint yield at 6.7 Mg ha⁻¹ with lowest at 2.2 Mg ha⁻¹. In corn, Sistani et al. (2008) reported similar grain yield at PL rates of 11 and 22 Mg ha⁻¹ in 3 years out of the 4-yr study (1998-2001). In another study conducted from 2006-2008 in central Kentucky, similar corn yields were reported between PL applied at 135 and 270 kg plant-available N ha⁻¹ indicating luxury consumption at higher N rates (Jn-Baptiste et al., 2012). These results suggest that high N rate applications may not guarantee elevated yields (Cassman et al., 2003), but can potentially deteriorate environmental quality with associated N losses (McSwiney and Robertson, 2005).

Research Objective

The use of PL as a soil amendment for row crop production continues to increase in row crop production systems. The efficacy of PL application is influenced by application rate and timing. There is plenty of scientific research about the impacts of PL on crop grain production; however, the results often lack consistency and vary with crop management practices and environmental conditions. In a meta-analysis of 90 studies investigating the yield response from PL vs. CF, Lin et al. (2019) concluded that the greatest benefits from PL can be attained when applied before planting as surface broadcast rather than incorporation with tillage, at the highest rate with consecutive applications over the years. However, adopting such management practices for PL may maximize grain yields but also increase soil NO₃ levels, NH₃ volatilization rates, and other forms of N losses. A general estimation of N losses will help adopt Best Management Practices (BMPs) to reduce the ENL. To our knowledge, no field-scale N balance study has been conducted under similar fertilization regimes in the southeastern United States. Therefore, the purpose of this study was to evaluate the agronomic and environmental impacts of PL fertilization as a function of application rate and timing relative to CF on dryland corn. In addition, there have also been limited reports on whether the application of PL enrich corn grain with mineral elements. Since the mineral composition of corn grain impacts its nutritional value and potential use as human food and animal feed, this work further investigated how PL addition affects grain mineral composition.

II. Comparison of Corn Grain Yield and Environmental Nitrogen Loss under Poultry Litter and Urea Application Systems

Abstract

For efficient use of PL as a nutrient source, it is critical to balance the amount of N needed to maximize crop yield while reducing N loss to the environment. This 2-year study (2018-19) investigated the effects of N source [PL and CF as urea], application rate [0 (control), 168 (low rate) and 336 (high rate) kg total N/ha], and time (single and split application) on corn grain yield and ENL in a randomized complete block design at three locations [E.V. Smith Research Center (EVS), Wiregrass Research and Extension Center (WREC), and Tennessee Valley Research and Extension Center (TVREC)]. With the single application, the target N rate was applied pre-plant, whereas in a split application, one-fourth of the target N rate was applied pre-plant and the remaining three-fourth was side dressed at the V6 stage. A field-scale partial N budget was generated to quantify ENL. Nitrogen inputs included N contribution from N source treatments, and the soil background, while N outputs included plant N uptake, residual soil inorganic N ($\text{NH}_4\text{-N} + \text{NO}_3\text{-N}$) and total N lost to the environment (i.e. ENL). Plant and soil samples (0-15 and 15-30 cm depths) were taken at harvest each year for measuring aerial dry matter and residual inorganic N content, respectively. Relative to CF, application of PL increased grain yield only at EVS. No response to application timing for grain yield was observed at WREC but the split application of PL reduced grain yield at EVS and TVREC. Increased application of PL resulted in greater grain yield at WREC and TVREC compared to a low rate. Averaged across EVS and WREC, drier growing conditions of 2019 lowered grain yield by 29% compared to 2018. Aerial plant dry biomass followed similar patterns as grain yield at EVS and WREC whereas the highest whole plant N concentration was attained from urea relative to PL, regardless of the study site, application rate, and timing. We found no consistent results for ENL among N sources with significantly greater ENL reported from PL at WREC but no differences at TVREC. At EVS, urea had greater ENL than

PL at a single application whereas the opposite was true at a split application. Application timing did not influence ENL at WREC and TVREC. However, ENL increased with increasing N rate ranging from 32 to 52% of the total N input.

Introduction

Poultry (*Gallus gallus domesticus*) industry has witnessed rapid growth over the past decade in the southeast United States (MacDonald, 2008). Among the U.S. states, Alabama ranks second in terms of broiler production generating about 1.68 million tons of PL annually (USDA-NASS, 2019). Poultry litter, a mixture of mainly chicken feces and bedding material such as wood shavings of pine (*Pinus palustris* P. Miller or *Pinus elliottii* Englem.), or hulls of rice (*Oryza sativa* L.) or peanut (*Arachis hypogaea* L.), is regarded as a cheap source of plant nutrients including N (Poffenbarger et al., 2015) and an effective alternative to chemical fertilizers for row crop production including corn (Sistani et al., 2014; Endale et al., 2008). In northeast Alabama, corn grain yield was consistently increased by the addition of PL in 8 out of 9 yr studied (Watts and Torbert, 2011). Additionally, the use of PL positively influences many soil physical, chemical, and microbiological properties (Entry et al., 1997), thus improving overall soil health (Kingery et al., 1994; Mitchell and Tu, 2005; Schomberg et al., 2009).

Due to abundant availability, and to reduce cost on conventional fertilizers, many farmers choose to apply PL, often based on crop N demand (Pote et al., 1996). However, like every other N source, PL application may stimulate N losses such as ammonia (NH₃) volatilization, nitrous oxide (N₂O) release, and nitrate (NO₃) leaching. Many studies have evaluated environmental implications of excessive PL application (Kingery et al., 1994; Ritter, 2000; Cabrera and Sims, 2000). Over-application of PL has been reported to reduce yields (Shortall et al., 1975, Weil et al., 1979) and enhance leaching of nitrate into the groundwater (Liebhardt et al., 1979). To effectively utilize its nutrient value and maintain a balance between agronomic, economic, and environmental goals, we must understand and determine major N loss pathways from PL applications. Quantification of N loss from individual pathways is complex and involves time, additional equipment, and labor. For

instance, it is often difficult to accurately measure N leaching and involves costly lysimeter field trials (Uhlen, 1994). However, one cost-effective, simple, and robust way of measuring total N loss to the environment (including all possible leaks) is using a N budget. A general estimation of N losses will help identify Best Management Practices (BMPs) to reduce total N loss to the environment.

A N budget is defined as the record of N inputs and outputs at the field level (Onenema et al., 2003) which is based on the principle of mass conservation where the difference in N inputs into, and outputs from the system must equal the change in the system's total N (Legg and Meisinger, 1982; Meisinger and Randall, 1991). Nitrogen budgets are often used to evaluate the efficiency of a production system at the soil, field, or farm-scale (Prasad and Hochmuth, 2016). The difference between total N input and output has been established as an indicator of ENL (OECD, 2001). A N mass balance tool has been successfully used to account for N losses such as NH_3 volatilization, N_2O emission, and the potential NO_3 leaching based on changes in soil total N storage (Huang et al., 2017). Karlen et al. (1998) used a field-scale N budget in four watersheds farmed under continuous corn to estimate excessive residual soil $\text{NO}_3\text{-N}$ and reported an average of 50% of the fertilizer N was potentially lost to the environment. On the other hand, Liu et al. (2003) found that NO_3 leaching was the dominant process for N losses in the winter wheat–maize cropping system via N budgeting. Other studies have also used agricultural N balance as an indicator of the risk of N loss from specific farm units (Lord et al., 2002; Constantin et al., 2010; Duan et al., 2016).

Applying the optimum N rate that coincides with crop uptake is critical to maximize grain yield, minimize cost, and reduce negative environmental impacts. However, the correct N rate is often variable within a site and among years for the same site (Jaynes et al. 2011). It has been established that in regions with warm falls and mild winters such as

Alabama, PL applied in the spring result in higher corn grain yield than fall application, regardless of application rate (Jn-Baptiste et al., 2013; Tewolde and Sistani, 2010) which also holds true for CF. However, splitting application of CF between pre-plant and in-season has shown to maintain or increase corn yield (Jaynes, 2013) while reducing NO₃ leaching (Mitchell et al., 2000; Bakhsh et al., 2002). There is limited information on the optimum application rate and timing of PL for maximizing corn grain yield in Alabama. In addition, documentation of research on N loss comparisons between PL and CF is absent altogether. Therefore, the primary objective of this study was to evaluate the effects of N source, application rate, and time on corn grain yield, and ENL using a partial N mass balance approach with an aim to develop BMPs for PL focused on minimizing N losses without reducing economic productivity. The study also investigated the influence of N source, application rate, and time on corn dry matter accumulation and whole-plant N concentration.

Materials and Methods

Study sites

Field experiments were conducted in 2018 and 2019 at the E.V. Smith Research Center (EVS) near Shorter, AL (32° 25'N, 85° 53'S) and Wiregrass Research and Extension Center (WREC) near Headland, AL (31° 22' N, 85° 18'S) representing central and south production regions of Alabama. An additional site, Tennessee Valley Research and Extension Center (TVREC) near Belle Mina, AL (34° 41'N, 86° 53'S) representing north production region was included in 2019 for a total of five site-years. The soil types were Compass loamy sand (coarse-loamy, siliceous, sub active, thermic Plinthic Paleudults) with 1 to 3% slope at EVS, Dothan fine sandy loam (fine-loamy, kaolinitic, thermic Typic Kandiudults) with 0 to 2% slope at WREC, and Decatur silty clay loam (clayey, kaolinitic, thermic, Rhodic Paleudults) with 1 to 2% slope at TVREC.

Annual and 5-yr historic weather data (2013-2017) were collected from an automated weather station located at the field sites (Table 2.1). With a humid subtropical climate, the 5-yr mean annual rainfall was 1229, 1235, and 1215 mm at EVS, WREC, and TVREC, respectively. Growing degree days (GDD) for each site-year from planting till harvest were calculated as $GDD = \left[\frac{T_{max} + T_{min}}{2} \right] - 10$, where T_{max} and T_{min} are the daily maximum and minimum temperatures (°C), respectively. When $T_{max} > 30^{\circ}\text{C}$, T_{max} was set to 30°C. When $T_{min} < 10^{\circ}\text{C}$, T_{min} was set to 10°C.

Two of the three sites (EVS & WREC) were fallow the previous year while the TVREC site was cropped to soybean [*Glycine max* (L.) Merr.] with no manure application history at any of the sites. Prior to planting in the first year, plots at EVS and WREC were strip-tilled. However, in the subsequent year, plots were seeded with a no-tillage planter. Four to five soil cores (80-mm diameter) were randomly taken from the 0- to 15-cm and

15- to 30-cm depths across each study site before starting the experiment for baseline soil characteristics (Table 2.2).

Treatments

The experiment consisted of 2×2×2 factorial treatments arranged in a randomized complete block design with four replications. Factors included N source, application rate, and time. Poultry litter and urea were used as fertilizer N sources each applied at two target N rates, a low rate (168 kg total N ha⁻¹), and a high rate (336 kg total N ha⁻¹) in the single vs. split application. In a single application, the target N rate was applied within 7 d before planting in the spring, whereas in a split application, one-fourth of the target N rate was applied as pre-plant and the remaining three-fourth of the N was side dressed at the V6 stage (approximately 41 d after planting). In addition to these eight treatments, an untreated control (0 kg N ha⁻¹) was also tested to quantify the background contribution of soil on corn grain yield, whole plant dry matter, tissue N concentration, and potential environmental N loss (ENL).

Crop Management

Poultry litter was obtained from two broiler houses one week before single application. The average nutrient concentration of the litter applied each year is presented in Table 2.3. The calculated amount of PL and urea was weighed for individual plots and surface-broadcasted by hand without incorporation. Litter was applied based on total N analysis without any assumptions regarding N availability from PL (Mitchell and Tu, 2006). Individual plots of size 6.1 × 3.7 m (four-rows wide) were planted to dryland corn (cv. Pioneer P1662YHR) at a row spacing of 0.76- (TVREC) or 0.91-m (EVS and WREC) in the first 15 days of April each year (Table 2.4). All treatment plots received a blanket application of phosphorus (P₂O₅) and potassium (K₂O) each year at a rate of 45 kg ha⁻¹

based on soil test recommendations. This was done to mimic a commercial production system where farmers apply P and K based on soil test recommendations and still apply PL as an additional nutrient source. It also helped to nullify the effects of additional P and K applied with the manure. Weed control during each growing season was achieved following locally established management practices.

The 2018 cash crop was followed by winter rye (*Secale cereal* L., cv. Wrens Abruzzi) cover crop planted on 24 October at a seeding rate of 67 or 101 kg ha⁻¹ at WREC and EVS, respectively in 0.20 m row spacing. The cover crop was chemically terminated a week before replanting the same plots with corn in the subsequent year.

Sampling and Analyses

Total aboveground or aerial corn biomass (stover + grain) was measured at physiological maturity by cutting two to three plants randomly selected per treatment plot at the soil level. Cover crop aboveground biomass was also collected on 11 March 2019 (EVS) and 15 March 2019 (WREC) using two 0.25 m² quadrants placed randomly in each plot (Table S1). The harvested biomass was dried at 60°C in a forced-air oven for at least 72 h till constant weight was attained, weighed, and then ground with a Wiley Mill to pass a 2-mm screen.

Total carbon (C) and N concentration in PL and biomass subsamples was determined using a dry combustion LECO C/N analyzer (LECO Corp., St. Joseph, MI). Soil concentrations of P, K, calcium (Ca) and magnesium (Mg) were measured using an inductively coupled plasma-atomic emission spectrometry (ICP-AES; Spectro Ciros, Spectro Analytical Instruments Inc. Mahwah, NJ) with Mehlich-1 (M1) extractant in a 1:4 soil/M1 extractant ratio. Three to four PL samples were drawn each year during procurement and analysed for total P, K, Mg, Ca, B, Zn, Mn, Fe, and Cu concentrations by

ICP-AES following dry-ashing and acid-digestion of litter subsamples as outlined by Donohue and Aho (1992).

Corn grain yield was determined by combine harvesting the middle two rows of each plot and adjusting moisture content to 155 g kg⁻¹. Two soil samples from each treatment plot were also taken during harvesting each year at 0- to 15- and 15- to 30- cm depths to measure residual inorganic N (NH₄-N and NO₃-N). Soil samples were air-dried, ground with a Dynacrush soil crusher (Custom Laboratory Inc., Orange City, IA) to pass a 2-mm mesh screen, and measured for inorganic N (NH₄-N and NO₃-N) with a 1:10 soil/2M KCL (potassium chloride) ratio (Keeney and Nelson, 1982). Extracts were then analyzed using the FIAlyzer-1000 flow injection analyzer (FIALab Instruments Inc, Seattle, WA).

Nitrogen balance

A partial N balance was calculated by accounting for known inputs and outputs. The total known N input ($N_{\text{total input}}$) in this experiment was calculated as:

$$N_{\text{total input}} = N_{\text{input from fertilizer or manure}} + N_{\text{background contribution}} + N_{\text{carry over from the 2018 corn}} \quad (1)$$

Where, $N_{\text{input from fertilizer or manure}}$ is N via fertilizer applications, $N_{\text{background contribution}}$ is N via soil N mineralization, crop seed, biological N fixation (symbiotic + non-symbiotic fixation), and atmospheric N depositions (wet and dry deposition), which was estimated from the plant uptake in the control plots (Huang et al., 2017), and $N_{\text{carry over from the 2018 corn}}$ is N via the residual effects of manure application, which was estimated from the cover crop uptake in the treatment plots, minus N removed in the control plots.

Total N output ($N_{\text{total output}}$) was calculated as:

$$N_{\text{total output}} = N_{\text{crop removal}} + N_{\text{soil residual inorganic N}} + N_{\text{total N loss to the environment}} \quad (2)$$

Where, $N_{\text{crop removal}}$ is plant uptake from the soil, $N_{\text{soil residual inorganic N}}$ is soil inorganic N status at physiological maturity, and $N_{\text{total N loss to the environment}}$ is N lost via all possible pathways such as NH_3 volatilization, denitrification, leaching, surface runoff, erosion and other gaseous losses (N_2O , NO_x).

Applying the principle of mass conservation, the sum of N inputs should be equal to the sum of N outputs (Legg and Meisinger, 1982; Meisinger and Randall, 1991, Sainju, 2017).

$$N_{\text{input from fertilizer or manure}} + N_{\text{background contribution}} + N_{\text{carry over from the 2018 corn}} = N_{\text{crop removal}} + N_{\text{soil residual inorganic N}} + N_{\text{total N loss to the environment}} \quad (3)$$

The total N loss to the environment or environmental N loss (ENL) was calculated from rearranging eq. (3) as:

$$N_{\text{total N loss to the environment}} = N_{\text{input from fertilizer or manure}} + N_{\text{background contribution}} + N_{\text{carry over from the 2018 corn}} - N_{\text{crop removal}} - N_{\text{soil residual inorganic N}} \quad (4)$$

Statistical analysis

Analysis of variance (ANOVA) was determined for treatment effects of N source, application rate, and time using PROC GLIMMIX as an augmented factorial design (Piepho et al., 2006) in SAS 9.4 (SAS Institute, 2013). The experiment comprised an untreated control in addition to the $2 \times 2 \times 2$ factorial treatment structure (i.e. 8 + 1). The analysis compared the control to the treatments and also analyzed the factorial structure. Data for each location was analyzed separately with treatment effects, year, and their interactions as fixed effects. Blocks were treated as random effects. Locations were analyzed independently due to significant location \times treatment interaction for the responses. Treatment means were separated using Fisher's protected LSD at $\alpha = 0.05$ probability level (Littell et al., 2006).

Results and Discussion

The results are presented and discussed for individual sites.

Weather

Rainfall received and temperature conditions during the growing season (April to September) were the key weather factors affecting dryland corn production and ENL. Total monthly precipitation fluctuated greatly within the growing season and among years, and locations (Table 2.1). The total growing season rainfall for 2018 was 704 and 918 mm at EVS and WREC, respectively whereas 522, 515, and 586 mm for 2019 at EVS, WREC and TVREC, respectively. The 2019 growing season received 26 and 44% less precipitation than 2018 at EVS and WREC, respectively. As a result, corn at EVS and WREC suffered water and heat stress in 2019. Therefore, to avoid the risk of crop failure, supplemental irrigation of 152 mm and 25 mm was provided at EVS and WREC, respectively (Endale et al., 2008).

Monthly average air temperatures at the experimental sites did not deviate much among years (i.e. not greater than 2–3°C). Average monthly air temperature during the growing season was lowest in April (ranging from 16.1 to 19.2°C) and highest in July (26.6 to 27.9°C) across the locations with a growing season average of 24.9, 25.2, and 23.9°C at EVS, WREC and TVREC, respectively. The accumulation of GDD by corn plants was highest in the months of July and August for all three sites.

Corn grain yield

(a) EVS: Corn grain yield at EVS varied with treatments and years with no significant interaction between treatment and year (Table 2.5). Results showed that although the main effect of N source on corn grain yield was statistically significant ($P = 0.0002$), the source \times rate and source \times time interactions greatly influenced grain yield

(Table 2.6). Averaged across application timings, PL produced 18% ($P>0.05$) and 70% ($P\leq 0.05$) higher grain yield than urea at a low and high rate, respectively (Figure 2.1a). Positive yield response to PL, relative to equivalent conventional fertilizer, have also been reported by Sistani et al. (2008) and Endale et al. (2008). However, increasing N fertilization, regardless of source, did not result in higher grain yields. Also, there was no significant grain yield difference between urea-treated and the control plots suggesting that N was not the yield-limiting factor. Soil P and K would have met the crop demand in both urea and control treatments since they were applied based on Auburn University soil test recommendations while soil test Ca ($>250 \text{ mg kg}^{-1}$) and Mg ($>13 \text{ mg kg}^{-1}$) at this site were rated “high”, indicating adequate fertility levels (Mitchell and Huluka, 2012; Table 2.2). Yield response to PL application could be related to some other factors such as availability of other essential mineral elements (Sistani et al., 2008) or improved soil biological properties (Acea and Carballas, 1996; Pratt and Tewolde, 2009). Averaged across application rates, PL resulted in a 107% higher corn grain yield ($P\leq 0.05$) compared to urea with a single application ($4.96 \text{ vs. } 2.40 \text{ Mg ha}^{-1}$) (Figure 2.1b). This could be attributed to the rapid early growth of corn from PL applied at the time of planting, translating into relatively greater grain yield compared with the urea (Figure 2.2). These findings were also reported by Endale et al. (2008). However, grain yield differences were non-significant with the split application of PL or urea. Looking at individual N sources averaged across application rates, PL had a 34% lower grain yield ($P\leq 0.05$) when split-applied relative to a single application ($3.26 \text{ vs. } 4.96 \text{ Mg ha}^{-1}$) whereas split application of urea produced 45% higher grain yield ($P\leq 0.05$) than single application ($3.47 \text{ vs. } 2.40 \text{ Mg ha}^{-1}$). Comparing both years, corn grain yield was significantly lower in 2019 than in 2018 ($2.85 \text{ vs. } 4.03 \text{ Mg ha}^{-1}$) (Table 2.7). This difference was most likely due to deficit rainfall received during the 2019 growing season compared to 2018 (522 vs. 703 mm).

(b) WREC: Corn grain yield at WREC varied between treatments and years, but with a significant interaction for treatment \times year reflecting the impact of year-to-year variability in weather conditions (Table 2.5). Thus, treatments are presented and discussed separately for each year. In 2018, the main effects of source and rate were significant ($P \leq 0.05$) on corn grain yield with no influence of application time ($P = 0.3305$); however, the interaction of source \times rate was also significant (Table 2.6). Averaged across application timings, urea gave 115% higher yield ($P \leq 0.05$) than PL at a low rate (6.89 vs. 3.20 Mg ha^{-1} ; Figure 2.3). However, there were no significant differences in yield at high rates of urea or PL. The application rate of urea did not affect grain yield but applying PL at a high rate greatly increased grain yield compared to a low rate. Furthermore, grain yield from the plots which received PL at a low rate was not statistically different from that in the control treatment. These results indicate that litter applied at 168 kg N ha^{-1} did not supply enough plant-available N to match the grain yield equal to that of urea. This difference was also reflected in the lower whole-plant N concentration in the PL-treated plots (data not shown). Since N nutrition of corn plant increased with application rate (Table 2.10), litter application at a high rate might have provided adequate plant-available N to produce yield equivalent to urea fertilization. The results support Tewolde et al. (2013) who reported lower grain yield from corn fertilized with PL than conventional fertilizer in the first growing season due to limited supply of plant-available N from the litter. In 2019, main effects of source and rate were significant ($P \leq 0.05$) on corn grain yield without application time and their interaction effects (Table 2.6). Among N sources, urea produced the highest grain yield of 4.85 Mg ha^{-1} followed by PL (3.79 Mg ha^{-1}) and control (2.66 Mg ha^{-1}). This is in contrast with Tewolde et al. (2013) who reported a positive residual effect of PL on grain yield in subsequent years. We speculate that N losses such as NH_3 volatilization, NO_3 leaching, and denitrification may have played a role (Tewolde et al., 2009a; Motavalli et

al., 2008; Sistani et al., 2011) and is consistent with ENL results (Table 2.12). Sistani et al. (2014) also reported no yield response to PL applied each year at a rate of 168 kg plant available-N ha⁻¹ in a 3-yr field study (2009-2011) and concluded losses of N as one of the many factors responsible for the lack of significant corn grain production from PL compared to chemical fertilizer. Increasing the application rate increased corn grain yield, irrespective of application source, and timing (Table 2.6). Corn grain yield was increased 78% and 47% from the application of high and low N rate, respectively compared with the control (2.66 Mg ha⁻¹). Although, ENL also increased with increasing N rate (Table 2.12) the excess N supplied by a high rate may have been adequate for crop need and compensated for the losses. Also, splitting the N application gave no yield benefits either year, regardless of source or rate. Zhang et al. (1993) in a 3-yr field experiment also found that corn grain yield increased with increasing N application and was not significantly affected by different application times. In another study, Miller et al. (1975) reported a significant effect of N rate on corn yield, however, yield differences were similar between spring and summer side-dress applications on two alluvial soils in Ohio County, Kentucky.

(c) TVREC: At TVREC, there was a significant effect of treatments (Table 2.5). Corn grain yield varied with N source ($P < 0.0001$) and rate ($P = 0.0001$) but not with application time (Table 2.6). However, there was a significant impact of source \times time interaction on grain yield ($P < .0001$). Grain yield increased greatly with an increase in the N application rate. The control treatment had the lowest grain yield ($P < 0.05$) among all fertilizer treatments. Averaged across application rates, PL did not result in higher grain yield compared to urea, regardless of application timing. Split application of urea produced 60% greater grain yield ($P < 0.05$) than PL (9.49 vs. 5.94 Mg ha⁻¹) and 21% higher yield ($P < 0.05$) than single application (9.49 vs. 7.82 Mg ha⁻¹; Figure 2.4). It has been long known

that side-dress fertilizer applications result in higher N-use efficiencies (NUEs) (Aldrich, 1984).

Aerial plant dry matter

(a) EVS: The aerial dry matter of corn plants at EVS varied with treatments and years (Table 2.5). Although the main effects of N source, rate, and application time on aerial plant dry matter were not significant ($P > 0.05$), there was a significant interaction for source \times time (Table 2.8). Like grain yield, plant dry matter followed similar patterns with PL producing significantly greater plant biomass under a single application than urea, which was reflective of field observations (Figure 2.5). The corn plants grown on the plots which received a single application of PL showed more vigorous early season growth than those on urea plots (Figure 2.2). A similar trend in corn growth was also reported by Endale et al. (2008). These results agree with Jn-Baptiste et al. (2013) and Schomberg et al. (2011) who reported higher plant biomass from PL than chemical fertilizer (ammonium nitrate, NH_4NO_3 or ammonium sulphate, $(\text{NH}_4)_2\text{SO}_4$) applied before planting at equivalent amounts of plant-available N. On the other hand, Sistani et al. (2014) and Cooperband et al. (2002) reported similar aboveground plant dry matter with PL and urea applied at a rate of 168 kg plant available-N ha^{-1} . With the split application, urea increased plant biomass although not significantly ($P > 0.05$), while PL reduced plant biomass ($P < 0.05$) compared to a single application, resulting in similar plant biomass among sources under the split application.

(b) WREC: Aerial plant biomass at WREC varied with treatments and years (Table 2.5). Although the main effect of N source was significant ($P = 0.0121$) and that of application rate was non-significant ($P = 0.0601$) on plant dry matter accumulation, there was a significant interaction for source \times rate (Table 2.8). However, application timing, regardless of the N source and rate, did not greatly influenced corn biomass ($P > 0.05$).

Averaged across application timings, urea application at a low rate resulted in significantly greater aboveground plant biomass relative to the PL, and control treatment (Figure 2.6). However, both N sources had a similar dry matter yield at a high rate. at WREC. Such similarities between biomass and corn yield were also reported by Cooperband et al. (2002) and observed at EVS. As previously mentioned, biomass and yield differences between PL and urea-treated plots may be explained by low N availability from the PL during the growing season as evidenced by whole plant N concentrations (Table 2.10). Among years, higher plant biomass was reported in 2018 than in 2019 at both EVS and WREC likely due to greater rainfall in 2018 (Tables 2.7 and 2.9).

(c) TVREC: At TVREC, only the main effect of application rate was significant on aerial plant dry matter (Table 2.8). The aboveground plant dry matter increased significantly with an increase in the N application rate. Jn-Baptiste et al. (2012) also found a 10% increase in plant biomass by increasing PL application from 9 to 18 Mg ha⁻¹. However, no significant effects of N source and application timing were observed on plant biomass. This could be attributed to the trends in ENL among the treatment factors (Table 2.12). Sistani et al. (2014) also found no significant plant biomass difference between PL and urea in Central Kentucky and speculated that N losses might have played a role.

Whole-plant N concentration

(a) EVS: At EVS, whole-plant N concentration varied with rainfall amount and distribution causing significant interaction for treatment × year (Table 2.5). In 2018, all the main effects (i.e. N source, application rate, and time) and rate × time interaction were significant on whole-plant N concentration (Table 2.10). Averaged across N sources, regardless of N rate, the split application had lower whole-plant N concentration, whether differences were statistically significant or not (Figure 2.7). And, increasing the N rate

increased the whole-plant N concentration only when applied in a single application. In 2019, only the main effect of N source had a significant ($P = 0.0212$) impact on whole-plant N concentration (Table 2.10). In both years, urea produced the highest whole-plant N concentration that was significantly different from the PL and the control. Interestingly, urea resulted in better plant N nutrition than PL in both years, but it did not translate into yield or dry matter differences. This is in agreement with Endale et al. (2008), who observed similar mean N content of leaves and stalks between PL and CF but grain yield and dry matter production were greater by 18% and 26%, respectively with PL. Jn-Baptiste et al. (2013) also observed that corn plants seemed greener in the NH_4NO_3 (146 kg N ha^{-1}) treatment but had a similar dry matter or grain yield than those at the low PL application (68 kg N ha^{-1}). This suggests that there may be yield-limiting factors other than N which PL addition can overcome (Adeli et al., 2005).

(b) WREC and TVREC: At WREC and TVREC, whole-plant N concentration varied among treatments with no influence of years, treatment \times year interactions, but significant main effects of N source and rate (Tables 2.5 and 2.10). However, no significant influence of application time was observed on whole-plant N concentration. At both locations, urea had the highest whole plant N concentration followed by PL and the control treatments which reported similar concentrations. Whole-plant N concentration increased with increasing N rate and was not affected by application timing of N fertilization. These results do not agree with Sistani et al. (2014) who reported similar whole plant N concentration in a 3-yr study evaluating corn response to urea and PL applied at 168 kg N ha^{-1} .

Environment Nitrogen Loss (ENL)

(a) EVS: At EVS, ENL varied with treatments with no influence of years or treatment \times year interactions (Table 2.5). Among treatments, the main effects of application rate ($P = <.0001$) and time ($P = 0.0243$) were significant on ENL along with source \times time and rate \times time interactions (Table 2.11). Averaged across application rates, urea had significantly higher ENL than PL when applied in a single application (Figure 2.8a). The high-water solubility of urea and significant rainfall event (25 mm) on the day of the single application in 2018 might have greatly increased NO_3 leaching (Figure S1a). However, 2019 was mostly dry (with a total rainfall of 19.56 mm within 10 days of single application; Figure S1b) indicating low leaching and denitrification losses, but high potential for NH_3 volatilization losses from surface-applied urea which increases with increasing soil temperature and decreasing soil water content (Clay et al., 1990). Also, similar ENL values were reported for both single- and split-applied urea. The reduced NUE efficiency from split-applied urea was again probably due to climatic conditions following application like significant rainfall events ($>20\text{mm}$) that occurred on the same or next day of split application in both years (Figure S2). On the other hand, PL had greater N loss to the environment than urea under split application but reported similar grain yield and plant biomass; thus, further supporting the non-limiting effect of N on corn growth. The higher ENL from the split application of PL might have occurred due to a mismatch between N release resulting from mineralization and crop N demand. Averaged across N sources, increasing N rate increased ENL, regardless of application timing (Figure 2.8b). Furthermore, at a low rate, both application timings reported similar ENL. However, at a high rate, the split application had significantly greater ENL than a single application. These results suggest that higher N rates are likely less effective when applied late in the season than applied at planting. Similar conclusions were drawn by Miller et al. (1975)

when 224 kg N/ha side-dressed (4-6 weeks after planting) at the soil surface gave similar corn grain yield as N applied at the same rate at planting.

(b) WREC: At WREC, ENL varied among treatments with source and rate (Tables 2.5 and 2.11). However, there was no significant effect of application time on ENL ($P = 0.0793$). The PL-treated plots had 23% higher ENL than urea (191 vs. 155 kg ha⁻¹; Table 2.12). We suspect that higher ENL from PL could be due to greater volatilization losses because litter N was surface-applied to a dry soil under no-till. Under similar conditions, Sharpe et al. (2004) reported an NH₃ loss of 24% of total N applied from PL with maximum volatilization rate occurring within 48 h of litter application. Also, the high N application rate increased ENL from 38% (at a low rate) to 52% of the total N applied. Although split applications of N fertilizers are recommended for coarse-textured soils with low nutrient holding capacity and high leaching potential (Murrel, 2006), no such response was observed on ENL which was consistent with corn yield and plant biomass data. This was likely due to greater rainfall received within 1 d after or before split application (74 and 28 mm in 2018 and 2019, respectively) compared to a single application (1 and 4 mm in 2018 and 2019, respectively) which could have led to significant nitrate leaching from urea (Figure S3).

(c) TVREC: Like EVS and WREC, a similar trend in ENL for N rate was observed at TVREC (Tables 2.11 and 2.12). However, the effects of N source and time on ENL were not significant ($P > 0.05$). Many studies, including ¹⁵N work, showed that 50-60% of applied N is utilized by growing crops (Janzen et al., 1990; Aulakh et al., 1992; Smil, 1999; Tran and Tremblay, 2000). In the United States, Snyder (2012) reported that corn uses 37 to 51% of total fertilizer N applied annually. Overfertilization may lower NUE (Shanahan, 2011), and increase N losses (Raun and Johnson, 1999). For instance, in our study, the application of N at a high rate did not result in additional grain yield at EVS while crop productivity

was lower at TVREC compared to a low rate; thereby, increasing the risk of N losses to the environment. Jaynes et al. (2001) also reported higher NO₃-N loss which increased from 29 to 48 kg ha⁻¹ with an increase in N fertilizer rates (57 to 202 kg ha⁻¹) from a 22-ha subsurface drained field near central Iowa. No influence of N source and application timing on ENL at TVREC was likely due to similar aerial plant biomass among treatment factors (Table 2.8) and consequently plant N uptake (data not shown), which is the major N output in mass balance calculations.

The total amount of N lost to the environment i.e. ENL, expressed as a percentage of total N applied, for main treatment effects at each location, is shown in Table 2.12; ranging from 39 to 50%, 38 to 52%, and 32 to 48% at EVS, WREC, and TVREC, respectively. These results are similar to with Huang et al. (2017) who estimated that 24 to 48% of the total N input was lost to the environment using N balance for various fertilizer treatments. Furthermore, this agrees with a large body of scientific literature showing estimates of NUE around 50%, as mentioned above.

Conclusion

The study showed that under adequate soil N levels such as at EVS, corn grain yield was increased by PL. The response was probably due to the supply of other essential mineral elements (i.e. micronutrients) or potential soil health benefits. However, where corn grain yield was limited by the plant-available N (PAN) levels in the soil as in most cases (WREC and TVREC in our study), urea outperformed PL and grain yields were increased with increasing N application rate, regardless of the source. Although no response to application timing on corn grain yield was observed at WREC but the split application of PL reduced grain yield compared to the pre-plant application at EVS and TVREC. Averaged across EVS and WREC, drier growing conditions of 2019 lowered grain yield by about 29% compared to 2018. Aerial plant dry biomass followed similar patterns as of grain yield at EVS and WREC with the highest whole-plant N concentration attained from urea application, regardless of the study site, N source, application rate, and timing. Conflicting results for ENL related to N source were reported at the study sites with significantly greater ENL reported from PL at WREC but similar values reported at TVREC. At EVS, urea had higher ENL than PL at a single application whereas the opposite was true at a split application. Application timing did not influence ENL at WREC and TVREC. However, ENL increased with increasing N rate from 38 to 49, 38 to 52, and 32 to 48% of the total N input at EVS, WREC, and TVREC, respectively.

Findings from this study provided valuable information needed to make BMPs decisions for PL use in Alabama's corn production systems. For efficient use, PL should be applied prior to planting in a single application at a low rate. High rate and side dress applications of PL should be discouraged due to the potential N losses and reduced crop productivity. However, future work is required to investigate the N release dynamics,

improvements in soil fertility and other benefits (both physical and biological) from PL additions and how they influence corn production at different locations in Alabama.

Table 2.1. Monthly average air temperature, total precipitation, and corn growing degree days (GDD) during the growing season (April to September) at the experimental sites.

	EVS [†]			WREC			TVREC	
	2018	2019	5-yr avg.	2018	2019	5-yr avg.	2019	5-yr avg.
Air temperature, °C								
April	16.1	17.9	17.8	16.5	19.2	19.9	16.5	17.5
May	23.8	24.0	20.9	23.7	25.5	23.3	22.8	21.2
June	26.7	26.4	25.3	26.3	27.7	26.5	24.9	25.9
July	27.4	27.7	26.4	26.8	27.9	27.6	26.6	26.8
August	26.7	27.8	26.2	26.3	27.8	27.3	26.1	26.2
September	27.1	27.3	24.1	26.9	27.9	25.2	26.2	23.9
Precipitation, mm								
April	90	174	149	127	122	147	159	114
May	119	144	120	196	42	94	108	103
June	127	109	127	110	85	102	113	103
July	80	51	115	225	113	95	120	138
August	148	41	81	180	142	146	66	66
September	140	3	31	80	11	90	20	50
April-September	704	522	623	918	515	674	586	574
January-December	1521	1148	1229	1886	1033	1235	1637	1215
Cumulative growing degree days (GDD), °C								
April	165	231	201	128	249	256	176	198
May	579	642	542	540	695	528	535	545
June	1043	1099	977	1003	1178	1122	945	989
July	1541	1592	1444	1497	1679	1619	1410	1461
August	2027	1915	1909	1970	2152	2111	1854	1920
September	2170	- ^{††}	-	2062	-	-	1962	2046

†E.V. Smith Research Center (EVS); Wiregrass Research and Extension Center (WREC); Tennessee Valley Research and Extension Center (TVREC).

††Harvesting was done in August (For cumulative GDD at EVS and WREC, corn growing period considered was from 5 April to 31 August).

Table 2.2. Baseline soil properties for 0- to 15- and 15- to 30-cm before treatment applications at each experimental site.

Property	EVS [†]		WREC		TVREC	
	0-15 cm	15-30 cm	0-15 cm	15-30 cm	0-15 cm	15-30 cm
pH ^{††}	6.2 (0.4 ^{†††})	6.0 (0.2)	6.4 (0.2)	6.3 (0.2)	6.2 (0.1)	6.2 (0.2)
Total C (g kg ⁻¹)	14.4 (0.8)	13.8 (1.7)	13.6 (1.2)	13.1 (1.4)	16.6 (1.1)	13.8 (0.4)
Total N (g kg ⁻¹)	3.5 (0.5)	3.7 (0.7)	0.4 (0.1)	0.35 (0.1)	4.3 (0.3)	4.3 (0.5)
Inorganic N (NH ₄ -N + NO ₃ -N; mg kg ⁻¹)	45.3 (12.0)	45.1 (8.9)	19.0 (8.3)	17.2 (7.1)	20.6 (3.5)	14.0 (3.5)
Mehlich-1 P (mg kg ⁻¹)	14.1 (10) M*	14.6 (8) M	26.2 (17.3) H	23.0 (17.8) M	24.8 (5.5) H	12.6 (1.7) M
Mehlich-1 K (mg kg ⁻¹)	42.3 (10) M	57.0 (30) M	80.2 (38.8) H	63.2 (21.9) H	120.4 (19.1) H	75.7 (8.9) M
Mehlich-1 Ca (mg kg ⁻¹)	603 (150) H	509 (56) H	434 (87) H	436 (92) H	979 (56.2) H	942 (100.9) H
Mehlich-1 Mg (mg kg ⁻¹)	42.2 (9) H	44.7 (12) H	107.8 (27.6) H	98.5 (21.1) H	75.5 (3.2) H	64.5 (5.9) H

[†]E.V. Smith Research Center (EVS; n = 4); Wiregrass Research and Extension Center (WREC; n = 4); Tennessee Valley Research and Extension Center (TVREC; n = 5).

^{††}Measured using a glass electrode in a 1:1 soil/water ratio.

^{†††}Standard deviation of the mean given in parenthesis.

*Rating codes were derived using the fertility recommendatins for Alabama soils (Mitchell and Huluka, 2012): L (low), M (medium), and H (high).

Table 2.3. Chemical properties of poultry litter (PL) applied to corn each year at the experimental sites.

Year	Moisture	Total C	Total N	P	K	Ca	Mg	B	Zn	Mn	Fe	Cu
	g kg ⁻¹	g kg ⁻¹						mg kg ⁻¹				
2018	262 (9 [†])	351.1	26.7	8.8 (1.5)	16.4	11.8	6.0	26	174.7	227 (10)	1498	59.3 (6.8)
		(10.0)	(1.0)		(1.7)	(0.8)	(0.3)	(2)	(10.1)		(306)	
2019	267	300.0	31.8	20.2	28.1	17.6	4.1	25	250.0	400	600 (0)	300.0
	(157)	(53.0)	(6.0)	(12.0)	(8.7)	(6.4)	(1.3)	(7)	(71.0)	(141)		(141.0)

[†]Standard deviation of the mean given in parenthesis (n = 3).

Table 2.4. Cultural practices adopted in the study at each experimental site.

Location	Pre-plant N application	Planting	Row spacing (m)	Seed rate ha ⁻¹	Side-dress N application	Harvesting
2018						
EVS [†]	29 March	5 April	0.91	75,335	29 June	10 September
WREC	6 April	13 April	0.91	71,729	28 June	7 September
2019						
EVS	29 March	5 April	0.91	75,335	16 May	21 August
WREC	1 April	5 April	0.91	71,729	14 May	30 August
TVREC	28 March	5 April	0.76	70,395	17 May	9 September

[†]E.V. Smith Research Center (EVS); Wiregrass Research and Extension Center (WREC); Tennessee Valley Research and Extension Center (TVREC)

Table 2.5. Test of significance for treatment, year, and treatment \times year interaction on corn grain yield, aerial plant dry matter, whole-plant nitrogen (N) concentration, and environmental N loss (ENL) at the experimental sites. Effects are statistically significant at $P \leq 0.05$.

Effect	Grain yield	Aerial plant dry matter	Whole plant N concentration	ENL
$P > F$				
EVS[†]				
Treatment (T)	<.0001	0.0009	<.0001	<.0001
Year (Y)	<.0001	0.0002	<.0001	0.3285
T \times Y	0.1454	0.8816	0.0241	0.1666
WREC				
Treatment (T)	<.0001	<.0001	<.0001	<.0001
Year (Y)	<.0001	<.0001	0.0113	0.4291
T \times Y	0.0073	0.0648	0.9238	0.3231
TVREC				
Treatment (T)	<.0001	0.0036	0.0010	<.0001

[†]E.V. Smith Research Center (EVS); Wiregrass Research and Extension Center (WREC); Tennessee Valley Research and Extension Center (TVREC).

Table 2.6. Corn grain yield measured across fertilizer treatments, and the analysis of variance (ANOVA) at each experimental site.

Effect	EVS [†]	WREC		TVREC
	----- 2018–2019	2018	2019	2019
Source (S)				
Urea	2.94b ^{††} (1.14 ^{†††})	7.03a (1.39)	4.86a (1.04)	8.66a (1.38)
Poultry litter	4.11a (1.46)	4.80b (1.82)	3.79b (0.92)	6.76b (1.25)
Control	2.75b (1.47)	2.25c (0.98)	2.66c (0.54)	4.39c (1.17)
Rate (R)				
Low	3.57a (1.57)	5.05b (2.23)	3.92b (1.16)	7.15b (1.42)
High	3.48a (1.29)	6.78a (1.14)	4.73a (0.83)	8.26a (1.65)
Control	2.75a (1.47)	2.25c (0.98)	2.66c (0.54)	4.39c (1.17)
Time (T)				
Single	3.68a (1.71)	6.12a (1.90)	4.64a (1.15)	7.70a (0.89)
Split	3.36ab (1.08)	5.72a (2.04)	4.01a (1.0)	7.71a (2.15)
Control	2.75b (1.47)	2.25b (0.98)	2.66b (0.54)	4.39b (1.17)
ANOVA (P>F)				
S	0.0002	<.0001	0.0037	<.0001
R	0.7672	0.0002	0.0204	0.0002
S × R	0.0396	0.0012	0.1227	0.3654
T	0.2933	0.3305	0.0627	0.9469
S × T	<.0001	0.7003	0.6147	<.0001
R × T	0.3437	0.1829	0.4019	0.1286
S × R × T	0.7041	0.2396	0.1572	0.1300

[†]E.V. Smith Research Center (EVS); Wiregrass Research and Extension Center (WREC); Tennessee Valley Research and Extension Center (TVREC).

^{††}Values followed by the same letters within a column for the same effect are not significantly different at $P \leq 0.05$.

^{†††}Standard deviation of the mean given in parenthesis.

Table 2.7. Effect of year on corn grain yield and aerial plant dry matter averaged over 2 years (2018–2019) at E.V. Smith Research Center (EVS).

Year	Yield (Mg ha ⁻¹)	Aerial plant dry matter (Mg ha ⁻¹)
2018	4.03a [†] (1.48 ^{††})	14.91a (4.0)
2019	2.85b (1.16)	11.42b (4.6)

[†]Values followed by the same letters within a column are not significantly different at $P \leq 0.05$.

^{††}Standard deviation of the mean given in parenthesis.

Table 2.8. Aerial plant dry matter averaged over 2 years (2018–2019) across fertilizer treatments, and the analysis of variance (ANOVA) at each experimental site.

Effect	EVS [†]	WREC	TVREC
	----- Mg ha ⁻¹ -----		
Source (S)			
Urea	11.97a ^{††} (4.54 ^{†††})	13.53a (3.16)	15.12a (2.59)
Poultry litter	13.56a (3.86)	11.53b (3.41)	14.60a (2.16)
Control	8.77b (4.24)	9.71b (2.08)	9.84b (1.70)
Rate (R)			
Low	12.50a (3.92)	11.80b (3.53)	13.89b (2.06)
High	13.03a (4.62)	13.26a (3.18)	15.83a (2.30)
Control	8.77b (4.24)	9.71b (2.08)	9.84c (1.70)
Time (T)			
Single	13.51a (5.13)	11.82ab (11.82)	15.54a (2.23)
Split	12.02a (3.02)	13.24a (13.24)	14.17a (2.36)
Control	8.77b (4.24)	9.71b (2.08)	9.84b (1.70)
ANOVA (P>F)			
S	0.2111	0.0121	0.4923
R	0.6202	0.0601	0.0167
S × R	0.1718	0.0044	0.1929
T	0.2929	0.0637	0.0813
S × T	0.0017	0.8592	0.7229
R × T	0.0523	0.1877	0.7241
S × R × T	0.8469	0.7388	0.7641

[†]E.V. Smith Research Center (EVS); Wiregrass Research and Extension Center (WREC); Tennessee Valley Research and Extension Center (TVREC).

^{††}Values followed by the same letters within a column for the same effect are not significantly different at $P \leq 0.05$.

^{†††}Standard deviation of the mean given in parenthesis.

Table 2.9. Effect of year on aerial plant dry matter and whole-plant N concentration at Wiregrass Research and Extension Center (WREC).

Year	Aerial plant dry matter (Mg ha ⁻¹)	Whole-plant N concentration (g kg ⁻¹)
2018	13.99a [†] (4.03 ^{††})	12.41b (1.76)
2019	11.18b (2.23)	13.22a (1.87)

[†]Values followed by the same letters within a column are not significantly different at $P \leq 0.05$.

^{††}Standard deviation of the mean given in parenthesis.

Table 2.10. Whole-plant N concentration measured across fertilizer treatments, and the analysis of variance (ANOVA) at each experimental site.

Effect	EVS [†]		WREC	TVREC
	g kg ⁻¹			
	2018	2019	2018–2019	2019
Source (S)				
Urea	12.24a ^{††} (2.02 ^{†††})	14.42a (1.56)	13.99a (1.65)	13.53a (1.82)
Poultry litter	9.94b (2.12)	12.97b (1.52)	11.98b (1.51)	11.42b (0.90)
Control	9.70b (0.67)	13.09ab (1.08)	11.46b (0.96)	11.74b (1.38)
Rate (R)				
Low	10.10b (1.82)	13.31a (1.59)	12.13b (1.70)	11.95b (1.48)
High	12.08a (2.33)	14.09a (1.74)	13.84a (1.63)	13.01a (1.93)
Control	9.70b (0.67)	13.09a (1.08)	11.46b (0.96)	11.74ab (1.38)
Time (T)				
Single	12.23a (2.42)	13.84a (1.54)	12.91a (1.95)	12.72a (1.72)
Split	9.95b (1.62)	13.56a (1.85)	13.06a (1.80)	12.24a (1.85)
Control	9.70b (0.67)	13.09a (1.08)	11.46b (0.96)	11.74a (1.38)
ANOVA (P>F)				
S	<.0001	0.0212	<.0001	<.0001
R	0.0001	0.1961	<.0001	0.0158
S × R	0.8064	0.9718	0.9591	0.1179
T	<.0001	0.6314	0.6510	0.2615
S × T	0.8164	0.7003	0.6843	0.3583
R × T	0.0073	0.5681	0.8944	0.7123
S × R × T	0.3525	0.2751	0.3973	0.6573

[†]E.V. Smith Research Center (EVS); Wiregrass Research and Extension Center (WREC); Tennessee Valley Research and Extension Center (TVREC).

^{††}Values followed by the same letters within a column for the same effect are not significantly different at $P \leq 0.05$.

^{†††}Standard deviation of the mean given in parenthesis.

Table 2.11. Analysis of variance (ANOVA) for environmental N loss (ENL) at each experimental site. Effects are statistically significant at $P \leq 0.05$.

Effect	EVS [†]	WREC	TVREC
	2018–2019	2018–2019	2019
	-----P > F-----		
Source (S)	0.9889	0.0018	0.0603
Rate (R)	<.0001	<.0001	<.0001
S × R	0.2291	0.2362	0.0647
Time (T)	0.0243	0.0709	0.0793
S × T	0.0025	0.9339	0.5214
R × T	0.0301	0.1441	0.3602
S × R × T	0.8838	0.7760	0.8847

[†]E.V. Smith Research Center (EVS); Wiregrass Research and Extension Center (WREC); Tennessee Valley Research and Extension Center (TVREC).

Table 2.12. Two-year (2018–2019) nitrogen (N) balance for different fertilization treatments at the experimental sites. The Tennessee Valley Research and Extension Center (TVREC) site included only one-year data.

Treatment	N inputs (kg ha ⁻¹)				N outputs (kg ha ⁻¹)			ENL (%)
	N _{input} from fertilizer or manure	N _{background} contribution	N _{carry over} from the 2018 corn	N _{total} input	N _{crop} removal	N _{soil residual} inorganic N	N _{total} N loss to the environment	
E.V. Smith Research Center (EVS)								
Source								
PL	252	104	9 (7 [†])	365	166 (63)	43 (16)	164a ^{††} (82)	46
Urea	252	104	12 (3)	368	169 (62)	48 (32)	161a (90)	44
Rate								
Low	168	104	9 (6)	281	154 (55)	40 (21)	105b (56)	38
High	336	104	11 (5)	451	182 (67)	50 (29)	216a (73)	49
Time								
Single	252	104	13 (3)	369	182 (73)	48 (26)	143b (82)	39
Split	252	104	7 (6)	363	153 (45)	43 (25)	179a (86)	50
Wiregrass Research and Extension Center (WREC)								
Source								
PL	252	114	1 (1)	367	145 (55)	31 (23)	191a (76)	52
Urea	252	114	1 (2)	367	195 (54)	24 (18)	155b (85)	42
Rate								
Low	168	114	0 (1)	282	151 (61)	26 (19)	107b (50)	38
High	336	114	2 (2)	452	190 (52)	29 (22)	234a (52)	52
Time								
Single	252	114	3 (2)	369	160 (61)	29 (20)	183a (89)	50
Split	252	114	0 (0)	366	180 (58)	26 (21)	164a (74)	45
Tennessee Valley Research and Extension Center (TVREC)								
Source								

PL	252	117	–	369	168 (33)	29 (10)	171a (86)	46
Urea	252	117	–	369	207 (53)	24 (8)	135a (64)	37
Rate								
Low	168	117	–	285	166 (34)	24 (8)	91b (35)	32
High	336	117	–	453	208 (51)	29 (10)	217a (54)	48
Time								
Single	252	117	–	369	199 (48)	29 (11)	135a (69)	37
Split	252	117	–	369	175 (46)	23 (7)	171a (85)	46

†Standard deviation of the mean given in parenthesis.

††Values followed by the same letters within a column for the same effect are not significantly different at $P \leq 0.05$.

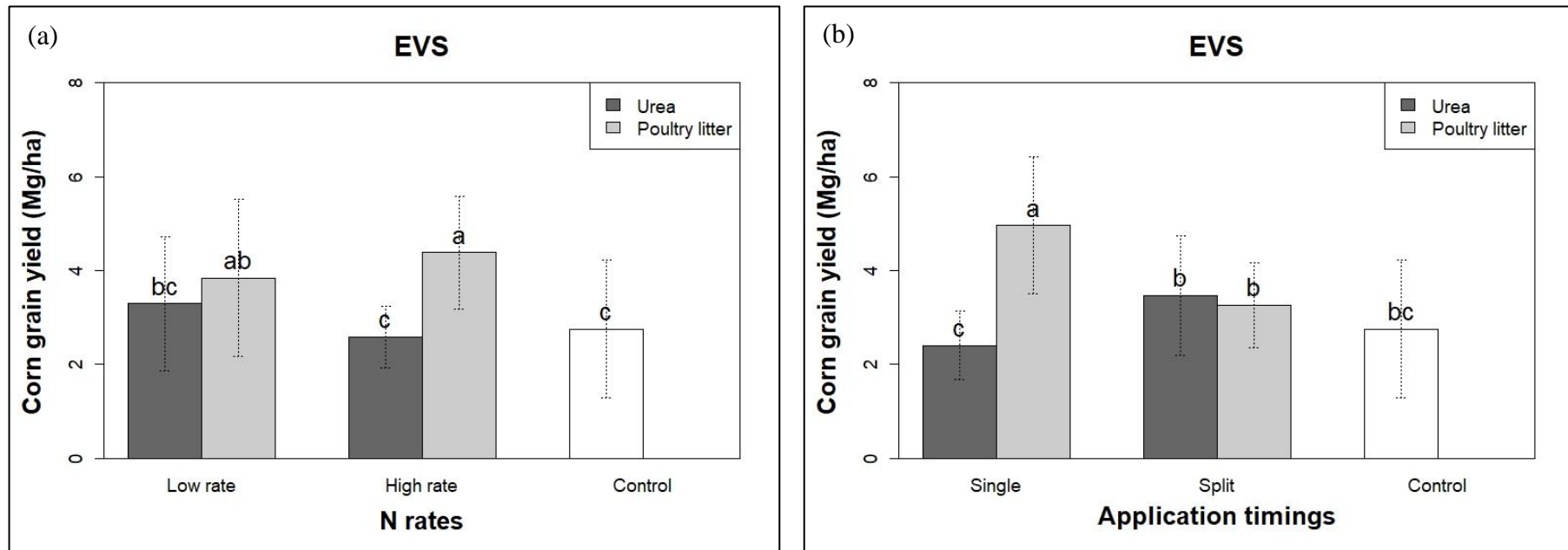


Figure 2.1. Effects of nitrogen (N) source (urea and poultry litter) × application rate (168 kg N ha⁻¹, a low rate and 336 kg N ha⁻¹, a high rate) interactions (2.1a) and N source × application time (100% target N applied as pre-plant, single application and 25% target N applied as pre-plant + 75% side dressed at the V6 growth stage, split application) interactions (2.1b) on corn grain yield averaged for 2 years (2018–2019) at E.V. Smith Research Center (EVS). An unfertilized control received no N, but P and K based on soil test recommendations. Grain yield values followed by the same letter are not statistically different from each other at $P \leq 0.05$. Error bar is the standard deviation of the mean.



Figure 2.2. Rapid early season growth of corn fertilized with poultry litter (PL) at the time of planting in 2019 at E.V. Smith Research Center (EVS).

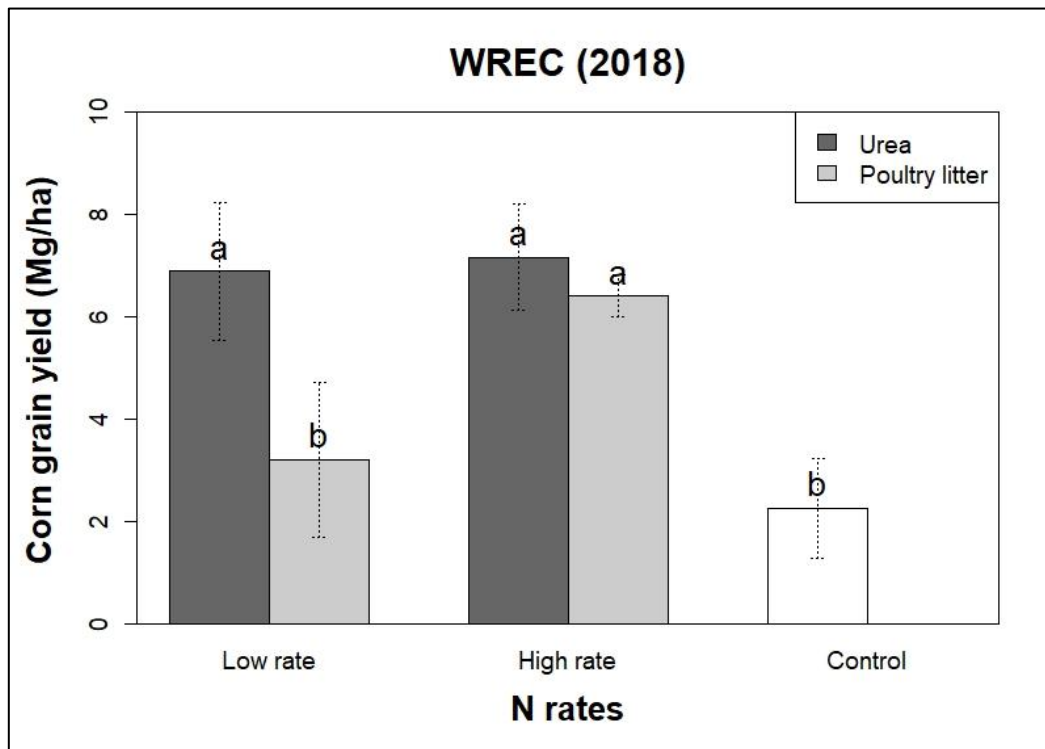


Figure 2.3. Effect of nitrogen (N) source (urea and poultry litter) \times application rate (168 kg N ha⁻¹, a low rate and 336 kg N ha⁻¹, a high rate) interactions on corn grain yield in 2018 at Wiregrass Research and Extension Center (WREC). An unfertilized control received no N, but P and K based on soil test recommendations. Grain yield values followed by the same letter are not statistically different from each other at $P \leq 0.05$. Error bar is the standard deviation of the means.

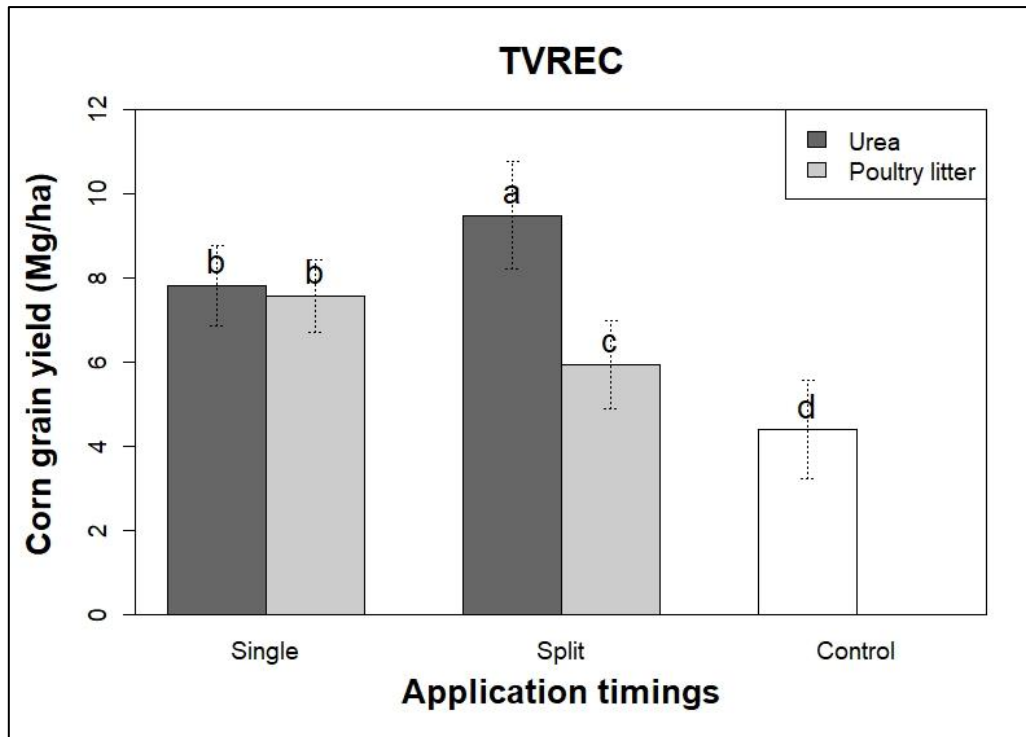


Figure 2.4. Effect of nitrogen (N) source (urea and poultry litter) × application time (100% target N applied as pre-plant, single application and 25% target N applied as pre-plant + 75% side dressed at the V6 growth stage, split application) interactions on corn grain yield at Tennessee Valley Research and Extension Center (TVREC). An unfertilized control received no N, but P and K based on soil test recommendations. Grain yield values followed by the same letter are not statistically different from each other at $P \leq 0.05$. Error bar is the standard deviation of the means.

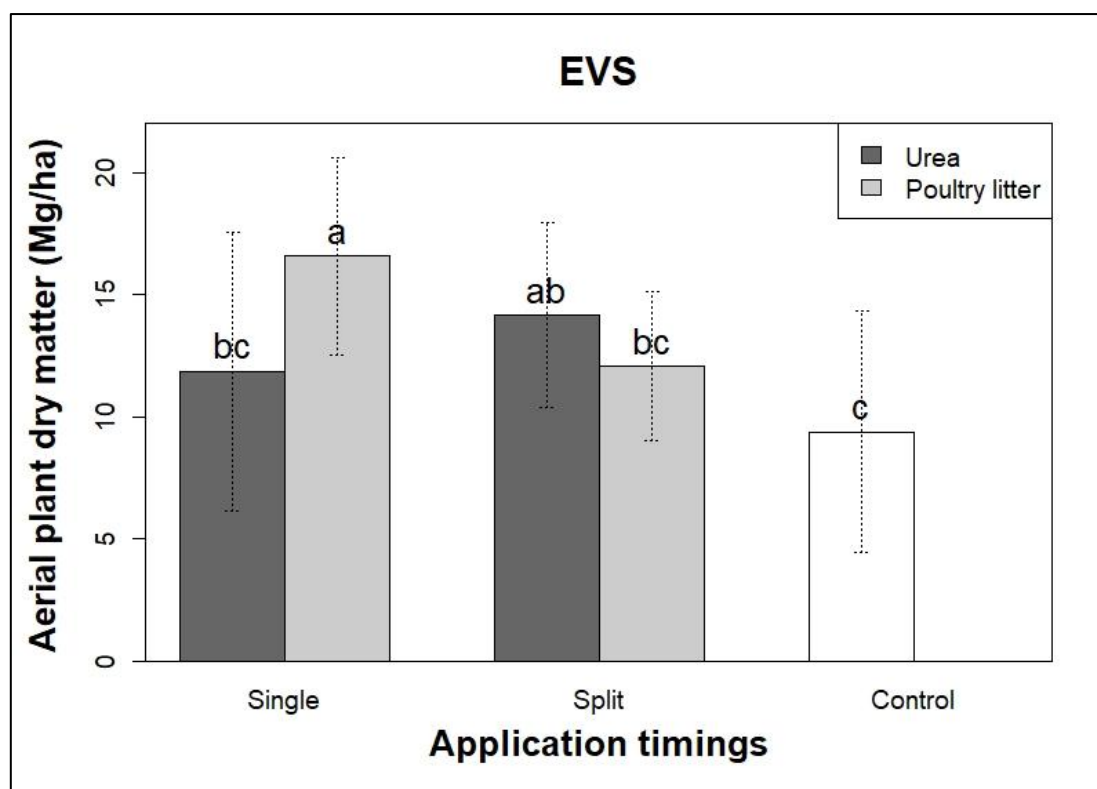


Figure 2.5. Effect of nitrogen (N) source (urea and poultry litter) × application time (100% target N applied as pre-plant, single application and 25% target N applied as pre-plant + 75% side dressed at the V6 growth stage, split application) interactions on aerial plant dry matter averaged for 2 years (2018–2019) at E.V. Smith Research Center (EVS). An unfertilized control received no N, but P and K based on soil test recommendations. Dry biomass values followed by the same letter are not statistically different from each other at $P \leq 0.05$. Error bar is the standard deviation of the means.

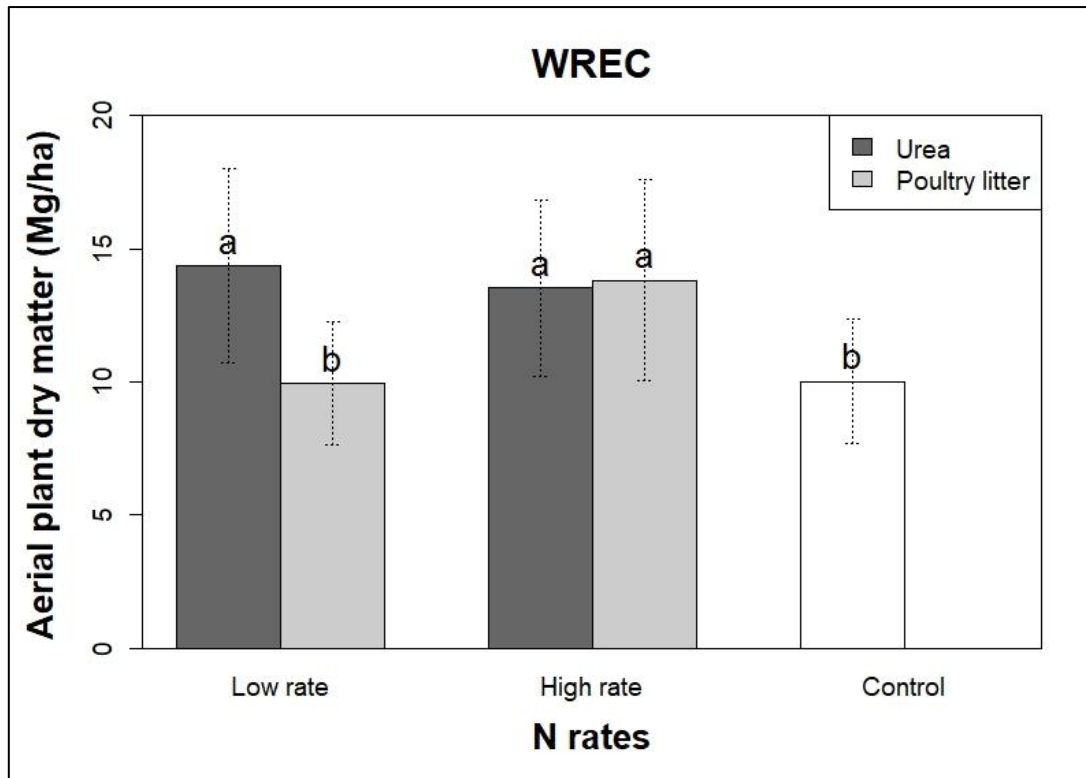


Figure 2.6. Effect of nitrogen (N) source (urea and poultry litter) \times application rate (168 kg N ha⁻¹, a low rate and 336 kg N ha⁻¹, a high rate) interactions on aerial plant dry matter averaged for 2 years (2018–2019) at Wiregrass Research and Extension Center (WREC). An unfertilized control received no N, but P and K based on soil test recommendations. Dry biomass values followed by the same letter are not statistically different from each other at $P \leq 0.05$. Error bar is the standard deviation of the means.

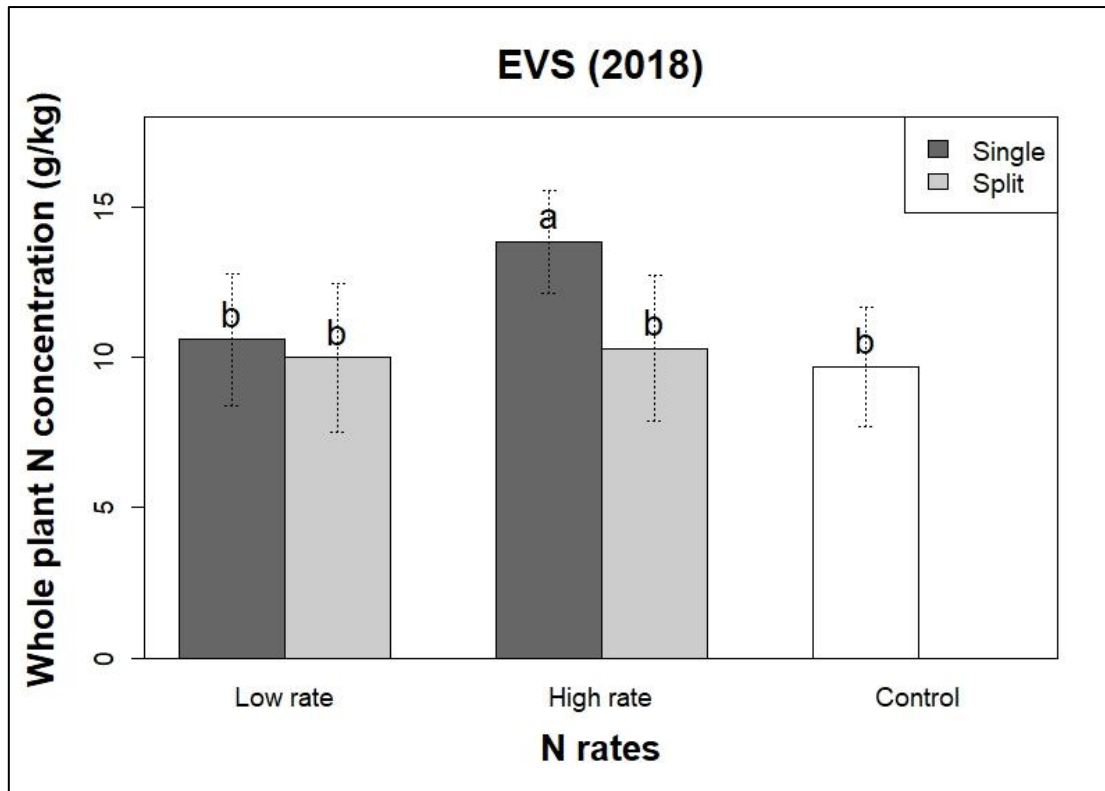


Figure 2.7. Effect of nitrogen (N) rate (168 kg N ha⁻¹, a low rate and 336 kg N ha⁻¹, a high rate) × application time (100% target N applied as pre-plant, single application and 25% target N applied as pre-plant + 75% side dressed at the V6 growth stage, split application) interactions on whole plant N concentration in 2018 at E.V. Smith Research Center (EVS). An unfertilized control received no N, but P and K based on soil test recommendations. Tissue N values followed by the same letter are not statistically different from each other at $P \leq 0.05$. Error bar is the standard deviation of the means.

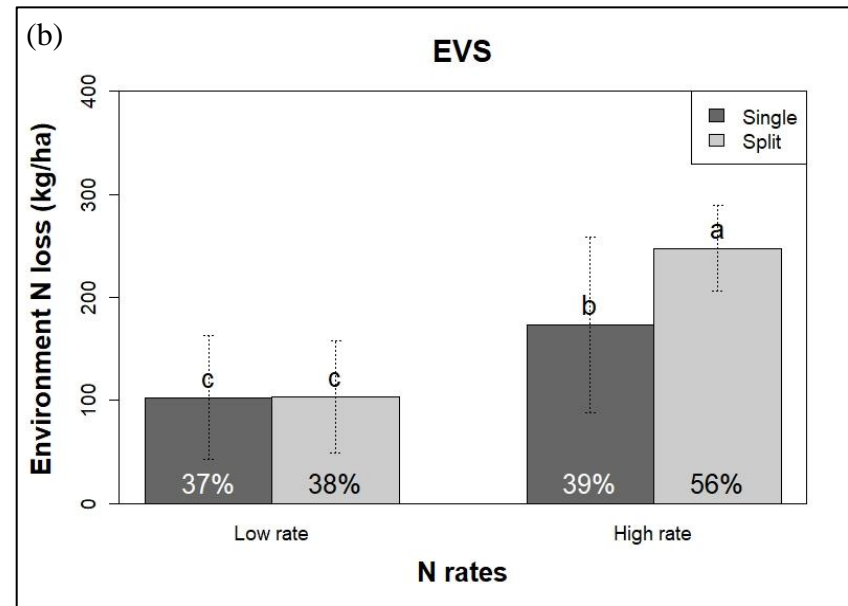
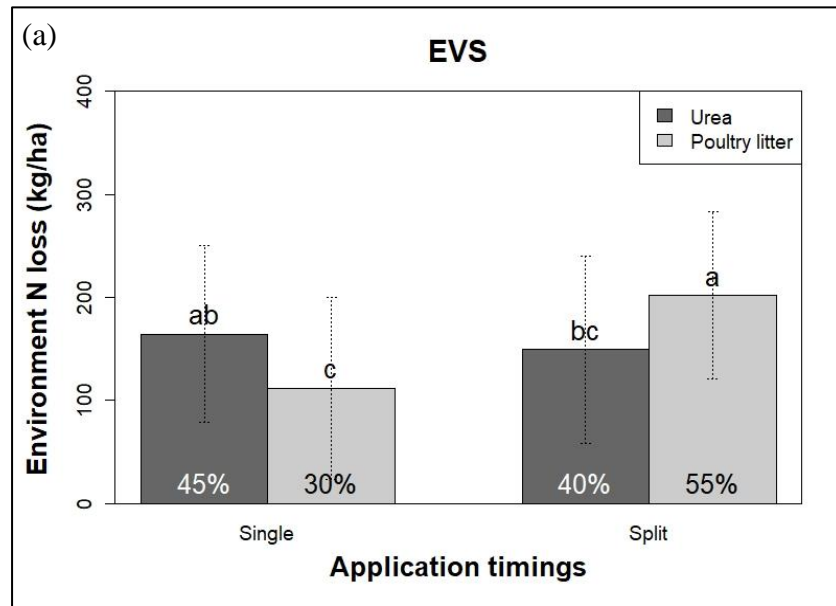


Figure 2.8. Effects of nitrogen (N) source (urea and poultry litter) × application time (100% target N applied as pre-plant, single application and 25% target N applied as pre-plant + 75% side dressed at the V6 growth stage, split application) interactions (2.7a) and application rate (168 kg N ha⁻¹, a low rate and 336 kg N ha⁻¹, a high rate) × application time (2.7b) on environmental N loss (ENL) averaged for 2 years (2018–2019) at E.V. Smith Research Center (EVS). Total N loss values followed by the same letter are not statistically different from each other at $P \leq 0.05$. Error bar is the standard deviation of the means. Percentages inside the bars are the ENL expressed as a percentage of the total N input.

III. Mineral Composition of Corn Ear as Affected by Poultry Litter Fertilization

Abstract

Mineral composition of corn ear (including grain, cob and husk plant parts) impacts its nutritional value and potential use as human food and animal feed. There are limited reports on whether application of poultry litter (PL) enrich corn ear with any of the mineral elements it supplies such as nitrogen (N), phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg) etc. This study investigated mineral composition of corn ear with no fertilization (control) or fertilization with 168 (low rate) or 336 kg (high rate) total N ha⁻¹ applied through PL or urea as conventional fertilizer (CF) in the single vs. split application. In single application, the target N rate was applied pre-plant whereas in split application, one-fourth of the target N rate was applied pre-plant and the remaining three-fourth was side dressed at the six-leaf stage. Field experiments were conducted in 2018 and 2019 at three locations [E.V. Smith Research Center (EVS), Wiregrass Research and Extension Center (WREC), and Tennessee Valley Research and Extension Center (TVREC)] in a randomized complete block design with four replications for a total of five site-years. Ear samples were harvested at physiological maturity each year and analyzed for the contents of 11 mineral elements namely N, P, K, Ca, Mg, sulphur (S), boron (B), zinc (Zn), manganese (Mn), iron (Fe), and copper (Cu). Poultry litter regardless of application rate, time, and location did not increase concentration of the selected mineral elements in the ear. Corn fertilized with CF had the highest ear concentrations of N, P, K, Ca, Mg, Zn, Mn, Fe, and Cu at all locations which increased with increasing N rate. Ear N concentration was dependent on the level of its plant available forms in the soil. However, ear levels of P, K, and other mineral elements were not in proportion to their soil levels, but dependent on ear N concentration. Results indicated that adequate plant-available N (PAN) in the soil, irrespective of whether derived from PL or CF, ensures maximum accumulation of ear N along with P, K, and other mineral nutrients.

Introduction

Broiler (*Gallus gallus domesticus*) industry has expanded rapidly in the United States, especially in the southeastern region increasing from 10 billion pounds of broilers produced in 1968 to about 57 billion pounds in 2018 (USDA-NASS, 2019). Currently, Alabama ranks second nationally in broiler production behind Georgia with 1.12 billion birds produced in 2018 generating 1.68 million tons of PL annually. Poultry litter consists of chicken manure mixed with bedding material (1.5 kg litter/broiler; USDA-NASS, 2019; Mitchell and Tu, 2005). Although perceived as an industrial by-product, PL is a valuable source of mineral plant nutrients (Endale et al., 2008; Watts and Torbert, 2011; Mitchell and Tu, 2005; Tewolde et al., 2010). Much of the litter is used in row crop production systems and with the increasing realization of its value, further adoption of PL is expected to increase.

Applying PL to row crops has been reported to increase the content of mineral elements such as P and K in the soil (Adeli et al., 2010; Tewolde et al., 2011). These increased soil nutrient levels may further enrich plant tissues such as leaves and stem (He et al., 2013; Tewolde et al., 2005b, 2007, 2010b). Fertilizing cotton (*Gossypium hirsutum* L.) with PL has been found to increase seed P, K, Mg, and Cu concentrations compared to CF (He et al., 2013). Benefits of PL application have also been reported in horticultural crops. For example, in cabbage (*Brassica oleracea*), the application of organic manures including PL has been shown to increase mineral contents (P, Ca, Zn) of the edible part relative to CF (Citak and Sonmez, 2010). In tomato (*Lycopersicon esculentum* Mill.), PL elevated levels of Zn (critical element for human nutrition) and lowered bromine (Br; potentially harmful) concentration in the fruit highlighting its value in tomato production (Demir et al., 2010).

Corn grain is used both as animal feed and for human consumption. As such, the mineral composition of corn ear impacts its nutritional value and potential use. When used as livestock feed, inadequate grain mineral content adversely affects animal physiology and

reduce growth (Schutte, 1964; Underwood and Somers, 1969). According to Olson and Frey (1987), corn grain fails to meet the dietary mineral requirements of P, Ca, sodium (Na), Mn, Zn, and Cu in pigs and poultry and require mineral supplements from external sources. Sauberlich et al. (1953) found that increasing corn grain N content improved its nutritive quality. In places where corn is consumed as a food, enrichment of the corn grain with mineral elements such as Fe and Zn could improve human health and nutrition.

Numerous studies have documented the effect of PL on corn grain yield (Adeli et al., 2012; Tewolde et al., 2013; Endale et al., 2008; Watts and Torbert, 2011; Liebhardt, 1976; Nyakatawa and Reddy, 2002). However, there is limited research on the effect of PL application rate and timing on the ear mineral composition of corn (Tewolde et al., 2019). Therefore, the objective of this study was to investigate mineral concentrations of corn ear as affected by nutrient source (PL vs. CF) including their application rates and timings. The study was part of a larger project that compared corn grain yield and quantified environmental nitrogen loss using a partial nitrogen budget for PL and urea application systems.

Materials and Methods

Study sites

Field experiments were conducted in 2018 and 2019 at the Tennessee Valley Research & Extension Center (TVREC) near Belle Mina, AL (34° 41'N, 86° 53'S), E.V. Smith Research Center (EVS) near Shorter, AL (32° 25'N, 85° 53'S), and Wiregrass Research and Extension Center (WREC) near Headland, AL (31° 22' N, 85° 18'S) representing north, central, and south production regions of Alabama, respectively. The soil types were Decatur silty clay loam (clayey, kaolinitic, thermic, Rhodic Paleudults) with 1 to 2% slope at TVREC, Compass loamy sand (coarse-loamy, siliceous, sub active, thermic Plinthic Paleudults) with 1 to 3% slope at EVS, and Dothan fine sandy loam (fine-loamy, kaolinitic, thermic Typic Kandiudults) with 0 to 2% slope at WREC. Two of the three sites (EVS & WREC) were fallow the previous year while the TVREC site was cropped to soybean [*Glycine max* (L.) Merr.] with no manure application history at any of the sites. Prior to planting in the first year, plots at EVS and WREC were strip-tilled. However, in the subsequent year, plots were seeded with a no-tillage planter. Annual and 5-yr historic weather data (2013–2017) were collected from an automated weather station located at the field sites (Table 3.1). With a humid subtropical climate, total growing season rainfall (April to September) across the study sites ranged from 486 mm to more than 900 mm with a mean air temperature of 24.7°C. Four to five soil samples (80-mm probe diameter) were randomly taken from the 0- to 15-cm and 15- to 30-cm depths across each study site before initiating the experiment for routine soil test analysis (Table 3.2).

Treatments

The experiment consisted of 2×2×2 factorial treatments arranged in a randomized complete block design with four replications. Factors included N source, N rate, and application time. Poultry litter and urea were used as fertilizer N sources. Poultry litter and urea

were used as fertilizer N sources each applied at two target N rates, a low rate (168 kg total N ha⁻¹), and a high rate (336 kg total N ha⁻¹) in the single vs. split application. In a single application, the target N rate was applied within 7 d before planting in the spring, whereas in a split application, one-fourth of the target N rate was applied as pre-plant and the remaining three-fourth of the N was side dressed at the V6 stage (approximately 41 d after planting). In addition to these eight treatments, a control (without N fertilization) was also included to understand background nutrient contribution of each soil to corn ear nutrient concentrations.

Crop Management

Poultry litter was obtained one week before single application to the plots. The average nutrient concentration of the litter applied each year is presented in Table 3.3. The calculated amount of PL and urea was weighed for individual plots and surface-broadcasted by hand without incorporation. Litter was applied based on total N analysis without any assumptions regarding N availability from PL (Mitchell and Tu, 2006). Individual plots of size 6.1 × 3.7 m (four-rows wide) were planted to dryland corn (cv. Pioneer P1662YHR) at a row spacing of 0.76- (TVREC) or 0.91-m (EVS and WREC) in April each year (Table 3.4). All treatment plots received a blanket application of phosphorus (P₂O₅) and potassium (K₂O) each year at a rate of 45 kg/ha based on soil test recommendations. This was done to mimic a commercial production system where farmers apply P and K based on soil test recommendations and still apply PL as an additional nutrient source. It also helped reduce the disparity in soil P and K levels between urea and PL plots since PL supplied additional P and K. Weed control during each growing season was achieved following locally established management practices.

The 2018 cash crop was followed by winter rye (*Secale cereal* L., cv. Wrens Abruzzi) cover crop planted on 24 October at a rate of 67 (WREC) or 101 (EVS) kg ha⁻¹ in

0.20m row spacing. The cover crop was chemically terminated a week before replanting the same plots with corn in the subsequent year.

Sampling and Analyses

Three to four PL samples were drawn during litter collection each year for analysis. Two to three corn plants were also randomly collected per treatment plot at physiological maturity (R6 stage). Ear samples (including grain, husk and cob) were harvested from selected plants, dried at 60°C in a forced-air oven for at least 72 h till constant weight was attained, and ground with a Wiley Mill to pass a 2-mm screen. Concentrations of total P, K, Ca, Mg, S, B, Zn, Mn, Fe, and Cu in the litter and corn ear were determined by inductively coupled plasma-atomic emission spectrometry (ICP-AES; Spectro Ciros, Spectro Analytical Instruments Inc. Mahwah, NJ) following dry-ashing and acid-digestion laboratory procedure outlined by Donohue and Aho (1992). About 0.2g of the sample (dried and ground) was used for ashing in a muffle furnace at 500°C for 4 h and digested with 1.0 ml of 6M HCL and 40 ml of 0.0125 M H₂SO₄ + 0.05M HCL (double acid) individually each for 1 h. Total N content in the litter and corn ear was estimated by a dry combustion LECO C/N analyzer (LECO Corp., St. Joseph, MI).

Statistical analysis

Analysis of variance (ANOVA) was determined for treatment effects of N source, application rate, and time using PROC GLIMMIX as an augmented factorial design (Piepho et al., 2006) in SAS 9.4 (SAS Institute, 2013). The analysis compared the control to the treatments and also analyzed the factorial structure. Data for each location was analyzed separately with treatment effects, year, and their interactions as fixed effects. Blocks were treated as random effects. Locations were analyzed independently due to significant location × treatment interaction for the responses. Treatment means were separated using Fisher's protected LSD at

$\alpha = 0.05$ probability level (Littell et al., 2006). Pearson correlation coefficient was also calculated for the different mineral nutrients in the ear.

Results

At physiological maturity, the year \times treatment interactions were significant for most corn ear nutrient concentrations reflecting the influence of deficient rainfall in 2019 (Tables 3.5 and 3.7). Thus, the results for each nutrient are presented separately for each year by location.

Macronutrients

Nitrogen

(a) EVS: Corn ear N concentration, at EVS varied with N source, application rate, and time but with a significant three-way interaction of source \times rate \times time in 2018 (Table 3.5). With a single application, PL resulted in a 37% lower ear N concentration than urea when applied at a low rate, but the ear N concentration difference was not significant at the high rate (Table 3.6). With the split application, N source did not influence ear N concentration regardless of application rate. In 2019, ear N concentration was not affected by the fertilizer treatments.

(b) WREC: At WREC, application rate and time were significant on corn ear N concentration with no influence of N source ($P = 0.1974$) in 2018 (Table 3.7). Split application, irrespective of N source and application rate, increased ear N concentration by 12% compared to a single application (Table 3.8). In 2019, N source and application rate affected ear N concentration significantly. Corn fertilized with urea produced higher ear N concentration than corn fertilized with PL (Table 3.9). In both years, ear N concentration increased with the increasing rate of urea or PL.

(c) TVREC: At TVREC, N source and rate were significant on corn ear N concentration with no influence of application time ($P = 0.5971$) (Table 3.10). Compared to PL, urea produced 14% higher ear N concentration (Table 3.11). Like WREC, a high N rate, regardless

of application source and time, increased ear N concentration by 14% compared with a low N rate.

These results suggest that fertilizing corn with PL did not significantly increase N concentration in ear compared with urea when both were applied at the same target N rates. However, increasing the N rate did increase the ear N concentration at WREC and TVREC.

Phosphorus

(a) EVS: Although corn ear P concentration at EVS was affected by N source ($P = 0.0014$) and rate (0.0074) but not by application time ($P = 0.0789$), the interaction of source \times rate \times time had a significant impact on ear P concentration in 2018 (Table 3.5). The single application and low rate of PL produced 24% lower ear P concentration than plots that received urea (Table 3.6). However, at the high rate, ear P concentration was not significantly different between PL- and urea-treated plots (Table 3.6). Unlike ear N, with a split application, urea-treated plots had higher ear P concentration than PL-treated plots at both low and high N rates by 30% and 34%, respectively. In 2019, like ear N, ear P concentration was not affected by fertilizer treatments (Table 3.7).

(b) WREC: Corn ear P concentration, at WREC, was significantly affected only by application rate in both years (Table 3.7). Effect of N source and time on ear P concentration was not significant ($P > 0.05$). Corn fertilized with urea had higher ear P concentration than that of corn fertilized with PL by 13% and 10% in 2018 and 2019, respectively (Tables 3.8 and 3.9).

(c) TVREC: At TVREC, N source and rate were significant on corn ear P concentration with no influence of application time ($P = 0.7472$) (Table 3.10). Compared to PL, urea plots had 18% higher ear P concentration and high N rate, regardless of source and application time, increased ear P concentration compared to the low rate (Table 3.11).

Potassium

(a) EVS: Although corn ear K concentration at EVS varied with N source ($P = 0.0004$), application rate ($P < .0001$) and time ($P = 0.0001$), there were significant source \times rate, source \times time, and rate \times time interactions on ear K concentration in 2018 (Table 3.5). Averaged across application timings, urea had higher ear K concentration than PL at both N rates regardless of whether differences were statistically significant or not (Table 3.12). Ear K concentration was significantly greater (7.66 g ka^{-1}) only when urea was applied at a high rate than with PL (6.40 g kg^{-1}). Averaged across application rates, urea had significantly higher ear K concentration than PL at a single application (Table 3.13). However, both N sources produced a similar ear K concentration with the split application. Averaged across sources, high N rate produced greater ear K concentration compared to low rate, regardless of application time (Table 3.14). However, the difference was significant only when applied in a single application. In 2019, ear K concentration was not affected by fertilizer treatments (Table 3.5).

(b) WREC: At WREC, corn ear K concentration varied with N source ($P = 0.0436$) and time ($P = 0.0052$) with no effect of application rate ($P = 0.2053$); however, the source \times time interaction had a significant influence on ear K concentration in 2018 (Table 3.7). Unlike EVS, averaged across application rates, split application produced a significant difference in ear K concentration between sources (Table 3.15). Ear K concentration increased 15% with split-applied urea compared to PL. However, in 2019, ear K concentration was not affected by fertilizer treatments (Table 3.7).

(c) TVREC: At TVREC, only the main effect of N source was significant on corn ear K concentration (Table 3.10). Urea-treated plots produced 13% higher ear K concentration than PL-treated plots ($5.01 \text{ vs. } 4.43 \text{ g kg}^{-1}$) (Table 3.11).

Calcium

(a) EVS and WREC: Although the main effects of N source and rate were significant at EVS and non-significant at WREC, corn ear Ca concentration varied with a significant interaction of source \times rate at both locations in 2018 (Tables 3.5 and 3.7). Averaged across application timings, differences in ear Ca concentration among N sources were not significant at the low rate (Tables 3.12 and 3.16). However, urea-treated plots produced significantly greater ear Ca concentration than PL-treated and control plots at a high rate. In 2019, ear Ca content was not affected by fertilizer treatments at either location (Tables 3.5 and 3.7).

(b) TVREC: At TVREC, only the N source was significant on corn ear Ca concentration (Table 3.10). Urea produced 36% higher ear Ca concentration compared with PL (0.15 vs. 0.11 g kg⁻¹; Table 3.11).

Magnesium

(a) EVS: Like ear P, corn ear Mg concentration at EVS varied with N source ($P = <.0001$) and rate (0.0008) but not with application time ($P = 0.0870$) (Table 3.5). In addition, there was significant impact of source \times rate \times time interaction on ear Mg concentration in 2018. The single application of PL produced 30% lower ear Mg concentration at the low rate (Table 3.6). However, both sources produced similar ear Mg concentrations at the high rate. On the other hand, split application of urea produced higher ear Mg concentration, regardless of the application rate, but the difference was only significant at the high rate. In 2019, ear Mg concentration was not affected by fertilizer treatments (Table 3.5).

(b) WREC and TVREC: At WREC, corn ear Mg concentration was influenced by N source, rate, and application time in 2018 (Table 3.7). In 2019, at both WREC and TVREC, ear Mg concentration was influenced by N source and rate without significant impact of application time (Tables 3.7 and 3.10). In 2018, split application of urea or PL, irrespective of application rate, increased ear Mg concentration by 19% compared to the single application

(Table 3.8). At both locations, urea and high application rate produced significantly higher ear Mg concentration than the PL and low rate, respectively (Tables 3.8, 3.9 and 3.11).

Sulphur

(a) EVS: Although corn ear S concentration at EVS varied with application rate ($P = 0.0061$) but not with N source ($P = 0.6715$) and time ($P = 0.0747$), there was significant interaction of source \times rate \times time ($P = 0.0227$) on ear S concentration in 2018 (Table 3.5). However, unlike ear N, P, and Mg, PL produced 21% higher ear S concentration at the high rate for the single application but both N sources produced similar ear S concentrations at the low rate and also for the split application, regardless of application rate (Table 3.6). In 2019, ear S concentration was not affected by fertilizer treatments (Table 3.5).

(b) WREC: At WREC, corn ear S concentration in 2018 varied with application rate ($P = 0.0059$) and time ($P = 0.0012$) with no influence of N source (Table 3.7). Split application, irrespective of N source and application rate, increased ear S concentration 12% compared to the single application (Table 3.8). In 2019, N source along with application rate were significant for ear S concentration (Table 3.7). Among N sources, PL produced higher ear S concentration than urea although the difference was only 1% (Table 3.9). In both years, high rate treatments produced the greatest ear S concentration, which was different from the low rate and control treatments (Tables 3.8 and 3.9).

(c) TVREC: At TVREC, N source and application rate were significant for corn ear S concentration (Table 3.10). However, unlike WREC, compared to PL, urea increased ear S concentration by 16% (Table 3.11). Also, increasing applied N increased ear S concentration at both TVREC and WREC (Tables 3.8, 3.9, and 3.11).

Micronutrients

Boron

(a) EVS: At EVS, corn ear B concentration varied with application rate ($P = 0.0004$) and time ($P = 0.0011$) in 2018 (Table 3.5). However, there was no significant effect of N source ($P = 0.3789$) on ear B concentration. High rate, regardless of N source or application time, produced 19% greater ear B concentration than the low N rate (Table 3.17). On the other hand, single application increased ear B concentration by 17% compared to split application. In 2019, N source for ear B concentration was significant (Table 3.5). However, unlike 2018, ear B concentration was affected by N source and 51% greater for PL than urea in 2019 (data not shown).

(b) WREC and TVREC: At WREC, no treatment effect was observed for corn ear B concentration in both years (Table 3.7) while at TVREC, only the N source was significant for ear B concentration (Table 3.10). Corn fertilized with urea had 18% higher ear B concentration than corn fertilized with PL at TVREC (2.44 vs. 2.06 mg kg⁻¹; Table 3.11).

Zinc

(a) EVS: At EVS, corn ear Zn concentration varied with application rate ($P = 0.0002$) and time ($P = 0.0289$) in 2018 (Table 3.5). However, there was no significant effect of N source ($P = 0.0542$) on ear Zn concentration. The high application rate increased ear Zn concentration by 29% compared to the low rate and single application, irrespective of N source and application rate, caused a 14% increase in ear Zn concentration (Table 3.17). In 2019, however, ear Zn concentration was not affected by fertilizer treatments (Table 3.5).

(b) WREC and TVREC: At WREC, corn ear Zn concentration varied with only application time in 2018 (Table 3.7). Unlike EVS, the split application increased ear Zn concentration by 24% compared to a single application (Table 3.8). In 2019 and at TVREC, ear Zn concentration varied with N rate and source \times time interaction (Tables 3.7 and 3.10). Increasing the N application rate to 336 kg ha⁻¹ increased ear Zn concentration by about 14%

at both locations (Tables 3.9 and 3.11). Ear Zn concentration, averaged across application rates, was also significantly increased with a single application of urea compared to PL (Tables 3.18 and 3.19). However, both N sources reported similar corn ear Zn concentrations with the split application.

Manganese

(a) EVS: At EVS, corn ear Mn concentration varied with N source ($P = 0.0124$) and time ($P = 0.0102$) in 2018 (Table 3.5). However, there was no significant effect of application rate ($P = 0.0512$) on ear Zn concentration. The single application, regardless of N source and application rate, increased ear Mn concentration by 75% compared to the split application (Table 3.17). In 2019, like other mineral elements except for B, ear Mn concentration was not affected by fertilizer treatments (Table 3.5).

(b) WREC: At WREC, N source and rate were significant for corn ear Mn concentration with no influence of application time in both years (Table 3.7). The high N rate, regardless of source and application time, had greater ear Mn concentration than the low N rate in both years (Tables 3.8 and 3.9).

(c) TVREC: At TVREC, only the N source was significant for corn ear Mn concentration (Table 3.10). At all site-years except EVS in 2019, corn fertilized with urea had significantly higher ear Mg concentration compared to PL (Tables 3.8, 3.9, 3.11, and 3.17).

Iron

(a) EVS: At EVS, only the N source was significant for corn ear Fe concentration in 2018 whereas in 2019, ear Fe concentration was not affected by any of the fertilizer treatments (Table 3.5). Averaged across application rates and timings, urea resulted in a 36% higher ear Fe concentration compared with PL (Table 3.17).

(b) WREC and TVREC: In 2018, corn ear Fe concentration was not affected by fertilizer treatments (Tables 3.7). However, in 2019, N source was significant at both WREC and TVREC with urea resulting in significantly higher ear Fe concentration compared to PL (Tables 3.7 and 3.10).

Copper

(a) EVS: At EVS, corn ear Cu concentration varied with N source, application rate, and time in 2018 (Table 3.5). Compared to PL, urea produced 13% higher ear Cu concentration and increasing the application rate from 168 to 336 kg total N ha⁻¹ increased ear Cu concentration by 20% (Table 3.17). Also, single application, regardless of N source and rate, increased ear Cu concentration by 18% compared to split application. However, in 2019, ear Cu concentration was not affected by fertilizer treatments (Table 3.5).

(b) WREC: Although corn ear Cu concentration at WREC did not varied with N source ($P = 0.1548$), application rate ($P = 0.2560$), and time ($P = 0.6887$); however, source \times time and rate \times time had a significant impact on ear Cu concentration in 2018 (Table 3.7). Averaged across application rates, ear Cu concentration increased by 29% when fertilized with split-applied urea compared with PL (Table 3.15). However, both N sources reported similar ear Cu concentrations with the single application. Averaged across N sources, the high rate resulted in greater ear Cu concentration than a low rate only when split applied (Table 3.20). Like EVS, ear Cu concentration was not affected by fertilizer treatments in 2019.

(c) TVREC: At TVREC, corn ear Cu concentration varied with N source, rate, and source \times time interaction (Table 3.10). Unlike other mineral elements, the high N rate lowered ear Cu concentration 14% compared to the low rate (2.25 vs. 2.62 mg kg⁻¹; Table 3.11). Averaged across application rates, unlike WREC, single application of urea increased ear Cu

concentration by 35% compared to PL (Table 3.19). However, with split application, both N sources resulted in similar ear Cu concentrations.

Discussion

Poultry litter is regarded as a valuable source of plant mineral elements (Poffenbarger et al., 2015; Tewolde et al., 2005a, 2005b). Use of PL as a fertilizer in crop production even for short intervals of 2 to 3 years have increased the content of many of the elements in the soil (Adeli et al., 2008; Tewolde et al., 2018; Wood et al., 1996). Some studies have also reported the enrichment of corn leaves and stem with mineral elements such as N, P, Cu, and Zn applied from PL (Tewolde et al., 2019). We speculated that elevated levels of nutrients in the soil and corn vegetative parts from litter application would also enrich the corn ear with some of the mineral elements. However, we found no effect of PL irrespective of application rate and timing on ear nutrient concentrations relative to chemical fertilizer urea. Although concentrations of most mineral elements in corn ear were affected by treatments at all locations, the response was not specific to PL.

Poultry litter did not elevate ear N concentration compared to urea regardless of the site-year probably due to inadequate plant available-N (PAN) derived as reflected by the lower N concentration in leaves and stems of corn plants in the PL treatments (data not shown). This was also consistent with the grain yield results (Chapter 2). Corn fertilized with urea had higher grain yield at both WREC and TVREC. However, PL resulted in greater grain yield at EVS in both years, but the response was attributed to other soil-related benefits and not to the amount of PAN received from PL. Ear N content seems to be the key determinant for levels of other mineral elements in the ear. Urea treatments that caused greater ear N concentration also reported higher ear levels of P, K, Mg, S, Zn, Mn, Fe, and Cu. Correlation analysis revealed that ear P, K, Mg, S, Zn and Mn concentrations were highly and positively correlated with ear N concentration at TVREC with correlation coefficients (r) of 0.85, 0.62, 0.87, 0.92, 0.84 and 0.64 ($n=36$), respectively, but to a lesser extent and consistency, at other locations (Table 3.21). For instance, at EVS in 2018, ear N concentration was highly and positively correlated with

ear P and Mg concentrations ($r=0.64$, $n=36$). Ear N concentration was reflective of N nutrition of the corn plant, as evidenced by high positive correlation ($r =0.56$) between ear N and leaf + stem N concentrations (data combined for all locations ($n = 180$) (data not shown). This study also revealed that increasing the N application rate elevated ear concentrations of N and other linked mineral elements (as discussed above). A close association between ear N and P could be due to the presence of protein bodies called globoids in the seed (Epstein and Bloom, 2005). Globoids contains seed N in protein forms and seed P in phytate forms (Lott et al., 2000; Raboy, 2009). The high linear correlation of ear N with elements K, Mg, Zn, and Mn could be attributed to the complex formation of these elements with seed P due to the presence of six-negatively charged phosphate groups per molecule of phytic acid (Lott et al., 2000). Results indicate N is readily taken up by plants and translocated to the ears based on soil N levels, unlike P, K, and other elements (Ahmadi et al., 1993; Biswas and Ma, 2016).

Application timing affected ear N concentration at physiological maturity at EVS and WREC but only in the first year (2018). At EVS, applying the target N rate in a single application resulted in higher ear N concentration along with leaf and stem N concentrations (data not shown) compared with the split application. However, at WREC, the split application had a higher ear N concentration than the single application. Split-N applications are recommended for coarse-textured soils such as the soils at WREC with low nutrient holding capacity and high leaching potential (Murrel, 2006). Despite receiving additional amounts of P, K, and other mineral elements from PL, PL treatments failed to enrich the ear with any of these elements. No treatment or residual effect of PL on ear nutrient levels in 2019 was observed at EVS, likely because the corn plants suffered water and heat stress during the 2019 growing season. Average grain yield was reduced by 30% due to lower rainfall received in 2019 than 2018 (Chapter 2). Additionally, high N losses such as ammonia volatilization, N leaching, and denitrification exhausted N from the soil; thus, no residual effect of PL on ear N

or other mineral elements was observed in 2019 (Chapter 2). All correlations among mineral elements were positive at TVREC, which suggests that enrichment of corn ear with one element does not decrease the storage of any of the other elements (data not shown). It also appears that ear Ca and Cu concentrations do not depend on N availability, as indicated by their poor correlation with ear N across all locations ($r < 0.50$) (Table 3.21).

The study, in general, indicates that corn fertilization with a N source that readily supplies PAN can enhance ear N concentration along with other mineral elements. These results agree with Tewolde et al. (2019) who conducted a 2-yr study in Mississippi on a Leeper silty clay loam similar to Decatur silty clay loam at TVREC (Tables S2, S3, S4, and S5). He reported no enrichment of corn grain with mineral elements from PL compared to conventional fertilizer and that grain N nutrition influenced levels of other elements in the kernel. However, in our study, we also found that the treatment effect on ear nutrient composition varied by locations. This could be because ear nutrient composition can be affected by several factors including genetic and environmental variability (Kleese et al., 1968), soil nutrient status, and fertilizer management practices (Yin and Vyn, 2003; Ross et al., 2006; Farmaha et al., 2011). Heckman et al. (2003) reported variability in grain nutrient concentrations with a single corn hybrid grown across six different site-years.

The findings from this study have implications on human nutrition especially from corn being grown using organic fertilizers such as PL. Adequate grain N enhances grain nutrition of other mineral elements including P and K. Intake of human food rich in P and K can help alleviate malnutrition known to affect pregnant women and children less than 3 years old in many developing countries (Black et al., 2013).

Conclusion

Poultry litter contains all essential nutrients required for plant nutrition. In this study, we speculated that addition of PL would elevate corn ear concentrations of some of the mineral elements it supplies. However, the results showed that fertilization of corn even with high rates of PL did not enrich ear with the measured nutrient elements. Ear N accumulation was dependent on N availability in the soil and plant N nutrition. Accumulation of P, K, Mg, S, Zn, Mn, and Fe in the ear was not in proportion to their soil levels but relied on ear N concentration. Application of PL did not increase ear N concentration and consequently concentrations of other elements due to lower PL-derived PAN. This research demonstrated that ensuring optimal levels of PAN in the soil, regardless of whether derived from PL or urea, also ensures maximum accumulation of ear N, P, K, and other elements.

Table 3.1. Monthly average air temperature and total precipitation during the corn growing season (April to September) at the experimental sites: E.V. Smith Research Center (EVS), Wiregrass Research and Extension Center (WREC), and Tennessee Valley Research and Extension Center (TVREC).

	EVS			WREC			TVREC	
	2018	2019	5-yr avg.	2018	2019	5-yr avg.	2019	5-yr avg.
Air temperature, °C								
April	16.1	17.9	17.8	16.5	19.2	19.9	16.5	17.5
May	23.8	24.0	20.9	23.7	25.5	23.3	22.8	21.2
June	26.7	26.4	25.3	26.3	27.7	26.5	24.9	25.9
July	27.4	27.7	26.4	26.8	27.9	27.6	26.6	26.8
August	26.7	27.8	26.2	26.3	27.8	27.3	26.1	26.2
September	27.1	27.3	24.1	26.9	27.9	25.2	26.2	23.9
Precipitation, mm								
April	90	174	149	127	122	147	159	114
May	119	144	120	196	42	94	108	103
June	127	109	127	110	85	102	113	103
July	80	51	115	225	113	95	120	138
August	148	41	81	180	142	146	66	66
September	140	3	31	80	11	90	20	50
April-September	704	522	623	918	515	674	586	574
January-December	1521	1148	1229	1886	1033	1235	1637	1215

Table 3.2. Baseline soil properties for 0- to 15- and 15- to 30-cm before treatment applications at each experimental site.

Property	EVS [†]		WREC		TVREC	
	0-15 cm	15-30 cm	0-15 cm	15-30 cm	0-15 cm	15-30 cm
pH ^{††}	6.2 (0.4 ^{†††})	6.0 (0.2)	6.4 (0.2)	6.3 (0.2)	6.2 (0.1)	6.2 (0.2)
Total C (g kg ⁻¹)	14.4 (0.8)	13.8 (1.7)	13.6 (1.2)	13.1 (1.4)	16.6 (1.1)	13.8 (0.4)
Total N (g kg ⁻¹)	3.5 (0.5)	3.7 (0.7)	0.4 (0.1)	0.35 (0.1)	4.3 (0.3)	4.3 (0.5)
Inorganic N (NH ₄ -N + NO ₃ -N; mg kg ⁻¹)	45.3 (12.0)	45.1 (8.9)	19.0 (8.3)	17.2 (7.1)	20.6 (3.5)	14.0 (3.5)
Mehlich-1 P (mg kg ⁻¹)	14.1 (10)	14.6 (8) M	26.2 (17.3)	23.0 (17.8)	24.8 (5.5)	12.6 (1.7) M
	M*		H	M	H	
Mehlich-1 K (mg kg ⁻¹)	42.3 (10)	57.0 (30) M	80.2 (38.8)	63.2 (21.9)	120.4	75.7 (8.9) M
	M		H	H	(19.1) H	
Mehlich-1 Ca (mg kg ⁻¹)	603 (150)	509 (56) H	434 (87) H	436 (92) H	979 (56.2)	942 (100.9)
	H				H	H
Mehlich-1 Mg (mg kg ⁻¹)	42.2 (9) H	44.7 (12) H	107.8	98.5 (21.1)	75.5 (3.2)	64.5 (5.9) H
			(27.6) H	H	H	

[†]E.V. Smith Research Center (EVS; n = 4); Wiregrass Research and Extension Center (WREC; n = 4); Tennessee Valley Research and Extension Center (TVREC; n = 5).

^{††}Measured using a glass electrode in a 1:1 soil/water ratio.

^{†††}Standard deviation of the mean given in parenthesis.

*Rating codes were derived using the fertility recommendatins for Alabama soils (Mitchell and Huluka, 2012): L (low), M (medium), and H (high).

Table 3.3. Chemical properties of poultry litter (PL) applied to corn each year at the experimental sites.

Year	Moisture	Total C	Total N	P	K	Ca	Mg	B	Zn	Mn	Fe	Cu
	g kg ⁻¹	-----g kg ⁻¹ -----						-----mg kg ⁻¹ -----				
2018	262 (9 [†])	351.1 (10.0)	26.7 (1.0)	8.8 (1.5)	16.4 (1.7)	11.8 (0.8)	6.0 (0.3)	26 (2)	174.7 (10.1)	227 (10)	1498 (306)	59.3 (6.8)
2019	267 (157)	300.0 (53.0)	31.8 (6.0)	20.2 (12.0)	28.1 (8.7)	17.6 (6.4)	4.1 (1.3)	25 (7)	250.0 (71.0)	400 (141)	600 (0)	300.0 (141.0)

[†]Standard deviation of the mean given in parenthesis (n = 3).

Table 3.4. Cultural practices adopted in the study at each experimental site.

Location	Pre-plant N application	Planting	Row spacing (m)	Seed rate ha ⁻¹	Side-dress N application	Harvesting
2018						
EVS [†]	29 March	5 April	0.91	75,335	29 June	10 September
WREC	6 April	13 April	0.91	71,729	28 June	7 September
2019						
EVS	29 March	5 April	0.91	75,335	16 May	21 August
WREC	1 April	5 April	0.91	71,729	14 May	30 August
TVREC	28 March	5 April	0.76	70,395	17 May	9 September

[†]E.V. Smith Research Center (EVS); Wiregrass Research and Extension Center (WREC); Tennessee Valley Research and Extension Center (TVREC)

Table 3.5. Test of significance for effect of nitrogen (N) source, application rate, time, and their interactions on corn ear nutrient concentration at the physiological maturity, at E.V. Smith Research Center (EVS).

Effect	N	P	K	Ca	Mg	S	B	Zn	Mn	Fe	Cu
-----P>F-----											
2018											
Source (S)	0.0196	0.0014	0.0004	0.0008	<.0001	0.6715	0.3789	0.0542	0.0124	0.0118	0.0253
Rate (R)	0.0033	0.0074	<.0001	0.0390	0.0008	0.0061	0.0004	0.0002	0.0512	0.3896	0.0022
S × R	0.4950	0.0789	0.0168	0.0390	0.6106	0.1278	0.5741	0.8008	0.0920	0.8674	0.1217
Time (T)	0.0101	0.0789	0.0001	0.4084	0.0870	0.0747	0.0011	0.0289	0.0102	0.0715	0.0049
S × T	0.3166	0.0421	0.0169	0.1432	0.2749	0.1270	0.1172	0.5434	0.0932	0.1903	0.9533
R × T	0.8668	0.7279	0.0042	0.2505	0.8798	0.8883	0.7280	0.7836	0.0888	0.9468	0.4588
S × R × T	0.0067	0.0153	0.2456	0.6182	0.0129	0.0227	0.2567	0.6904	0.1688	0.5713	0.9665
2019											
Source (S)	0.1431	0.9346	0.5169	0.2039	0.6752	0.8811	0.0018	0.5666	0.1883	0.6384	0.8250
Rate (R)	0.4948	0.7186	0.8490	0.8291	0.9661	0.4235	0.4200	0.9842	0.5697	0.6215	0.6394
S × R	0.5578	0.1270	0.3787	0.7200	0.1847	0.7763	0.2940	0.2119	0.3565	0.1775	0.0766
Time (T)	0.2300	0.1630	0.3040	0.6662	0.1388	0.1359	0.1022	0.0978	0.2747	0.0727	0.0766
S × T	0.8615	0.9506	0.3904	0.4770	0.5951	0.6681	0.1625	0.6494	0.6927	0.6710	0.0309
R × T	0.6828	0.9525	0.8316	0.1795	0.7990	0.5757	0.6935	0.9677	0.8064	0.5662	0.1183
S × R × T	0.2138	0.5684	0.2537	0.2890	0.7445	0.2211	0.3213	0.3276	0.2370	0.9235	0.2603
2018–2019											
Treatment (T)	0.0018	0.0088	<.0001	0.0018	0.0021	0.1588	0.0013	0.0227	0.0008	0.0562	0.0026
Year (Y)	<.0001	0.0534	<.0001	<.0001	<.0001	0.3317	<.0001	<.0001	<.0001	<.0001	<.0001
T × Y	0.0549	0.0223	0.0004	0.0096	0.0147	0.2704	0.1805	0.0418	0.0046	0.1442	0.0025

Table 3.6. Effect of Source (S) × Rate (R) × Time (T) interaction on corn ear N, P, Mg, and S concentrations at the physiological maturity in 2018, at E.V. Smith Research center (EVS).

Rate	N (g kg ⁻¹)		P (g kg ⁻¹)		Mg (g kg ⁻¹)		S (g kg ⁻¹)	
	Low rate	High rate	Low rate	High rate	Low rate	High rate	Low rate	High rate
Single application								
Source								
U	14.37a [†]	13.96a	3.90a	3.60ab	1.67a	1.70a	1.10a	1.05b
PL	9.04b	14.08a	2.95b	4.07a	1.17b	1.65a	0.97a	1.27a
Control	11.09b	11.09b	3.02b	3.02b	1.25b	1.25b	0.97a	0.97b
Split application								
Source								
U	9.21a	13.4a	3.47a	4.12a	1.42a	1.85a	0.97a	1.15a
PL	9.81a	10.67a	2.67b	3.07b	1.15a	1.27b	0.92a	1.02ab
Control	11.09a	11.09a	3.07ab	3.07b	1.25a	1.25b	0.97a	0.97b

[†]Means followed by the same letter within a column for the same effect are not significantly different at $P \leq 0.05$.

Table 3.7. Test of significance for effect of nitrogen (N) source, application rate, time, and their interactions on corn ear nutrient concentration at the physiological maturity, at Wiregrass Research and Extension Center (WREC).

Effect	N	P	K	Ca	Mg	S	B	Zn	Mn	Fe	Cu
-----P>F-----											
2018											
Source (S)	0.1974	0.3367	0.0436	0.1329	0.0272	0.2763	0.6395	0.1822	0.0036	0.2496	0.1548
Rate (R)	0.0032	0.0115	0.2053	0.2548	0.0091	0.0059	0.0978	0.1579	0.0175	0.1743	0.2560
S × R	0.6577	0.3705	0.8149	0.0283	0.6886	0.6374	0.3204	0.1546	0.2632	0.9077	0.0587
Time (T)	0.0013	0.1107	0.0052	0.2548	0.0006	0.0012	0.9137	0.0013	0.1568	0.7767	0.6887
S × T	0.3874	0.2231	0.0436	0.4443	0.1805	0.4340	0.0604	0.8216	0.2658	0.8960	0.0011
R × T	0.7545	0.5268	0.5595	0.2548	0.4724	0.8749	0.6811	0.7963	0.5432	0.3868	0.0082
S × R × T	0.4941	0.2484	0.3532	1.0000	0.1805	0.2763	0.4217	0.6830	0.3745	0.3247	0.0738
2019											
Source (S)	<.0001	0.2324	0.1456	0.1467	0.0178	0.0319	0.5461	0.0188	0.0004	0.0070	0.7766
Rate (R)	0.0207	0.0386	0.0985	1.0000	0.0444	0.0319	0.5461	0.0042	0.0259	0.4744	0.0560
S × R	0.5335	0.9310	0.4807	1.0000	1.0000	0.6527	0.5461	0.1608	0.2717	0.3783	0.7766
Time (T)	0.5783	0.9310	0.8317	1.0000	0.6752	0.0808	0.0786	0.5610	0.7110	0.9663	0.3979
S × T	0.9596	0.1093	0.2933	1.0000	0.4045	0.3711	0.5461	0.0090	0.7110	0.2959	0.1643
R × T	0.8791	0.8625	0.7769	0.1467	0.6752	1.0000	0.0786	0.9577	0.7110	0.9663	0.3979
S × R × T	0.3401	0.1742	0.2630	0.1467	0.1026	0.3711	0.5461	0.1096	0.2717	0.7038	0.7766
2018–2019											
Treatment (T)	<.0001	0.0069	0.0031	0.1847	<.0001	0.0001	0.2487	0.0082	0.0002	0.6373	0.2638
Year (Y)	0.0625	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	0.0021
T × Y	0.0103	0.1173	0.0985	0.2076	0.0056	0.2983	0.2138	0.0024	0.2420	0.7715	0.0331

Table 3.8. Corn ear nutrient concentration measured across nitrogen (N) source, application rate, and time at the physiological maturity in 2018, at Wiregrass Research and Extension Center (WREC).

Effect	N	P	K	Ca	Mg	S	B	Zn	Mn	Fe	Cu
-----g kg ⁻¹ -----						-----mg kg ⁻¹ -----					
Source											
U	13.50a [†] (1.73 ^{††})	4.18a (0.61)	7.02a (0.68)	0.32a (0.12)	1.78a (0.32)	1.19a (0.16)	3.48a (0.47)	37.23b (8.65)	20.57a (4.66)	87.51a (64.90)	2.52a (0.53)
PL	12.94a (1.34)	4.00a (0.60)	6.57b (0.73)	0.27a (0.06)	1.60b (0.25)	1.23a (0.11)	3.56a (0.52)	40.34ab (7.45)	16.74b (2.41)	66.59a (28.71)	2.35a (0.33)
Control	12.90a (1.2)	3.95a (0.46)	6.02b (0.51)	0.30a (0.14)	1.72ab (0.21)	1.17a (0.13)	3.96a (0.59)	45.20a (13.06)	19.04ab (3.13)	87.07a (10.70)	2.35a (0.46)
Rate											
168	12.53b (1.56)	3.84b (0.61)	6.66ab (0.77)	0.27a (0.07)	1.58b (0.32)	1.15b (0.14)	3.38b (0.50)	37.14b (8.18)	17.15b (2.59)	64.63a (23.94)	2.37a (0.35)
336	13.92a (1.22)	4.34a (0.49)	6.94a (0.69)	0.31a (0.12)	1.80a (0.24)	1.27a (0.11)	3.67ab (0.44)	40.44ab (7.93)	20.17a (4.88)	89.47a (66.08)	2.50a (0.52)
Control	12.90ab (1.2)	3.95ab (0.46)	6.02b (0.51)	0.30a (0.14)	1.72ab (0.21)	1.17ab (0.13)	3.96a (0.59)	45.20a (13.06)	19.04ab (3.13)	87.07a (10.70)	2.35a (0.46)
Time											
Single	12.45b (1.61)	3.94a (0.64)	7.12a (0.61)	0.27a (0.08)	1.54b (0.28)	1.14b (0.14)	3.53a (0.50)	34.67b (6.22)	17.79a (3.91)	79.59a (67.04)	2.41a (0.30)
Split	13.99a (1.05)	4.24a (0.54)	6.47b (0.71)	0.31a (0.11)	1.84a (0.24)	1.28a (0.10)	3.51a (0.49)	42.90a (7.79)	19.52a (4.29)	74.51a (27.59)	2.46a (0.56)
Control	12.90ab (1.2)	3.95a (0.46)	6.02b (0.51)	0.30a (0.14)	1.72ab (0.21)	1.17ab (0.13)	3.96a (0.59)	45.20a (13.06)	19.04a (3.13)	87.07a (10.70)	2.35a (0.46)

[†]Means followed by the same letter within a column for the same effect are not significantly different at $P \leq 0.05$.

^{††}Standard deviation of the mean given in parenthesis.

Table 3.9. Corn ear nutrient concentration measured across nitrogen (N) source, application rate, and time at the physiological maturity in 2019, at Wiregrass Research and Extension Center (WREC).

Effect	N	P	K	Ca	Mg	S	B	Zn	Mn	Fe	Cu
	-----g kg ⁻¹ -----						-----mg kg ⁻¹ -----				
Source											
U	14.91a [†] (1.10 ^{††})	3.42a (0.48)	5.19a (0.56)	0.11a (0.03)	1.26a (0.20)	0.90b (0.08)	1.94a (0.25)	29.69a (5.15)	8.75a (1.95)	25.87a (4.41)	2.87a (0.72)
PL	13.10b (1.20)	3.25a (0.46)	4.92a (0.41)	0.10a (0.01)	1.11b (0.80)	0.96a (0.80)	2.00a (1.00)	26.75b (18.00)	6.69b (5.00)	21.56b (16.00)	2.81a (2.00)
Control	11.17c (0.30)	2.62b (0.33)	4.75a (0.58)	0.10a (0.01)	0.85c (0.10)	0.82b (0.05)	2.00a (0.01)	20.25c (2.87)	5.75b (0.50)	21.50b (4.51)	2.50a (0.58)
Rate											
168	13.55b (1.15)	3.18b (0.46)	4.91a (0.38)	0.11a (0.02)	1.12b (0.19)	0.90b (0.09)	1.94a (0.44)	26.37b (4.14)	7.15b (1.26)	23.19a (5.26)	2.62a (0.62)
336	14.46a (1.56)	3.49a (0.45)	5.21a (0.57)	0.11a (0.02)	1.25a (0.21)	0.96a (0.08)	2.00a (0.01)	30.06a (5.04)	8.31a (2.15)	24.25a (3.38)	3.06a (0.57)
Control	11.17c (0.30)	2.62c (0.33)	4.75a (0.58)	0.10a (0.01)	0.85c (0.10)	0.82b (0.05)	2.00a (0.01)	20.25c (2.87)	5.75b (0.50)	21.50a (4.51)	2.50a (0.58)
Time											
Single	13.90a (1.41)	3.33a (0.48)	5.04a (0.48)	0.11a (0.02)	1.17a (0.20)	0.91ab (0.08)	1.87a (0.34)	27.87a (5.03)	7.62ab (1.63)	23.69a (5.33)	2.94a (0.77)
Split	14.11a (1.48)	3.34a (0.49)	5.07a (0.54)	0.11a (0.02)	1.20a (0.22)	0.96a (0.10)	2.06a (0.25)	28.56a (4.92)	7.81a (2.07)	23.75a (3.33)	2.75a (0.45)
Control	11.17b (0.30)	2.62b (0.33)	4.75a (0.58)	0.10a (0.01)	0.85b (0.10)	0.82b (0.05)	2.00a (0.01)	20.25b (2.87)	5.75b (0.50)	21.50a (4.51)	2.50a (0.58)

[†]Means followed by the same letter within a column for the same effect are not significantly different at $P \leq 0.05$.

^{††}Standard deviation of the mean given in parenthesis.

Table 3.10. Test of significance for nitrogen (N) source, application rate, time, and their interactions on corn ear nutrient concentration at physiological maturity, at Tennessee Valley Research and Extension Center (TVREC).

Effect	N	P	K	Ca	Mg	S	B	Zn	Mn	Fe	Cu
-----P>F-----											
Source (S)	0.0001	0.0049	0.0063	0.0478	0.0014	0.0007	0.0365	0.0003	0.0007	0.0045	0.0481
Rate (R)	0.0002	0.0033	0.2055	0.7683	0.0040	0.0019	0.1527	0.0043	0.7991	0.2965	0.0481
S × R	0.1139	0.3788	0.9487	0.7683	0.3330	0.2567	0.4673	0.2485	0.1356	0.1552	0.4941
Time (T)	0.5971	0.7472	0.6529	0.3802	0.9135	0.4461	1.0000	0.3668	0.6114	0.3944	0.4941
S × T	0.2274	0.1786	0.6074	0.1492	0.1126	0.4461	0.4673	0.0462	0.7991	0.9474	0.0481
R × T	0.2802	0.2999	0.1315	0.1492	0.2389	0.2567	0.1527	0.6040	0.2104	0.8950	0.4941
S × R × T	0.9472	0.3378	0.2283	0.3802	0.2389	0.7019	0.4673	0.3668	0.3135	0.4313	0.4941
Treatment	0.0003	0.0077	0.0589	0.2468	0.0035	0.0034	0.1166	0.0018	0.0240	0.0807	0.0087

Table 3.11. Corn ear nutrient concentrations measured across nitrogen (N) source, and application rate at physiological maturity, at Tennessee Valley Research and Extension Center (TVREC).

Effect	N	P	K	Ca	Mg	S	B	Zn	Mn	Fe	Cu
	-----g kg ⁻¹ -----						-----mg kg ⁻¹ -----				
Source											
U	13.63a [†] (1.58 ^{††})	3.17a (0.55)	5.01a (0.69)	0.15a (0.08)	1.09a (0.20)	0.95a (0.11)	2.44a (0.51)	22.94a (3.53)	8.94a (1.61)	27.12a (5.08)	2.62a (0.50)
PL	11.93b (1.15)	2.69b (0.45)	4.43b (0.42)	0.11b (0.02)	0.89b (0.17)	0.82b (0.10)	2.06b (0.44)	18.94b (2.79)	7.06b (1.18)	21.25b (5.42)	2.25b (0.58)
Control	12.30b (0.57)	2.70ab (0.35)	4.22b (0.22)	0.12ab (0.05)	0.87b (0.13)	0.85ab (0.06)	1.75b (0.50)	21.25ab (2.63)	7.50ab (1.29)	21.00b (4.55)	1.50c (0.58)
Rate											
168	11.97b (1.18)	2.68b (0.40)	4.59a (0.63)	0.12a (0.08)	0.90b (0.16)	0.83b (0.09)	2.12ab (0.50)	19.44b (2.87)	7.94a (1.81)	23.19a (5.44)	2.62a (0.58)
336	13.59a (1.59)	3.18a (0.58)	4.84a (0.62)	0.13a (0.05)	1.08a (0.23)	0.94a (0.13)	2.37a (0.50)	22.44a (3.97)	8.06a (1.61)	25.19a (6.47)	2.25b (0.50)
Control	12.30b (0.57)	2.70ab (0.35)	4.22a (0.22)	0.12a (0.05)	0.87b (0.13)	0.85ab (0.06)	1.75b (0.50)	21.25ab (2.63)	7.50a (1.29)	21.00a (4.55)	1.50c (0.58)
Time											
Single	12.68a (1.80)	2.91a (0.61)	4.76a (0.54)	0.12a (0.04)	0.99a (0.26)	0.90a (0.14)	0.25a (0.58)	20.50a (3.95)	7.87a (1.54)	23.37a (5.84)	2.50a (0.52)
Split	12.88a (1.44)	2.96a (0.51)	4.67a (0.73)	0.14a (0.08)	0.99a (0.16)	0.87a (0.11)	0.25a (0.45)	21.37a (3.57)	8.12a (1.86)	25.00a (6.17)	2.37a (0.62)
Control	12.30a (0.57)	2.70a (0.35)	4.22a (0.22)	0.12a (0.05)	0.87a (0.13)	0.85a (0.06)	1.75a (0.50)	21.25a (2.63)	7.50a (1.29)	21.00a (4.55)	1.50b (0.58)

[†]Means followed by the same letter within a column for the same effect are not significantly different at $P \leq 0.05$.

^{††}Standard deviation of the mean given in parenthesis.

Table 3.12. Interaction between nitrogen (N) source and application rate on corn ear potassium (K) and calcium (Ca) concentrations at physiological maturity in 2018, at the E.V. Smith Research Center (EVS).

Source	Low rate	High rate
Ear K concentration, g kg ⁻¹		
U	6.06bc [†]	7.66a
PL	5.77c	6.40b
Control	5.70c	5.70c
Ear Ca concentration, g kg ⁻¹		
U	0.29b	0.45a
PL	0.22b	0.22b
Control	0.22b	5.70c

[†]Means followed by the same letter within a column and row are not significantly different at $P \leq 0.05$.

Table 3.13. Interaction between nitrogen (N) source and application time on corn ear potassium (K) concentration at physiological maturity in 2018, at the E.V. Smith Research Center (EVS).

Source	Single	Split
Ear K concentration, g kg ⁻¹		
U	7.54a [†]	6.19b
PL	6.27b	5.90b
Control	5.70b	5.70b

[†]Means followed by the same letter within a column and row are not significantly different at $P \leq 0.05$.

Table 3.14. Interaction between application rate and time on corn ear potassium (K) concentration at physiological maturity in 2018, at the E.V. Smith Research Center (EVS).

Rate	Single	Split
Ear K concentration, g kg ⁻¹		
168	6.05b [†]	5.79b
336	7.76a	6.30b
Control	5.70b	5.70b

[†]Means followed by the same letter within a column and row are not significantly different at $P \leq 0.05$.

Table 3.15. Interaction between nitrogen (N) source and application time on corn ear potassium (K) and copper (Cu) concentration at physiological maturity in 2018, at the Wiregrass Research and Extension Center (WREC).

Source	Single	Split
Ear K concentration, g kg ⁻¹		
U	7.12a [†]	6.92a
PL	7.12a	6.02b
Control	6.02b	6.02b
Ear Cu concentration, g kg ⁻¹		
U	2.28bc	2.77a
PL	2.54ab	2.15c
Control	2.35abc	2.35abc

[†]Means followed by the same letter within a column and row are not significantly different at $P \leq 0.05$.

Table 3.16. Interaction between nitrogen (N) source and application rate on corn ear calcium (Ca) concentration at physiological maturity in 2018, at the Wiregrass Research and Extension Center (WREC).

Rate	Low rate	High rate
Ear Ca concentration, g kg ⁻¹		
U	0.26b [†]	0.37a
PL	0.29ab	0.25b
Control	0.30ab	0.30ab

[†]Means followed by the same letter within a column and row are not significantly different at $P \leq 0.05$.

Table 3.17. Corn ear nutrient concentrations measured across nitrogen (N) source, application rate, and time at physiological maturity in 2018, at the E.V. Smith Research Center (EVS).

Effect	N	P	K	Ca	Mg	S	B	Zn	Mn	Fe	Cu
	-----g kg ⁻¹ -----						-----mg kg ⁻¹ -----				
Source											
U	12.76a [†] (2.93 ^{††})	3.77a (0.56)	6.86a (1.29)	0.37a (0.14)	1.66a (0.25)	1.07a (0.13)	2.68a (0.46)	29.09a (5.84)	20.11a (16.02)	79.71a (26.60)	3.03a (0.62)
PL	10.90b (2.68)	3.19b (0.62)	6.09b (0.66)	0.22b (0.06)	1.31b (0.24)	1.05a (0.17)	2.78a (0.49)	25.95a (5.62)	11.67b (2.64)	58.42b (17.74)	2.68b (0.49)
Control	11.09ab (0.32)	3.02b (0.35)	5.70b (0.58)	0.22b (0.19)	1.25b (0.31)	0.97a (0.17)	2.24b (0.52)	24.53a (5.09)	11.88ab (4.93)	52.80b (13.89)	2.02c (0.23)
Rate											
168	10.61b (3.62)	3.25b (0.64)	5.92b (0.61)	0.26b (0.06)	1.35b (0.25)	0.99b (0.11)	2.49b (0.32)	24.05b (3.07)	12.69a (3.32)	65.65a (20.90)	2.60b (0.37)
336	13.05a (1.60)	3.72a (0.57)	7.03a (1.16)	0.34a (0.16)	1.62a (0.28)	1.12a (0.15)	2.97a (0.49)	30.99a (5.75)	19.08a (15.90)	72.48a (27.53)	3.11a (0.63)
Control	11.09ab (0.32)	3.02b (0.35)	5.70b (0.58)	0.22b (0.19)	1.25b (0.31)	0.97b (0.17)	0.24b (0.52)	24.05b (5.09)	11.88a (4.93)	52.80a (13.89)	2.02c (0.23)
Time											
Single	12.86a (3.13)	3.63a (0.62)	6.91a (1.26)	0.31a (0.16)	1.55a (0.28)	1.10a (0.15)	2.94a (0.42)	29.32a (5.82)	20.24a (15.90)	76.41a (28.51)	3.09a (0.55)
Split	10.79b (2.48)	3.34ab (0.67)	6.04b (0.66)	0.28a (0.09)	1.42ab (0.31)	1.02a (0.14)	2.52b (0.41)	25.72b (5.50)	11.54b (2.82)	61.71a (18.42)	2.62b (0.52)
Control	11.09ab (0.32)	3.02b (0.35)	5.70b (0.58)	0.22a (0.19)	1.25b (0.31)	0.97a (0.17)	2.24b (0.52)	24.53b (5.09)	11.88ab (4.93)	52.80a (13.89)	2.02c (0.23)

[†]Means followed by the same letter within a column for the same effect are not significantly different at $P \leq 0.05$.

^{††}Standard deviation of the mean given in parenthesis.

Table 3.18. Interaction between nitrogen (N) source and application time on corn ear zinc (Zn) concentration at physiological maturity in 2019, at the Wiregrass Research and Extension Center (WREC).

Rate	Single	Split
Ear Zn concentration, mg kg ⁻¹		
U	31.00a [†]	28.37a
PL	24.75b	28.75a
Control	20.25c	20.25c

[†]Means followed by the same letter within a column and row are not significantly different at $P \leq 0.05$.

Table 3.19. Interaction between nitrogen (N) source and application time on corn ear zinc (Zn) and copper (Cu) concentrations at physiological maturity, at the Tennessee Valley Research & Extension Center (TVREC).

Source	Single	Split
Ear Zn concentration, mg kg ⁻¹		
U	23.50a [†]	22.37ab
PL	17.50c	20.37b
Control	21.25ab	21.25ab
Ear Cu concentration, mg kg ⁻¹		
U	2.87a	2.37ab
PL	2.12bc	2.37ab
Control	1.50c	1.50c

[†]Means followed by the same letter within a column and row are not significantly different at $P \leq 0.05$.

Table 3.20. Interaction between nitrogen (N) application rate and time on corn ear copper (Cu) concentration at physiological maturity in 2018, at the Wiregrass Research and Extension Center (WREC).

Rate	Single	Split
Ear Cu concentration, g kg ⁻¹		
168	2.51ab [†]	2.22b
336	2.31b	2.70a
Control	2.35ab	2.35ab

[†]Means followed by the same letter within a column and row are not significantly different at $P \leq 0.05$.

Table 3.21. Pearson correlation coefficients (n=36) of corn ear nitrogen (N) concentration with other grain mineral elements, by site year.

	N			
	EVS (2018)	WREC (2018)	WREC (2019)	TVREC (2019)
P	0.64	0.80	0.69	0.85
K	0.49	0.28	0.46	0.62
Ca	0.40	0.26	0.10	0.37
Mg	0.64	0.88	0.79	0.87
S	0.51	0.82	0.34	0.92
B	0.37	0.25	0.01	0.51
Zn	0.56	0.42	0.71	0.84
Mn	0.28	0.46	0.73	0.64
Fe	0.45	0.06	0.45	0.58
Cu	0.46	0.24	0.17	0.47

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Supplemental material

Table S1. Cover crop aboveground dry matter, tissue N concentration, and aboveground N uptake as affected by fertilizer treatments applied to the main crop during the 2018–19 growing season.

	Aboveground dry matter		Tissue N concentration		Aboveground N uptake	
	kg ha ⁻¹		g kg ⁻¹		kg ha ⁻¹	
	EVS [†]	WREC	EVS	WREC	EVS	WREC
Source						
PL	1122 (421 ^{††})	1087 (335)	24.7 (5.2)	16.3 (1.4)	27 (11)	18 (5)
Urea	1114 (372)	1057 (437)	27.1 (6.1)	16.9 (1.8)	30 (13)	17 (6)
Control	747 (213)	1022 (382)	25.6 (2.1)	16.4 (2.2)	19 (5)	17 (7)
Rate						
Low rate	1056 (354)	985 (296)	25.9 (5.4)	17.0 (1.5)	28 (13)	17 (5)
High rate	1181 (426)	1159 (447)	26.0 (6.2)	16.2 (1.7)	30 (11)	18 (6)
Time						
Single	1190 (472)	1173 (472)	27.2 (6.2)	16.8 (1.8)	32 (13)	19 (6)
Split	1047 (286)	971 (243)	24.6 (5.0)	16.4 (1.4)	26 (10)	16 (4)

[†]E.V. Smith Research Center (EVS); Wiregrass Research and Extension Center (WREC); Tennessee Valley Research and Extension Center (TVREC).

^{††}Standard deviation of the mean given in parenthesis.

Table S2. Comparison of nitrogen (N), phosphorus (P), and potassium (K) concentration in the corn grain fertilized with urea or ammonium nitrate (NH₄NO₃) as conventional fertilizer (CF) and organic manure (OM) including poultry litter (PL) among different studies.

Reference	N			P			K		
	CF [†]	OM ^{††}	Control	CF	OM	Control	CF	OM	Control
	-----g kg ⁻¹ -----								
Tewolde et al. (2019)	15.35	14.12	9.60	5.63	5.05	3.25	7.64	6.97	5.68
Hossain (2006)	15.55	14.35	11.50	3.00	3.07	2.90	3.16	3.39	3.30
Ahmadi et al. (1993)	12.72	–	–	2.60	–	–	4.15	–	–
This study [†]	14.05	12.73	12.36	3.66	3.41	3.21	6.05	5.61	5.44

[†]Corn grain also included cob and husk plant parts.

Table S3. Comparison of calcium (Ca), magnesium (Mg), and sulphur (S) concentration in the corn grain fertilized with urea or ammonium nitrate (NH₄NO₃) as conventional fertilizer (CF) and organic manures (OM) including poultry litter (PL) among different studies.

Reference	Ca			Mg			S		
	CF [†]	OM ^{††}	Control	CF	OM	Control	CF	OM	Control
	-----g kg ⁻¹ -----								
Hossain (2006)	0.08	0.08	0.09	0.94	0.96	0.93	1.27	1.25	1.11
Ahmadi et al. (1993)	0.04	–	–	0.99	–	–	–	–	–
Tewelde et al. (2019)	0.09	0.09	0.08	1.72	1.66	1.08	–	–	–
This study [†]	0.23	0.18	0.18	1.47	1.29	1.24	1.04	1.06	0.99

[†]Corn grain also included cob and husk plant parts.

Table S4. Comparison of boron (B), zinc (Zn), and manganese (Mn) concentration in the corn grain fertilized with urea or ammonium nitrate (NH₄NO₃) as conventional fertilizer (CF) and organic manures (OM) including poultry litter (PL) among different studies.

Reference	B			Zn			Mn		
	CF [†]	OM ^{††}	Control	CF	OM	Control	CF	OM	Control
	-----mg kg ⁻¹ -----								
Hossain (2006)	–	–	–	18.85	19.42	20.60	4.85	4.53	4.41
Ahmadi et al. (1993)	3.51	–	–	19.40	–	–	4.49	–	–
Tewelde et al. (2019)	–	–	–	42.60	38.92	27.20	9.3	7.82	4.6
This study [†]	2.39	2.59	2.42	29.59	28.82	28.24	14.39	10.41	10.86

[†]Corn grain also included cob and husk plant parts.

Table S5. Comparison of iron (Fe) and copper (Cu) concentration in the corn grain fertilized with urea or ammonium nitrate (NH₄NO₃) as conventional fertilizer (CF) and organic manures (OM) including poultry litter (PL) among different studies.

Reference	Fe			Cu		
	CF [†]	OM ^{††}	Control	CF	OM	Control
	-----mg kg ⁻¹ -----					
Hossain (2006)	27.43	27.97	21.96	3.28	3.19	2.41
Ahmadi et al. (1993)	20.67	–	–	2.63	–	–
Tewolde et al. (2019)	42.20	29.30	16.20	4.71	4.12	3.82
This study [†]	53.77	41.62	42.33	2.67	2.51	2.34

[†]Corn grain also included cob and husk plant parts.

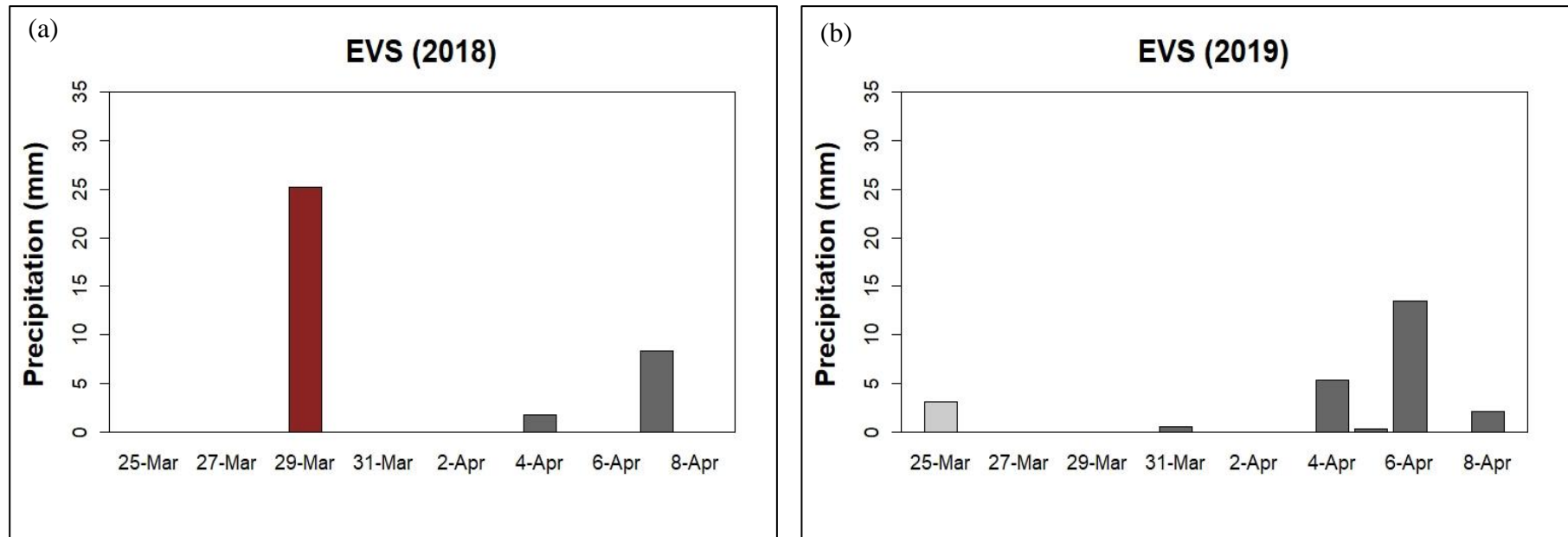


Figure S1. Rainfall events around the time of single application during the 2018 and 2019 growing seasons at E.V. Smith Research Center (EVS).

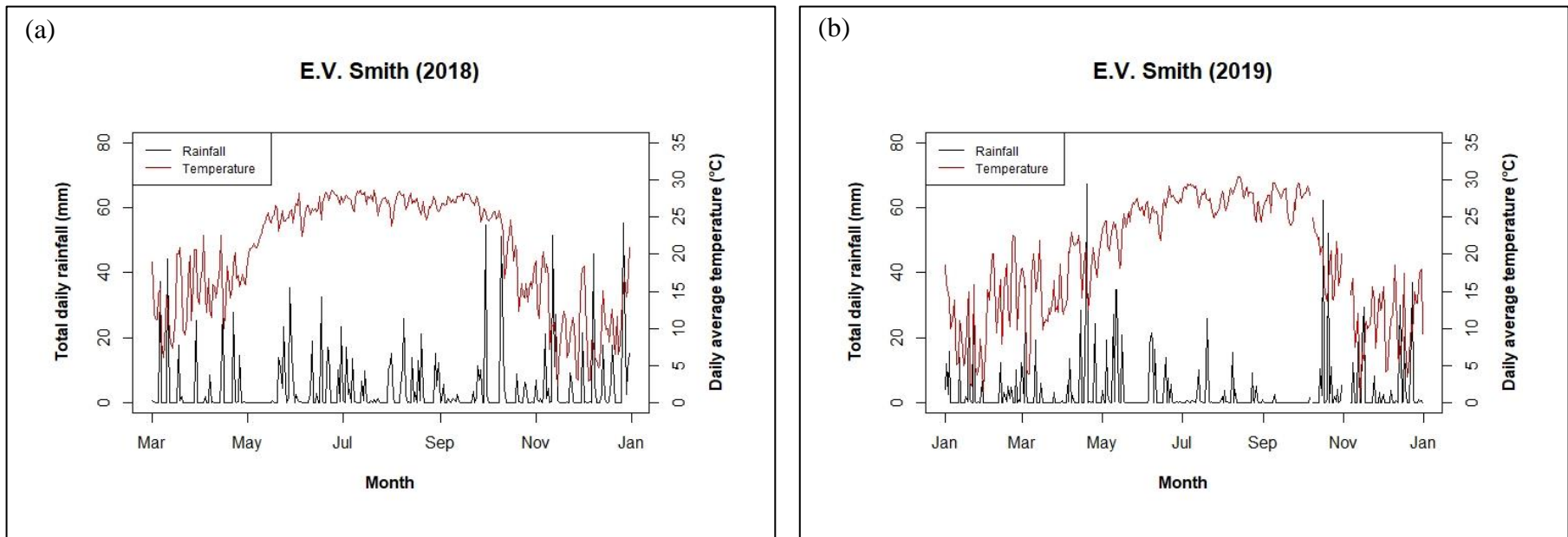


Figure S2. Daily rainfall and average air temperature during the 2018 and 2019 growing seasons at E.V. Smith Research Center.

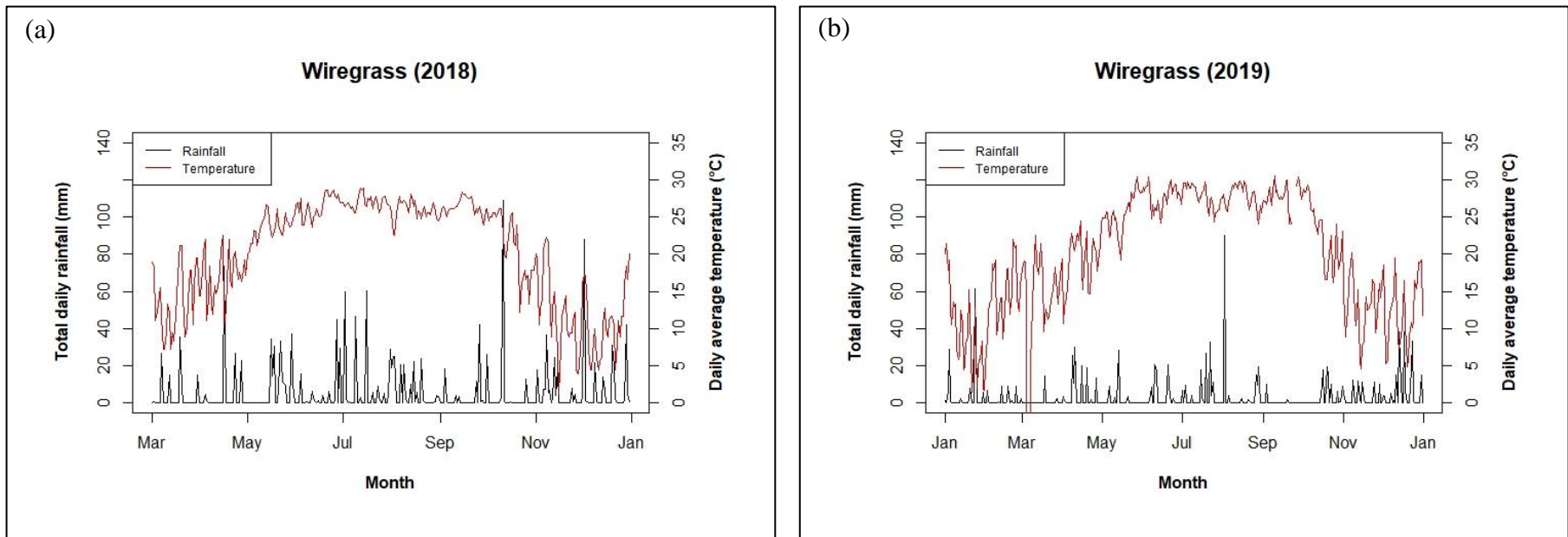


Figure S3. Daily rainfall and average air temperature during the 2018 and 2019 growing seasons at Wiregrass Research and Extension Center.

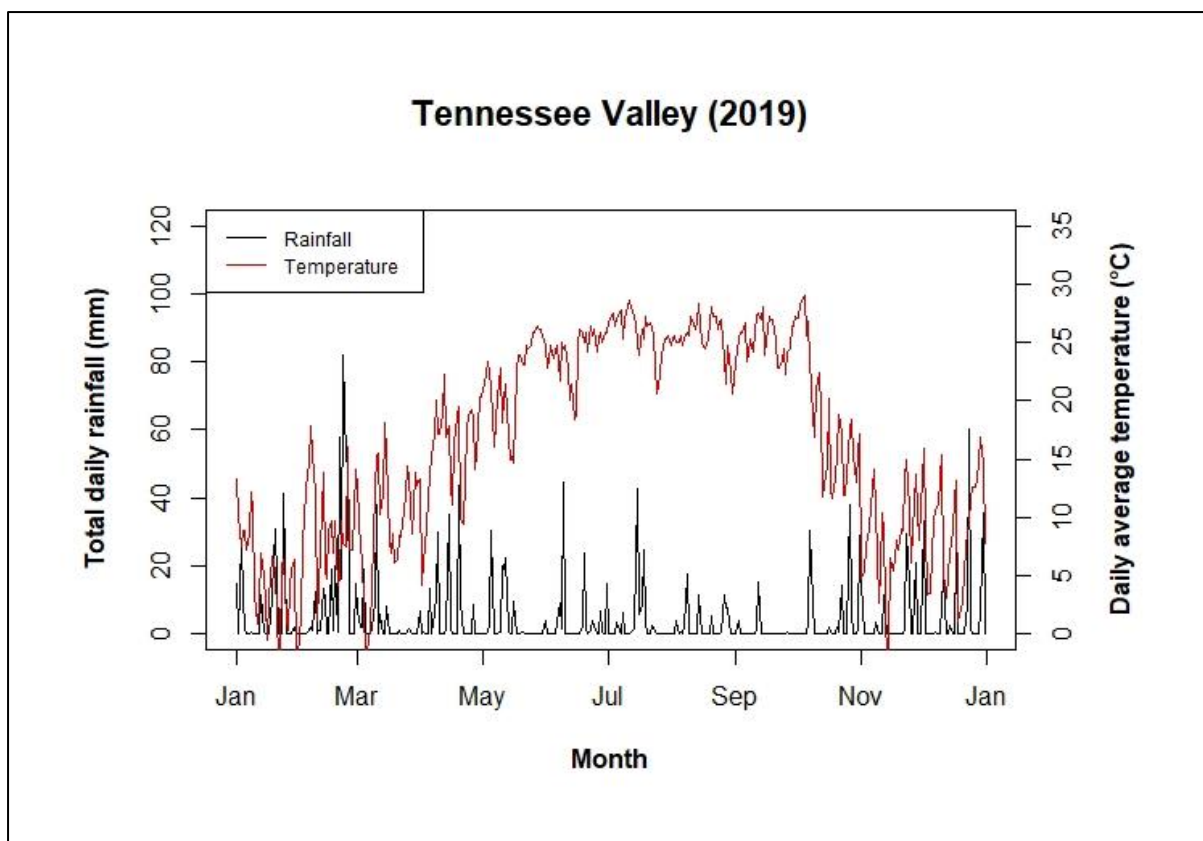


Figure S4. Daily rainfall and average air temperature during the 2019 growing season at Tennessee Valley Research and Extension Center.