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EVALUATION OF STABILIZED FERTILIZERS AND CROP CANOPY SENSORS AS
NEXT-GENERATION NITROGEN MANAGEMENT TECHNOLOGIES IN IRRIGATED
CORN

by

Leonardo Mendes Bastos

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CORN

Leonardo Mendes Bastos, Ph.D.

University of Nebraska, 2019

Advisor: Richard B. Ferguson

Nitrogen (N) is often the most limiting nutrient to corn. Once applied to the field, N can be lost through different pathways, which contributes to low N use efficiency (NUE) by plants. Increases in NUE and decreases in N losses can be potentially achieved by using management options that allow a better synchrony between N supply and demand, such as stabilized fertilizers, and spatially-variable sensor-derived in-season N application. Three studies were conducted in order to assess the effects of different stabilized fertilizers and crop canopy sensors on irrigated corn yield. The first study evaluated the effect of urease inhibitor on ammonia losses and corn grain yield. The use of urease inhibitors significantly reduced ammonia volatilization losses by 21 to 62%, but this did not translate into higher corn yields. The second study evaluated the effect of various management practices along with the use of a nitrification inhibitor and their interaction with weather on irrigated corn grain yield over 28 yrs. The use of a nitrification inhibitor had negative, neutral, and positive effects on corn grain yield, and the magnitude of its effect was less than other management practices. The most important weather variables in explaining different yield responses were year- yield potential, precipitation volume and distribution, and air temperature. The third study compared active and passive crop canopy sensors in assessing corn N deficiency and the accuracy of recommended side-dress N rates

compared to the economic optimum N rate. This study included eight field studies using different N fertilizer rates and the use of both active and passive crop canopy sensor during the mid-vegetative growth stage in corn. Active and passive sensors recommended comparable side-dress N rates given proper selection of algorithm inputs. Their recommendation was partially or fully accurate in four of six studies. Both stabilized fertilizers and crop canopy sensors are important management tool options for producers, and an understanding of their strengths and weaknesses is needed to guide proper adoption decisions.

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Chapter 1 - Literature Review

Corn Production and Fertilizer Consumption

The U.S. is the largest corn producer in the world, with ~371 million metric tons of the grain harvested in 2018 (USDA-NASS, 2018). The large area planted with the crop and high average yields are the main drivers of this level of production. To achieve high corn yields, farmers need to properly manage their crop, and fertilization is one key aspect. The main nutrient supplied via fertilization to corn in the U.S. is nitrogen (N). Corn fields alone received 47% of all N applied to crops in the U.S. in 2014 (USDA-NASS, 2018). Of all the N applied that year, 44% was in the form of N solutions, 24% as urea, and 14% as anhydrous ammonia (AA) (USDA-NASS, 2018).

Corn is an important crop in Nebraska, generating ~ US\$ 5.5 billion in the state economy in 2017 (USDA-NASS, 2018) which represented about 58% of all crop production value in the state. In 2017, Nebraska ranked third nationally in corn planted area (3.8 million ha, of which 57% were irrigated), third in corn production (43 million metric tons of grain) and fourth in yield [11.4 Mg ha^{-1} averaged over irrigated (13.1 Mg ha^{-1}) and non-irrigated (9 Mg ha^{-1})].

Nitrogen is often the most limiting nutrient to corn as soil supply can greatly vary and plant demand is high. To ensure high yielding conditions, farmers supplement N nutrition by the addition of fertilizers. In 2016, Nebraskan farmers applied an average rate of 160 kg N ha^{-1} on corn as different formulations. In that year, the three most utilized N fertilizers in corn production were urea-ammonium nitrate (UAN, 28-32% N), AA (82% N) and urea (46% N), representing 65%, 15.7% and 13.3% of total only-N fertilizer applied in the state, respectively (Nebraska Department of Agriculture, 2016). About 20% more fertilizer was applied as UAN

and 8% less as urea in Nebraska compared to average U.S. consumption. Regardless of the source, once applied to the environment, N is transformed into different forms and can be lost through different pathways, including ammonia (NH_3) volatilization (Pan et al., 2016), nitrate (NO_3^-) leaching (Quemada et al., 2013), and denitrification (Shcherbak et al., 2014).

Nitrogen Losses and Crop Nitrogen Use Efficiency

Ammonia volatilization is the emission of N as NH_3 gas to the atmosphere, and is an important loss mechanism for surface-applied, urea- or NH_4^+ -containing fertilizer sources, especially in cropping systems with high residue quantity on the soil surface. Ammonia volatilization losses from the field are affected by multiple factors and thus are highly variable, ranging from 0 to ~60% of applied fertilizer (Terman, 1980; McInnes et al., 1986a; b; Harrison and Webb, 2001; Pan et al., 2016; Silva et al., 2017), with an average loss of the order of 17.6% of applied fertilizer (Pan et al., 2016).

Nitrate leaching is the loss of N as NO_3^- to soil depths beyond the root system. Leaching can be a significant pathway of loss in both free-draining coarser-texture soils and artificially drained soils when soil NO_3^- concentration is high, excessive water is present, and evapotranspiration rates are limited (Dinnes et al., 2002; Quemada et al., 2013; Karimi and Akinremi, 2018). Under these conditions, reported NO_3^- leaching losses can occur even when no fertilizer is applied, and increase with N rate with up to 80% loss when N is applied (Bergström and Johansson, 1991; Randall et al., 2003; Quemada et al., 2013; Karimi and Akinremi, 2018).

Denitrification is a biologically-driven anaerobic process where NO_3^- is reduced in a step-wise chain of reactions following the sequence NO_3^- , NO_2^- , NO, N_2O , N_2 (Wrage et al., 2001). Different products during the denitrification reaction chain can escape to the atmosphere, including nitrous oxide (N_2O). Nitrous oxide is a greenhouse gas with a global warming potential

~300 times higher than CO₂ (Solomon et al., 2007). Furthermore, N₂O reacts with oxygen in the stratosphere to form nitric oxide (NO), which promotes ozone destruction (Ravishankara et al., 2009). Agricultural N management is the main source of national and global N₂O emissions (Forster et al., 2007; EPA, 2018), with N₂O losses ranging from ~0 to 8% of the applied fertilizer but normally not exceeding 1% (Kim et al., 2013; Fernández et al., 2014; Halvorson and Bartolo, 2014). The magnitude of N₂O loss increases with increasing N rates, especially at N rates exceeding the optimum N rate for plant production (Kim et al., 2013; Halvorson and Bartolo, 2014; Shcherbak et al., 2014).

Fertilizer losses from NH₃ volatilization, NO₃⁻ leaching and denitrification combined can be large and contribute to the low fertilizer N use efficiency (NUE) observed for various crops, including corn (Raun and Johnson, 1999). Cassman et al. (2002) reported that the regional N fertilizer recovery efficiency (RE_N) of corn, calculated as the percentage of applied N fertilizer accumulated on the crop aboveground biomass for 55 on-farm studies during the period 1995-1999 across eight U.S. Midwest states, was 37%. Despite this low value, Cassman et al. (2002) noted that the ratio of crop yield per unit of applied fertilizer (also known as the partial factor productivity of N fertilizer, PFP_N) for U.S. corn increased from 42 in 1980 to 57 kg grain kg⁻¹ N in 2000. This consistent increase in PFP_N was attributed to a significant increase in average corn yield of 109 kg ha⁻¹ yr⁻¹ combined with a stable fertilizer N application rate over time (Cassman et al., 2002). Thus, farmers are able to produce more grain with the same N input, thereby increasing crop NUE over time. Ferguson (2015) demonstrated a similar trend for corn production in Nebraska, with PFP_N values increasing from ~42 in 1965 to 67 kg kg⁻¹ in 2010.

Asynchrony in the timing of crop N demand and N application is a common reason for low observed NUE and high potential for environmental N losses. In addition to temporal asynchrony, spatial asynchrony contributes to N losses when the application of a single N rate to an entire field results in sub-field areas that are either over- or under-fertilized (Mamo et al., 2003). Therefore, further increases in NUE and decreases in N losses can be potentially achieved by using management options and technologies that allow a better synchrony between N supply and demand. In Nebraska, improvements in corn NUE have been proposed to be attainable by continued development and adoption of next-generation management practices such as fertigation, controlled release fertilizers, and spatially-variable sensor-derived in-season N application (Ferguson, 2015).

“4Rs” Framework

Producers are faced with a multitude of management to make the most efficient use of applied fertilizer, including those related to soil, crop, fertilizer, and their interactions. Given this complexity, Bruulsema et al. (2008) proposed a framework for fertilizer best management practices named the “4 Rs of nutrient stewardship”. The 4Rs are related to selecting the “right rate, source, timing, and placement” of fertilizers to achieve the objectives of productivity, profitability, sustainability and environmental health.

The literature on corn fertilizer management is vast, and many studies have summarized the effects of one or more of the 4Rs on corn yield, NUE, and N losses (Hergert and Wiese, 1980; Dinnes et al., 2002; San Francisco et al., 2011; Quemada et al., 2013; Halvorson and Bartolo, 2014; Anderson and Kyveryga, 2016; Pan et al., 2016; Silva et al., 2017; Tao et al., 2018). For example, Tao et al. (2018) conducted an extensive study that included 920 corn fields over four U.S. Midwest states during seven years assessing the effect of multiple variables on

corn N availability. Their work used the end-of-season corn stalk nitrate test (CSNT) as a measure of crop N availability during the growing season, where sample N levels were categorized as deficient, marginal, optimal, or excessive. The authors found that N rate, N source and timing, previous crop in rotation, tillage, and drainage class significantly impacted the probability of CSNT to be in a higher category. Although N rate was an important factor and normally receives the most attention, the most useful understanding of NUE, losses and yield considers N rate within the context of the other practices (e.g. placement, timing, source, previous crop) (Tao et al., 2018).

Hereafter, this review focuses on two next-generation N management technologies which are part of the 4Rs framework and proposed by Ferguson (2015). Those are: i) the use of different types of enhanced-efficiency fertilizers as a protective strategy; and ii) the use of crop canopy sensors for in-season N application as a reactive strategy.

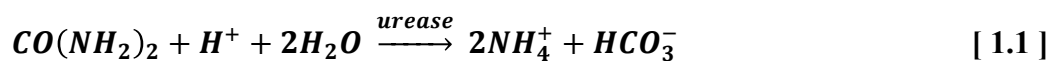
Protective Strategy – Enhanced Efficiency Fertilizers

The terminology and description for enhanced efficiency fertilizers (EEFs) and its different categories lack agreement in the literature. Trenkel (2010) summarized the most accepted terminology and proposed most of the concepts used herein. Enhanced efficiency fertilizers are defined as “fertilizers that reduce loss to the environment and/or increase nutrient availability compared with conventional fertilizers” (Olson-Rutz et al., 2011). Generally, EEFs can be categorized as slow- and controlled-release fertilizers (SCRFs), or stabilized fertilizers (SFs) (Trenkel, 2010). SCRFs decrease the rate of nutrient release by using a physical coating that creates a somewhat predictable release pattern (e.g. sulfur- and polymer-coated fertilizers), or by chemical formulations that reduce fertilizer solubility to moderate unpredictable N release due to soil and weather variability (e.g. urea-formaldehyde, magnesium ammonium phosphate)

(Trenkel, 2010). SFs are regular N fertilizers amended with a urease inhibitor (UI), nitrification inhibitor (NI), or both to decrease the reactivity of fertilizer N in chemical and biological processes that lead to N losses.

Urease Inhibitors

Urease is an environmentally ubiquitous enzyme that catalyzes the hydrolysis of urea to NH_4^+ in the soil, which increases the availability of N to subsequent volatilization loss. Urease inhibitors are molecules that bind to the active site of the urease enzyme to temporally reduce the onset and rate of urea hydrolysis (Eq. [1.1]). This reduction then decreases the potential for and magnitude of NH_3 losses (San Francisco et al., 2011; Silva et al., 2017; Sunderlage and Cook, 2018; Cantarella et al., 2018).

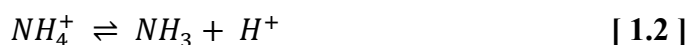


A large number of compounds and mixtures tested as UIs have shown varying levels of efficacy (Kiss and Simihaian, 2013). The most successful to inhibit urease are urea analogues. The compound N-(n-butyl) thiophosphoric triamide (NBPT, Agronomic Services, Wichita, KS) has the most proven efficacy and is the most utilized commercially since its market introduction in the mid-1990s (Cantarella et al., 2018). More recently, a new compound developed by BASF, N-(n-propyl) thiophosphoric triamide (NPPT), has been introduced to the UI market and sold in a mixture with NBPT under the trade name Limus (BASF Crop Protection). Both NBPT and NBPT+NPPT are commercialized for use with urea and UAN fertilizers. Another commercially available UI is NutriSphere-N (Specialty Fertilizer Products, LLC, Leawood, KS), which is a polymer-based product containing maleic and itaconic acid. Although the product

manufacturer's description claims that NutriSphere-N inhibits both nitrification and urea hydrolysis by complexing soil copper and nickel, respectively, the specific modes of action of this product have not been demonstrated in refereed literature and its efficacy frequently questioned (Goos, 2018; Sunderlage and Cook, 2018).

Ammonia Volatilization

Ammonia volatilization arises from the equilibrium reaction between NH_4^+ and NH_3 (Eq. [1.2]). All compounds containing NH_4^+ (e.g. AN, AS, UAN) or NH_4^+ -forming compounds (e.g. urea, UAN) undergo this reaction, which has a $\text{pK}_a = 9.25$ at 25C.



Ammonia volatilization can comprise a large percentage of applied N fertilizer under certain conditions. These losses are important from both an economic and environmental perspective. Pan et al. (2016) estimated that, with an average NH_3 loss of 17.6% of the fertilizer N applied and the estimated global demand for N fertilizer in 2014 of 112 million tons, the cost of global NH_3 losses from N application were in the order of US\$ 15 billion. This economic cost estimate increases substantially if subsequent crop yield declines due to limited N nutrition are included. The environmental cost of volatilized NH_3 includes the deposition of reactive N into non-target ecosystems, causing undesirable changes on reproductive success, herbivory, and competition patterns (Adams, 2003) and subsequent biodiversity impacts (Guthrie et al., 2018).

The potential and magnitude of NH_3 volatilization from NH_4^+ -containing and NH_4^+ -forming fertilizer is affected by multiple factors. Those include fertilizer management (source, rate, placement), soil properties (texture, H^+ buffering capacity, cation exchange capacity, pH, residue cover, moisture, temperature, competing processes), weather (rainfall/irrigation, wind speed/air exchange, air relative humidity (RH)), and their interactions (Hargrove, 1988; Harrison

and Webb, 2001; Gioacchini et al., 2002; Sommer et al., 2004; Kissel et al., 2008; San Francisco et al., 2011; Silva et al., 2017; Sunderlage and Cook, 2018). These factors are discussed below.

Important Factors

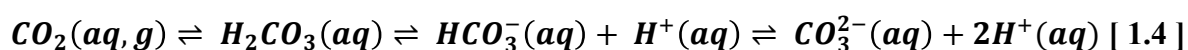
Fertilizer management plays an important role in NH_3 loss magnitude because the selection of N fertilizer type (i.e. N source), amount, placement, and use of inhibitor product will define the specific environmental reactions that may lead to loss. Fertilizer N sources can be categorized as acidic [e.g. ammonium sulfate (AS), ammonium nitrate (AN)] or alkaline (e.g. AA, urea) in relation to how they change soil pH after dissolution and decomposition (Sommer et al., 2004). Furthermore, N sources containing NH_4^+ salts can be categorized as Ca^{+2} precipitate-forming (e.g. AS) and nonprecipitate-forming (e.g. AN) in relation to the soil calcium carbonate (CC) content and soil pH (Fenn and Kissel, 1974; Sommer et al., 2004).

Generally, as the initial soil/residue pH rises, the proportion of total ammoniacal N (TAN) prone to NH_3 loss increases. More precisely, the NH_3 proportion of TAN prone to volatilization is 0.0026, 0.26, and 20.8% at initial pH values of 5, 7, and 9 at 14.5 °C, respectively.

Under acidic soil conditions (indicative of negligible CC), the potential and magnitude of NH_3 losses from fertilizer N source follows the order of urea > UAN > AN ~ AS (Keller and Mengel, 1986; Hargrove, 1988; Harrison and Webb, 2001; Sommer et al., 2004; San Francisco et al., 2011; Pan et al., 2016). Urea has the highest NH_3 loss potential because its hydrolysis consumes H^+ (Eq. [1.1]), which directly increases the pH of the surrounding soil and shifts the equilibrium in Eq. [1.2] to the right. For example, Keller and Mengel (1986) found that NH_3 losses from urea, UAN, and AN were 30, 9, and 4% of the total N fertilizer applied on an acidic

sandy loam. Similarly, AS under acidic soil conditions has a very low NH₃ loss potential since the initial low pH favors the NH₄⁺.

Under alkaline soil conditions (indicative of increased CC content), the sulfate anion of AS competes with soil carbonate anions (CO₃²⁻) to form a precipitate with Ca⁺² thereby removing Ca⁺² from the soil solution. This shifts Eq. [1.3] to the left, increasing CO₃²⁻ availability (Harrison and Webb, 2001). To compensate for excess CO₃²⁻, the equilibrium in Eq. [1.4] shifts to the right, causing a consumption of H⁺ and a concurrent increase in soil pH, which favors NH₃ formation (Eq. [1.2]) and loss (Fenn and Miyamoto, 1981). Therefore, NH₃ volatilization potential is the highest for urea and lowest for AN and AS under acidic soil conditions, whereas under alkaline soil conditions AN is the least prone to losses.



Fertilizer N rate interacts with soil pH, CC content, and H⁺ buffering capacity in determining the extent of NH₃ volatilization. On acidic (i.e. noncalcareous) soils, greater NH₃ volatilization occurs with increasing application rates of surface-applied fertilizers containing or forming NH₄⁺ (Fenn and Kissel, 1974; Hargrove, 1988; Wang et al., 2004; Ma et al., 2010; Pan et al., 2016; Cantarella et al., 2018). On alkaline (i.e. calcareous) soils, increasing N rate has no effect on NH₃ volatilization losses (Fenn and Kissel, 1974; Du Preez and Burger, 1988). For example, Pan et al. (2016) summarized a total of 824 observations from 145 published studies on ammonia volatilization and concluded that increasing N rate of various N sources increased average NH₃ loss up to 180% more as compared to the lowest N rate. The authors attributed this to the increased availability of NH₄⁺, and in the case of urea, a greater increase in soil pH. On the

other hand, Fenn and Kissel (1974) found that increasing AN application rate on a calcareous soil did not influence total NH_3 volatilization, which varied from 15 to ~28% of applied fertilizer for N rates varying from 33 to 550 kg N ha⁻¹ under temperatures ranging from 12 to 32C.

Fertilizer placement is an important management practice in controlling NH_3 loss potential. Generally, NH_4^+ -containing and NH_4^+ -forming fertilizers, and especially urea, are more prone to NH_3 volatilization when surface-applied compared to when incorporated (Ernst and Massey, 1960; Hargrove, 1988; Sommer et al., 2004; Pan et al., 2016). Greater potential for NH_3 loss from surface application results from an increase in TAN in solution, and in the case of urea, a concurrent hydrolysis-driven pH increase, favoring TAN in the NH_3 form. When incorporated, both TAN and pH are controlled by soil cation exchange capacity (CEC) and H⁺ buffering capacity (HBC), respectively, which reduces the potential for NH_3 loss. For example, Pan et al. (2016) observed a 55% reduction in NH_3 volatilization when various N fertilizers were deep-placed compared to surface application, and Rochette et al. (2013) found negligible NH_3 losses when urea was placed at 7.5 cm below the soil surface. The depth of injection plays an important role, with NH_3 losses decreasing the deeper the fertilizer is placed (Fenn and Miyamoto, 1981; Rochette et al., 2013). Surface application method, however, can affect NH_3 volatilization. Broadcasting urea fertilizer can both increase (Hargrove, 1988) or decrease (Bouwmeester et al., 1985; Sommer et al., 2004; Cantarella et al., 2018) volatilization compared to surface banding.

Various soil properties are known to influence the potential of NH_3 losses (Hargrove, 1988; Sommer et al., 2004; Silva et al., 2017; Sunderlage and Cook, 2018). In addition to soil pH effects (discussed previously), multiple studies have reported a significant correlation between soil texture and NH_3 losses, with larger losses observed under higher sand content (Martens and Bremner, 1989; San Francisco et al., 2011) and lower losses under higher silt and clay content

soils (San Francisco et al., 2011; Sunderlage and Cook, 2018), although this relationship has not always been observed (Silva et al., 2017).

Initial soil pH has been shown to impact NH_3 losses, with higher pH favoring higher losses (Ernst and Massey, 1960; Ferguson et al., 1984), especially in the case of AN application (San Francisco et al., 2011). In the case of urea application, perhaps a more important variable in explaining NH_3 loss magnitude is the soil HBC (Avnimelech and Laher, 1977; Ferguson et al., 1984; Sommer et al., 2004; Sunderlage and Cook, 2018). Higher soil HBC can both increase or decrease NH_3 loss, depending on whether buffering power is working against a decrease or increase in pH, respectively (Ferguson et al., 1984; Hargrove, 1988). For example, Ferguson et al. (1984) observed a decrease in NH_3 loss from 42 to 18% of 224 kg N ha^{-1} as HBC increased while keeping CEC and initial pH the same on a noncalcareous silt loam that buffered against an increase in pH. On the other hand, Avnimelech and Laher (1977) found an increase in NH_3 loss as HBC increased on a calcareous soil that buffered against a decrease in pH.

Another soil variable closely related to NH_3 loss is soil CEC. Generally, NH_3 losses decrease as CEC increases (Keller and Mengel, 1986; Hargrove, 1988; Sommer et al., 2004). Higher soil CEC allows greater adsorption of NH_4^+ , thus decreasing soil solution TAN and subsequent loss potential. However, the effect of CEC can be suppressed by HBC, as demonstrated by Ernst and Massey (1960). The authors observed that applying lime to a silt loam increased both CEC and NH_3 losses, and attributed this effect to a decrease in HBC due to liming.

Soil residue cover also affects NH_3 losses and is especially important for surface-applied urea fertilizer. Crop residue has both high pH and HBC (McInnes et al., 1986a); greater urease activity than soil (McInnes et al., 1986a); and creates a physical barrier between fertilizer and

soil (Silva et al., 2017), all of which increase the chances for NH_3 loss. For example, a literature review by Pan et al. (2016) observed that residue cover increased NH_3 losses by 25%. Similarly, San Francisco et al. (2011) found that NH_3 losses (as percentage of applied urea) increased by a median value of 60% in residue-covered soils compared to bare soils across 12 different soils.

Two other important aspects that control NH_3 losses and are related to residue cover are soil temperature and moisture. Soil temperature impacts NH_3 losses through multiple chemical and biochemical reasons. As soil temperature rises, it i) increases urea hydrolysis rate; ii) increases NH_3 proportion in the gas rather than liquid phase; iii) increases diffusion rates of both urea and NH_4^+ ; and iv) increases CaCO_3 solubility in alkaline, calcareous soils (Hargrove, 1988). For example, Ernst and Massey (1960) observed cumulative NH_3 volatilization of 6, 10, 15, and 24% of surface-applied urea fertilizer when under a Dickson silt loam incubated at temperatures of 7, 16, 24, and 32°C, respectively, during 11 days (initial soil pH of 6.5). However, field- and laboratory-measured NH_3 volatilization has been reported to be in the order of ~11-25%, ~82-92% and 93% of added ammonia via manure application under temperatures of -20, -3, and 10°C, respectively (Steenhuis et al., 1979), with lower temperatures having a longer emitting period than higher temperatures. Furthermore, soil temperature interacts with soil water content in determining NH_3 peak loss (Hargrove, 1988). Larger losses have been observed when soil temperature is increasing and soil is drying, but not at daily maximum temperature (Bouwmeester et al., 1985; McInnes et al., 1986a; b), because at this point soil surface reaches dryness, and the lack of water to drive urea hydrolysis becomes the controlling factor.

Soil water content is an important driver of NH_3 losses due to i) controlling urea hydrolysis rate; and ii) affecting diffusion and mass flow of surface-applied fertilizer both into the soil and upward thereafter as the surface dries (Hargrove, 1988). For example, Ernst and

Massey (1960) reported cumulative NH_3 loss of 3, 3, 12, and 19% of surface-applied urea on a Dickson silt loam with initial soil gravimetric moisture content of 1, 5, 21, and 38%, respectively, over 14 days. The significantly smaller losses at 1 and 5% soil moisture content were attributed to incomplete urea hydrolysis. Similar conclusions were reported by others (Bouwmeester et al., 1985; McInnes et al., 1986b). Other soil competing processes, such as nitrification, may play a role on the extent of NH_3 losses by controlling TAN concentration, and by reducing soil pH. For example, Flowers and O'Callaghan (1983) reported on a pH decrease of 1 unit after complete nitrification of 250 ppm of NH_4^+ -N.

Weather variables interact with both fertilizer management choices and soil properties to determine NH_3 losses. The most important weather variables impacting NH_3 volatilization are rainfall/irrigation, wind speed, and RH. Rainfall/irrigation volume and frequency are perhaps the most important variables when considering urea surface application because they determine the extent of water availability for both urea hydrolysis on the surface (increased loss potential) and urea movement into the soil (decreased loss potential) (Harper et al., 1983; Bouwmeester et al., 1985; McInnes et al., 1986a; b; Sommer et al., 2004; Holcomb et al., 2011; Pan et al., 2016). Some studies have reported a significant decrease in NH_3 volatilization from surface-applied urea after receiving a single-input event of 11-22 mm (Holcomb et al., 2011) and 24 mm (Bouwmeester et al., 1985). On the other hand, McInnes et al. (1986b) observed that rainfall volumes up to 9 mm promoted urea hydrolysis but were not enough to leach urea into the soil and ended up increasing volatilization losses. Rainfall frequency can impact the potential and magnitude of losses, with loss potential increases under high-frequency, small-volume events compared to a one-time, same-volume event (Bouwmeester et al., 1985; McInnes et al., 1986a; b). Changes in wind speed can impact NH_3 losses by promoting soil drying and air exchange.

Bouwmeester et al. (1985) reported on increased NH_3 losses at the lowest wind speed and attributed this effect to the concurrent slow drying of the soil. Additionally, higher wind speeds cause faster soil drying, which decreases the rate of urea hydrolysis and loss potential (Ernst and Massey, 1960; Bouwmeester et al., 1985). Air RH is also important in determining NH_3 losses. High RH (ca. 85%) can have a similar effect as high-frequency, small-volume rainfall events on increasing urea hydrolysis and NH_3 losses (Bouwmeester et al., 1985). Various studies have demonstrated a decrease in NH_3 volatilization with decreased RH (Ernst and Massey, 1960; Ferguson and Kissel, 1986; McInnes et al., 1986a), which is attributed to soil drying and concurrent decrease in the rate of urea hydrolysis.

Ammonia volatilization losses are very complex and respond to multiple fertilizer, soil, and weather variables, and their interactions. Generally, higher losses are expected from urea or urea-containing fertilizers on any type of soil and from AS on calcareous soils; when applied on the soil surface on top of moist crop residue on a coarser-texture soil; when CEC and HBC are low and in pH is high; under medium-to-high air temperatures and high RH; and when winds are slow.

UI Effect on Loss

When urea is surface-applied, the use of a UI can create large savings in N kept from being volatilized (Gioacchini et al., 2002; San Francisco et al., 2011; Soares et al., 2012; Pan et al., 2016; Sunderlage and Cook, 2018). A UI increases the efficacy of urea application under loss-conducive conditions by both delaying hydrolysis and decreasing the extent of peak loss rate (Soares et al., 2012; Silva et al., 2017). As a result, a UI allows more time for urea incorporation into the soil before significant hydrolysis occurs (Sommer et al., 2004). In an incubation study comprising 79 soils across the U.S., Sunderlage and Cook (2018) observed that 24.5% of

surface-applied urea-N was volatilized, but that the addition of NBPT+NPPT decreased losses to 6.3% of applied N (a 75% loss reduction). Similarly, an incubation study by San Francisco et al. (2011) found significant reductions in NH_3 volatilization from 10 of 12 soils with 2.4 t ha^{-1} of wheat residue and receiving surface application of urea alone or urea+NBPT. The authors reported NH_3 losses of untreated urea ranging from 28 to 59% of applied N (mean=44%, median=43%), while those from urea+NBPT were significantly reduced and ranged from 4 to 34% (mean=17.5%, median=20%). In a meta-analysis of 35 studies from 12 different countries, Silva et al. (2017) found that UIs decreased NH_3 losses by 50% compared to untreated urea across a wide range of soil pH, soil texture, soil organic carbon content, N application rates, and NBPT rates. Finally, Cantarella et al. (2018) found no difference in NH_3 loss reductions from using NBPT (53.2% reduction compared to urea alone) between laboratory or field studies when summarizing the results from four meta-analyses.

UI Effect on Yield

In spite of the demonstrated efficacy of NBPT in decreasing NH_3 losses from applied urea, limited yield responses to urea+NBPT application compared to untreated urea have been found (Gioacchini et al., 2002; Abalos et al., 2014; Silva et al., 2017; Cantarella et al., 2018). Cantarella et al. (2018) observed a yield increase averaged over multiple crops of 6% from NBPT use compared to untreated urea. These authors reported on crop-specific average yield increases of 10.2, 7.6, 4.1, and 1.8% for wheat, rice, corn, and cotton, respectively, from NBPT use compared to urea alone. In accordance, Abalos et al. (2014) summarized 27 studies and 160 observations related to the impact of UI on yield of multiple crops and reported an average yield increase of 10% from using NBPT compared to untreated fertilizer. Similarly, Silva et al. (2017) observed an average yield increase of 5.3% over multiple crops, and noted that application rates

< 80 kg N ha⁻¹ had the largest benefit from NBPT use (8% yield increase), and attributed the response to N likely being the most limiting factor under such conditions. Fox and Piekielek (1993) also found no significant corn yield increase at low N rates (56 kg N ha⁻¹) when comparing both untreated and NBPT-treated urea and UAN, but observed a 11 to 16% yield increase when NBPT was applied with urea at the highest rate (168 kg N ha⁻¹). The lack of or reduced yield response from NBPT use, even when N savings from volatilization are high, has been attributed to the large contribution of mineral N from other sources (e.g. mineralization) that end up supplying enough N and avoid untreated urea to cause crop N deficiency (Cantarella et al., 2018). Rose et al. (2018) further pointed out that many studies evaluating the effect of EEFs, including UIs, on grain or biomass yield do not include them at multiple N rates, and reported that the largest yield increase from their use (11% over untreated fertilizer) was observed at 50% of the optimal N rate. Therefore, excessive N supply from different sources, including N mineralization and high N rates, likely mask the positive effects of UI loss savings and thus are weakly translated into increased final yield.

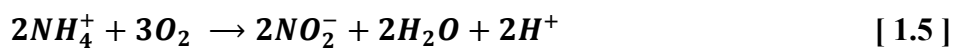
UI Effect on NUE

The effect of UI use on NUE has been reported by multiple studies and currently summarized in two meta-analyses (Abalos et al., 2014; Cantarella et al., 2018). Abalos et al. (2014) found that using inhibitors overall (UI, NI, and UI+NI) significantly increased NUE (12%) compared to no inhibitor use for multiple crops. This benefit was greatest when soil pH < 6 (~24%), under medium-texture soils (~22%), and when applied with N at rates > 300 kg N ha⁻¹ (~22%). Furthermore, the authors noted that UI alone had the largest variability among inhibitors (UI, NI, and UI+NI), with NUE confidence interval ranging from ~5 to ~25% compared to untreated fertilizer. Fox and Piekielek (1993) evaluated N uptake from urea and UAN with and

without NBPT at three fertilizer rates (56, 112, and 168 kg N ha⁻¹) over three years, and found that the use of NBPT averaged over years: i) had no effect on N uptake at the lowest N rate; ii) increased N uptake by 20 to 28% when both urea and UAN were applied at planting, but decreased N uptake at the medium rate by 12% when UAN was applied at side-dress at the medium rate; and iii) increased N uptake by 18% when urea was applied at both planting and side-dress at the highest N rate. Fox and Piekielek (1993) results partially disagree with those found by Abalos et al. (2014), in that the highest N uptake increase from NBPT use was found at an intermediate N rate on the former study, whereas the latter observed higher NUE increase from NBPT at higher N application rates.

Nitrification Inhibitors

Nitrification inhibitors (NIs) are compounds that block one or more of the steps in the nitrification process. Nitrification is the biologically-driven process where first NH₄⁺ is transformed into NO₂⁻ by *Nitrosomonas* spp. (Eq. [1.5]), followed by the transformation of nitrite to nitrate by *Nitrobacter* spp. (Eq. [1.6]).



Hauck (1980) stated that the ideal NI for agricultural use would “specifically block ammonia but not nitrite oxidation, does not adversely affect other beneficial soil organisms and higher plants, is not toxic to animals and humans in amounts used to effectively inhibit nitrification, remains effective in soil for several weeks after fertilizer application, and is economical to use”. Three commercial NIs have been the most utilized and researched. They are

nitrapyrin (2-chloro-6-trichloromethyl-pyridine), dicyandiamide (DCD), and DMPP (3, 4-dimethylpyrazole phosphate). Nitrapyrin was the first commercial NI to become available, introduced into the market in 1974 (Wolt, 2000) as N-Serve[®] (Dow Agrosiences LLC, Indianapolis, IN). Nitrapyrin is volatile (Briggs, 1975) and thus mostly used with AA applications, although a new encapsulated formulation with decreased nitrapyrin volatilization potential (Instinct, Dow Agrosiences LLC, Indianapolis, IN) has been recently launched for use with surface applied N fertilizer (Sassman et al., 2018). Various products in the U.S. contain DCD, but the most commercially recognized are Super-U[®] and Agrotain Plus SC[®] (Koch Agronomic Services, Wichita, KS). Super-U[®] is a urea formulation incorporated with both DCD (NI) and NBPT (UI), while Agrotain Plus SC[®] is a fertilizer stabilizer containing DCD designed to add to UAN. DMPP has not yet been commercialized in the U.S., and is sold under the trade name Entec[®] (BASF) in Europe.

Nitrate Leaching and Denitrification

Normally, losses from NO_3^- leaching are higher than as N_2O emissions (Gollany et al., 2004; Quemada et al., 2013; Maharjan et al., 2014; Pan et al., 2016). Under loss-prone conditions, NO_3^- leaching can represent a large portion of applied N, causing crop N deficiency, economic loss to the producer, and negative environmental impacts. For example, increased adoption of irrigation and fertilizer practices in Nebraska corn production from 1950 to 1970 resulted in groundwater contamination with agriculturally-derived NO_3^- in large areas, especially those with coarse soil and shallow aquifers (Ferguson, 2015). In contrast, denitrification losses as N_2O emissions are generally small and unlikely to be intentionally managed by a producer (Snyder et al., 2014). Nonetheless, management practices that improve NUE in general will likely decrease N losses, including N_2O (Snyder et al., 2009; Van Groenigen et al., 2010).

Several factors influence the loss potential and magnitude as NO_3^- leaching and denitrification (Bergström and Johansson, 1991; Dinnes et al., 2002; Hofstra and Bouwman, 2005; David et al., 2009; Cameron et al., 2013), including soil [texture, organic matter (OM), drainage, tillage, pH for denitrification only], management (N fertilizer timing and rate, irrigation timing and rate, crop rotation, tillage), and weather (rainfall, temperature). The majority of these factors impact NO_3^- leaching and denitrification in the same direction. Soil drainage is perhaps the main variable that has a significant different directional effect on leaching as compared to denitrification, with the former increasing as drainage increases (Dinnes et al., 2002; Hofstra and Bouwman, 2005), and the latter increasing as drainage decreases (David et al., 2009).

To reduce or mitigate these N losses, various management practices can be implemented, including adjusting N rates to optimum yield, better irrigation management, and the use of a NI (Dinnes et al., 2002; Hofstra and Bouwman, 2005; Cameron et al., 2013; Quemada et al., 2013).

NI Effect on Loss

The use of a NI can significantly decrease NO_3^- leaching and denitrification losses under certain environmental and edaphic conditions (Wolt, 2004; Akiyama et al., 2009; Quemada et al., 2013; Qiao et al., 2015). Wolt (2004) summarized seven studies from the U.S. Midwest region that compared NO_3^- leaching loss from NI-treated and untreated fertilizer under different crops; N fertilizer timing, application method, N rate, and N source; and different soil types. The use of NI decreased N lost via leaching by 15.8% as compared to fertilizer alone in 19 out of 24 observations, though results varied widely and ranged from a maximum reduction of 42.6% to increased N losses of 31.7%. Similarly, a meta-analysis by Quemada et al. (2013) found an 18% reduction in leaching losses when NI was used compared to untreated fertilizer, but noted that NIs were less effective than improved water management, which reduced N leaching by 58%.

However, Quemada et al. (2013) further noted that the use of NI was the strategy that had the least impact on NO_3^- leaching, with improved water management having the largest impact in reducing leaching (58% less than the control).

Wolt (2004) also reported that NIs reduced N_2O losses by 51.2% compared to untreated fertilizer alone. In a larger meta-analysis of EEFs, Akiyama et al. (2009) concluded that the overall effect of NI on N_2O losses was -38% compared to untreated fertilizer, specifically where losses were -30, -50, and -50% using DCD, nitrapyrin and DMP, respectively.

NI Effect on Yield

The reported effects of NI on grain yield are variable, with positive, neutral, and negative outcomes compared to fertilizer alone (Touchton et al., 1979; Hergert and Wiese, 1980; Hoefl, 1984; Blackmer and Sanchez, 1988; Cerrato and Blackmer, 1990; Ferguson et al., 1991, 2003; Wolt, 2004; Randall and Vetsch, 2005; Quemada et al., 2013; Burzaco et al., 2014; Qiao et al., 2015; Sassman et al., 2018). It is often expected that the use of a NI will be reflected in higher grain yield in case a response to N fertilizer exists and N loss pressure is high to the point of limiting N availability to crops (Hergert and Wiese, 1980). For example, Hergert and Wiese (1980) reported a 10-40% frequency of positive yield response from NI for irrigated corn in fine textured soils in Kansas and Nebraska. Wolt (2004) reported a 75% frequency of positive yield from NI use, with an average positive yield effect of 7%. However, NIs can also negatively impact grain yield as a result of: i) drier soil conditions causing N positional unavailability (Hoefl, 1984; Sassman et al., 2018); ii) NI-induced N immobilization (Ferguson et al., 1991, 2003); and/or iii) adverse effects of NI on plant growth (Blackmer and Sanchez, 1988). Hoefl (1984) reported a yield decrease from the use of AA at 67 kg N ha^{-1} with NI on a dry year, and attributed this to positional unavailability since roots were likely extracting water from deeper

soil layers whereas N was positioned on layers closer to the dry surface. Sassman et al. (2018) observed the use of NI to decrease corn grain yield in two out of three years, and increase agronomic and economic optimum N rate in one year when UAN was applied at multiple N rates. The authors suggested that the negative impact of NI could be due to the high efficacy of the inhibitor, thus maintaining more N as ammonium in a small soil volume, decreasing the chances of fertilizer interception by roots. Ferguson et al. (1991) observed a decrease in inorganic N in NI-treated AA injection bands in three years of field corn studies, and suggested that this was due to NI-induced temporary N immobilization. Blackmer and Sanchez (1988) observed that most of the site-year-rate data points that increased corn leaf, stover, and grain N concentration yet produced grain yields below plateau levels were NI-treated, and attributed this to a negative effect of the inhibitor on plant growth.

NI Effect on NUE

Similarly to its variable effect on yield, NI effect on crop NUE can be positive, neutral, and negative (Chancy and Kamprath, 1982; Walters and Malzer, 1990; Ferguson et al., 1991; Burzaco et al., 2014; Qiao et al., 2015), depending on NO_3^- leaching loss pressure, N deficiency extent, and N immobilization. Chancy and Kamprath (1982) observed no effect of NI on NUE of corn grown on coarse-texture soils in a year when NO_3^- leaching losses were negligible (42 vs. 46% recovery of applied fertilizer from urea and urea+NI, respectively), but NI increased NUE and grain yield in a wet year when significant NO_3^- leaching occurred (17 and 53% of applied fertilizer from urea and urea+NI, respectively). Walters and Malzer (1990) observed an increase in corn NUE from urea+NI only at a low N rate (90 kg N ha^{-1}) when high NO_3^- leaching conditions existed, but not when NO_3^- leaching was negligible compared to urea alone. Burzaco et al. (2014) reported on the RE_N for both a 2-year study with UAN alone and with NI applied to

corn at two different timings, as well as a meta-analysis including 112 treatment means from eight studies. The authors found that RE_N for the 2-year field study increased 10% when NI was used, and to a greater extent when applied pre-plant (20% increase) vs. side-dress (negligible), but their meta-analysis indicated no effect of NI on RE_N . Ferguson et al. (1991) reported on RE_N from three years of corn field studies receiving different N rates of AA with and without NI applied at V6 to V9 stages. The authors found that NI reduced RE_N at two N rates in two years, and had a positive, neutral, and negative effect in RE_N in the third year depending on the N rate. These authors attributed the overall negative impact of NI on corn RE_N to a temporary immobilization of N caused by the presence of NI.

Urease plus Nitrification Inhibitors

Fertilizer products containing both UI and NI have been suggested as an option to reducing all three major N losses (Xu et al., 2000), and further protecting yield and improving NUE (Zaman et al., 2008). However, the effect of UI+NI on N losses, crop yield and NUE has been variable in the literature (Gioacchini et al., 2002; Soares et al., 2012; Abalos et al., 2014; Pan et al., 2016).

Combined Inhibitor Effect on Loss

Studies evaluating the effect of UI+NI have found: i) NH_3 volatilization generally being intermediate compared to the inhibitors individually (Gioacchini et al., 2002; Soares et al., 2012; Pan et al., 2016); ii) N_2O emissions being larger, equal, and smaller with UI+NI compared to UI alone (Akiyama et al., 2009; Khalil et al., 2009; Sanz-Cobena et al., 2012; Drury et al., 2017); and iii) NO_3^- leaching losses being larger with UI+NI compared to UI only (Gioacchini et al., 2002; Sanz-Cobena et al., 2012).

Adding UI only appears more effective at reducing NH_3 losses compared to combining UI+NI. Soares et al. (2012) conducted a laboratory study evaluating the effect of UI and UI+NI when surface applied with urea and their effects on NH_3 volatilization. Of the total applied N, 37% was lost as NH_3 from unprotected urea, 15% lost for urea+UI, 44% lost for urea+NI, and intermediate losses (28-33%) for urea+UI+NI. The authors observed higher soil pH, higher NH_4^+ and lower NO_3^- as a result of nitrification inhibition, and attributed these effects as the reason for higher NH_3 losses from NI use. Pan et al. (2016) summarized 145 studies on the effect of different management practices on NH_3 volatilization, and found that fertilizer+NI increased NH_3 loss by 38% compared to untreated fertilizer, except for DMPP (no effect). Gioacchini et al. (2002) studied the effect of urea alone, urea+UI, and urea+UI+NI on NH_3 volatilization and NO_3^- leaching losses from a clay loam and sandy loam, reporting that urea+UI+NI increased NH_3 volatilization from both soils but to a larger extent from the clay loam compared to urea+UI. These studies indicate that adding NI to UI-containing fertilizer offsets some of the NH_3 volatilization savings from UI alone.

Conversely, studies have found that adding UI to NI-containing fertilizer has limited or no effectiveness on reducing N_2O losses, and in many cases increased N_2O losses. Khalil et al. (2009) conducted a laboratory study evaluating the effect of urea, urea+NI, urea+UI, and urea+NI+UI on N_2O losses over 45 d, and found that urea+UI significantly increased N_2O losses by ~7 times compared to urea+NI or urea+UI+NI, and by ~2 times to unprotected urea when fertilizers were surface-applied. Similarly, Woodley et al. (2018) reported an average increase in N_2O losses of ~17 and 19% from UAN+UI compared to both UAN+UI+NI and unprotected UAN, respectively, over two years of a field study. Drury et al. (2017) also found that broadcasting urea with UI significantly increased N_2O losses compared to urea alone, and

numerically more than urea+UI+NI in one out of two years of field study. During this same year, injecting UAN+UI emitted significantly 2.4 times more N₂O than both unprotected UAN and UAN+UI+NI. Sanz-Cobena et al. (2012) observed significantly less N₂O cumulative losses under urea+UI as compared to urea alone, but no difference from urea+UI+NI in one year; and no difference among urea, urea+UI, and urea+UI+NI on the second year of a field study with corn. The authors attributed this result to a negative effect of one inhibitor on the other. The overall UI-effect of stimulating N₂O loss may reflect that urea hydrolysis is not directly linked to N₂O production as is nitrification, but that eventually all urea will be hydrolyzed and undergo nitrification regardless of hydrolysis rate (Akiyama et al., 2009).

Although NI alone has been demonstrated to decrease NO₃⁻ leaching losses (Wolt, 2004; Quemada et al., 2013), combining NI and UI appears to have no effect or increases NO₃⁻ leaching losses compared to using inhibitors individually. Gioacchini et al. (2002) found that the inclusion of any of the inhibitors (UI, NI) enhanced both fertilizer-derived and total (fertilizer-plus soil-derived) nitrate leaching compared to untreated fertilizer in both a sandy loam and clay loam soils, and that UI+NI had significantly higher nitrate leaching losses than UI alone. The authors attributed this effect to an increase in N mineralization through an additive priming effect derived from the use of inhibitors. Sanz-Cobena et al. (2012) also observed a significant increase in NO₃⁻ leaching during the growing season when both UI+NI were used as compared to UI-only in one year (17 vs. 12 kg NO₃⁻ -N ha⁻¹), but no difference between them in the second year. Interestingly, authors observed a 6-month lag in inhibitor effects, such that effects were noted during the fallow period between growing seasons when leaching losses were significantly higher and losses were greatest in urea+UI+NI, then urea alone, then urea+UI (75, 60, and 43 kg NO₃⁻ -N ha⁻¹, respectively). These studies provide some evidence that adding NI to UI-treated

fertilizers leads to higher NO_3^- leaching losses compared to using UI alone. The addition of NI may change immobilization/mineralization dynamics due to the presence of both inhibitors, though the exact mechanisms are unclear and warrant further study.

Combined Inhibitor Effect on Yield

Combining inhibitors appears to provide limited or no benefit to crop yields compared to untreated fertilizers or fertilizers treated with inhibitors individually. In a meta-analysis including 27 studies, Abalos et al. (2014) reported an average yield increase of 10% with NBPT (UI), 6% with DCD and ~2% with DMPP (NIs), and ~7% with NBPT+DCD (UI+NI), with the only significant difference being that from NBPT and DMPP alone. Gioacchini et al. (2002) observed that the inclusion of UI or UI+NI with urea did not affect wheat grain yield, in spite of increased leaching losses when inhibitors were used. They attributed this effect due to a large portion of the N taken up by the plants being soil-derived. Sanz-Cobena et al. (2012) observed no corn yield differences from the use of UI or UI+NI over untreated urea in two years of a field study. On a two-year corn study receiving side-dress application of UAN, UAN+UI, and UAN+UI+NI at 130 kg N ha^{-1} rate, Woodley et al. (2018) found that no inhibitor treatment was able to significantly increase grain yield over untreated UAN, and attributed this lack of effect to small loss magnitude. In spite of large reductions in NH_3 losses from the use of UI+NI as compared to untreated fertilizer, Drury et al. (2017) did not observe significant grain yield differences between untreated fertilizer, fertilizer+UI and fertilizer+UI+NI in any given year of a two-year corn study, but noted that when averaged over years, fertilizer+UI+NI increased yield from 5 to 7% over untreated fertilizer. Overall, the use of UI or UI+NI produced similar yields to that from untreated fertilizer most of the time, and when a yield gain existed it was about 5 to 10% greater than fertilizer alone.

Combined Inhibitor Effect on NUE

Combining inhibitors has promoted higher, lower, and similar NUE compared to untreated fertilizer or fertilizers amended with a single inhibitor type. In the same meta-analysis study by Abalos et al. (2014), the authors reported that applying NBPT+DCD was the inhibitor combination that increased NUE the most (14.7% compared to no inhibitor), but that high variability precluded any significant differences between inhibitors alone or combined. Conversely, Gioacchini et al. (2002) reported on a significant decrease in fertilizer-derived N uptake by wheat when both UI+NI were used compared to UI-only. This effect was due to an increase in N mineralization from the use of inhibitors, and concurrent increased proportion of N uptake from this source. Sanz-Cobena et al. (2012) reported no differences in N uptake between UI+NI and UI-only, but both were higher than untreated urea in two years of corn field studies. Woodley et al. (2018), however, found no significant differences in N uptake between UAN, UAN+UI, and UAN+UI+NI in two years of corn field studies. Similar findings were reported by Drury et al. (2017), who concluded that neither fertilizer+UI nor fertilizer+UI+NI significantly improved corn N uptake compared to fertilizer alone in a 2-yr study including broadcast urea and injected UAN. The use of UI and UI+NI has demonstrated limited efficacy in increasing NUE, even under high N loss conditions like those observed by Drury et al. (2017). Nonetheless, the retention of N due to inhibitors may still benefit cropping systems in the long-term, as this N can be incorporated into soil organic matter (SOM) and conserved as N reserves (Cantarella et al., 2018).

Reactive Strategy – Crop Canopy Sensors

Properly managing fertilizer practices to achieve optimum yields without generating surplus N is difficult and greatly influenced by N rate decisions. To assist producers on N rate decisions, researchers have created algorithms based on the mass balance theory developed by Stanford (1973), that follows simple mathematical logic of needed inputs vs. generated outputs. This approach remains as the dominant corn N recommendation approach in 34 U.S. states today (Morris et al., 2018). Briefly, these algorithms calculate a pre-plant N rate by first estimating crop N needs as the product of expected yield (EY) multiplied by an efficiency coefficient (e.g. 1.2 bu lb⁻¹ N), and then deducting N credits based on field-specific information, such as expected N mineralized from SOM, soil residual NO₃⁻, manure application, previous crop (legume vs. non-legume), N from irrigation water, and others.

Historically, mass balance approaches have not considered soil-plant N resiliency (i.e. the capacity of soil-plant system to vary plant available N according to growing conditions). Furthermore, mass balance approaches seldom include economic considerations. One example of a mass balance approach that does consider both of these factors is the University of Nebraska-Lincoln corn N algorithm (Shapiro et al., 2008). In this algorithm, soil-plant N resiliency is accounted for by correcting the SOM contribution to N mineralization by considering EY the term (0.14 x EY x OM). This algorithm also incorporates a correction factor for price adjustment based on the cost of N and the price of corn.

Although the input-output approach of mass balance algorithms is easy to convey and has led to its widespread adoption, the approach uses only pre-existing information to predict crop N need. As a result, in-season conditions (i.e. weather) that add variability to crop N demand, soil N dynamics, and N fertilizer loss are not considered. Furthermore, while total N uptake (e.g.

from fertilizer, soil N mineralization, residual NO_3^- , etc.) is strongly correlated with grain yield in corn (Cassman et al., 2002; Nyiraneza et al., 2009), it has been shown that yield and economic optimum N fertilizer rate (EONR), which can be calculated only after the crop is harvested, are uncorrelated (Cassman et al., 2002; Nyiraneza et al., 2009).

Regardless of the method used to determine fertilizer N rate, the optimum rate needed to economically maximize yield varies in both space (i.e. across and within fields) and time (i.e. across and within years). As a result, applying a single N rate to an entire field at a single time point can create areas of under-fertilization (e.g. lower realized yield) and over-fertilization (e.g. lost input costs, greater environmental risk) (Raun et al., 2002; Mamo et al., 2003; Scharf et al., 2005). Given these limitations, researchers have been investigating a reactive approach to N management using crop canopy sensors during the growing season to estimate crop N status and fertilization needs that vary in space (Raun et al., 2002; Teal et al., 2006; Barker and Sawyer, 2010; Holland and Schepers, 2010; Solari et al., 2010; Scharf et al., 2011). Because crop growth integrates soil and weather effects on N nutrition from time of planting to time of sensing, in-season sensor use on crop canopy color can diagnose N status and N application rate.

A series of steps and decisions need to occur before crop canopy data can be translated into an N rate recommendation. Translation of canopy data depends on: i) the type of sensor and what platform carries it; ii) spectral bands available from the sensor; iii) vegetation indices that can be calculated from those bands; iv) the standardization procedure used to overcome confounding effects (e.g. planting date, hybrid); and v) selection of sensor-based N rate algorithm to generate a prescription.

Sensor Type

Sensor types can be classified according to different characteristics, such as their light source (active vs. passive), number of bands (multispectral vs. hyperspectral), and distance from the target (remote vs. proximal vs. contact). Active sensors are those that emit their own modulated light, making sensor performance theoretically independent of atmospheric conditions, such as cloud cover and time of day. Active sensors have been used “on-the-go”, capable of assessing crop N status and directing variable rate N (VRN) application on the same pass. Due to these advantages, commonly used algorithms for VRN recommendation have been developed for active sensors (Holland and Schepers, 2010; Solari et al., 2010). Because they emit their own light source, active sensors require a certain proximity to their target, and thus are mostly limited to ground-based platforms (e.g. tractor, application implement boom) and low-flying (0.5 to 1.5 m above canopy) unmanned aerial systems (Krienke et al., 2015). The most commonly studied active sensors are the Crop Circle family of sensors (e.g. ACS-430, ACS-470, RapidSCAN CS-45) (Holland Scientific, Lincoln, NE), and the GreenSeeker sensor (NTech Industries, Inc., Ukiah, CA).

Passive sensors, in contrast, rely on sunlight as the energy source and thus may be limited by atmospheric conditions like time of day and cloud cover (de Souza et al., 2010). Furthermore, the use of passive sensors to generate VRN application is a two-step process, where first the field is imaged, and only after data correction and processing can a prescription map be generated and fed into a variable rate applicator software. Historically, passive sensors have been mostly employed in agriculture via satellite or aircraft. Recently, unmanned-aerial systems (UAS) have become a popular platform for carrying passive sensors both in research and commercially. UAS-mounted passive sensors have the flexibility of sensing independently of field conditions

(e.g. wet soil). A variety of different passive sensors have been utilized in agriculture, including MicaSense RedEdge (MicaSense Inc., Seattle, WA, USA), and Parrot Sequoia (Parrot Inc., San Francisco, CA, USA).

Bands

Plants absorb most radiation in the visible light range (400 to 700 nm) for photosynthesis, and the relative amount absorbed depends on the concentration of different pigments in leaf material, such as chlorophyll a and b, carotenoids, and anthocyanins (Chappelle et al., 1984). Reflectance in the near-infrared (NIR) part of the spectrum (800 to 1100 nm) is related to crop biomass and leaf structure. As a result, gross plant N responses in leaf reflectance, leaf density, vegetation cover and biomass can be assessed with both visible and NIR wavelengths instead of relying on physical sampling and chemical analysis of plant N concentration (Fox et al., 2008).

Crop canopy sensors often have one to two bands in the visible range, and one band in the NIR range. In the visible range, sensors commonly have bands centered on the green (G, 500 to 600 nm) and red (R, 600 to 700 nm) regions. The R band is the most common in sensors due to its inclusion in the classic normalized difference vegetation index (NDVI, Eq [6]), which was the first vegetation index (VI) developed for vegetation monitoring based on satellite-mounted sensors (Rouse Jr et al., 1974).

However, reflectance in the R band saturates when leaf area index (LAI) values are > 2 (Gitelson et al., 1996; Viña et al., 2011) and when chlorophyll concentrations are as low as 3 to 5 $\mu\text{g cm}^{-2}$ (Gitelson and Merzlyak, 1997), values which are common for corn during the period when sensors would be utilized (e.g. after \sim V8 growth stage). Thus, the saturation of the R band renders it insensitive in differentiating between N deficient or N sufficient crop conditions (Blackmer and Schepers, 1994; Holland and Schepers, 2013).

Because of this limitation, other bands in the visible range have been used in place of the R band, including the G and red-edge (RE, 700 to 800 nm) bands. For example, Blackmer and Schepers (1994) found that reflectance in the G region was significantly more sensitive than reflectance in the R region in differentiating leaves from corn plants at tasseling that had received different levels of N fertilization (0, 40, 80, 120, and 160 kg N ha⁻¹). Similarly, Gitelson et al. (2003) demonstrated that reflectance in the G and RE bands was significantly more sensitive to increases in chlorophyll levels than those in the R and blue (400 to 600 nm) bands. Given that different bands contribute different plant-related information, individual bands are rarely used alone but instead combined in multiple different formulations (i.e. vegetation index).

Vegetation Index

Vegetation indices (VIs) are combinations of different single-band reflectance values. Their development allowed normalizing single-band reflectance measurements with respect to atmospheric conditions, crop structure, soil reflectance, sensor calibration, illumination angle, among others (Bannari et al., 1995). Moreover, VIs allow combining bands related to different plant biophysical characteristics. For example, NDVI (Rouse Jr et al., 1974; Eq [6]), one of the most common VIs used, combines both the R band (related to chlorophyll content) and NIR band (related to leaf structure) (Eq. [1.7]). This unitless index can range in value from -1.0 to 1.0, with very low values indicating bare soil and higher positive values correlated with greater canopy density or greenness. Saturation of the R band under higher biomass conditions promoted the study and development of other VIs that overcome this limitation. The R band can be replaced by the RE band in the NDVI formula to create the Normalized Difference Red Edge (NDRE), which is more sensitive to high-biomass crop conditions such as those observed in corn.

$$NDVI = \frac{(NIR-Red)}{(NIR+Red)} \quad [1.7]$$

Various VIs have been developed for assessing different vegetation parameters (Mulla, 2013). For example, the Visible Atmospherically Resistant Index [VARI, Gitelson et al. (2002)] was shown to behave linearly in a range of 10 to 100% vegetation fraction, while NDVI became unresponsive >65% vegetation fraction in corn and >50% in wheat. Another VI developed to address the saturation of R band at higher chlorophyll content was the green NDVI [GNDVI, Gitelson et al. (1996)], which uses the G band instead of the R band in the NDVI formula. The use of GNDVI can provide increased power to differentiate N status in high-chlorophyll situations over NDVI (Shanahan et al., 2001; Solari et al., 2008), especially when a sensor lacks a RE band but has a G band.

The assessment of N status can be confounded if other stresses are also present (e.g. drought, disease, other nutrient deficiencies). Given that limitation, the canopy chlorophyll content index (CCCI), calculated as the ratio between NDVI and NDRE, was developed to assess N and water deficiency independently (Barnes et al., 2000), but the ability of CCCI to make this differentiation is inconsistent (Fitzgerald et al., 2006; El-Shikha et al., 2008). In short, VIs combine crop-related information from multiple bands into a single value, but the differential effects of other non-N related variables on VIs complicate any generalizations of VI-based applications to different hybrids, growth stages, and locations (Schepers et al., 1992).

Reference

To account for some of the non-N related factors confounding VIs, different approaches based on an N-rich reference strip (NRS) have been developed. The NRS is an area of the field that receives a non-limiting N rate to assure total N-sufficiency at the time of sensing (Biggs et

al., 2002). The NRS then serves as the basis of comparison for the remainder of the field where N status is unknown. Two different approaches relate field VI and NRS VI. The first is the response index (RI, Eq. [1.8]), where the VI of the reference (e.g. NRS) is divided by the VI of the unknown-N area (Raun et al., 2002). The second is the sufficiency index (SI, Eq. [1.9]), which is the inverse of RI (Blackmer and Schepers, 1994). Both RI and SI normalize non-N related confounding effects and have widespread effectiveness indicating N-related crop responses. For example, Hawkins et al. (2007) analyzed 102 site-year studies of corn in Iowa with multiple N rates (0 to 270 kg N ha⁻¹) and over different soil and growing conditions. The authors used a SPAD (Soil-Plant Analyses Development) unit to measure plant chlorophyll at different growth stages, and reported that lower variability in NRS-normalized SPAD measurements improved the prediction of EONR independent of previous crop phase (i.e. corn or soybeans) and growth stage.

$$RI = \frac{VI_{Reference}}{VI_{Unknown}} \quad [1.8]$$

$$SI = \frac{VI_{Unknown}}{VI_{Reference}} \quad [1.9]$$

Both RI and SI approaches rely on proper NRS establishment such that the only difference between the reference and the rest of the field is their N sufficiency level. In other words, a valid NRS should be planted the same date, with the same hybrid, receiving the same fertility management (except for N), under a similar soil type, topography, etc. as the remainder of the field. If this is not attainable due to any of these aspects, each differing management group should have its own NRS.

Establishing a NRS can be inconvenient and even restricted in commercial sensor-derived N applications (Holland and Schepers, 2013). Given this limitation, Holland and Schepers (2013) proposed the use of a virtual reference (VR), defined as the 95th cumulative percentile of a histogram from a given VI data collected over a subfield area which best represents the range of crop N status levels in the entire field. The use of a VR solves a problem where high-N-induced sulfur deficiency may occur when high N rates are applied to create the NRS (Franzen et al., 2016). Regardless of which normalization approach (SI vs. RI) or what reference type is used, the last step in the process is to translate SI or RI into a recommended N rate through the use of an algorithm.

Recommendation Algorithm

After normalizing field VI by a reference VI, either SI or RI values serve as input to an algorithm to calculate an N rate recommendation that overcomes observed N stress. Available algorithms vary in number and type of inputs in translating sensor-derived plant nutrient deficiency status into a recommended N rate (Holland and Schepers, 2010; Scharf et al., 2011; Solie et al., 2012; Franzen et al., 2016). The algorithm developed by Holland and Schepers (2010) calculates an N rate based on a quadratic plateau response function between SI and N rate, and then uses a mass approach to deduct N credits. Input variables include optimum N rate, management zone scalar, different sources of N credits (i.e. previous crop, organic matter, irrigation water NO_3^- , manure application, fertilizer applied prior to sensing), SI, and delta SI (DSI, defined below) (Eq. [1.10]). Of these, the minimum required inputs to generate a VRN prescription are optimum N rate and SI.

$$N_{app} = (MZ_i \cdot N_{opt} - N_{credits}) \cdot \sqrt{\frac{(1-SI)}{\Delta SI}}, \text{ where} \quad [1.10]$$

MZ_i = management zone scalar (unitless)

N_{app} = calculated recommended side-dress N rate (kg ha^{-1})

N_{opt} = optimum N rate defined by producer (kg ha^{-1})

$N_{credits}$ = N credits (kg ha^{-1})

SI = sufficiency index (unitless)

ΔSI = difference between 1 and SI for an unfertilized area [SI(0)] (unitless).

Another popular algorithm used for sensor-based N rate recommendations was developed by Solie et al. (2012) for both wheat and corn, also referred to as the N fertilization optimization algorithm (NFOA). This algorithm assumes that yield potential and N response are independent, thus requiring each to be separately estimated (Raun et al., 2011). This nine-step algorithm includes defining the maximum yield potential (e.g. by producer); choosing a crop-specific potential yield curve (wheat, corn, or combined wheat-corn); calculating an NDVI-based RI; calculating the curve inflection and curvature based on NRS NDVI; calculating the N-unfertilized yield potential; calculating yield with additional N fertilizer; and finally, outputting a recommended N rate based on previous calculations.

Current commercially-available variable N rate systems use active sensors to detect crop N status, calculate a specified VI, use it to calculate a SI considering a reference VI, inputs the SI into an algorithm, calculates and applies to the field a given N application rate, which changes as the applicator moves through the field and senses new areas. The algorithms developed by Holland and Schepers (2010) and Solie et al. (2012) are the two most prominent in commercial use due to their inclusion into AgLeader OptRx and GreenSeeker systems, respectively.

The use of algorithms to translate sensor information into N rate is an important development for N site-specific management. However, this sensor-based approach relies on the efficiency and accuracy of N recommendations, and the extent to which these recommendations can reduce N input costs so as to increase producer profit margins.

Total N Rate, Grain Yield, and Use Efficiency

The adoption of sensor-based variable N rate application is generally low (<10%) and attributed to inconsistent production, environmental, and economical benefits (Scharf et al., 2011; Colaço and Bramley, 2018). Many studies have evaluated these aspects of sensor-based N management, with performance metrics reported as being positive, neutral, and negative, compared to a non-sensor based standard practice (Raun et al., 2002; Kitchen et al., 2010; Ma et al., 2010; Scharf et al., 2011; Colaço and Bramley, 2018).

In a recent review, Colaço and Bramley (2018) summarized 24 publications that included at least one standard non-sensor based N practice and a sensor-based N management option with the use of Crop Circle and/or GreenSeeker and that utilized either Holland-Schepers or NFOA as the implemented algorithm. The authors reported that, on average, sensor-based N approaches increased yield by 3.3%, decreased N rates by 26.7%, increased PFP_N by 41%, and increased profit by US\$ 30.40 ha⁻¹. While average outcomes were positive, individual observations per metric varied widely. For example, yield varied from -4 to 17.5% compared to the standard practice, with 25% of the studies reporting a negative yield outcome from the use of sensors. Furthermore, profit varied from -26 to 196 US\$ ha⁻¹, with 33% of the studies reporting a negative profit value from the use of sensors. Similarly, PFP_N varied from -18 to 554%, with 33% of the studies reporting a negative outcome from the use of sensor-based N management. Nitrogen rate

was the metric that improved most frequently and varied from -82 to 31% compared to the standard practice, with only 12% of studies reporting higher N rates in sensor-based approaches.

The use of crop canopy sensors for in-season N management is a promising tool to decrease N rates to levels that maximize yield and profit and minimize environmental losses. This level of N management fine-tuning is expected to be adopted first by producers that recognize the need for more sustainable N practices, and that are already relatively efficient with their current N management. However, the level of improvement from the use of sensor-based N management depends not only on the degree of field spatial variability, but also on the current management as a starting point. Thus, farmers that are already highly efficient in N management are likely the ones that will see the least relative benefit from implementing such technologies.

Summary

Once applied to the environment, N fertilizer is prone to different losses. Ammonia volatilization, NO_3^- leaching and N_2O emission can vary from 0 to 80% of the applied fertilizer, and their magnitude depends on various soil, management, and weather variables. Larger losses are observed when N supply in the soil greatly surpasses N demand, coupled with the fact that soil N balance varies spatially and temporally.

The mismatch between N supply and demand at a given moment and place may lead to increased N losses, which in turn can impact plant productivity and N use efficiency. Because N dynamics in the soil is greatly affected by weather, especially rainfall and temperature, management practices adopted to decrease losses and improve yield need to address the unpredictability of weather. For that, protective and reactive approaches have been studied as next-generation N management practices.

Protective approaches are used in order to protect N fertilizer from possible loss-conducive weather events, and thus are implemented before losses occur. Protective approaches include the use of stabilized fertilizers like those containing UI, NI, or both. The use of UI with broadcast urea-based fertilizers has been shown to decrease NH_3 losses by 50-75%, increase yield in 4-10%, and promote NUE ranging from -12 to 28% compared to untreated fertilizer. The use of NI has been shown to decrease NO_3^- leaching by 17%, decrease N_2O losses by 38-52%, promote yield ranging from -30 to 7%, and NUE ranging from -20 to 200% compared to untreated fertilizer. The combined use of UI+NI has been shown to decrease NH_3 losses to a lesser extent than UI alone, have little effect on N_2O emissions compared to NI-only, and promote higher NO_3^- leaching compared to fertilizer alone, while having an impact on yield that ranged from 0 to 7%, and NUE ranging from 0 to 15% compared to untreated fertilizer.

The lack of consistent response of stabilized fertilizers on yield and NUE has been attributed to many factors. Those include i) lack of yield response to N application (i.e. N was not the limiting factor); ii) large contribution of N from soil organic matter mineralization or other sources including excessively high N rates; iii) conditions not conducive to loss; iv) N positional unavailability in relation to root active uptake region; v) negative effect of inhibitor on crop growth.

Reactive approaches are used in-season in order to react to a nutritional stress early on its development. This type of approach allows for soil, management, and especially weather, to affect the crop nutritional condition, assesses the level of stress through crop color, and reacts to it by recommending a fertilizer rate to mitigate the stress. Reactive approaches are more complex as they require some type of crop vigor measurement via a sensor (proximal or remote), formulation of a VI, calculation of an SI, which is then used as input into an algorithm to

translate it to a fertilizer rate. The use of sensor-based N management has been shown to increase yield in 3.3%, decrease total N application rates in 27%, increase N use efficiency as measured by PFP_N by 41%, and increase profit in US\$30, on average, compared to a non-sensor based management approach. Nonetheless, there were occasions where sensor-based management failed compared to conventional, non-sensor management.

Although protective and reactive approaches have been demonstrated to decrease N losses, improve N use efficiency, and maintain or increase yield, negative results have also been observed. The success of these tools to manage N more sustainably largely depends on whether they are the correct tool for a given situation. Therefore, while the current knowledge can assist in selecting the correct tool for a specific site, future studies will aid in elucidating different conditions that may or may not benefit from the use of one or more of these technologies. The following chapters of this dissertation will summarize studies evaluating protective and reactive approaches for irrigated corn N management in Central Nebraska.

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Chapter 2 - Urease Inhibitor Decreases Ammonia Loss without Affecting Corn Grain Yield

Introduction

Once applied to soil, N fertilizer is dynamic with high risk of loss through different pathways including ammonia (NH_3) volatilization (Pan et al., 2016), nitrate (NO_3^-) leaching (Quemada et al., 2013), and denitrification (i.e. emission as nitrous oxide, N_2O) (Shcherbak et al., 2014). Losses via NH_3 volatilization are of particular interest in Nebraska, where all three of the most utilized N fertilizer sources [urea-ammonium nitrate (UAN), anhydrous NH_3 (AA) and urea] contain or convert to ammonium (NH_4^+) and are susceptible to volatilization. In-field NH_3 losses are affected by multiple factors and thus are highly variable, ranging from 0 to ~60% of applied fertilizer (Terman, 1980; McInnes et al., 1986a; b; Harrison and Webb, 2001; Pan et al., 2016; Silva et al., 2017), with an average loss of 18% (Pan et al., 2016).

Urea is hydrolyzed to NH_4^+ and bicarbonate through the urease enzyme, which is found both in the soil and on crop residue (McInnes et al., 1986). During hydrolysis, a proton is consumed causing a temporary increase in pH around the urea granule. This increase in pH can shift the equilibrium between NH_3 and NH_4^+ towards the gaseous, loss-prone NH_3 form and enhance volatilization loss.

Producers can decrease NH_3 loss potential and magnitude by exerting different levels of control over fertilizer, soil, and weather variables. Among the fertilizer-related variables, N placement and source are most important as they relate to NH_3 loss potential, followed by N rate and timing (Ma et al., 2010; Pan et al., 2016; Silva et al., 2017). The application of N fertilizer below the surface is one of the simplest ways to avoid NH_3 losses from urea-containing

fertilizers. This practice places fertilizer in contact with soil colloids and allows for NH_4^+ to be removed from solution by cation exchange capacity (CEC) sites (Sommer et al., 2004). For example, Pan et al. (2016) observed a 55% reduction in NH_3 volatilization when various N fertilizers were deep-placed compared to surface applied, and Rochette et al. (2013) observed negligible NH_3 losses when urea was placed 7.5 cm below the soil surface.

Fertilizer source choice plays an important role, especially if surface-applied. For example, urea, UAN and ammonium nitrate (AN) have a high, intermediate and low potential for NH_3 loss if surface applied, respectively (Keller and Mengel, 1986; Hargrove, 1988; San Francisco et al., 2011). The rapid hydrolysis of urea-based fertilizers increases soil NH_4^+ availability for volatilization and promotes volatilization potential by increasing pH in the vicinity of the fertilizer (Sommer et al., 2004). In contrast, AN transformations do not lead to immediate increases in soil pH, limiting risk losses to when initial soil pH is alkaline (Sommer et al., 2004).

Another way of decreasing the magnitude and potential NH_3 loss from surface-applied urea-containing fertilizers is to impede urea hydrolysis using a urease inhibitor (UI) (Silva et al., 2017; Sunderlage and Cook, 2018; Cantarella et al., 2018). The most common, commercially available UI is N-(n-butyl) thiophosphoric triamide (NBPT, Koch Agronomic Services, Wichita). NBPT bonds with the urease active site to reduce the rate of urea hydrolysis, thus decreasing the potential for both pH to increase and for NH_4^+ to build up in solution. Delaying these reactions also allows more time for rainfall to occur and incorporate fertilizer into the soil where fertilizer is better protected from volatilization loss. The use of NBPT has been demonstrated to both delay the peak NH_3 loss from surface-applied urea and lower overall loss rates compared to untreated fertilizer (Soares et al., 2012; Silva et al., 2017), such that

cumulative NH_3 loss reductions range from 15 to 75% (San Francisco et al., 2011; Silva et al., 2017; Sunderlage and Cook, 2018; Cantarella et al., 2018).

In 2015, a new compound developed by BASF, N-(n-propyl) thiophosphoric triamide (NPPT), was introduced in the UI market and is sold in a mixture with NBPT (5.6% NPPT, 16.9% NBPT) under the trade name Limus® (BASF Crop Protection). Given its recent commercial availability, published evaluations of Limus are limited (Li et al., 2015; Sunderlage and Cook, 2018), and no published studies were found where Limus was compared to the industry standard NBPT. Sunderlage and Cook (2018) conducted a laboratory study comparing NH_3 losses from untreated urea and urea+Limus on 79 soils from the U.S., and found that urea+Limus reduced 75% of the NH_3 losses observed with urea alone but that product effectiveness decreased in acidic soil conditions due to UI degradation. The effect of soil pH on UI efficiency is not unique to Limus and has been observed for NBPT-only products (San Francisco et al., 2011). Additional studies are needed to understand how Limus compares to NBPT-only products in both NH_3 loss mitigation and its effect on grain yield, and to assess its efficacy under different soil and weather conditions.

Fertilizer products containing both UI and nitrification inhibitors (NI) have been suggested as an option to reducing all three major N losses (Xu et al., 2000), and further protecting yield and improving NUE (Zaman et al., 2008). However, various studies have reported on increased NH_3 losses from UI+NI application compared to UI only (Gioacchini et al., 2002; Soares et al., 2012; Abalos et al., 2014; Pan et al., 2016). The increase in NH_3 volatilization from NI use is attributed to higher soil pH, higher NH_4^+ and lower NO_3^- as a result of nitrification inhibition (Soares et al., 2012).

Different approaches have been developed to measure field NH_3 losses, including mass balance, tracer techniques, enclosures (static and dynamic, semi-open and closed chambers), micrometeorological methods, gradient diffusion methods, eddy correlation, relaxed eddy accumulation, and modeling (McGinn and Janzen, 1998; Sommer et al., 2004). Among these, enclosures are one of the most used methods (Nõmmik, 1973; Schlegel et al., 1986; Grant et al., 1996; Rawluk et al., 2001; Gioacchini et al., 2002; Ma et al., 2010; Jantalia et al., 2012) due to their level of portability and overall simplicity, applicability to multiple experimental units in small plot studies, low cost, and high sensitivity (McGinn and Janzen, 1998; Sommer et al., 2004; Smith et al., 2007).

The amount of measured NH_3 volatilized is dependent upon the measurement methodology. Enclosures may alter the measured environment compared to plot-level conditions, especially regarding gas concentration gradient, air movement, water flux, and temperature regime (Marshall and Debell, 1980; McInnes et al., 1986; Schlegel et al., 1986; Martha et al., 2004; Smith et al., 2007). Closed static chambers generally recover less volatilized NH_3 (22 to 96%) compared to semi-open chambers (Marshall and Debell, 1980; Wang et al., 2004), wind tunnel, and micrometeorological approaches (Smith et al., 2007).

Although UIs can effectively decrease NH_3 losses from urea application, crop yield and NUE responses to UI applications do not match the quantities of N conserved (Fox and Piekielek, 1993; Gioacchini et al., 2002; Abalos et al., 2014; Silva et al., 2017; Cantarella et al., 2018). Compared to untreated urea, positive yield responses under urea+NBPT range from only 1.8% to 12% across multiple crops (Abalos et al., 2014; Cantarella et al., 2018). In addition, crop NUE variability is highest in UIs compared to a variety of inhibitor treatments (i.e. UI, nitrification inhibitors (NI), UI+NI), with confidence intervals ranging from ~5 to 25% compared

to untreated fertilizer (Abalos et al., 2014). The lack of larger yield and NUE improvements from NBPT use has been attributed to crop N demand being met through other N sources like soil N mineralization (Cantarella et al., 2018), factors other than N being limiting to crop growth and yield (Silva et al., 2017), and studies evaluating UI at N rates close to the required amount for optimum grain yield (Rose et al., 2018).

We hypothesize that i) the use of an UI with pre-plant broadcast applied fertilizer will decrease NH_3 volatilization compared to untreated fertilizer; ii) Limus will promote NH_3 loss reductions similar to other NBPT-only products; iii) the use of UI+NI will promote NH_3 loss reductions that are intermediate between fertilizer+UI and untreated fertilizer; and that iv) the use of UI and UI+NI will promote greater grain yield than untreated fertilizer if weather conditions are conducive to N losses, such as no rainfall during the first 3-5 days after fertilizer application. The objectives of this study were to i) compare different UI and UI+NI products on how they affect sealed-chamber measured NH_3 volatilization losses from surface-applied UAN, ii) compare NH_3 volatilization losses of Limus at different rates to other inhibitors in reducing NH_3 loss, and iii) assess the impact of different UI and UI+NI products on corn growing season vigor and grain yield.

Material and Methods

This project was comprised of experiments conducted from 2014 through 2017 at two sites for a total of five studies (Table 2.1). The soils were classified as Crete silt loam (fine, smectitic, mesic Pachic Udertic Argiustolls) at SCAL14 and SCAL15, Novina sandy loam (coarse-loamy, mixed, superactive, mesic Fluvaquentic Haplustolls) at CC15, and Hastings silt loam (fine, smectitic, mesic Udic Argiustolls) at SCAL16 and SCAL17.

Table 2.1. Characterization of each site-year study.

Study	Site	Year	Soil Type	Soil Properties (0-20 cm) [†]					Planting Date	N App. Date
				pH	OM (%)	CEC (me 100g ⁻¹)	K (ppm)	P-M3 (ppm)		
SCAL14	SCAL	2014	Silt Loam	-	-	-	-	-	5-May	7-May
CC15	Central City	2015	Loamy Sand	7.2	1.1	7	104	12	15-Apr	22-Apr
SCAL15	SCAL	2015	Silt Loam	6.6	3.6	14	458	33	24-Apr	30-Apr
SCAL16	SCAL	2016	Silt Loam	7.0	3.1	22	406	20	12-May	18-May
SCAL17	SCAL	2017	Silt Loam	7.6	3.5	14	348	25	8-May	22-May

[†]pH (1:1 soil:water), OM = organic matter, CEC = cation exchange capacity, K = potassium, P-M3=Mehlich-3 phosphorus.

The treatment design at each study was one-way with different combinations of N rate, inhibitor type (UI alone, UI+NI) and inhibitor rate. Different inhibitor types and rates were used depending on the study (Table 2.2). The inhibitors used were Agrotain Ultra (AgU, Koch Agronomic Services, LLC, Wichita, KS), Agrotain Plus (AgP, Koch Agronomic Services, LLC, Wichita, KS), Limus (BASF Corp., Research Triangle Park, NC), DMP (3, 4-dimethylpyrazole phosphate) nitrification inhibitor (BASF Corp., Research Triangle Park, NC), and NutriSphere-N (NS, Verdesian, Cary, NC). AgU contains the UI N-(n-butyl) thiophosphoric triamide (NBPT, 26.7%); AgP contains both a UI (NBPT, 1-5%) and an NI (dicyandiamide, DCD, 35-55%); Limus contains two types of UI compounds (NBPT, 16.9%, and N-(n-propyl) thiophosphoric triamide, NPPT, 5.6%); NS is a polymer-based product containing maleic and itaconic acid (40%) and purportedly inhibits both nitrification and urea hydrolysis by complexing soil copper and nickel, respectively (mechanisms have not been demonstrated in refereed literature). All inhibitors were applied at manufacturer recommended rates, except for Limus, which was applied at multiple rates including the manufacturer recommended rate. Inhibitor application rates were 1.5 L Mg⁻¹ UAN for AgU, 7 kg Mg⁻¹ UAN for AgP, variable rates for Limus (from 0.8 to 2.1 L Mg⁻¹ UAN; Table 2.2), 0.7 L ha⁻¹ for DMP, and 0.5% v/v for NS. When not explicitly noted, the Limus rate used was the label-recommended rate of 1.5 L Mg⁻¹ UAN.

Table 2.2. Description of applied treatments, sensor information (type and timing), response to N fertilizer, and agronomic optimum N rate (AONR, in kg ha⁻¹) for each study.

Study	Treatments*	Sensor	Sensor timing	N-responsive	AONR (kg ha ⁻¹)
SCAL14	0N , 136N, 136N+AgU, 136N+L-0.8, 136N+L-1.2, 136N+L-1.5, 182N, 182N+AgU , 182N+L-0.8 , 182N+L-1.2 , 182N+L-1.5 , 182N+L-2.2 , 226N	SPAD	V10, VT	Yes	208
CC15	0N , 96N, 96N+L, 130N, 130N+AgP , 130N+AgU , 130N+L , 161N, 161N+L	RapidScan	V6, V12, VT, R4	No**	146
SCAL15	0N, 96N, 96N+L, 96N+L+DMP, 130N, 130N+AgP, 130N+AgU, 130N+L, 130N+L+DMP, 161N	RapidScan	V6, V13, VT, R4	No	0
SCAL16	0N , 173N, 173N+AgP , 173N+AgU , 173N+L-0.5 , 173N+L-1 , 173N+L-1.5 , 173N+NS , 215N, 215N+L-1.5	RapidScan	V6, V7, V9, V12, V16, VT, R3, R5	Yes	160
SCAL17	0N , 45N, 90N, 133N, 133N+AgP , 133N+AgU , 133N+L-0.5 , 133N+L-1 , 133N+L-1.5 , 178N, 178N+L-1.5 , 268N	RapidScan	V7, V9, V12, V16, R1, R3, R4	Yes	173

*Only treatment names in bold were measured for ammonia volatilization.

**Yield response to N fertilizer at CC15 was marginally significant (p=0.077).

AONR = agronomic optimum N rate.

Treatment labels include information on N rate (0, 45, 90, 96, 130, 133, 136, 161, 173, 178, 182, 215, 226, 268 kg N ha⁻¹), inhibitor type (none, AgU=Agrotain Ultra, AgP=Agrotain Plus, L=Limus, DMP), and Limus rate (0.5, 0.8, 1, 1.2, 1.5, 2.2 L Mg⁻¹ fertilizer).

Each study was conducted as a randomized-complete block design with four replicates.

Corn was planted at a target population of 84,000 plants ha⁻¹ with a row spacing of 0.76 m (dates in Table 2.1). Plots were 20-24 m long and 3 m wide, comprising four rows. Corn population at harvest varied from 80,000 to 82,000 plants ha⁻¹ except for SCAL14, which had a windstorm on June 14th causing plant breakage and reducing harvest population to 52,000 plants ha⁻¹. All studies were irrigated fields with a central pivot system. Irrigation frequency was determined by soil moisture balance monitoring with the use of Watermark matric potential sensors for all studies but CC15, where irrigation frequency was determined by the cooperator based on his experience. Irrigation single-event volume ranged from 25 to 40 mm.

The N source utilized for all studies was UAN solution (32% N) surface broadcast after planting and before emergence through a boom attached to a tractor. Treatment mixtures were performed directly in the tractor mixing tank connected to the boom, at volumes that matched both N and inhibitor application rates. Fertilizer N rates were determined by first calculating the full rate through the University of Nebraska-Lincoln corn N algorithm (Shapiro et al., 2008), which considers yield goal and N credits from organic matter and soil residual nitrate. Thereafter, other N rates were selected as a percentage (from 0 to 200%) of the full N rate, with different levels over different studies. Weather data was collected from automated weather station installed in proximity to the study area.

Volatilization losses of NH₃ were assessed only at specific treatment levels (Table 2.2, bold text). Losses of NH₃ were measured by using sealed polyvinyl chloride chambers with 0.55 m height and 0.1 m inner radius. Two weeks prior to fertilizer application, NH₃ traps were

prepared by syringe-applying 35 mL of phosphoric acid-glycerol solution (40 mL glycerol, 50 mL concentration H_3PO_4 acid, 910 mL deionized water) to foam disks (0.03 m height, 0.1 m radius), which were thoroughly kneaded to incorporate solution within the foam. Each foam trap was then placed inside a Ziploc bag, sealed and stored at 4°C until deployed in the field. On the same day following fertilizer application, one chamber was installed per plot in the mid row position between rows 2 and 3 by driving the chamber base ~ 10 cm into the soil. The chambers were equipped with four equally-spaced 3-cm long screws on the inner walls to suspend the foam trap at ~ 0.4 m above the soil surface. After chamber installation, one foam trap was deployed per chamber, then the chamber top sealed by covering it with impermeable plastic held in place by a 2-cm wide rubber band to minimize water exchange between the enclosed chamber headspace and the external atmosphere. This chamber design is similar to that employed by Marshall and Debell (1980), except the current study did not have an umbrella protecting from rainfall.

Ammonia volatilization losses generally peak at two to three days after fertilizer application (DAA), with 75% of cumulative losses occurring up until six and 11 DAA for untreated and NBPT-treated urea, respectively (Silva et al., 2017). For this study, foam traps were exchanged at 5, 10, 15, 20, and 30 DAA, with eventual departure of ± 1 day, comprising the main period of NH_3 losses. Used foam traps were sealed individually in Ziploc bags and stored at 4°C until analysis for NH_4^+ concentration. Losses of NH_3 were estimated by extracting the phosphoric acid-glycerol solution from the foam trap followed by analysis for NH_4^+ . Total NH_4^+ mass in solution was then standardized to the chamber area and summed over all sampling dates to calculate cumulative NH_3 volatilized ($\text{kg N ha}^{-1} 30 \text{ d}^{-1}$).

Crop vigor was assessed at different crop stages using a handheld sensor. For SCAL14, the sensor utilized was a SPAD meter (Konica Minolta, Japan), whereas all other studies were sensed with the active sensor RapidScan CS-45 (Holland Scientific, Lincoln, NE, USA). The SPAD meter is a contact sensor that measures light transmittance through an area of 6 mm² of plant leaf in the spectral regions of 650 and 940 nm. SPAD measurements were taken from the upper-most fully expanded leaf before tasseling (V10) and from the ear-leaf after tasseling (VT), midway between the leaf tip and base and also between leaf margin and midrib, from 20 plants in the middle rows. SPAD measurements were internally averaged and one value was recorded per plot.

The RapidScan CS-45 is an active handheld sensor equipped with a modulated light source and three photodetector measurement channels at 670, 730 and 780 nm. The RapidScan was oriented in the nadir position and approximately 0.6 m above the crop canopy. The two central rows of each plot were scanned individually, producing one average value from each measurement channel per row. RapidScan readings were taken directly over the corn row. Values generated for each row were averaged to create one value for each wavelength per plot. Active sensor bands of red-edge and near infrared were used to calculate the normalized difference red-edge (NDRE) index.

Both SPAD and NDRE values were used to calculate a sufficiency index (SI), obtained by dividing the sensor value of each plot by a virtual reference sensor value. Virtual reference sensor values were determined for each study-replicate-growth stage combination as the 95th percentile of the sensor measurement (SPAD or NDRE) histogram including data from all N treatments (Holland and Schepers, 2013). The use of an SI provides crop N status information

that is normalized and independent of crop growth stage in comparison to non-normalized sensor measurements (SPAD or NDRE).

Corn grain yield was determined by combine-harvesting the middle two rows of each plot and correcting grain moisture content to 155 g kg⁻¹ moisture content. The agronomic optimum N rate (AONR, kg N ha⁻¹) for each study was estimated by regressing N rate against grain yield data using linear, linear-plateau, quadratic, and quadratic-plateau models. The model with the lowest Akaike information criterion was chosen for AONR calculation.

All statistical analyses were conducted in R (R Core Team, 2017). The data were analyzed by performing study-specific ANOVAs for each of the response variables cumulative ammonia volatilization, SI, and grain yield. Studies were analyzed separately because i) UI effect on N loss, N nutrition, and grain yield is site- and year-specific, and ii) treatment design was not consistent. Analysis of variance for the fixed-effect sources and further pairwise comparisons (Fisher's least-significant differences) were deemed significant at $\alpha = 0.05$. Cumulative ammonia volatilization and SI were analyzed by using the function *gls* from the *nlme* package (Pinheiro et al., 2017) with block, treatment, DAA and treatment \times DAA as fixed effects and a linear spatial error correlation structure to account for the unevenly-spaced repeated measure nature of the response variables. Grain yield was analyzed by using the *lm* function from the *stats* package (R Core Team, 2017) with block and treatment as fixed effects. Model assumptions were visually assessed by constructing fitted vs. residual, residual quantile-quantile and residual histogram plots. All models residuals satisfactorily met the assumptions.

Results

In order to address the objectives of this study, water input (precipitation and irrigation) and air temperature were monitored throughout the growing season (April-September) and were

used to interpret the effect of UI and UI+NI on plot-level NH_3 losses. Sealed-chamber measured NH_3 losses were monitored from the date of fertilizer application through 30 days thereafter. Measured NH_3 volatilization were affected by the chamber environment and may not represent the plot-level NH_3 loss, and thus cannot be directly related to plot-observed grain yield. Corn vigor was assessed throughout the growing season by the use of both SPAD and RapidScan. This data was used to determine if stressing factors such as N affected crop color, and whether the use of inhibitors mitigated N stress magnitude.

Weather

Weather data for each study is shown on Figure 2.1, including growing season (April through September) monthly cumulative rainfall, irrigation, and total water input; and average air temperature; and average daily temperature and rainfall in a window of -5 to +10 days surrounding N fertilizer application timing. Average growing season total precipitation ranged from 476 to 578 mm, and average growing season total irrigation ranged from 120 to 318 mm, with an average growing season total water input that ranged from 661 to 798 mm. Average growing season air temperature was generally higher at CC15 and SCAL15. Average daily air temperature ranged from 8 to 22°C over the 15-d window surrounding N application. Daily precipitation patterns varied over different studies during the 15-d window surrounding N application. During the period preceding N application, CC15, SCAL16, and SCAL17 received ~4 to 40 mm, while SCAL14 and SCAL15 did not receive precipitation. During the 10-day period after N application, all studies received some precipitation volume at different frequencies. While SCAL14 and CC15 received a total of 72 and 21 mm within five DAA, respectively, SCAL15, SCAL16, and SCAL17 only received significant amounts of rain after five DAA, totaling 91, 59 and 5 mm, respectively during the 5 to 10 DAA period.

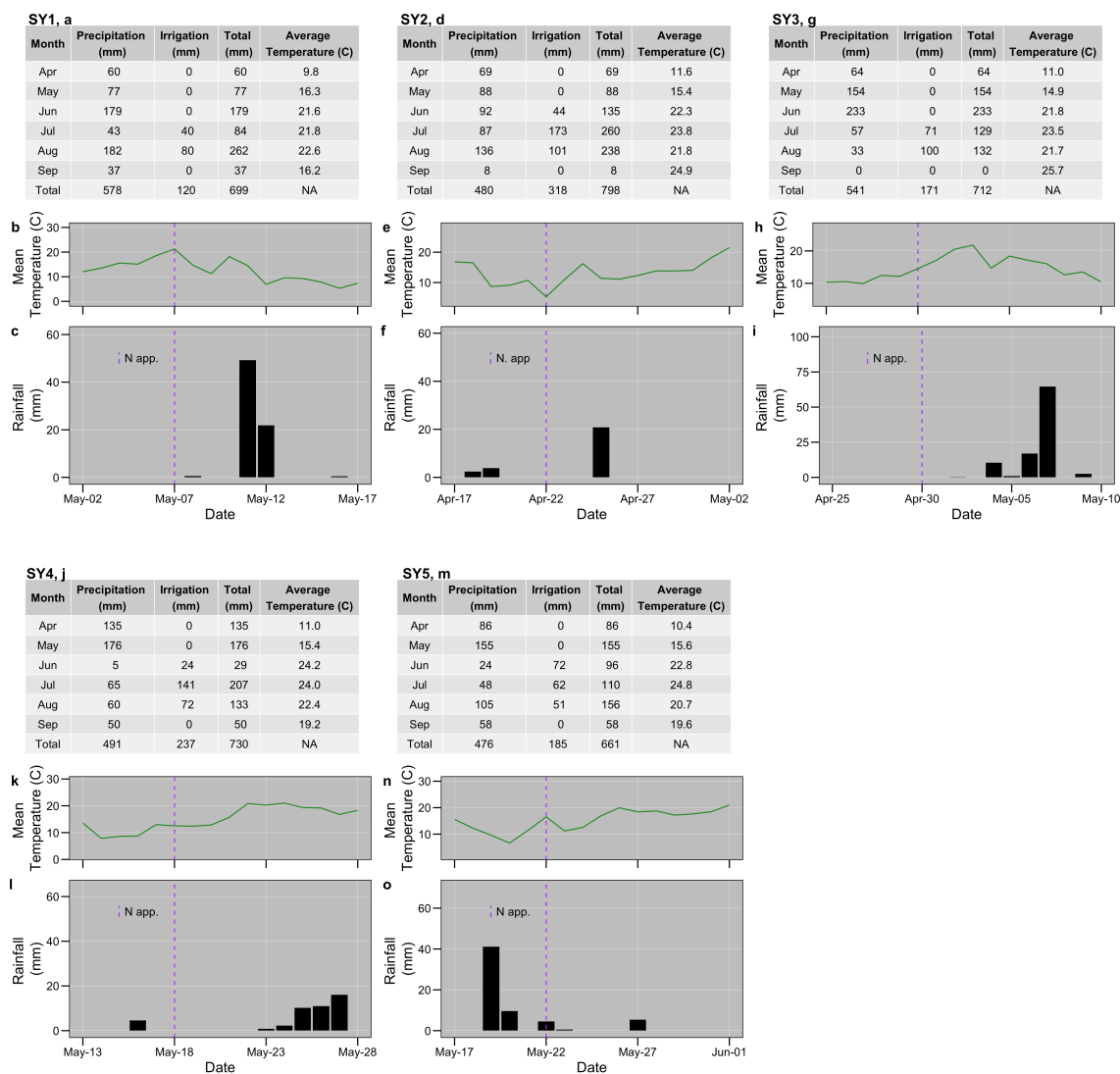


Figure 2.1. Growing season weather summary for each study. Tables include cumulative precipitation, irrigation, and total water input (mm), and mean air temperature (°C). Figures show mean air temperature (°C) and cumulative rainfall (mm) for a window of -5 to +10 days after N application (purple dashed line = N application date).

Ammonia Volatilization

Although NH_3 loss comparisons among treatments in this study are valid, they do not represent the loss conditions to which the entire plot area was subjected. This limitation is an artifact of the methodology employed, where NH_3 losses were measured from a sealed chamber

that did not allow for moisture exchange, wind movement and likely had a different daily temperature pattern than the exterior environment. Therefore, NH_3 volatilization losses measured on these studies should be interpreted with caution especially in regard to effects on final grain yield.

Thirty-day NH_3 volatilization after N fertilizer application was assessed at all studies except for SCAL15, and for a subset of treatments (Table 2.2). Over all studies, measured NH_3 volatilization losses ranged from 0 to 26 $\text{kg NH}_3\text{-N ha}^{-1}$. The maximum NH_3 loss observed varied from 16 to 26 $\text{kg NH}_3\text{-N ha}^{-1}$ and represented 10 to 15% of total fertilizer N. The use of inhibitors decreased NH_3 losses from 21 to 62% compared to untreated fertilizer. Even though a UI only protects the urea portion of UAN (44% of total N), relative losses will be expressed as percent of total fertilizer N in order to facilitate comparison with other studies that used different N sources.

At SCAL14, cumulative NH_3 volatilization was affected by N rate and inhibitor addition (Figure 2.2). Background soil NH_3 loss (0N) was 2 $\text{kg NH}_3\text{-N ha}^{-1}$. Applying fertilizer increased losses (19 $\text{kg NH}_3\text{-N ha}^{-1}$), with no differences between untreated 182N or 182N treated with AgU, or L-2.2. Losses from these treatments were ~10% of total applied fertilizer. Ammonia losses in the L-0.8 and L-1.5 treatments (15 $\text{kg NH}_3\text{-N ha}^{-1}$) were lower by 21%. Intermediate losses in 182N-L-1.2 did not differ from any other treatment. All UI treatments showed some reduction in NH_3 loss soon after N application (through 15 DAA), but treatments that significantly reduced volatilization did so throughout the 30-d measurement period.

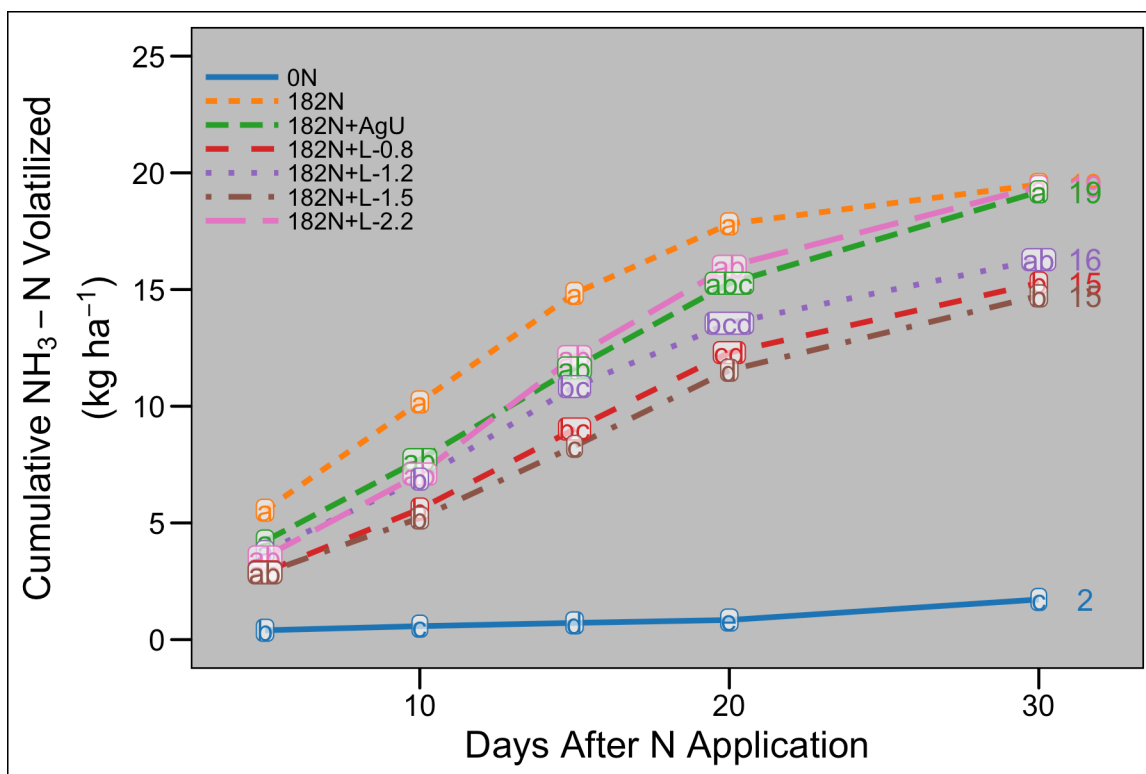


Figure 2.2. Mean cumulative ammonia volatilization (kg ha^{-1}) from 0 to 30 days after N application (DAA) at SCAL14. Treatment means within a date sharing the same letter are not statistically different at $\alpha=0.05$. Numbers on the right are the cumulative ammonia losses at 30 DAA. AgU = Agrotain Ultra; L = Limus at 0.8, 1.2, 1.5 and 2.2 L Mg^{-1} UAN rates.

At CC15, cumulative NH_3 volatilization was affected by N rate and inhibitor addition (Figure 2.3). Background soil NH_3 loss (0N) was $1 \text{ kg NH}_3\text{-N ha}^{-1}$. Total ammonia losses increased with fertilizer rate and were highest for untreated fertilizer (18 and $23 \text{ kg NH}_3\text{-N ha}^{-1}$ for 130N and 161N, respectively), with respective losses of 14 and 10% of applied N. Relative to controls, adding Limus reduced NH_3 losses by 30 to 33% at both 130N and 161N rates (12 and $16 \text{ kg NH}_3\text{-N ha}^{-1}$, respectively). At the 130N level, total NH_3 losses were greatest for UI-only amendments (AgU, Limus; 10, $12 \text{ kg NH}_3\text{-N ha}^{-1}$, respectively), and to a lesser extent for AgP (UI+NI; $15 \text{ kg NH}_3\text{-N ha}^{-1}$), which did not differ from control. Ammonia loss reductions

corresponded to 44%, 33%, and 17% of untreated control, respectively. Inhibition activity was greatest for all products for 10 to 15 DAA, then persisted until the end of the 30-d period.

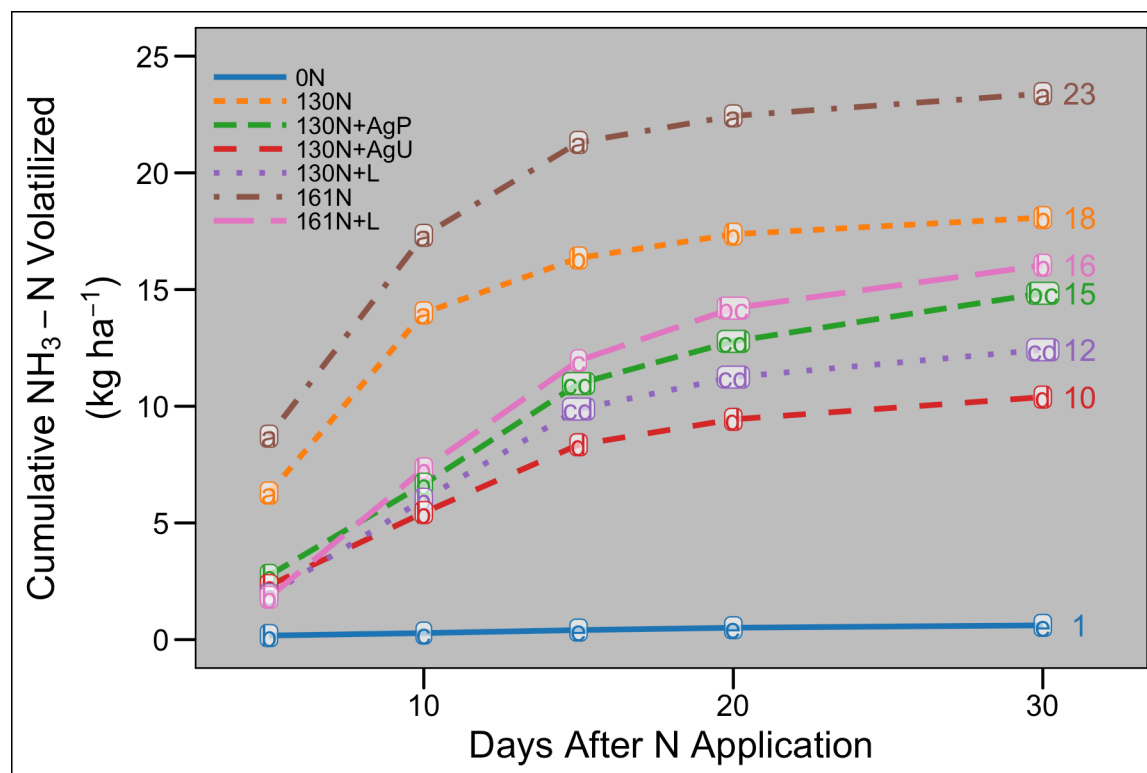


Figure 2.3. Mean cumulative ammonia volatilization (kg ha^{-1}) from 0 to 30 days after N application (DAA) at CC15. Treatment means within a date sharing the same letter are not statistically different at $\alpha=0.05$. Numbers on the right are the cumulative ammonia losses at 30 DAA. AgP = Agrotain Plus; AgU = Agrotain Ultra; L = Limus.

At SCAL16, cumulative NH_3 volatilization was affected by N rate and inhibitor addition (Figure 2.4). Background soil NH_3 loss (0N) was $0.23 \text{ kg NH}_3\text{-N ha}^{-1}$ over the 29-d sampling period. Untreated fertilizer (173N) showed the highest total losses ($26 \text{ kg NH}_3\text{-N ha}^{-1}$, 15% of applied fertilizer), with no effect of Limus at rates below the manufacturer recommendation (L-0.5, L-1), ranging from 24 to 21 $\text{kg NH}_3\text{-N ha}^{-1}$. All other inhibitors (AgP, AgU, L-1.5, NS) decreased NH_3 losses to 10 to 13 $\text{kg NH}_3\text{-N ha}^{-1}$, representing 50 to 62% loss reduction

compared to untreated control. Inhibition activity for effective products was established by 15 DAA, and then increased to the end of the 29-d sampling period.

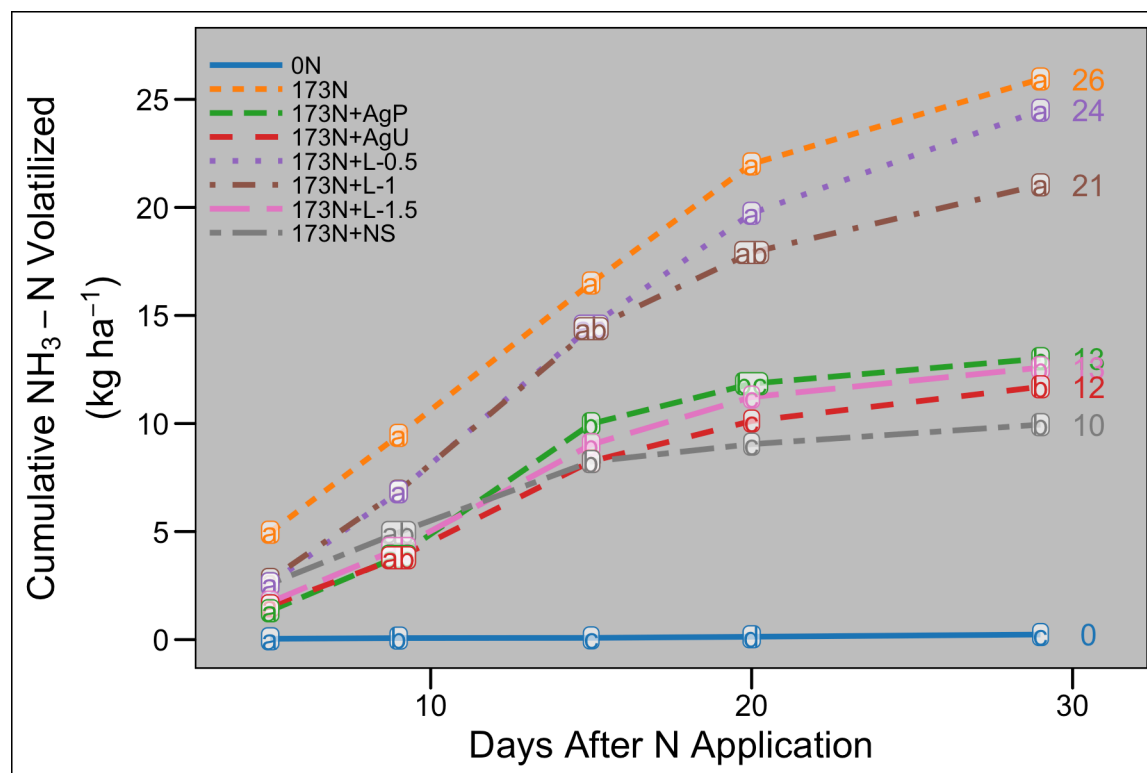


Figure 2.4. Mean cumulative ammonia volatilization (kg ha^{-1}) from 0 to 29 days after N application (DAA) at SCAL16. Treatment means within a date sharing the same letter are not statistically different at $\alpha=0.05$. Numbers on the right are the cumulative ammonia losses at 30 DAA. AgP = Agrotain Plus; AgU = Agrotain Ultra; L = Limus at 0.5, 1 and 1.5 L Mg^{-1} UAN rates, NS = NutriSphere-N.

At SCAL17, cumulative NH_3 volatilization was affected by the addition of N and inhibitors (Figure 2.5). Total background soil NH_3 loss (0N) was $0.24 \text{ kg NH}_3\text{-N ha}^{-1}$ by the end of the 31-day sampling period. Total ammonia losses were greatest in untreated N regardless of N rate. Although losses did not differ between untreated N rates (16 and $15 \text{ kg NH}_3\text{-N ha}^{-1}$ for 133N and 178N, respectively), respective proportional losses of applied N were concomitantly larger at the lower rate (12%, 8%). At 133N, reductions in NH_3 losses increased with Limus-amendment rate (8 to $12 \text{ kg NH}_3\text{-N ha}^{-1}$), equivalent to 25 to 50% reduction in N loss relative to

untreated controls. There was no effect of Limus at the higher 178N rate. At the 133N rate, a reduction in total NH_3 losses occurred for AgP (UI+NI; 11 $\text{kg NH}_3\text{-N ha}^{-1}$) to a similar extent as Limus amendments, but no reductions occurred using AgU (UI; 14 $\text{kg NH}_3\text{-N ha}^{-1}$). For effective products, inhibitor action was established by 10 DAA and continued to the end of the 31-d sampling period.

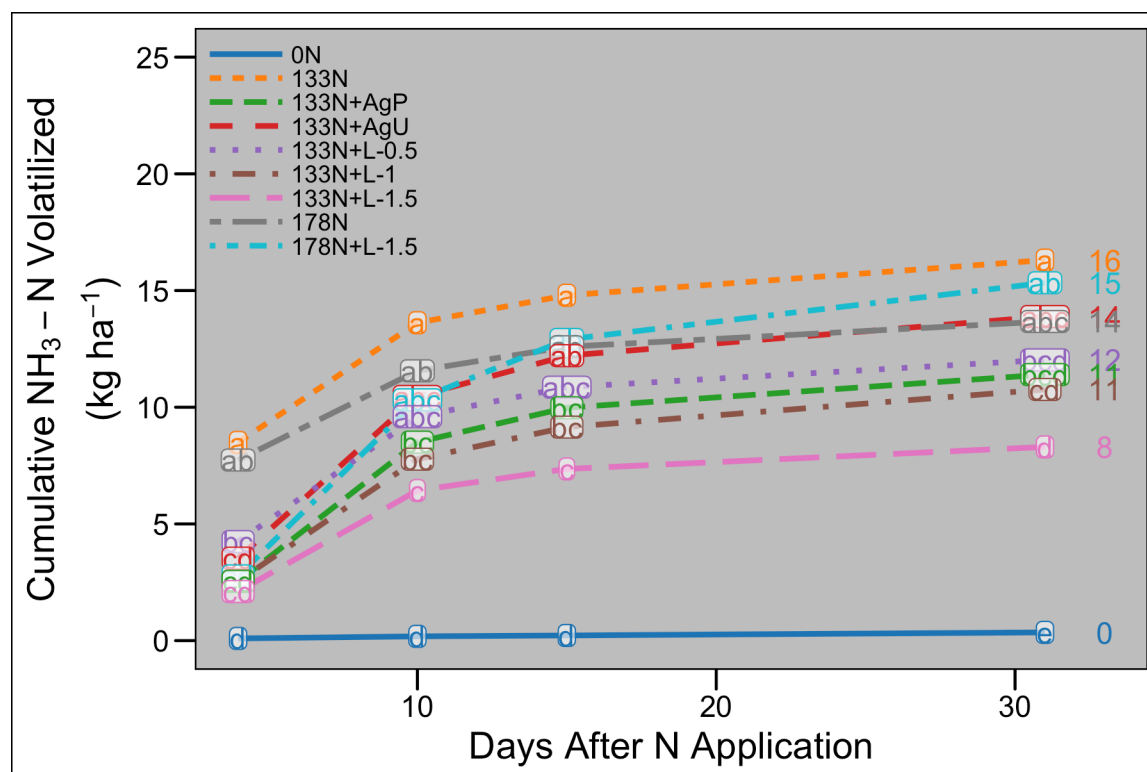


Figure 2.5. Mean cumulative ammonia volatilization (kg ha^{-1}) from 0 to 31 days after N application (DAA) at SCAL17. Treatment means within a date sharing the same letter are not statistically different at $\alpha=0.05$. Numbers on the right are the cumulative ammonia losses at 30 DAA. AgP = Agrotain Plus; AgU = Agrotain Ultra; L = Limus at 0.5, 1 and 1.5 L Mg^{-1} UAN rates.

Ammonia volatilization loss maximum, extent of protection by using an UI, and effect of UI+NI varied across studies (Table 2.3). The greatest loss across studies was observed at SCAL16 under untreated fertilizer applied at 173 kg N ha^{-1} (26 $\text{kg NH}_3\text{-N ha}^{-1}$). The use of AgU

(NBPT only) and Limus (NBPT/NPPT) reduced NH₃ volatilization compared to untreated fertilizer in two and four studies, respectively. The addition of NI to fertilizer+UI only significantly increased NH₃ volatilization compared to fertilizer+UI at CC15.

Table 2.3. Cumulative ammonia volatilization summary across all studies except for SCAL15 for treatments 0N, N (untreated fertilizer, study-rate specified in parenthesis on first column), N+AgU (NBPT), N+L-1.5 (NBPT/NPPT at 1.5 L Mg⁻¹ UAN), and N+UI+NI.

	0N	N	N+AgU	N+L-1.5	N+UI+NI
	----- Cumulative Ammonia Volatilization (kg NH ₃ -N ha ⁻¹) -----				
			--		
SCAL14 (182N)	2 c	19 a	19 a	15 b	NA
CC15 (130N)	1 e	18 b	10 d	12 cd	15 bc
SCAL16 (173N)	0 c	26 a	12 b	13 b	13 b
SCAL17 (133N)	0 e	16 a	14 abc	8 d	11 bcd

Reflectance

For all studies, in-season crop canopy sensor data showed limited treatment differences in crop vigor, though differences were clearer at earlier growth stages (V6 to V9). Sensor data obtained at VT for SCAL14 and SCAL15, at V6 for CC15, from V9 through V16 and R5 for SCAL16, and R3 for SCAL17, more closely followed the differences observed on final grain yield (shown and discussed later in the chapter).

At SCAL14, SPAD SI values at V10 and VT generally were below SI=0.95 (Figure 2.6), where 0.95 is a threshold between sufficient (>0.95) and deficient (<0.95) crop status (Blackmer and Schepers, 1995). At V10, SPAD SI was highest for 182N+AgU, which was greater than 182N, 136N+L0-1.2, and 0N only. At VT, SPAD SI was highest for 182N+L-1.2, which was greater than 182N+L-0.8, 182N, 136N+L-1.2, and 0N only, the latter being the lowest value. SPAD SI decreased numerically (i.e. not significant) from V10 to VT for most treatments.

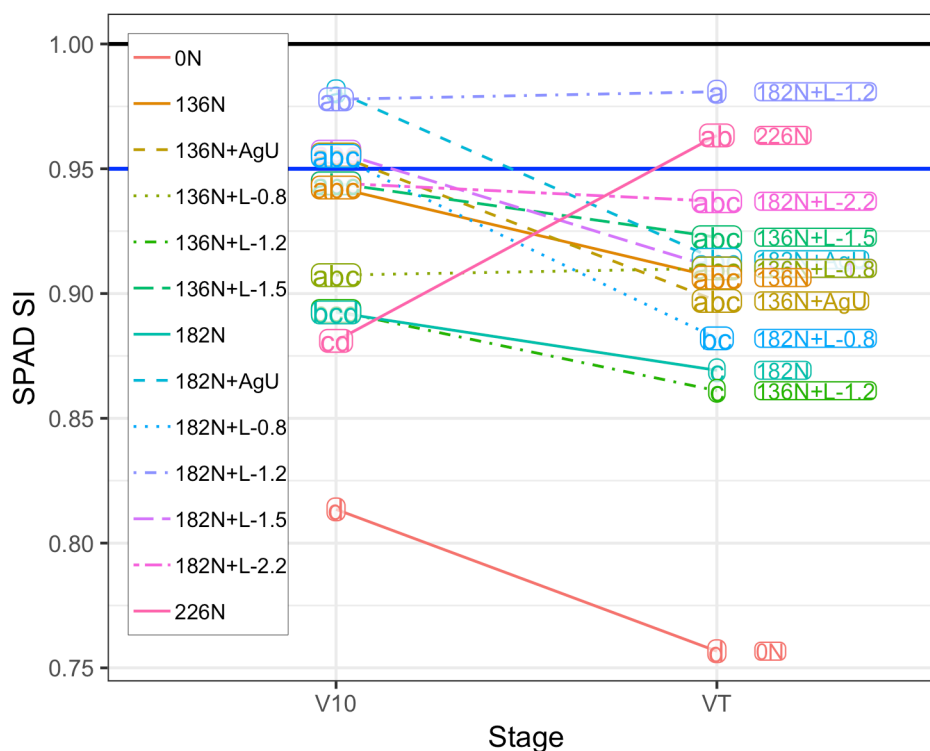


Figure 2.6. Mean SPAD sufficiency index (SI) at SCAL14. Treatment means within a growth stage with the same letter are not statistically different ($\alpha=0.05$). AgU = Agrotain Ultra; L = Limus at 0.8, 1.2, 1.5, and 2.2 L Mg⁻¹ UAN rates. Blue and black lines show SI of 0.95 and 1, respectively.

At CC15, NDRE SI values varied over the growing season by N treatments (Figure 2.7). Overall, variability due to N treatments was greatest at V6 and V12, then treatment values tended to converge at VT and R4. Most treatment means were below SI=0.95 for all growth stages. The numerically highest NDRE SI value occurred during V6 and V12 for 130N+AgP, and the lowest value at these same stages was for 0N. No treatment differences were observed at VT, and R4 NDRE SI values were highest for 161N and lowest for 0N.

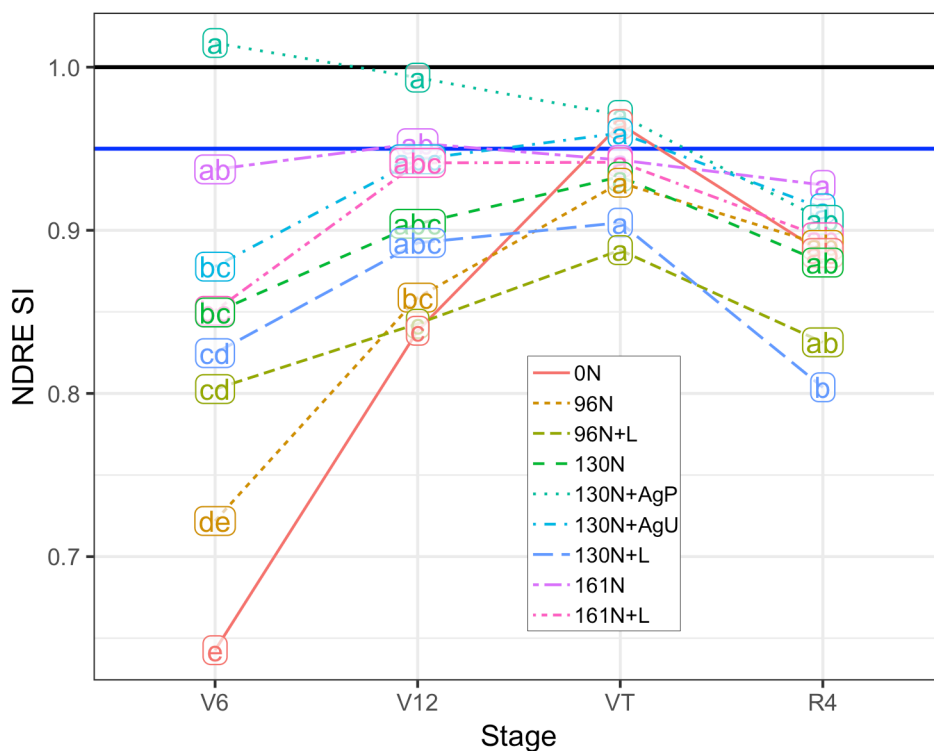


Figure 2.7. Mean NDRE sufficiency index (SI) at CC15. Treatment means within a growth stage with the same letter are not statistically different ($\alpha=0.05$). AgP = Agrotain Plus; AgU = Agrotain Ultra; L = Limus. Blue and black lines show SI of 0.95 and 1, respectively.

At SCAL15, NDRE SI values varied over the growing season for different N treatments (Figure 2.8). Overall, more variability due to N treatments occurred at V6 and R4, whereas treatment means converged at V12 and VT. Only 0N NDRE SI was <0.95 at V6 and remained lower than other N treatments at all stages. All other SI treatment means were >0.95 . The numerically highest NDRE SI value at V6 was for 96N, which was greater than 96N+L+DMP and 0N only. At V12, the highest NDRE SI values were for 130N+L+DMP, 130N+L, 130N+AgU and 96N, which were greater than 0N only. At VT, no treatment differences were

found. At R4, the highest NDRE SI value was for 130N+AgU, which was greater than 130N+L+DMP, 130N+L, and 0N only.

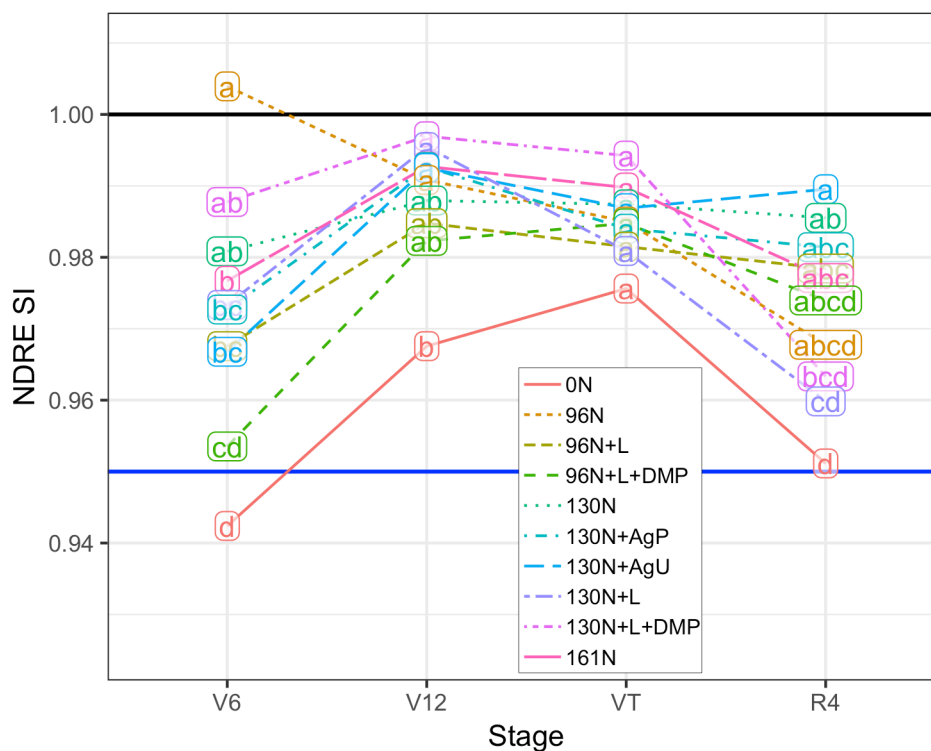


Figure 2.8. Mean NDRE sufficiency index (SI) at SCAL15. Treatment means within a growth stage with the same letter are not statistically different ($\alpha=0.05$). AgP = Agrotain Plus; AgU = Agrotain Ultra; L = Limus. Blue and black lines show SI of 0.95 and 1, respectively.

At SCAL16, NDRE SI values varied over the growing season for different N treatments (Figure 2.9). Treatment variability was greatest at V6, VT, and R3. Mean NDRE SI was lowest for 0N, which remained <0.95 throughout the season. All other treatment means were greater than 0N with values >0.95 and converged towards similar values after V6. At V6, the highest NDRE SI values were for 173N, 173N+AgP, 173N+AgU, and 173N+NS, ranging from 0.94 to 0.97, and the lowest value for 0N (0.81). At V7, the highest NDRE SI occurred for 215N+L-1.5,

which was greater than 215N, 173N+L-0.5, and 0N only. At V9, V12, V16, VT and R5, all treated fertilizers showed similar NDRE SI and were greater than 0N. At R3, the highest NDRE SI was for 215N+L-1.5, which was greater than 173N+L-0.5, 173N+L-1.5, and 0N only.

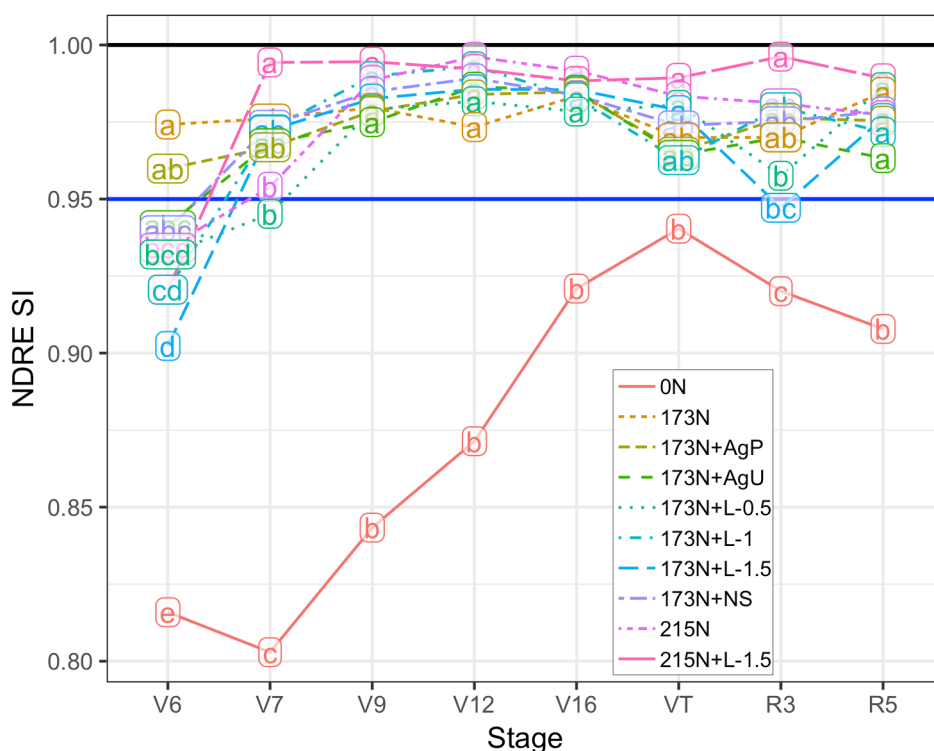


Figure 2.9. Mean NDRE sufficiency index (SI) at SCAL16. Treatment means within a growth stage with the same letter are not statistically different ($\alpha=0.05$). AgP = Agrotain Plus; AgU = Agrotain Ultra; L = Limus at 0.5, 1 and 1.5 L Mg⁻¹ UAN rates; NS=NutriSphere-N. Blue and black lines show SI of 0.95 and 1, respectively.

At SCAL17, NDRE SI values varied over the growing season for different N treatments (Figure 2.10). Variability due to N treatments was highest at V7, V9, and R3. Mean NDRE SI was lowest for 0N, which remained <0.95 throughout the season. All other treatment means were greater than 0N with values >0.95 and converged towards similar values after V7. Most SI means for treated fertilizers were >0.95 after V12, except for 0N and 45N (only >0.95 at V16).

At V7, the highest NDRE SI was for 133N+L-1, which was greater than 133N+L-0.5, 90N and 0N only. At V9, the highest NDRE SI was for 133N, which was greater than 90N and 0N only. At V12, VT, and R4, all SI values were similar and greater than 0N. At R1 and R3, 45N was lower than other N treatments.

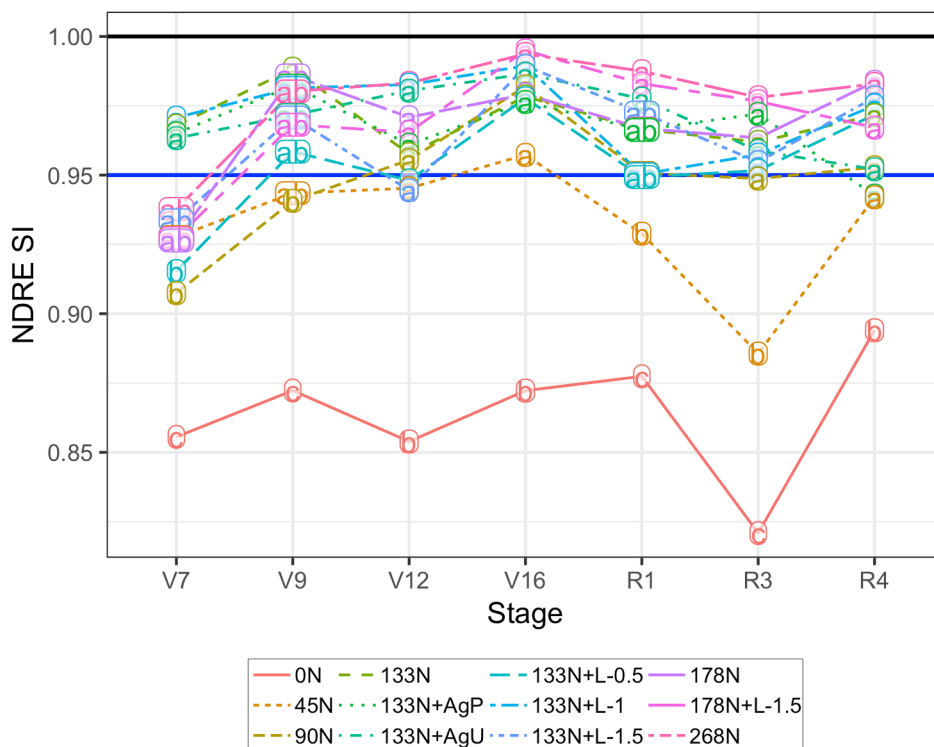


Figure 2.10. Mean NDRE sufficiency index (SI) at SCAL17. Treatment means within a growth stage with the same letter are not statistically different ($\alpha=0.05$). AgP = Agrotain Plus; AgU = Agrotain Ultra; L = Limus at 0.5, 1 and 1.5 L Mg⁻¹ UAN rates. Blue and black lines show SI of 0.95 and 1, respectively.

Yield

Corn grain yield responded to N fertilizer treatments for only three of five studies, and AONR varied from 160 to 208 kg N ha⁻¹ among the N-responsive studies (Table 2.2). Yield ranged from 5.7 (SCAL14) to 15.2 Mg ha⁻¹ (SCAL17). Inhibitor use (UI, UI+NI) did not

increase yield compared to untreated fertilizer at any study, and decreased yield in one case (182N-AgU at SCAL14).

At SCAL14, grain yield responded to N fertilizer, with a significant AONR of 209 kg N ha⁻¹ estimated using a linear-plateau model. The lowest grain yield occurred in unfertilized control (0N; 8.7 Mg ha⁻¹). The highest yield occurred at 226N which was not different from 182N+L-1.5, 182N+L-1.2, and 182N, ranging from 12.1 to 12.7 Mg ha⁻¹ (Figure 2.11).

Compared to fertilizer alone, using any UI did not improve yield for either 136N or 182N rates, and yield decreased for AgU-amended N.

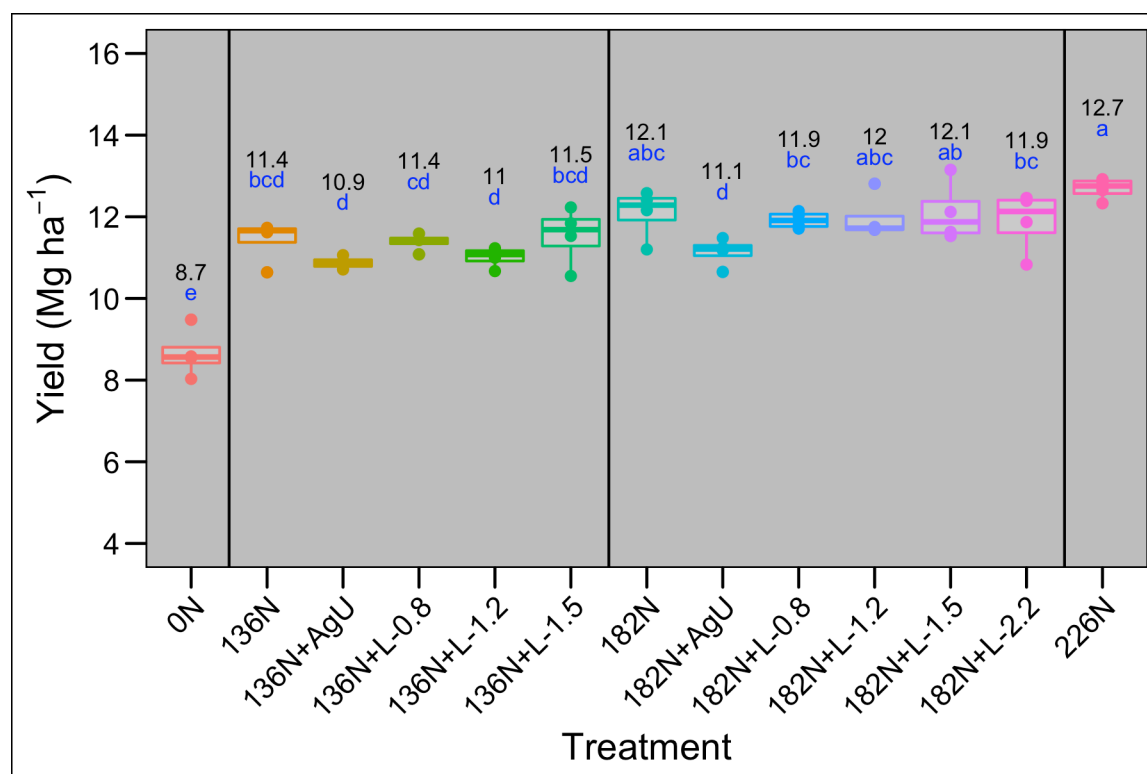


Figure 2.11. Boxplot (n=4) of corn grain yield (Mg ha⁻¹) at SCAL14. Numbers above the boxplots are the mean yield for that treatment. Treatment means sharing the same letter are not statistically different at $\alpha=0.05$. Vertical bars separate different N rate groups. AgU = Agrotain Ultra; L = Limus at 0.5, 1 and 1.5 L Mg⁻¹ UAN rates.

At CC15, grain yield responded marginally to N fertilizer application ($p=0.077$), with an AONR of 146 kg N ha^{-1} estimated using a quadratic-plateau model. The lowest grain yield occurred at 0N, which was not different from any other N-added treatments except for 161N and 130N+AgP (Figure 2.12). The highest grain yield occurred for 130N+AgP which was not different than 161N, 161N+L, 130N, and 130N+L, ranging from 6.3 to 7.1 Mg ha^{-1} . Using UIs did not improve grain yield at any N rate (96, 130, 161N). However, adding AgP (UI+NI) at the 130N rate increased yield compared to adding AgU (UI only).

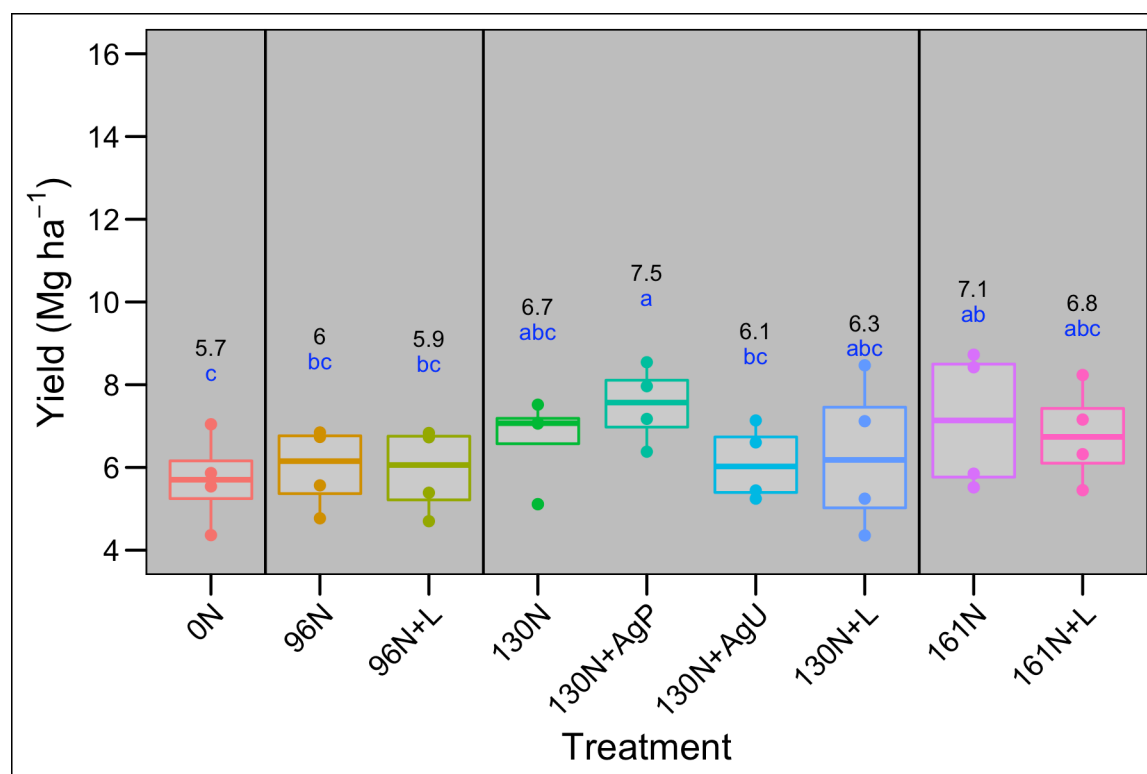


Figure 2.12. Boxplot (n=4) of corn grain yield (Mg ha^{-1}) at CC15. Numbers above the boxplots are the mean yield for that treatment. Treatment means sharing the same letter are not statistically different at $\alpha=0.05$. Vertical bars separate different N rate groups. AgP = Agrotain Plus; AgU = Agrotain Ultra; L = Limus.

At SCAL15, grain yield did not respond to N fertilizer or inhibitor amendments ($p=0.8$), and AONR was set to 0 kg N ha^{-1} . Grain yield varied from 13.7 (0N) to 14.3 Mg ha^{-1} (130N) (Figure 2.13), with overall site mean of 13.9 Mg ha^{-1} .

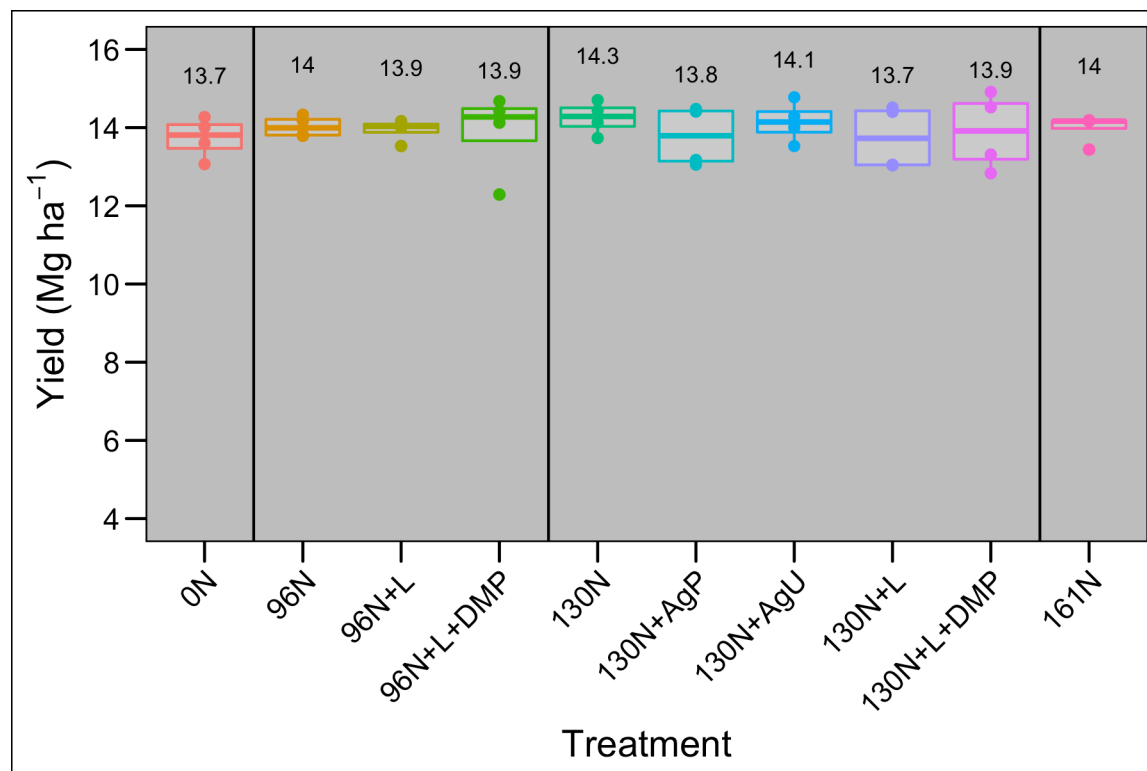


Figure 2.13. Boxplot ($n=4$) of corn grain yield (Mg ha^{-1}) at SCAL15. Numbers above the boxplots are the mean yield for that treatment. Vertical bars separate different N rate groups. AgP = Agrotain Plus; AgU = Agrotain Ultra; L = Limus.

At SCAL16, grain yield responded to N fertilizer, with a significant AONR of 160 kg N ha^{-1} estimated using a quadratic model. The lowest grain yield occurred under 0N (13.1 Mg ha^{-1} ; Figure 2.14). Yield increased after any fertilizer addition regardless of N rates or inhibitor addition, with an overall mean of 15 Mg ha^{-1} .

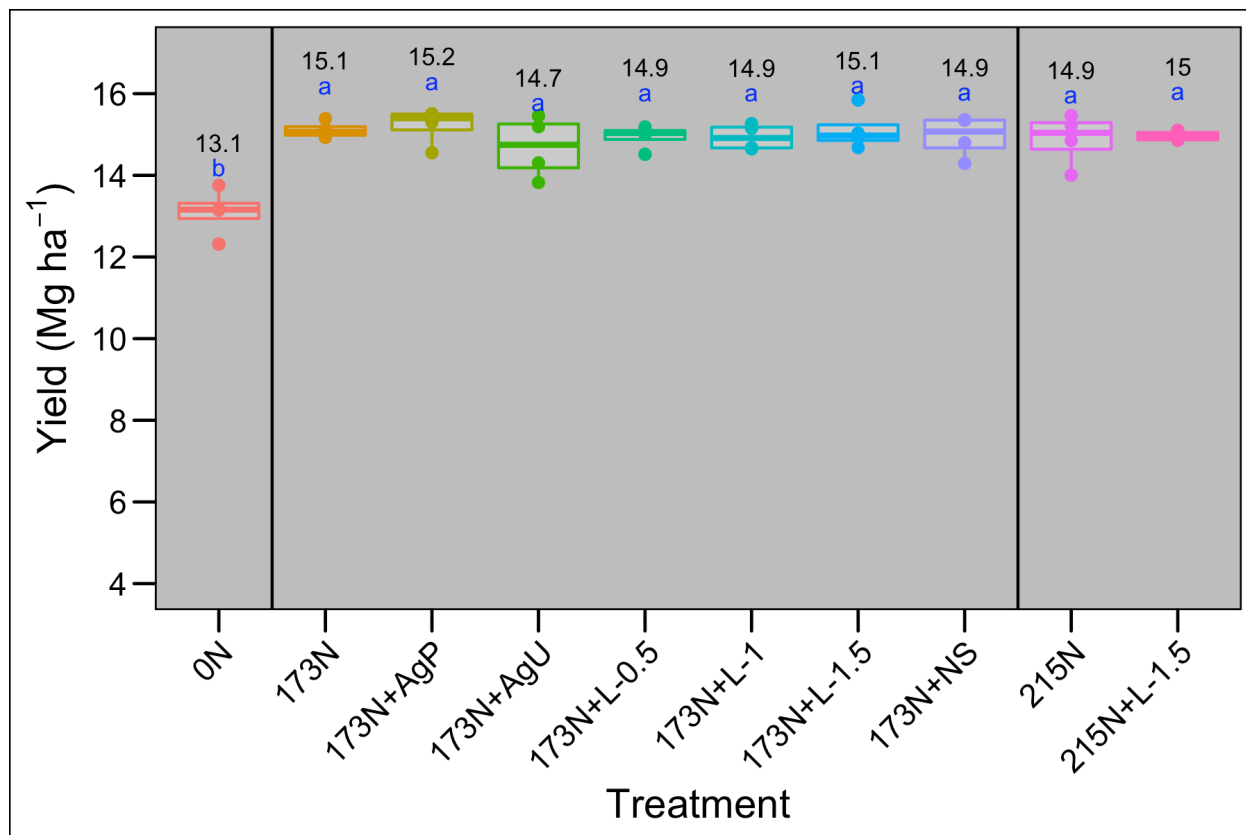


Figure 2.14. Boxplot (n=4) of corn grain yield (Mg ha⁻¹) at SCAL16. Numbers above the boxplots are the mean yield for that treatment. Treatment means sharing the same letter are not statistically different at $\alpha=0.05$. Vertical bars separate different N rate groups. AgU = Agrotain Ultra; L = Limus at 0.5, 1 and 1.5 L Mg⁻¹ UAN rates; NS= NutriSphere-N.

At SCAL17, grain yield responded to N fertilizer, resulting in an AONR of 173 kg N ha⁻¹ estimated using a quadratic-plateau model. The lowest grain yield occurred under 0N (10.4 Mg ha⁻¹) (Figure 2.15). Yield increased significantly with N rate from 0 to 133N, followed by limited to no yield improvements for higher N rates. The highest grain yield occurred for 268N which did not differ from 178N+L-1.5, 178N, 133N+L-1.5, 133N+AgU and 133N, and ranged from 14.4 to 15.2 Mg ha⁻¹. The use of any inhibitor did not improve grain yield compared to fertilizer alone at 133N and 178N.

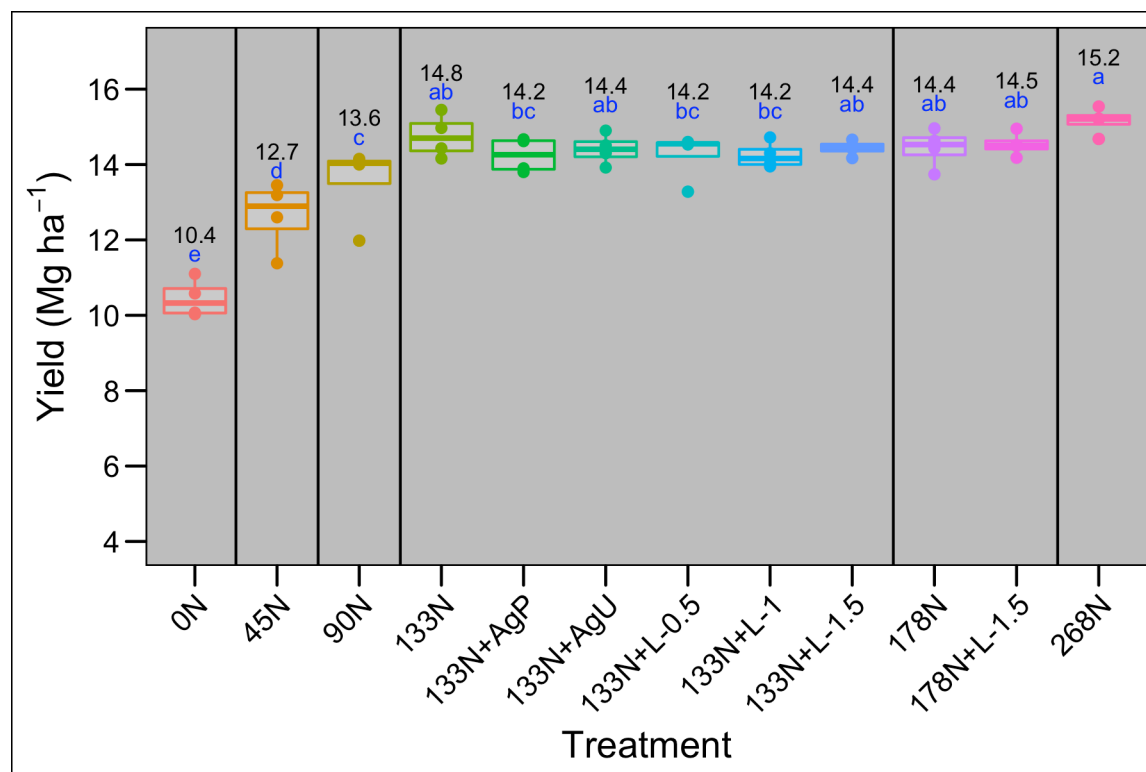


Figure 2.15. Boxplot (n=4) of corn grain yield (Mg ha⁻¹) at SCAL17. Numbers above the boxplots are the mean yield for that treatment. Treatment means sharing the same letter are not statistically different at $\alpha=0.05$. Vertical bars separate different N rate groups. AgU = Agrotain Ultra; L = Limus at 0.5, 1 and 1.5 L Mg⁻¹ UAN rates.

Discussion

Ammonia Volatilization

For four of five studies measured, total NH₃ volatilization losses for the 30 d following fertilizer application varied from ~0 (when no fertilizer was applied) to 26 kg NH₃- N ha⁻¹, with maximum losses ranging from 10 to 15% of applied fertilizer. Measured values here agreed with other published studies reporting 5 to 16% losses (McInnes et al., 1986a, Keller and Mengel 1986). Relatively low loss rates here are also consistent for surface-applied UAN compared to urea because the lower urea content of UAN decreases potential hydrolysis-driven pH increases that promote NH₃ volatilization (Keller and Mengel, 1986; Hargrove, 1988).

Inhibitor use reduced volatilization losses by 21 to 62% compared to untreated fertilizer in five out of six study-N rate combinations, saving 4 to 16 kg NH₃- N ha⁻¹ from being volatilized. The loss reductions from UI reported here are comparable to other studies reporting 25 to 89% reductions in volatilization losses by adding UI to urea fertilizer (San Francisco et al., 2011; Silva et al., 2017; Sunderlage and Cook, 2018). Ammonia volatilization from UAN is generally lower than that from urea, since UAN only has 44% of its total N in the urea form. Nonetheless, studies evaluating the effect of UI on NH₃ losses from UAN application have reported similar loss reduction percentages (from 37 to 84%) to those found for urea (Grant et al., 1996; Goos, 2013).

The highest N savings from UI use were observed at SCAL16, while the other studies had similar N savings from UI use (from 4 to 7 kg NH₃-N ha⁻¹). The largest reduction in NH₃ volatilization from UI use in SCAL16 is likely related to this study having the highest observed cumulative NH₃ loss of unprotected fertilizer (26 kg NH₃-N ha⁻¹ under 173N), coupled with lower volatilization observed when a UI was used. Other studies had either lower maximum volatilization potential of unprotected fertilizer (SCAL14 and SCAL17), and/or decreased efficacy of UI in protecting N from being volatilized (SCAL14, CC15, SCAL17).

The degree of inhibitor effectiveness varied by study and by product. When applied at the manufacturers' recommended rates, all products evaluated (AgP, AgU, L, NS) showed effectiveness in reducing volatilization losses, except for AgU (UI) at SCAL14 and, to a lesser extent, AgU at SCAL17 and AgP (UI+NI) at CC15. Limus (NBPT+NPPT) was the only product tested at rates in addition to the manufacturer recommended rate in effort to perform a more thorough evaluation of this new UI product. The efficacy of Limus in decreasing NH₃ loss compared to other UI products depended on the study. At least one Limus rate was more

efficient than AgU or AgP in decreasing NH_3 volatilization at SCAL14 and SCAL17, and Limus had comparable efficiency with AgU or AgP at CC15 and SCAL16. Over all studies, Limus applied at the 1.5 L Mg^{-1} UAN rate (the manufacturer recommended rate) was the most consistent in decreasing NH_3 losses, whereas both lower and higher Limus rates had mixed results. This result is consistent with a recent meta-analysis reporting no benefit of increasing NBPT above the manufacturer-recommended rate on NH_3 loss reduction (Silva et al., 2017).

Adding an NI to a UI (i.e. AgP) had mixed effects on NH_3 losses. At CC15, AgP enhanced cumulative NH_3 losses compared to UI only (L, AgU; 15 vs. $10 \text{ kg NH}_3\text{-N ha}^{-1}$, respectively), but no product differences were observed at SCAL16 and SCAL17. This effect likely resulted from differences in soil properties, where CC15 soils were coarser-textured with lower CEC than SCAL16 and SCAL17. Because soils with lower CEC have lower capacity to adsorb NH_4^+ (Keller and Mengel, 1986; Hargrove, 1988; Gioacchini et al., 2002; Sommer et al., 2004), CC15 soils likely favored the maintenance of N in ammoniacal form for a longer period and promoted higher NH_3 losses. Gioacchini et al. (2002), however, found that adding NI to UI overrode the UI savings of NH_3 in a clay loam to a greater extent than in a sandy loam, in contrast to our findings. Nonetheless, our results confirm reported increases in NH_3 losses from adding NI to UI+fertilizer (Gioacchini et al., 2002; Soares et al., 2012; Peng et al., 2015; Pan et al., 2016).

A major limitation of the results reported here is that the sealed chamber-based method does not represent the loss conditions of the entire treatment area. The chambers themselves altered soil microsite conditions because they were deployed continuously, protecting the measurement area from water inputs and wind and potentially altering daily temperature patterns compared to the exterior environment. The absence of water inputs to incorporate surface-

applied fertilizer into the soil plus reduction in soil drying due to reduced wind speeds could both enhance soil NH_3 losses compared to ambient plot areas (Bouwmeester et al., 1985; McInnes et al., 1986b; a; Holcomb et al., 2011). As a result, crop vigor and yield data presented here are heavily dependent on antecedent soil conditions prior to chamber deployment. Because chamber-based methods are common in many studies, our results are comparable to published reports and relative treatment effects within a study are valid, but data should be interpreted with caution in regard to crop vigor and final grain yield (below).

Weather

The weather patterns during the 15-day window surrounding N application date (-5 to +10 DAA) varied in both volume and frequency of rainfall, both of which potentially affect NH_3 losses. As mentioned above, chamber-based volatilization measurements protected soils from inputs following N application, suggesting greater impacts of pre-deployment soil moisture status on NH_3 losses (5 d prior to N application vs. 10 d post N application). Pre-fertilization precipitation inputs occurred at CC15, SCAL16, and SCAL17 only. At SCAL17, precipitation events of 41, 10, and 5 mm happened three, two, and zero days before fertilizer application, followed by days with increasing air temperature. These conditions could have been optimum for NH_3 volatilization as fertilizer was applied on wet residue followed by a drying period. For example, Bouwmeester et al. (1985) reported increased NH_3 losses at a low wind speed and attributed this effect to the concurrent slow drying of the soil.

Small-volume high-frequency water additions can avoid TAN buildup in the soil, resulting in half the NH_3 losses compared to applying the same total volume in larger, less frequent events (Bouwmeester et al., 1985). Similarly, Holcomb et al. (2011) estimated a 95% reduction in NH_3 losses from surface-applied urea using a single small irrigation event (19 mm)

shortly after fertilizer application. In contrast, other studies have found that rainfall volumes <25 mm can enhance NH_3 losses by providing enough water to promote urea hydrolysis on the surface yet not enough to move fertilizer into the soil (McInnes et al., 1986ab). SCAL14 and CC15 received >20 mm rainfall at four and three DAA, which were likely sufficient to move the fertilizer into the soil and reduce losses. In contrast, SCAL15 had a 10-mm rainfall event at four DAA, and SCAL16 received precipitation every day from five to nine DAA, with each event being of small magnitude. The only rain event at SCAL17 to happen within 10 DAA was at five DAA and of only 5 mm, which could have further enhanced NH_3 losses.

Sensor-based Crop Vigor

Normalized sensor data in the form of a sufficiency index (SI) was used to infer in-season crop vigor as affected by different N treatments. At SCAL14, SPAD was used at V10 and VT, with 182N+L-1.2 ranking high (>0.95) at both stages whereas 182N ranked low (<0.95) in both stages. This could be an indication that using a UI at the 1.2 L Mg^{-1} UAN rate was able to keep NH_3 from being volatilized compared to untreated fertilizer to the point of being expressed in crop vigor. However, both higher and lower rates of UI applied at 182N had SPAD SI values lower than 0.95, indicating that there must exist an optimum UI rate.

At all other studies, an active sensor was used and crop vigor was assessed at multiple growth stages. At CC15, the use of UI at 96, 130 and 161N did not create significantly higher SI than their untreated counterparts at any growth stage. The only treatment combination with higher SI than untreated fertilizer was 130N+AgP, which contains both UI and NI. This study was a sandy loam soil and received 315 and 318 mm of precipitation and irrigation, respectively, from June through August. The excessive water input on a coarse-texture soil likely promoted nitrate leaching losses, evidenced by visual assessment during active sensor data collection.

Under these circumstances, the use of an NI likely protected N fertilizer against leaching losses and thus created higher SI values. This can be observed on the SI data as almost all SI values are below 0.95, including both unfertilized and fertilized treatments. In this case, NH_3 volatilization played a secondary role on N losses, which was likely dominated by nitrate leaching.

At SCAL15, little difference was observed in SI over the growing season. The use of UI+NI (Limus+DMP) decreased NDRE SI at 96N compared to untreated fertilizer at V6, and UI at 130N had significantly lower SI at R4 compared to 130N alone. Although these differences were significant, SI values were all above 0.95 including the unfertilized treatment, indicating that crop vigor and N nutrition differences may not have been of agronomic significance.

At SCAL16, the use of UI at 215N created SI values that remained among the highest throughout the growing season, even though treatments were similar and above 0.95 at almost all crop stages. The only treatment that had consistently lower SI was 0N. This is evidence that N fertilization promoted crop vigor, but the crop was sufficiently provided with N at 173N and above. A similar pattern was found at SCAL17. No differences in SI were observed throughout the growing season when UI alone or in combination with NI was added at 133N and above. Lower SI values were observed consistently with 0N and occasionally with 45N. This indicates that crop vigor responded to N fertilizer up to a certain rate and became unresponsive thereafter, regardless of inhibitor use, partially due to the fact that inhibitors were used at N rates above the responsive threshold.

Grain Yield

Grain yield integrates the effects of N management on N losses and plant N sufficiency over the growing season. In spite of differences observed from chamber-measured NH_3 losses

and crop vigor throughout the growing season, crop responses to N fertilizer were measurable in only three of the five studies, with resulting AONR values varying from 160 to 208 kg N ha⁻¹.

Overall, the use of UIs at different rates reduced potential NH₃ losses, which varied from 4 to 16 kg NH₃-N ha⁻¹. However, in no occasion was a UI able to improve grain yield compared to untreated fertilizer. Limited yield response to UI-treated fertilizer has been demonstrated in the literature (Gioacchini et al., 2002; Abalos et al., 2014; Silva et al., 2017; Cantarella et al., 2018). Cantarella et al. (2018) summarized multiple studies on the impact of NBPT on the yield of various crops and found that, on average, NBPT increased corn grain yields by 4.1%. These authors attributed the limited or lack of yield response to NBPT to the large contribution of mineral N from other sources (e.g. mineralization) that end up supplying enough N and avoid untreated fertilizer loss causing crop N deficiency. Rose et al. (2018) further pointed out that many studies evaluating the effect of EEFs, including UIs, on grain or biomass yield do not include them at multiple N rates, and reported that the largest yield increase from their use (11% over untreated fertilizer) was observed at 50% of the optimal N rate.

Therefore, the use of a UI is expected to increase grain yield when i) conditions conducive to fertilizer-derived NH₃ volatilization exist, ii) fertilizer is applied at below-optimum N rates (N rate < AONR), and iii) the amount of N kept from being lost is large in comparison to loss from untreated fertilizer. None of the studies included in this study fulfilled all three of these conditions. At SCAL14 and SCAL17, inhibitors were applied at N rates below AONR, but potential NH₃ volatilization from UI use was decreased by only 4 kg NH₃-N ha⁻¹ compared to untreated fertilizer. At CC15, SCAL15, and SCAL16, inhibitors were applied at N rates in excess of AONR.

The three conditions for observing a UI effect on yield are manageable to different degrees. All studies herein reported received N applications in the form of urea-containing N fertilizer, broadcast on the soil surface with varying amounts of crop residue, and with no weather forecast of rainfall following fertilizer application. Even then, potential NH_3 losses measured by the sealed chamber method demonstrated limited room for UIs to save N, mostly due to low potential losses from untreated fertilizer. In three of five studies, UIs were only applied at N rates that were greater than AONR. Future studies evaluating the efficacy of UI on losses and grain yield should include UI treatments at lower N rates, perhaps closer to at least half of the full pre-plant recommended N rate, so the extent of N saved from UI use can contribute towards plant N demand.

These constraints should create N fertilizer management scenarios where UI savings on volatilization could benefit N nutrition and have an impact on grain yield. Moreover, results from such studies could better inform producers and policymakers on the extent of how excessive current N rates can be and by how much they can be decreased given that fertilizer is protected against NH_3 volatilization.

Conclusions

The efficacy of different inhibitors (UI, UI+NI) in decreasing NH_3 volatilization losses and increasing corn grain yield was evaluated. All inhibitors were tested with broadcast application of UAN fertilizer at different rates. Volatilization losses from these studies were relatively low, ranging from 10 to 15% of applied fertilizer. The use of a UI decreased NH_3 losses from 21 to 62%, and Limus applied at the label rate was comparable to AgU and/or AgP in decreasing NH_3 losses. Although using UIs decreased sealed-chamber measured NH_3 losses, no inhibitor was able to improve grain yield over untreated fertilizer. That was because i) the

amount of fertilizer lost via volatilization was small, and ii) other sources of N (e.g. soil mineralization), likely provided N in excess of crop needs.

Although limited losses were observed in our study, NH_3 volatilization can be significant for urea-containing, surface-applied N fertilizer. The extent of loss is directly dependable on rainfall volume and frequency surrounding fertilizer application. Therefore, using a UI can assist a producer in managing fertilizer loss risk associated with application window constraints and weather unpredictability by conserving time, soil, and economic resources.

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Chapter 3 - Nitrogen Management Practices and Yearly Weather Impact

Long-term Yield Variability in Irrigated Corn

Introduction

The application of nitrogen (N) fertilizer is a key management practice for achieving high yields in corn (*Zea mays* L.). Vast areas of the U.S. are planted to corn annually. In 2018, 89 million ha in the U.S. was planted to corn (USDA-NASS, 2017). As a result, national fertilizer use for corn is also high, with 5.7 million tons of N fertilizer applied in 2014, or ~48% of all N fertilizer utilized in the country (USDA-NASS, 2017). However, once applied to the soil, fertilizer N is at risk of being lost to the environment through multiple pathways and in varying forms, including gas emissions as ammonia (NH₃) (i.e. volatilization) and nitrous oxide (N₂O) (i.e. byproducts of soil microbial N processes such as denitrification), and as N dissolved in solution (i.e. nitrate (NO₃⁻) leaching) (Motavalli et al., 2008). In addition to causing negative environmental impacts, fertilizer N losses decrease N availability for crop uptake and conversion to grain yield, resulting in lower fertilizer returns on investment. Krupnik et al. (2004) estimated only 39% of fertilizer N was recovered in North American corn crops, comparable to the 37% estimated by Cassman et al. (2002) from 55 corn on-farm studies in the U.S. Midwest.

To achieve greater crop fertilizer use efficiency and yield, best practices developed for fertilizer management often focus on adaptive approaches that address field-specific characteristics (i.e. soil type, local climate). The most widespread best practices are the “4Rs” of nutrient stewardship, where each “R” represents the right source, right rate, right placement and right timing (Bruulsema et al., 2008). Although there exists a general understanding of cause and effect between N management practices and their effects on yield and losses, yield outcomes are not always assured due to the complexity of the N cycle and the interactions between

management, soil, and weather. For an example of N timing, the widely accepted best practice is to minimize the time between N application and when crops begin their growing season. In other words, N applied in the fall after grain harvest is expected to be more prone to loss and thus less favorable for yield and profit compared to N applied in the spring (Hendrickson et al., 1978; Touchton et al., 1979; Torbert et al., 2001; Randall et al., 2003; Randall and Vetsch, 2005; Tao et al., 2018). Some studies, however, have reported no differences (Torbert et al., 2001) or even higher yields from fall vs. spring applications in certain conditions (Randall and Vetsch, 2005).

In the case of selecting N rate, predictive approaches (i.e. mass balance, maximum return on N) are the historical standard, where a single N rate is recommended for whole-field use. For a given fertilizer source, newer technological advances can now address multiple aspects of the 4R paradigm simultaneously (i.e. rate, timing, placement) by using in-season reactive approaches through crop sensors (Raun et al., 2002; Scharf and Lory, 2009; Holland and Schepers, 2010; Solari et al., 2010), models (Melkonian et al., 2008; Setiyono et al., 2011), and the development of new products such as stabilized fertilizers.

Stabilized fertilizers are defined by the American Plant Food Control Officials (Association of American Plant Food Control Officials, 2013) as "...a fertilizer to which nitrogen stabilizer has been added...[to extend] the time the nitrogen component of the fertilizer stays in the soil in the urea-N or ammoniacal-N form." Nitrification inhibitors (NIs) and urease inhibitors (UIs) are both categorized as stabilizers for N fertilizer. The use of these inhibitors with conventional fertilizer sources is often effective in decreasing average N losses and increasing average crop yield (Hergert and Wiese, 1980; Wolt, 2004; Abalos et al., 2014). The range of N losses and yield responses to inhibitors, however, vary widely, with reports of no or

negative effects on N losses (Gioacchini et al., 2002; Soares et al., 2012) and yield (Blackmer and Sanchez, 1988; Cerrato and Blackmer, 1990).

Researchers and producers are continually exploring the 4Rs for N fertilizer management to better identify specific soil and weather conditions that favor yield and reduce loss outcomes. Most studies evaluating N management practices are relatively short (<5 yr) and have limited assessment of how other management practices and interannual weather variability interact with N management to affect grain yield. Such comprehensive evaluations require long-term datasets with static combinations of different management treatments. To identify the individual or suite of best practices that optimized yield level while reducing yield variability and N losses, we assessed a 28-yr field study of irrigated corn in south central Nebraska USA. Specifically, we: i) evaluated the effects of N rate, tillage, N application timing, and the use of nitrapyrin (an NI) on grain yield; and ii) assessed how weather affected grain yield responses under these different N management practices over time. Soils were also measured to evaluate the effectiveness of different management practices on soil fertility status (organic matter, nutrient availability).

Material and Methods

Site Description

This study was conducted at the University of Nebraska-Lincoln South Central Agricultural Laboratory (SCAL), near Clay Center, Nebraska (40.571297° N, 98.134988° W). The predominant soil type is a Crete silt loam (Fine, smectitic, mesic Pachic Udertic Argiustolls). The study was established in 1986. While study location remained constant, corn hybrid changed over time as new hybrids were released in the region to better represent relevant

producer practice. Management treatments were also adjusted periodically to maintain relevancy (see below).

Corn was planted in 0.76-m row spacing at a target population of 74,000 plants ha⁻¹, with planting dates varying from April 17th to May 21st over the 28 years of study. All treatments below received the same irrigation inputs. Herbicides and/or pesticides were applied as needed. Since 2010, 112 kg ha⁻¹ of monoammonium phosphate (11-52-0) was surface broadcasted during winter to the entire study area. Tillage occurred shortly after 11-52-0 application. Grain yield was measured by combine-harvesting the middle 2 rows of each plot and reported on a 15.5% moisture basis.

Experimental and Treatment Design

This study was established in a randomized complete block with four replicates, with minor treatment changes over the full 31-yr period (1986 to 2017). From 1986 to 1988, treatments were in a split-split-split-plot arrangement with the main factor of N fertilizer rate applied as anhydrous ammonia (0, 75, 150, 300 kg N ha⁻¹, hereafter 0N, 75N, 150N, 300N, respectively), the split-plot factor of tillage [conventional chisel/disk (CT), reduced tillage (RT)], the split-split-plot factor of corn hybrid (Pioneer 3377, 3475, 3551), and split-split-split plot factor of nitrification inhibitor (with NI, without NI) as N-Serve[®] (0.5 kg nitrapyrin ha⁻¹; Dow Agrosciences LLC, Indianapolis, IN). Reduced tillage is defined as intermittent no-till and ridge-till. From 1989 to 1992, fertilization timing treatment factor was added as early side-dress (SD, approximately V4) and late SD (approximately V8). In 1990, the corn hybrid treatment factor was dropped, and a single hybrid adapted to the region was planted thereafter. From 1993 to 2013, fertilization timing was changed to spring pre-plant (PP) (from four weeks to one day

before planting) and SD (approximately V4). From 2014 through present, fertilizer timing was dropped as a variable, and all fertilizer has been applied PP in the spring.

Soil Data

Soil was sampled for fertility characterization in the fall of 2000 and 2006 after harvest. Sampling in 2000 characterized 14-yr changes in surface soils (0-20 cm) since study establishment, for which the last 10 yrs (1990 to 2000) covered the period using only one corn hybrid throughout the study. In 2000, samples were taken from the N rate treatment plots (0N, 75N, 150N and 300N) from blocks 1 and 4 at the 0-20 cm depth. Samples were pooled by plot, mixed and analyzed for pH, soil organic matter (SOM; % loss on ignition), nitrate (NO_3^- -N; ppm), Bray phosphorus (P; ppm), potassium (K; ppm), and zinc (Zn; ppm).

Sampling in 2006 characterized 20-yr soil changes throughout the whole soil profile (to 91-cm depth) since study establishment. In 2006, samples were taken from 10 treatments in blocks 1, 2 and 3. Treatments sampled were all combinations of three N rates (0N, 150N, 300N), two tillage types (CT, RT) and Nserve (with vs. without), except for 0N-CT-Nserve and 0N-RT-Nserve, which were not treatments in the field design. Six cores per plot from a radius of 3 m around a sampling point were taken to 91-cm depth and split into 15-cm increments, for a total of six depths. Three cores were randomly selected and composited by depth for fertility characterization (pH, SOM, P, K; described above). In addition, particulate organic matter (POM; Mg ha^{-1}) was also measured. The remaining three cores were kept intact for determination of bulk density (Mg m^{-3}) (data not shown).

Weather Data

Daily weather data for the 28-yr period from 1990 to 2017 was obtained from a weather station located within 1 km of the study site. Weather variables included solar radiation

(Solar_MJm2d, in MJ m⁻² d⁻¹); air relative humidity (RH_pct, in %); precipitation (Precip_mm) and evapotranspiration (ET_mm); wind speed (WindSp_ms, in m s⁻¹); soil temperature at 10 cm depth (SoilT10cm_C) and minimum (Tmin_C), and maximum air temperature (Tmax_C), in °C. Secondary variables were calculated from the measured variables and included growing degree days (GDD, Eq. [3.1]) with a base temperature of 10°C, corn heat units (CHU, Eq. [3.2]), precipitation Shannon diversity index (SDI, Eq. [3.3]) and abundant well-distributed rainfall (AWDR, Eq. [3.4]) (Tremblay et al., 2012).

$$GDD = \frac{(T_{max} + T_{min})}{2} - 10, \text{ where} \quad [3.1]$$

T_{max} = maximum daily air temperature in °C. If $T_{max} > 30^\circ\text{C}$, then T_{max} is set to 30°C .

T_{min} = minimum daily air temperature in °C. If $T_{min} < 10^\circ\text{C}$, then T_{min} is set to 10°C .

$$CHU = \frac{(Y_{max} + Y_{min})}{2}, \text{ where} \quad [3.2]$$

$Y_{max} = 3.3(T_{max} - 10) - 0.084(T_{max} - 10)^2$. If $T_{max} < 10^\circ\text{C}$, then Y_{max} is set to 0.

$Y_{min} = 1.8(T_{min} - 4.44)$. If $T_{min} < 4.44^\circ\text{C}$, then Y_{min} is set to 0.

T_{max} is the maximum daily temperature, in °C.

T_{min} is the minimum daily temperature, in °C.

$$SDI = \frac{-\sum pi \ln(pi)}{\ln(n)}, \text{ where} \quad [3.3]$$

$$pi = \frac{\text{daily rainfall in mm}}{\text{cumulative rainfall in mm}}$$

n is the number of days in the period used to calculate the cumulative rainfall.

$$AWDR = \textit{Cumulative precipitation} \times SDI \quad [3.4]$$

Statistical Analysis

Soil properties measured in 2000 were analyzed separately using a mixed model analysis of variance (ANOVA), with Nrate as a fixed effect and Block as random effect. Soil properties measured in 2006 were analyzed separately for each soil depth using a mixed model ANOVA, with Nrate, tillage, Nserve and their two- and three-way interactions as fixed effects. Block and all interaction terms including it were considered random and were appropriately pooled at each experimental unit size to represent experimental error. Means for significant fixed treatment responses were compared with Fisher's least-significant differences. All statistical outcomes were significant at $\alpha = 0.05$, unless noted.

Although this study has been ongoing for 31 years, only data from the last 28 years (1990 to 2017) are evaluated here. Grain yield responses were analyzed for two time periods. Period one (1994 to 2013) corresponded to the study period that included fertilizer application timing as a variable. Analyses for period one excluded 1993 because no SD application was applied that year, and excluded 1999 due to hail damage. Period two (1990 to 2017) corresponded with the study period when only one corn hybrid was used across treatments. Yield data were pooled across timing treatments from 1990 to 1993 before joining data to the full 28-year dataset.

For each time period, 0N grain yields were excluded from statistical analysis to evaluate the full factorial nature of the design (0N with Nserve was not an applied treatment). Period one grain yield was analyzed with a mixed model ANOVA, where year, N rate, tillage, application timing, and Nserve were considered as fixed effects. Block, block within year and all interaction terms including these effects were considered random and were appropriately pooled at each

experimental unit size to represent experimental error. Period two grain yield was analyzed similarly, except with no application timing treatment included. For each time period, yield differences for significant fixed treatment responses were evaluated with Fisher's least-significant differences. All statistical outcomes were significant at $\alpha = 0.05$, unless noted.

Post-hoc analyses for significant treatment effects showed a high range of interannual variability in grain yield. To facilitate statistical analyses of weather (described below), mean grain yields for year were grouped into three year-yield potential categories: high-yielding years (12.6 to 14.7 Mg ha⁻¹), medium-yielding years (10 to 12 Mg ha⁻¹), and low-yielding years (8.5 to 9.8 Mg ha⁻¹). Years were grouped by utilizing the k-means unsupervised classification algorithm. The number of groups (k, i.e. three in this case) was chosen based on the value of k that most parsimoniously maximized inter-group variance and minimized intra-group variance. For period one, significant treatments that included both year and application timing (Ntiming) were selected for further weather assessment. For period two, weather assessment was conducted for significant treatments with the most interaction terms that included year.

For the significant treatments identified above, years were classified as having a negative (less than), neutral (equal to) or positive (greater than) yield group response in relation to a given significant interaction between year and other treatment factors. For example, for the interaction between Year and Nserve, a year that using Nserve created higher yield than untreated fertilizer was categorized as "Positive"; a year that using Nserve created lower yield than untreated fertilizer was categorized as "Negative", and a year where no yield difference existed was categorized as "Neutral".

Thereafter, weather data were summed or averaged into increments of 1 to 15 weeks, starting with the N fertilizer application date(s) of each year. Weather variables summed were

Precip_mm, ET_mm, CHU_sum, and GDD_sum, and those averaged were Solar_MJm2d, RH_pct, WindSp_ms, SoilT10cm_C, Tmin_C, Tmax_C, SDI, and AWDR. Incremental weather data and year-yield potential (i.e. high, medium, low) were then input into a conditional inference tree (CIT; described below) to identify which variables had the strongest ability to predict the effectiveness of significant management treatments. In addition, the CIT identified the optimum time windows (1 to 15 weeks) after fertilizer application for which these weather variables had the strongest predictive power.

Conditional inference trees have been increasingly used for unstructured agricultural data such as weather (Mourtzinis et al., 2018a; b). The main advantages for using CIT are unbiasedness; avoidance of overfitting; robustness to outliers, missing data, multicollinearity and heteroscedasticity; handling of both continuous and categorical data; accounting of interactions among variables and of variables measured at different scales; and consideration of distributional properties of the measures (Hothorn et al., 2006).

Here, the CIT is used to identify significant weather predictors for yield level (i.e. variable selection), then partition those significant predictors in order of strongest to weakest effect (i.e. splitting). The effect level is pre-defined (here, $\alpha = 0.1$; see below) such that for all combinations of predictor variables (weather), the strongest predictor is identified (i.e. lowest p-value) and the CIT splits (i.e. node one), with each subsequent weather predictor branching the tree into subsequent nodes (i.e. nodes two through X) in order of predictive strength. These independence tests are conducted through a permutation test framework where all steps are recursively repeated until no significant independent predictors remain and the tree is complete (Hothorn et al., 2006).

For this work, the independence test was performed by using a Bonferroni-adjusted Monte-Carlo p-value ($\alpha = 0.1$). Further, each terminal node was ensured to account for at least 14% of the total observations to protect against overfitting and power loss. To prioritize simpler tree models, the maximum tree depth here was set to 10 nodes. After performing CIT, selected weather variables were fit into a logistic (two categorical outcomes) or ordinal multinomial (three categorical outcomes) regression algorithm to assess the probability of yield group responses to CIT-selected weather variables.

All statistical analyses were performed in R (R Development Core Team, 2017). The `lme` function from *nlme* package (Pinheiro et al., 2017) was used for soil and grain yield analysis. The `kmeans` function from *stats* package (R Core Team, 2017) was used for year-yield potential grouping. The `ctree` function from *partykit* package (Hothorn et al., 2006; Hothorn and Zeileis, 2015) was used for weather CIT analysis. The functions `glm` and `polr` from packages *stats* (R Core Team, 2017) and *MASS* (Venables and Ripley, 2002) were used to fit logistic and ordinal multinomial regression analysis, respectively.

Results

Soil Characterization

Soil properties at the 0-20 cm layer varied among different N rate treatments in 2000 (Figure 3.1). Nitrogen rate affected NO_3^- ($p < 0.001$), K ($p = 0.035$), and pH ($p < 0.001$). Nitrate did not differ among 0N, 75N, and 150N (average of 6.1 ppm) but was highest under 300N (17.3 ppm). Adding N fertilizer increased soil K relative to 0N, but no pairwise comparison between N rates were different at $\alpha = 0.1$. This is likely a result of the low number of replicates ($n=2$) and high degree of variability. Soil pH tended to decrease as N rate increased, with lowest pH under 300N (5.8) and pH 6.5 for all other N rates (range 6.45 to 6.65).

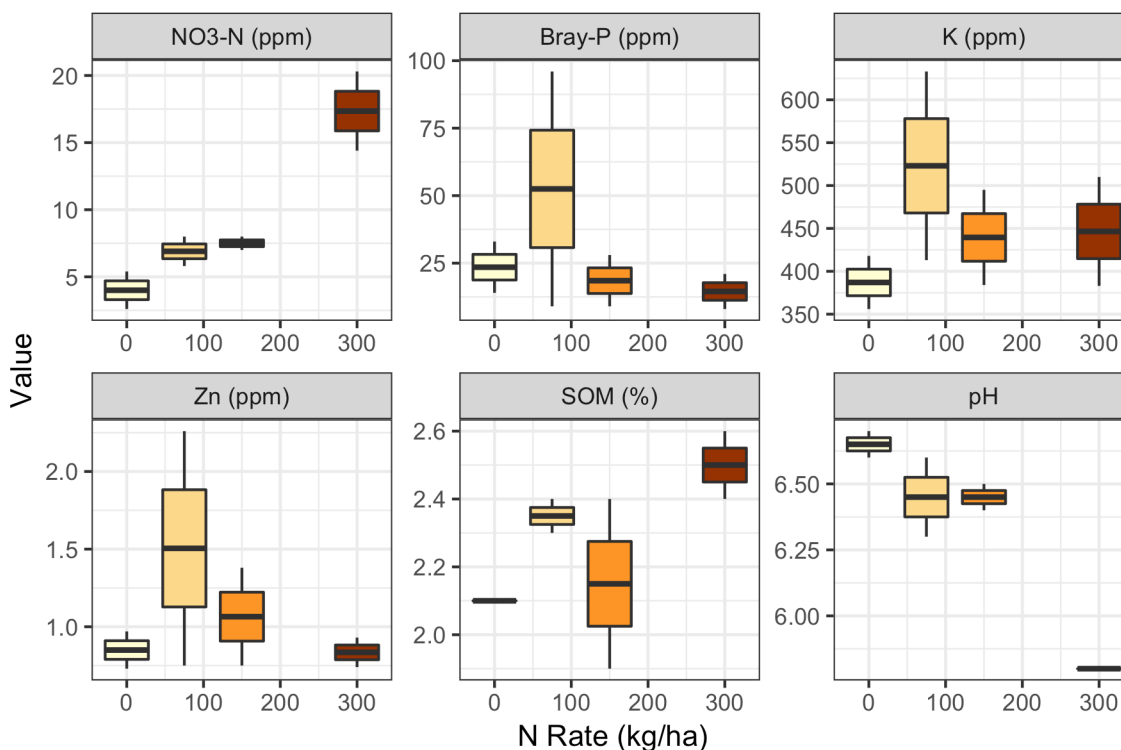


Figure 3.1. Soil properties boxplots (n=2) from N rate treatments (0N, 75N, 150N and 300N) at 0-20 cm depth in 2000.

Soil properties varied as a function of N rate, tillage type and Nserve at different soil depths in 2006 (Figure 3.2). Soil pH was only affected by one or more treatment factors at the 0-15, 15-30, 30-46, and 46-61 cm depths. Soil pH at the 0-15 cm depth was affected by N rate \times tillage ($p=0.02$), with the lowest pH under CT-300N. Soil pH at the 15-30, 30-46 and 46-61 cm depths was affected by N rate only, with 300N < 150N and differences between N rate treatments decreasing as depth increased.

Soil POM was affected by one or more treatment factors at the 0-15, 30-46 and 46-61 cm depths only. Soil POM at the 0-15 cm depth was affected by N rate \times Tillage \times Nserve ($p=0.008$), where POM was highest for 300N-RT-noNserve (7.35 Mg ha^{-1}) and lowest for both 300N-CT-noNserve (5.4 Mg ha^{-1}) and 300N-RT-Nserve (5.8 Mg ha^{-1}). Soil POM at the 30-46

cm depth was affected by Nserve only ($p=0.0002$), where POM was lower with vs. without Nserve (0.72 vs. 0.81 Mg ha^{-1}). Soil POM at the 46-61 cm depth was affected by the main effects of tillage ($p=0.007$) and Nserve ($p=0.04$), where POM was marginally higher in CT than RT (0.74 vs. 0.61 Mg ha^{-1} , $p=0.054$), and marginally lower with vs. without Nserve (0.63 vs. 0.73 Mg ha^{-1} , $p=0.073$).

Soil OM was affected by one or more treatment factors at the 15-30, 30-46, and 76-91 cm depths only. Soil OM at the 15-30 cm depth was marginally affected by tillage, with CT < RT (2.3 vs. 2.42% , $p=0.063$). Although SOM at the 30-46 cm depth was affected by tillage \times Nserve ($p=0.02$), no pairwise comparison was significant at $\alpha =0.1$ (varying from 1.93% for RT without Nserve to 2.15% for RT with Nserve). Soil OM at the 76-91 cm depth was affected by the main effect of Nserve ($p=0.01$) and N rate \times tillage ($p=0.03$), where SOM was lower with vs. without Nserve (1.24 vs. 1.32%), and lower in CT-150N (1.22%) vs. RT-150N (1.39%).

Soil K was only affected by one or more treatment factors at the 0-15, 15-30, 30-46, and 46-61 cm depths. Soil K at the 0-15 and 15-30 cm depths was affected by N rate ($p=0.005$ and 0.01 , respectively), with 300N numerically higher than 150N at both depths. Soil K at the 30-46 and 46-61 cm depths was affected by N rate \times Nserve ($p=0.042$ and 0.05 , respectively). In spite of the significant effect on the ANOVA, the N rate \times Nserve pairwise comparisons at these depths were not significant at $\alpha =0.1$, with numerically higher K under 300N without Nserve (488 and 524 ppm at the 30-46 and 46-61 cm depths, respectively) and lower under 150 without Nserve (407 and 478 ppm at the 30-46 and 46-61 cm depths, respectively).

Soil P was only affected by one or more treatment factors at the 15-30, 30-46 and 61-76 cm depths. Soil P at the 15-30 cm depth was affected by N rate \times tillage ($p=0.046$), and though no pairwise comparison was significant at $\alpha =0.1$, P was numerically highest at 300N under RT

only. Soil P at the 30-46 cm depth was significantly affected by N rate \times Nserve ($p=0.02$), however no pairwise comparison was significant at $\alpha =0.1$, with 300N with Nserve having the numerically greatest P concentration (6.7 ppm) and 150 N with Nserve having the lowest (5.2 ppm). Soil P at 61-76 cm depth was affected by N rate \times tillage \times Nserve ($p=0.048$), with adding Nserve decreased soil P in the 150N-RT treatment only (21.6 ppm with vs 8.4 ppm without).

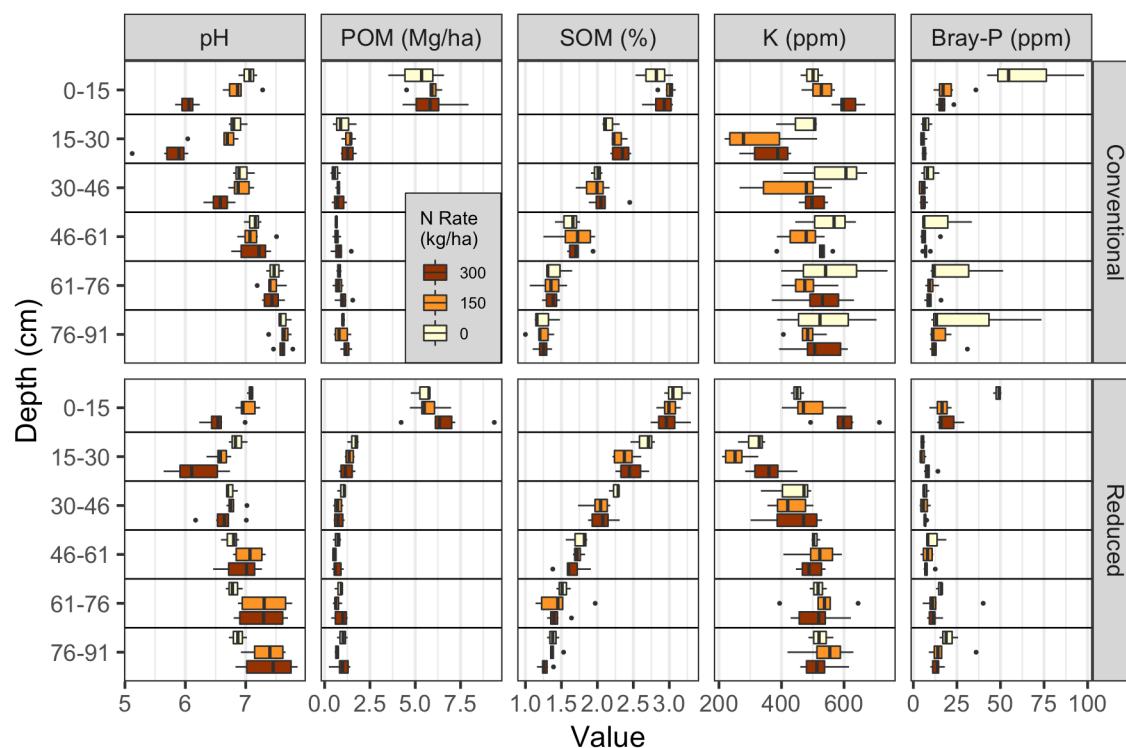


Figure 3.2. Soil properties boxplots (n=3 for 0N, n=6 for all others) at six depths from N rate (0N, 150N and 300N) and tillage (conventional and reduced) treatments across block and Nserve (with and without) treatment levels in 2006.

Weather Summary

Weather conditions were highly variable during the 28 yrs of study (Figure 3.3). For analysis purposes, weather variables were summarized [either summed (Precip_mm, ET_mm, CHU_sum, GDD_sum) or averaged (all others)] from 1 to 13 weeks after fertilizer application. Because each summarizing window creates unique data, weather variables from a period of 3

and 13 weeks after fertilizer application have been chosen for display. These two periods were approximately the ones that best explained most of the weather-related yield variability observed under different treatment factors and interactions (discussed below).

For the 3-week window after fertilizer application over all years, daily air minimum temperature ranged from 0.6 to 18°C and averaged 11°C. Cumulative corn heat units ranged from 137 to 497 and averaged 372. Daily solar radiation ranged from 11.4 to 24.5 and averaged 20 MJ m⁻² d⁻¹. Cumulative precipitation ranged from 16 to 182 mm and averaged 70.7 mm. SDI ranged from 0.14 (more concentrated) to 0.75 (more distributed) and averaged 0.46.

For the 13-week window after fertilizer application over all years, daily air minimum temperature ranged from 7.9 to 16.2°C and averaged 13.8°C over the years. Cumulative corn heat units (CHU_sum) ranged from 1296 to 2158 and averaged 1723. Daily solar radiation ranged from 17.5 to 23.2 and averaged 20.5 MJ m⁻² d⁻¹. Cumulative precipitation ranged from 114.4 to 543.8 mm and averaged 274.2 mm. SDI ranged from 0.48 to 0.69 and averaged 0.58.

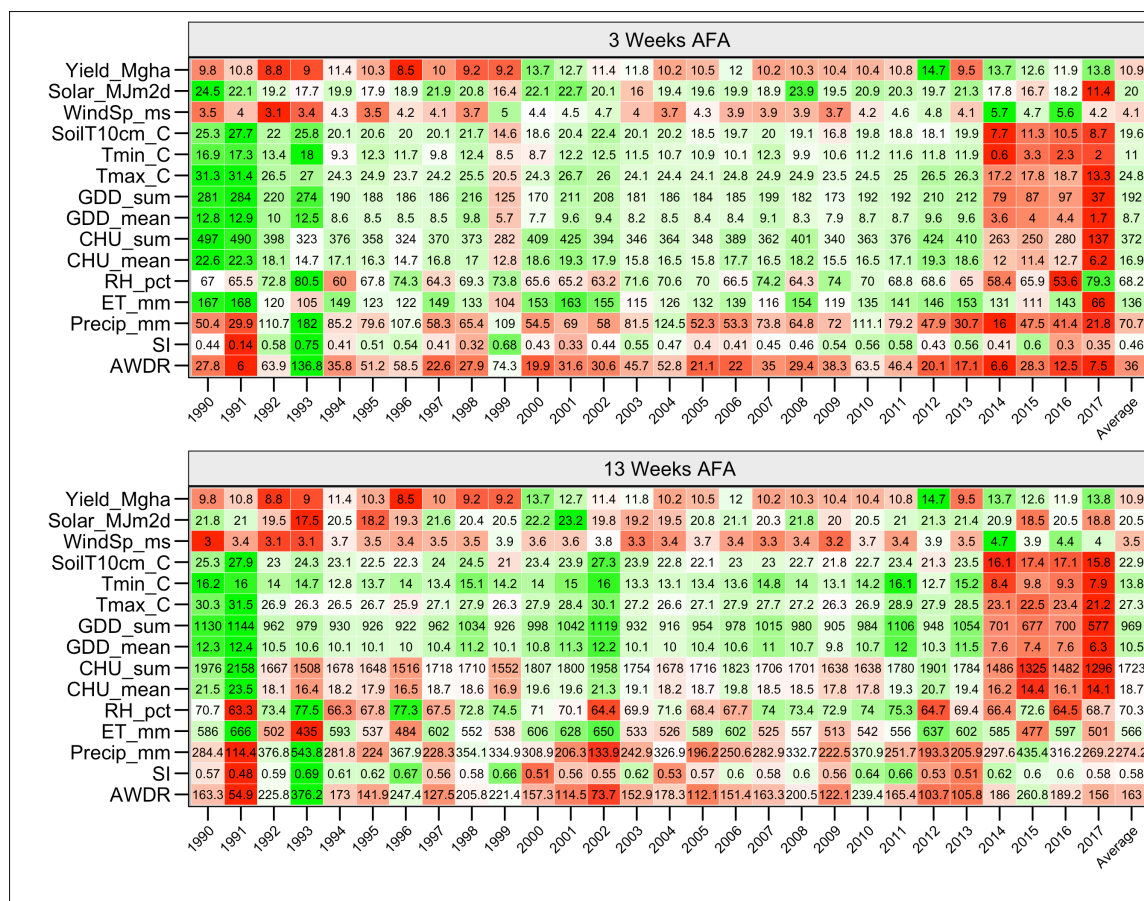


Figure 3.3. Daily weather variables and yield summary for windows of 3 and 13 weeks after fertilizer application. Red and green shades represent values that are closer to the minimum and maximum for a given variable over all years, respectively.

Grain Yield and Weather from Period One (1994 to 2013)

ANOVA

Corn grain yield from period one (1994 to 2013 except 1999, 19 years) was significantly affected by year, N rate, tillage, timing, Nserve, and many two- and three-way interactions between these variables at $\alpha=0.05$ (Table 3.1). Given that the main focus of period one was to assess the impact of timing on grain yield, alone and as part of interactions, expected marginal means were calculated for Year \times Ntiming \times Nserve ($p<0.05$), Year \times Nrate \times Ntiming ($p<0.01$),

and Year \times Ntiming ($p < 0.001$). Other significant three-way interactions not including Ntiming (i.e. Year \times Nrate \times Nserve) were explored using the period two dataset (discussed later).

Table 3.1. Grain yield analysis of variance table for period one (1994 to 2013 except 1999, 19 years).

Source	NumDF	DenDF	Pr(>Chisq)	Significance
(Intercept)	1	684	<0.001	***
Year	18	54	<0.001	***
Nrate	2	114	<0.001	***
Tillage	1	171	<0.001	***
Ntiming	1	342	<0.001	***
Nserve	1	684	<0.001	***
Year \times Nrate	36	114	<0.001	***
Year \times Tillage	18	171	<0.001	***
Nrate \times Tillage	2	171	<0.001	***
Year \times Ntiming	18	342	<0.001	***
Nrate \times Ntiming	2	342	<0.001	***
Tillage \times Ntiming	1	342	0.51	
Year \times Nserve	18	684	<0.001	***
Nrate \times Nserve	2	684	<0.01	**
Tillage \times Nserve	1	684	<0.001	***
Ntiming \times Nserve	1	684	<0.001	***
Year \times Nrate \times Tillage	36	171	0.91	
Year \times Nrate \times Ntiming	36	342	<0.01	**
Year \times Tillage \times Ntiming	18	342	0.83	
Nrate \times Tillage \times Ntiming	2	342	0.26	
Year \times Nrate \times Nserve	36	684	<0.01	**
Year \times Tillage \times Nserve	18	684	0.18	
Nrate \times Tillage \times Nserve	2	684	<0.05	*
Year \times Ntiming \times Nserve	18	684	<0.05	*
Nrate \times Ntiming \times Nserve	2	684	0.17	
Tillage \times Ntiming \times Nserve	1	684	0.13	
Year \times Nrate \times Tillage \times Ntiming	36	342	1	
Year \times Nrate \times Tillage \times Nserve	36	684	0.81	
Year \times Nrate \times Ntiming \times Nserve	36	684	0.23	
Year \times Tillage \times Ntiming \times Nserve	18	684	0.88	
Nrate \times Tillage \times Ntiming \times Nserve	2	684	0.84	
Year \times Nrate \times Tillage \times Ntiming \times Nserve	36	684	0.84	

Year \times Ntiming \times Nserve

The use of Nserve significantly increased grain yield (from 0.37 to 0.73 more Mg ha^{-1}) in six out of 19 years when fertilizer was PP applied, compared to one occasion (0.36 Mg ha^{-1}) when fertilizer was SD applied (Figure 3.4). The use of Nserve decreased yield in one occasion for each fertilizer timing, 0.35 and 0.41 less Mg ha^{-1} for PP and SD applied, respectively, compared to not using Nserve.

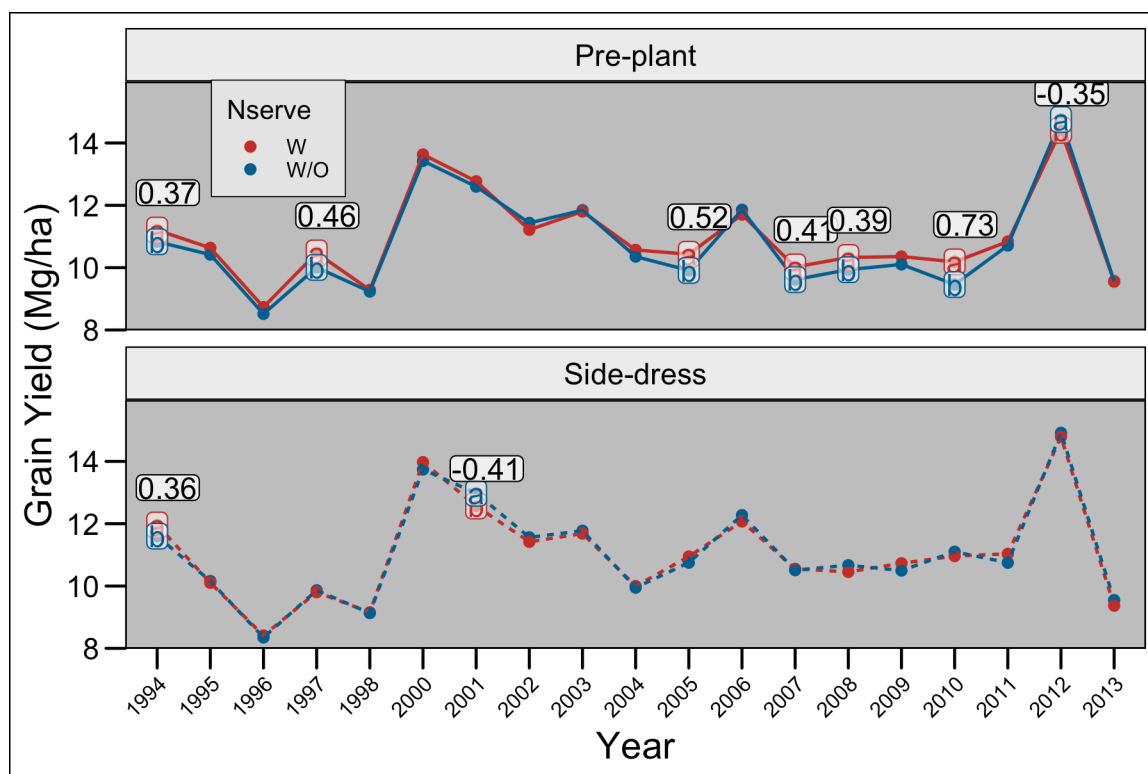


Figure 3.4. Mean (n=24) corn grain yield as affected by year, fertilizer timing (pre-plant vs. side-dress) and Nserve (W=with vs. W/O=without) for period one (1994 through 2013 except for 1999). Means within a given year and fertilizer timing followed by a common letter are not significantly different at $\alpha=0.05$. Numbers in the plot area represent Δ yield calculated as $\text{Yield}_{\text{withNserve}} - \text{Yield}_{\text{withoutNserve}}$, in Mg ha^{-1} .

Based on the yield responses observed for Nserve (with, W; without, W/O) for its 3-way interaction with Year and Ntiming (Figure 3.4), annual yield outcomes ($\text{Yield}_{\text{withNserve}} - \text{Yield}_{\text{withoutNserve}}$) were classified as $W < W/O$ (2001, 2012), $W > W/O$ (1994, 1997, 2005, 2007,

2008, 2010) and $W = W/O$ (all other years). The optimum prediction window for assessing weather impacts on Nserve effectiveness was 3 weeks after fertilizer application. The CIT-selected weather variables with the strongest predictive power for Nserve effectiveness were: year-yield potential (high vs. medium and low categories) and air minimum temperature ($T_{min_C} = 6^{\circ}C$; Figure 3.5), with a collective predictive accuracy of 86%. Using Nserve during high-yielding years either had no effect (78% of years) or a negative effect (22% of years) on yield, but never a positive effect (0% of years). For medium/low-yielding years, using Nserve had a positive effect on yield when $T_{min_C} < 6^{\circ}C$ during the 3-week window after fertilizer application for 60% of years and no effect for 40% of years. When $T_{min_C} > 6^{\circ}C$, Nserve benefits dropped to only 8% of years, with no effect occurring for 92% of year

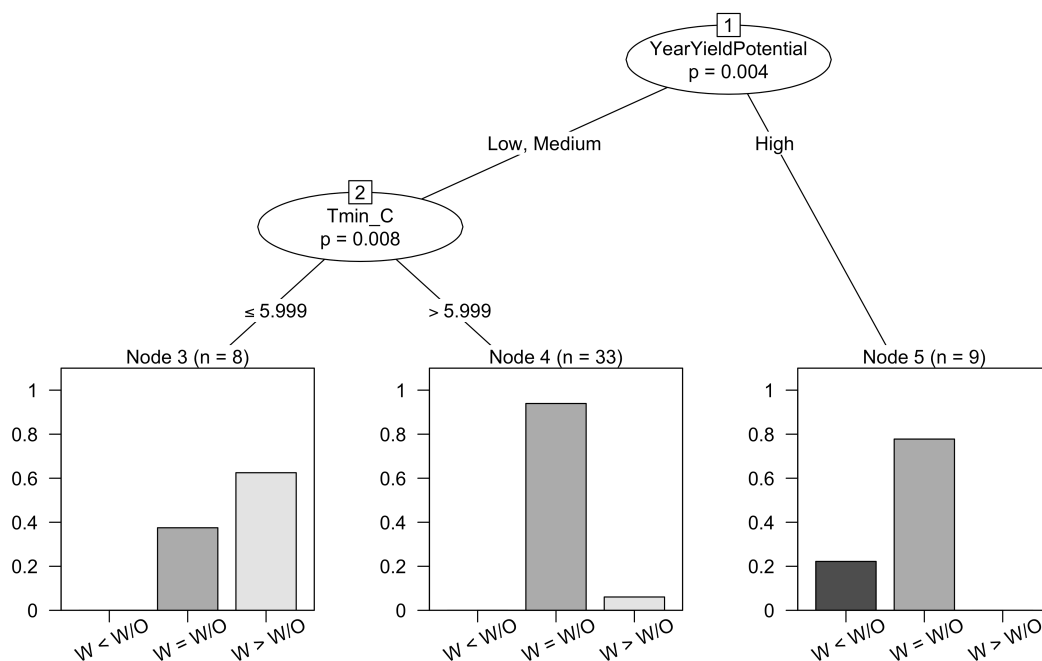


Figure 3.5. Conditional inference tree for the response of Nserve (negative, neutral or positive based on $\text{Yield}_{\text{withNserve}} - \text{Yield}_{\text{withoutNserve}}$ from the Year \times Ntiming \times Nserve interaction) across year and timing over three weeks after fertilizer application. Each terminal node contains the ratio of cases in each yield response category. Note: n= number of observations, YearYieldPotential is the year-yield potential grouping (High from 12.6 to 14.7 Mg ha⁻¹, Medium from 10 to 12 Mg ha⁻¹ and Low from 8.5 to 9.8 Mg ha⁻¹).

The probabilities of observing Nserve yield responses to Tmin_C based on the Year \times Ntiming \times Nserve interaction was calculated for each year-yield potential group (Figure 3.6). For high-yielding years, the probability of observing a negative Nserve yield response increased as Tmin_C increased. For medium-yielding years, the probability of observing a positive Nserve yield response decreased as Tmin_C increased. For low-yielding years, using Nserve had a 100% probability of having no effect on yield (i.e. neutral).

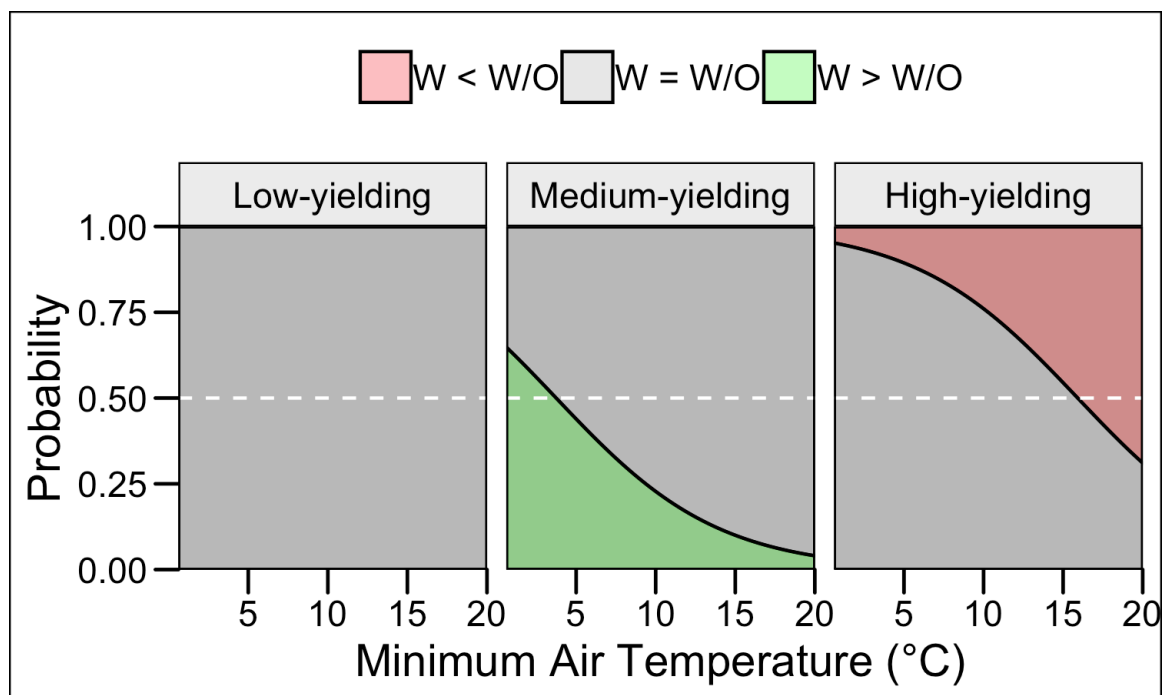


Figure 3.6. Ordinal multinomial regression for the response of Nserve (negative, neutral or positive based on $\text{Yield}_{\text{withNserve}} - \text{Yield}_{\text{withoutNserve}}$ from the $\text{Year} \times \text{Ntiming} \times \text{Nserve}$ interaction) as impacted by year-yield potential and average air minimum temperature over three weeks after fertilizer application.

Year × Nrate × Ntiming

The effect of fertilizer application timing and N rate on grain yield varied among years (Figure 3.7). Side-dressed N fertilizer increased grain yield compared to PP application nine years at 75N, three years at 150N, and one year at the 300N rate. Pre-planting applied N fertilizer increased grain yield compared to SD for one year at 150N and three years at 300N.

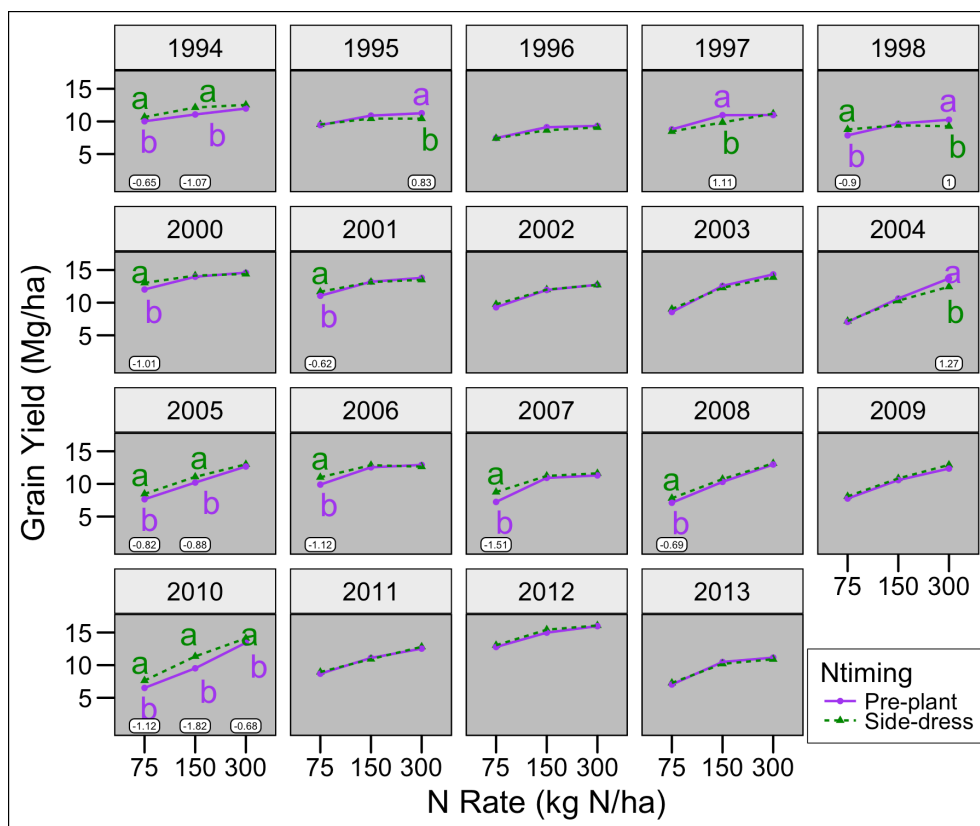


Figure 3.7. Mean ($n=16$) corn grain yield as affected by year, N rate (75, 150 and 300 kg N ha^{-1}) and fertilizer timing (pre-plant vs side-dress) for period one (1994 through 2013 except for 1999). Means within a given year and N rate followed by a common letter are not significantly different at $\alpha=0.05$. Numbers in the plot area represent Δ yield calculated as $\text{Yield}_{\text{pre-plant}} - \text{Yield}_{\text{side-dress}}$, in Mg ha^{-1} .

Based on the yield responses observed for Ntiming (pre-plant, PP; side-dress, SD) for its 3-way interaction with Year and Nrate (Figure 3.7), annual yield outcomes ($\text{Yield}_{\text{pre-plant}} - \text{Yield}_{\text{side-dress}}$) were classified as $\text{PP} > \text{SD}$ (1995, 1997, 1998, 2004), $\text{PP} < \text{SD}$ (1994, 2000, 2001, 2005, 2006, 2007, 2008, 2010) and $\text{PP} = \text{SD}$ (all others). The optimum prediction window for assessing weather impacts on Ntiming effectiveness was 13 weeks after fertilizer application. The CIT-selected weather with the strongest predictive power for Ntiming effect were: solar radiation ($\text{Solar_Mjm2d} = 19 \text{ MJ m}^{-2} \text{ d}^{-1}$), year-yield potential (low vs. medium and high categories) and precipitation ($\text{Precip_mm} = 211 \text{ mm}$, Figure 3.8), with a collective predictive

accuracy of 70%. Positive yield response from PP fertilizer application was observed with the highest frequency (70% of years) when solar radiation was below $19.4 \text{ MJ m}^{-2} \text{ d}^{-1}$. Negative yield response from PP fertilizer application was observed when solar radiation was above $19.4 \text{ MJ m}^{-2} \text{ d}^{-1}$ in high- and medium-yielding years, and at a higher frequency (60% of years) when cumulative precipitation was above 211 mm. Neutral response (no yield difference between PP and SD) was observed in all scenarios, with the highest probability (80% of years) when solar radiation was above $19.4 \text{ MJ m}^{-2} \text{ d}^{-1}$ under low-yielding years.

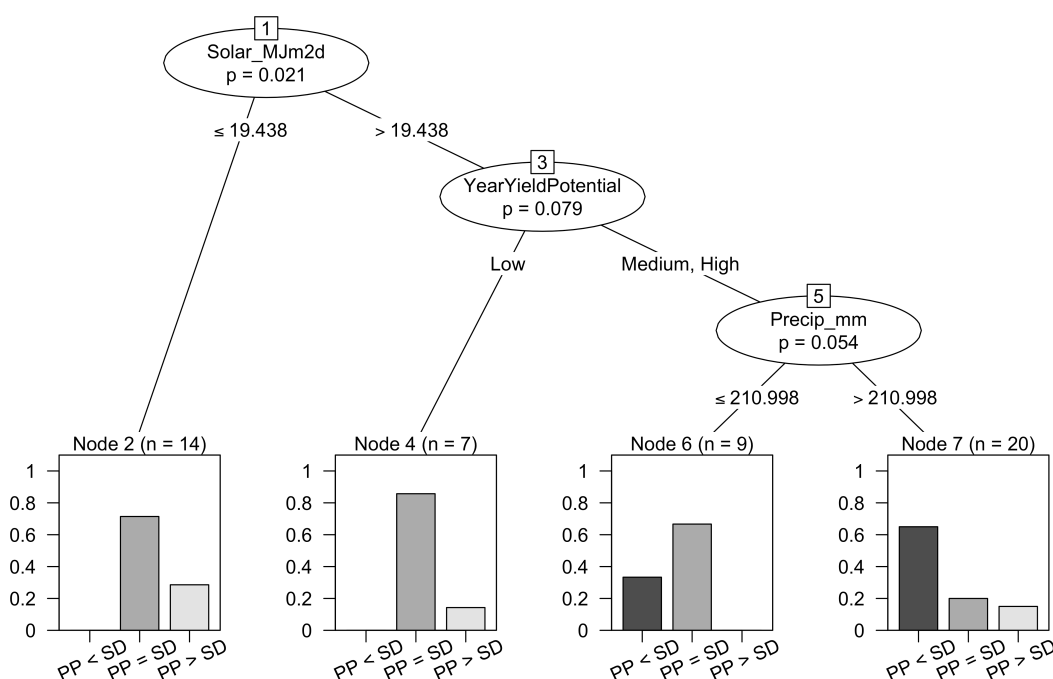


Figure 3.8. Conditional inference tree for the response of application timing (negative, neutral or positive based on $\text{Yield}_{\text{pre-plant}} - \text{Yield}_{\text{side-dress}}$ from the Year \times Nrate \times Ntiming interaction) across year and N rate over 13 weeks after fertilizer application. Each terminal node contains the ratio of cases in each yield response category. Note: n= number of observations, Solar_MJm2d is the average solar radiation in $\text{MJ m}^{-2} \text{d}^{-1}$, YearYieldPotential is the year-yield potential grouping (High from 12.6 to 14.7 Mg ha^{-1} , Medium from 10 to 12 Mg ha^{-1} and Low from 8.5 to 9.8 Mg ha^{-1}), Precip_mm is the cumulative precipitation in mm.

The probabilities of observing different yield response groups based on the Year \times Nrate \times Ntiming interaction as affected by solar radiation, year-yield potential and cumulative precipitation are shown on Figure 3.9. In general, the probability of observing a decrease in yield from PP compared to SD (negative) increased as both cumulative precipitation and solar radiation increased. The highest probability of a negative yield response (PP lower than SD) was at higher cumulative precipitation and higher average solar radiation conditions under medium-

and high-yielding years. The highest probability of a positive yield response (PP higher than SD) was at lower cumulative precipitation and lower average solar radiation conditions under low-yielding years.

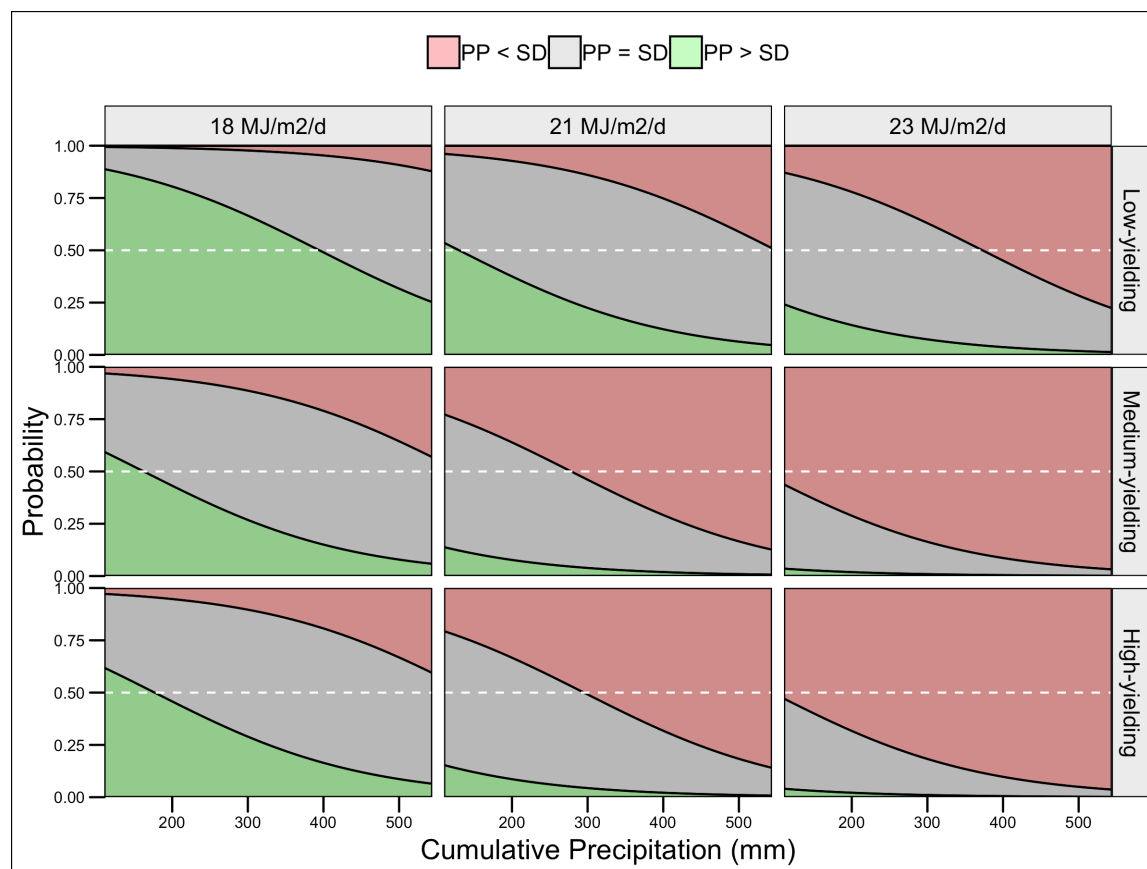


Figure 3.9. Ordinal multinomial regression for the response of application timing (negative, neutral or positive based on $\text{Yield}_{\text{pre-plant}} - \text{Yield}_{\text{sid-edress}}$ from the $\text{Year} \times \text{Nrate} \times \text{Ntiming}$ interaction) as impacted by year-yield potential, solar radiation quantile levels and cumulative precipitation over 13 weeks after fertilizer application.

Year × Ntiming

Grain yield was significantly affected by application timing over the years (Figure 3.10). SD N fertilizer produced higher grain yield (from 0.38 to 1.21 Mg ha⁻¹ more grain) than PP applications in seven out of 19 years. PP fertilizer application produced higher grain yield (from 0.39 to 0.49 Mg ha⁻¹ more grain) than SD in three out of 19 years.

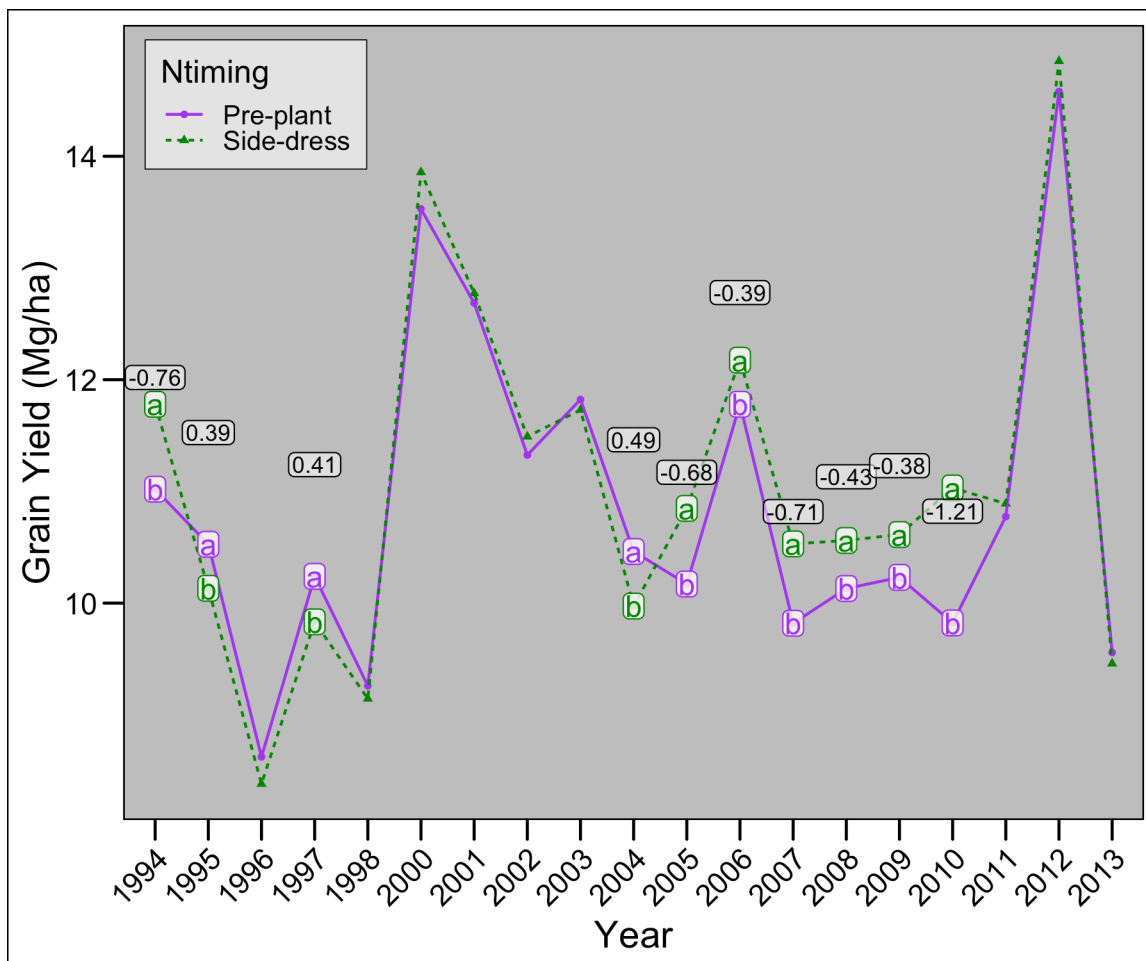


Figure 3.10. Mean ($n=48$) corn grain yield as affected by year, and fertilizer timing (pre-plant vs. side-dress) for period one (1994 through 2013 except for 1999). Means within a given year followed by a common letter are not significantly different at $\alpha=0.05$. Numbers in the plot area represent Δ yield calculated as $\text{Yield}_{\text{pre-plant}} - \text{Yield}_{\text{side-dress}}$, in Mg ha^{-1} .

Based on the yield responses observed for the Ntiming (pre-plant, PP; side-dress, SD) for its 2-way interaction with Year (Figure 3.10), annual yield outcomes ($\text{Yield}_{\text{pre-plant}} - \text{Yield}_{\text{side-dress}}$) were classified as PP > SD (1995, 1997, 2004), PP < SD (1994, 2005, 2006, 2007, 2008, 2009, 2010) and PP = SD (all others). The optimum prediction window for assessing weather impacts on Ntiming effect was 15 weeks after fertilizer application. The CIT-selected weather variables with the strongest predictive power for Ntiming effect were: year-yield potential (low and high

vs. medium categories) and precipitation (Precip_mm = 219 mm, Figure 3.11), with a collective predictive accuracy of 78%. Negative yield response from PP application was observed only under medium-yielding years, with highest frequency (60% of years) when cumulative precipitation was above 219 mm. Positive yield response from PP application was also observed in medium-yielding years, and at a higher frequency (20% of years) when cumulative precipitation was above 219 mm. Neutral response (no yield difference between PP and SD) was observed in all scenarios, with the highest probability (100% of years) occurring under low- and high-yielding years.

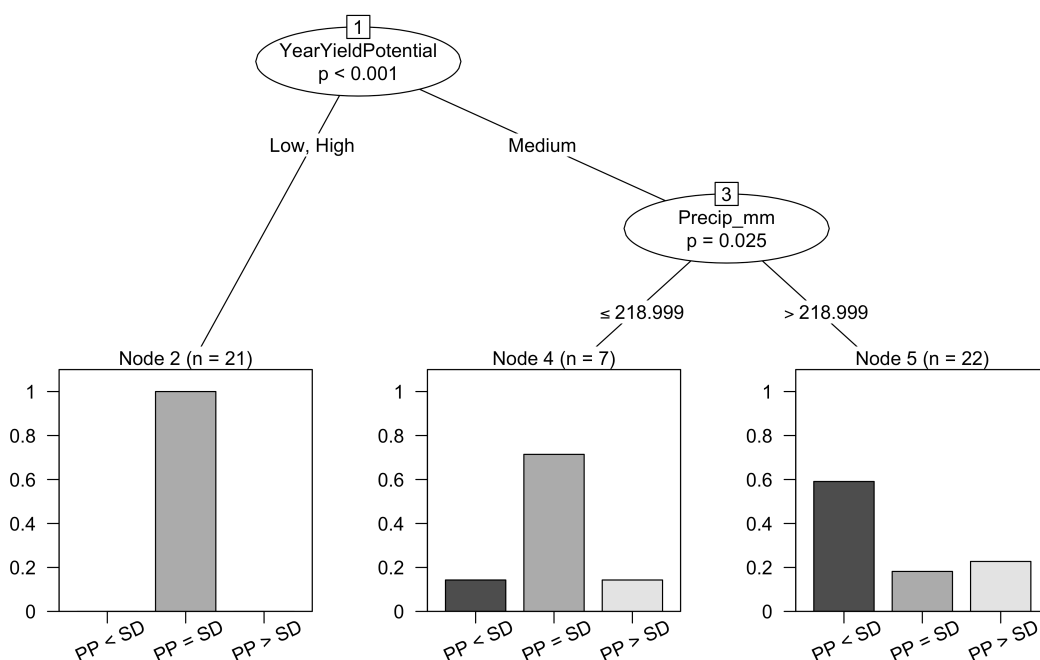


Figure 3.11. Conditional inference tree for the response of application timing (negative, neutral or positive based on Yield_{pre-plant} - Yield_{side-dress} from the Year × Ntiming interaction) across year over 15 weeks after fertilizer application. Each terminal node contains the ratio of cases in each yield response category. Note: n= number of observations, YierYieldPotential is the year-yield potential grouping (High from 12.6 to 14.7 Mg ha⁻¹, Medium from 10 to 12 Mg ha⁻¹ and Low from 8.5 to 9.8 Mg ha⁻¹), Precip_mm is the cumulative precipitation in mm.

The probabilities of observing different yield response groups based on the Year \times Ntiming interaction as affected by year-yield group and cumulative precipitation are shown on Figure 3.12. In general, the probability of observing a decrease in yield from PP compared to SD (negative) increased as cumulative precipitation increased for all year-yield groups. However, the probability of a negative impact of PP vs. SD increased at a faster rate under medium-yielding years. The highest probability of a positive yield response (PP higher than SD) was at lower cumulative precipitation under low and high-yielding years.

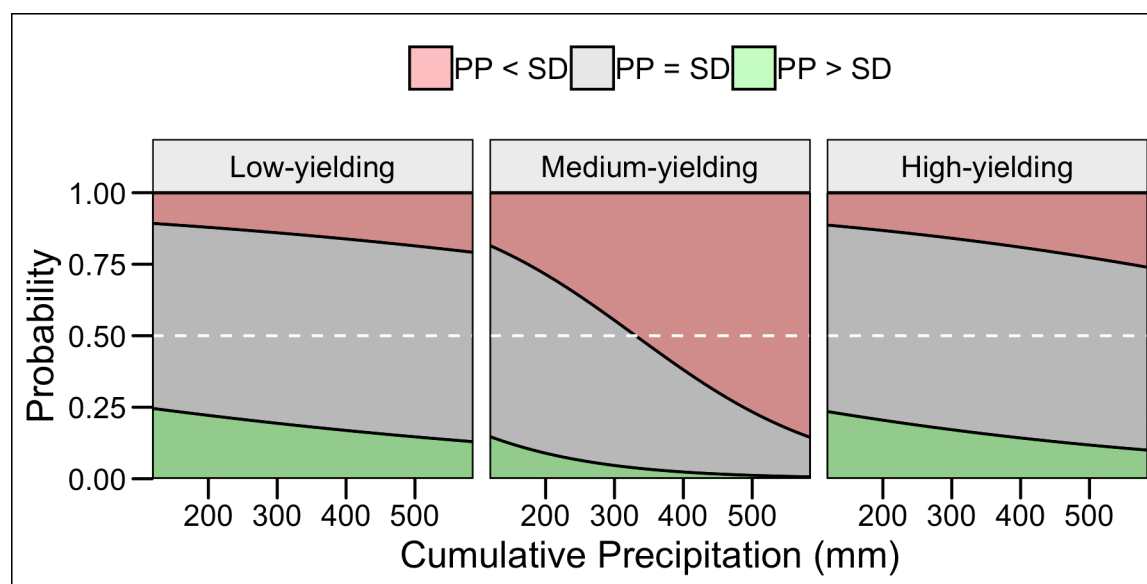


Figure 3.12. Ordinal multinomial regression for the response of application timing (negative, neutral or positive based on $\text{Yield}_{\text{pre-plant}} - \text{Yield}_{\text{side-dress}}$ from the Year \times Ntiming interaction) as impacted by year-yield potential and cumulative precipitation over 15 weeks after fertilizer application.

Grain Yield from Period Two (1990 to 2017, 28 years)

ANOVA

Corn grain yield from period two (1990 to 2017, 28 years) was significantly affected by year, N rate, tillage, Nserve, many two-way interactions between these variables and one three-way interaction at $\alpha=0.05$ (Table 3.3). Expected marginal means were calculated for Year \times Nrate \times Nserve ($p<0.001$), Year \times Tillage ($p<0.001$), Year \times Nrate ($p<0.001$), Tillage \times Nserve ($p<0.001$), and Nrate \times Tillage ($p<0.001$).

Table 3.2. Grain yield analysis of variance table for period two (1990 to 2017, 28 years).

Source	NumDF	DenDF	Pr(>Chisq)	Significance
(Intercept)	1	504	<0.001	***
Year	27	81	<0.001	***
Nrate	2	168	<0.001	***
Tillage	1	252	<0.001	***
Nserve	1	504	<0.05	*
Year \times Nrate	54	168	<0.001	***
Year \times Tillage	27	252	<0.001	***
Nrate \times Tillage	2	252	<0.001	***
Year \times Nserve	27	504	<0.001	***
Nrate \times Nserve	2	504	0.12	
Tillage \times Nserve	1	504	<0.001	***
Year \times Nrate \times Tillage	54	252	0.93	
Year \times Nrate \times Nserve	54	504	<0.001	***
Year \times Tillage \times Nserve	27	504	0.05	
Nrate \times Tillage \times Nserve	2	504	0.29	
Year \times Nrate \times Tillage \times Nserve	54	504	0.99	

Year \times Nrate \times Nserve

Grain yield was affected by the interaction between Year \times Nrate \times Nserve (Figure 3.13). Positive responses to Nserve were observed 4, 2 and 3 times at 75N, 150N and 300N, respectively, ranging from 0.4 to 0.6 Mg ha⁻¹ more grain compared to fertilizer alone. Negative responses were observed 2, 1 and 1 times at 75N, 150N and 300N, respectively, ranging from 0.5

to 0.7 Mg ha^{-1} less grain compared to fertilizer alone. All other years for all N rate treatments showed no response to Nserve.

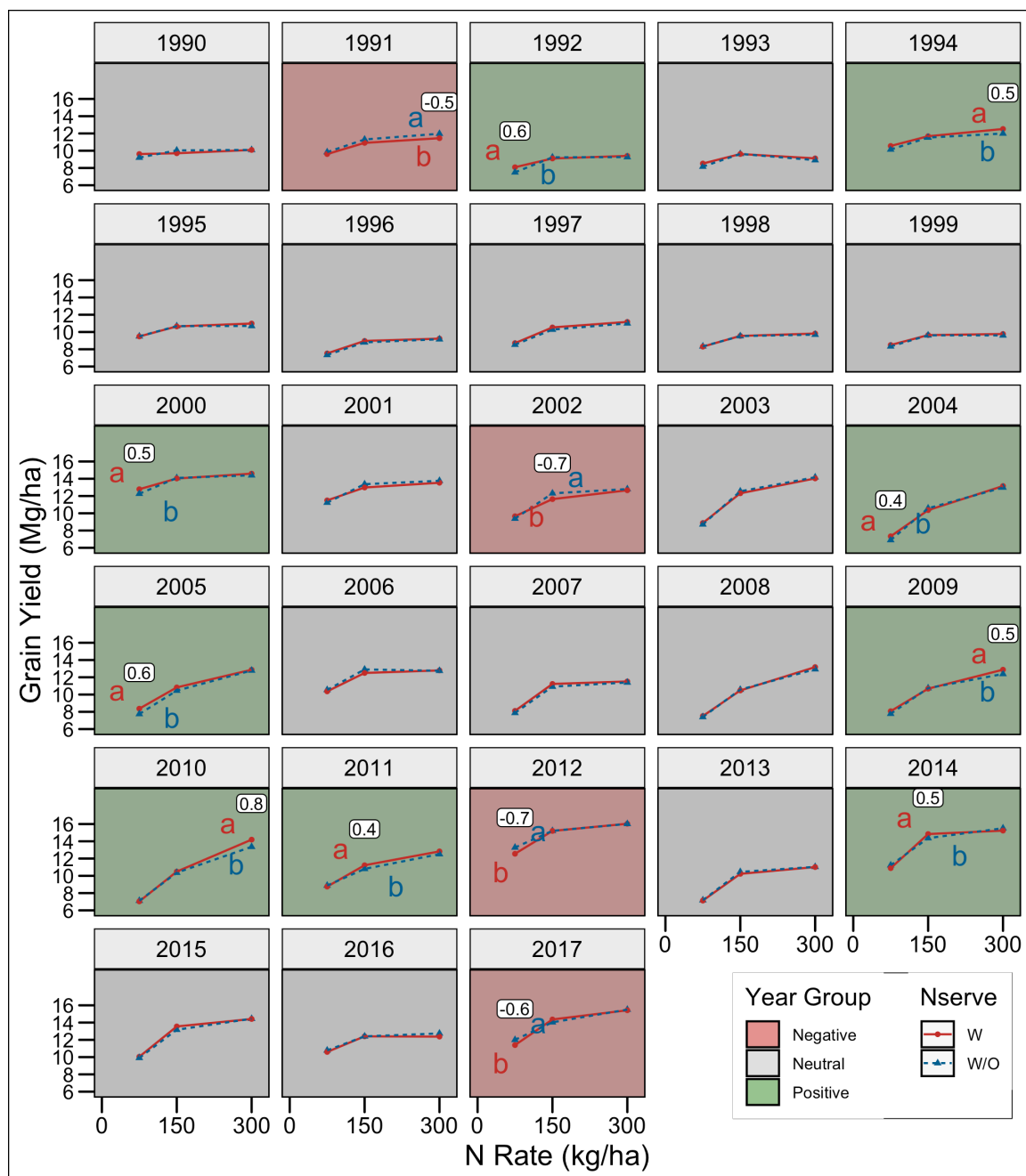


Figure 3.13. Mean ($n=8$) corn grain yield as affected by N rate (75, 150 and 300 kg N ha⁻¹), year and Nserve (W=with vs. W/O=without) for period two (1990 through 2017). Means within a given year followed by a common letter are not significantly different at $\alpha=0.05$. Green-shaded panels indicate that the use of NI statistically increased yield in at least one N rate. Red shaded panels indicate that the use of NI statistically decreased yield in at least one N rate. Numbers in the plot area represent Δ yield calculated as Yield_{withNserve} - Yield_{withoutNserve}, in Mg ha⁻¹.

Based on the yield responses observed for the Year \times Nrate \times Nserve interaction (Figure 3.13), years were classified as $W < W/O$ (1991, 2002, 2012, 2017), $W > W/O$ (1992, 1994, 2000, 2004, 2005, 2009, 2010, 2011, 2014) and $W = W/O$ (all others) based on $Yield_{withNserve} - Yield_{withoutNserve}$. The optimum prediction window for assessing weather impacts on Nserve effectiveness was 10 weeks after fertilizer application. The CIT-selected weather variables with the strongest predictive power for Nserve effectiveness were: precipitation (Precip_mm = 142 mm), year-yield potential (high and medium vs. low categories), and cumulative heat units (CHU_sum = 15) (Figure 3.14), with a predictive accuracy of 66%. For all yield potential groups, yield responses to Nserve were predominantly negative when Precip_mm < 142 mm (65% of years) or no N serve effect (35% of years). When Precip_mm > 142 mm, there was no Nserve effect for 85% of low-yielding years and a positive effect in 15% of those years, but no negative Nserve responses under these conditions. For Precip_mm > 142 mm, positive Nserve yield responses occurred in 60% of medium/high-yielding years when CHU_sum > 15, with no effect of Nserve in 40% of those years and no negative Nserve responses occurring under these conditions. When CHU_sum < 15, there was no response to Nserve in 55% of medium/high-yielding years, a positive response in 30% of those years, and a negative response in 15% of those years.

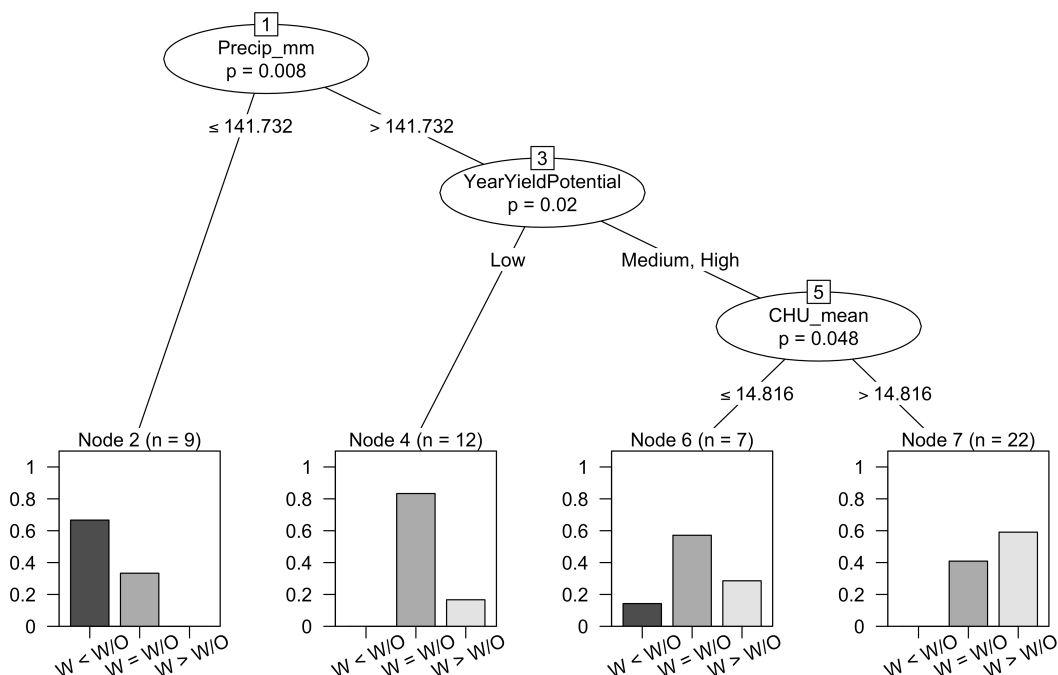


Figure 3.14. Conditional inference tree for the response of Nserve (negative, neutral or positive based on $\text{Yield}_{\text{withNserve}} - \text{Yield}_{\text{withoutNserve}}$ from the Year \times Nrate \times Nserve interaction) across year and N rate over 10 weeks after fertilizer application. Each terminal node contains the ratio of cases in each yield response category. Note: n= number of observations, Precip_mm is the cumulative precipitation in mm, YearYieldPotential is the year-yield potential grouping (High from 12.6 to 14.7 Mg ha⁻¹, Medium from 10 to 12 Mg ha⁻¹ and Low from 8.5 to 9.8 Mg ha⁻¹), and CHU_mean is the average corn heat units.

The probabilities of observing yield responses to Precip_mm and CHU_sum based on the Year \times Nrate \times Nserve interaction was calculated for each year-yield potential group (Figure 3.15). In general, the probability of negative yield responses to Nserve was greatest under i) low CHU, high Precip, and ii) high CHU, low Precip. The probability of positive yield responses to Nserve was greatest under i) low CHU, low Precip, and ii) high CHU, high Precip.

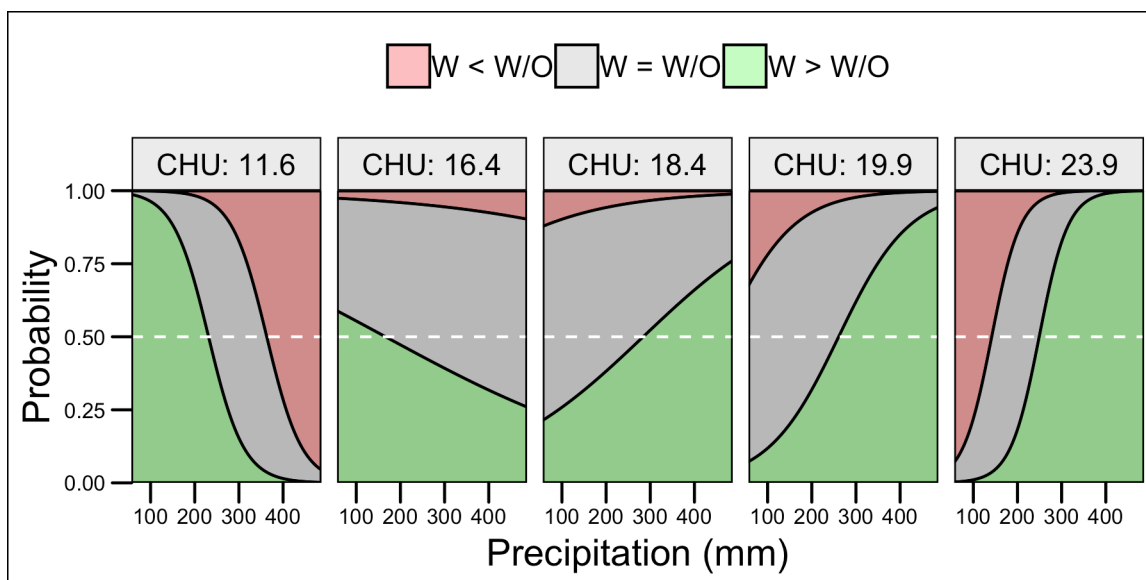


Figure 3.15. Ordinal multinomial regression for the response of Nserve (negative, neutral or positive based on $\text{Yield}_{\text{withNserve}} - \text{Yield}_{\text{withoutNserve}}$ from the Year \times Nrate \times Nserve interaction) as impacted by average corn heat units (CHU) and cumulative precipitation over 10 weeks after fertilizer application.

Year \times Tillage

Grain yield was significantly affected by tillage over the years (Figure 3.16). Grain yields were higher under CT in 11 out of 28 years, ranging from 0.5 to 2 Mg ha⁻¹ more grain than RT. There was no tillage difference in yields for the remaining study years (17 yrs), and no years when grain yield in RT was greater than CT.

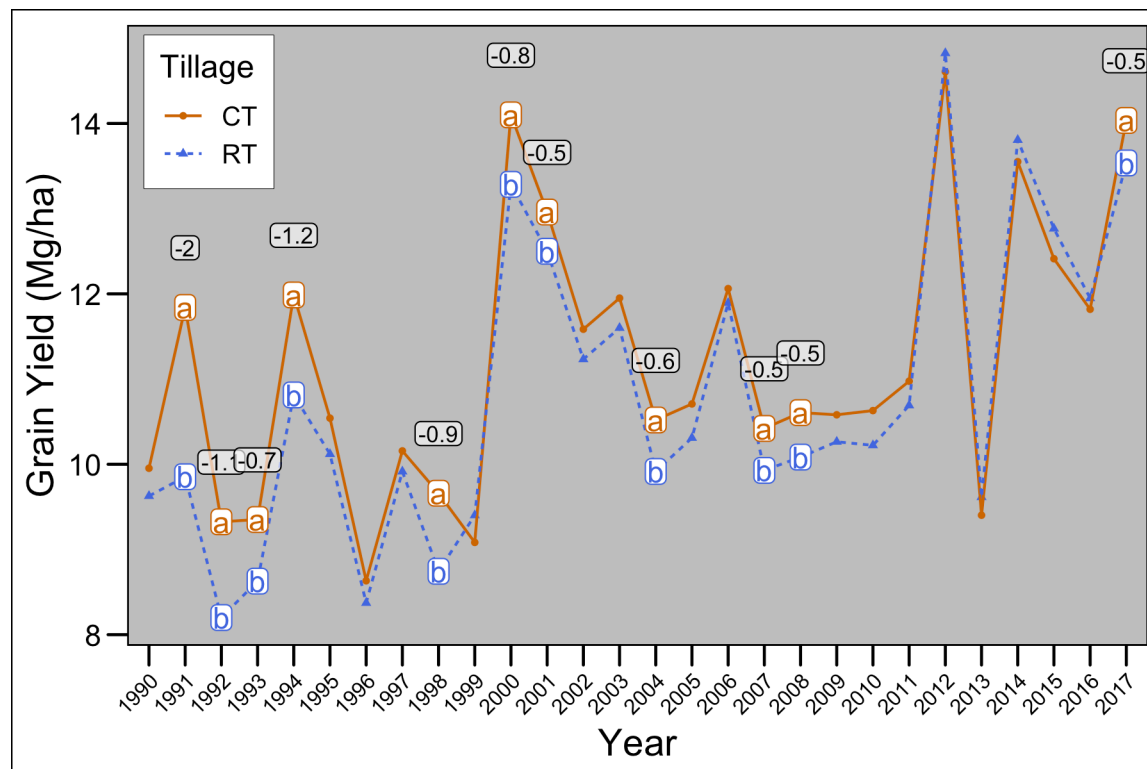


Figure 3.16. Mean (n=24) corn grain yield as affected by tillage (CT=conventional tillage vs. RT=reduced tillage) for period two (1990 through 2017). Means within a given year followed by a common letter are not significantly different at $\alpha=0.05$. Numbers in the plot area represent Δ yield calculated as $\text{Yield}_{\text{RT}} - \text{Yield}_{\text{CT}}$, in Mg ha^{-1} .

Based on the yield responses observed for the Year \times Tillage interaction (Figure 3.16), years were classified as RT < CT (1991, 1992, 1993, 1994, 1998, 2000, 2001, 2004, 2007, 2008, 2017) or RT = CT (all others) based on $\text{Yield}_{\text{RT}} - \text{Yield}_{\text{CT}}$. The optimum prediction window for assessing weather impacts on tillage response was two weeks after fertilizer application. The CIT-selected weather variables with the strongest predictive power for tillage response were: average wind speed ($\text{WindSp}_{\text{ms}} = 3.6 \text{ m s}^{-1}$), precipitation Shannon diversity index ($\text{SDI} = 0.32$), and average air minimum temperature ($\text{Tmin}_{\text{C}} = 4.5^{\circ}\text{C}$) (Figure 3.17), with a predictive accuracy of 80%. Using RT when $\text{WindSp}_{\text{ms}} < 3.6 \text{ m s}^{-1}$ resulted in negative yield responses

80% of years and no response in 20% of years compared to CT. When $\text{WindSp_ms} > 3.6 \text{ m s}^{-1}$ and $\text{SDI} < 0.32$, negative yield response to RT occurred in 75% of years and no response occurred in 25% of years. When $\text{WindSp_ms} > 3.6 \text{ m s}^{-1}$ and $\text{SDI} > 0.32$, negative yield responses occurred in 50% of years when the Tmin_C of the 2-week window after N application was $< 4.5^\circ\text{C}$, but the negative effect of RT was greatly diminished to no effect in 95% of years when $\text{Tmin_C} > 4.5^\circ\text{C}$.

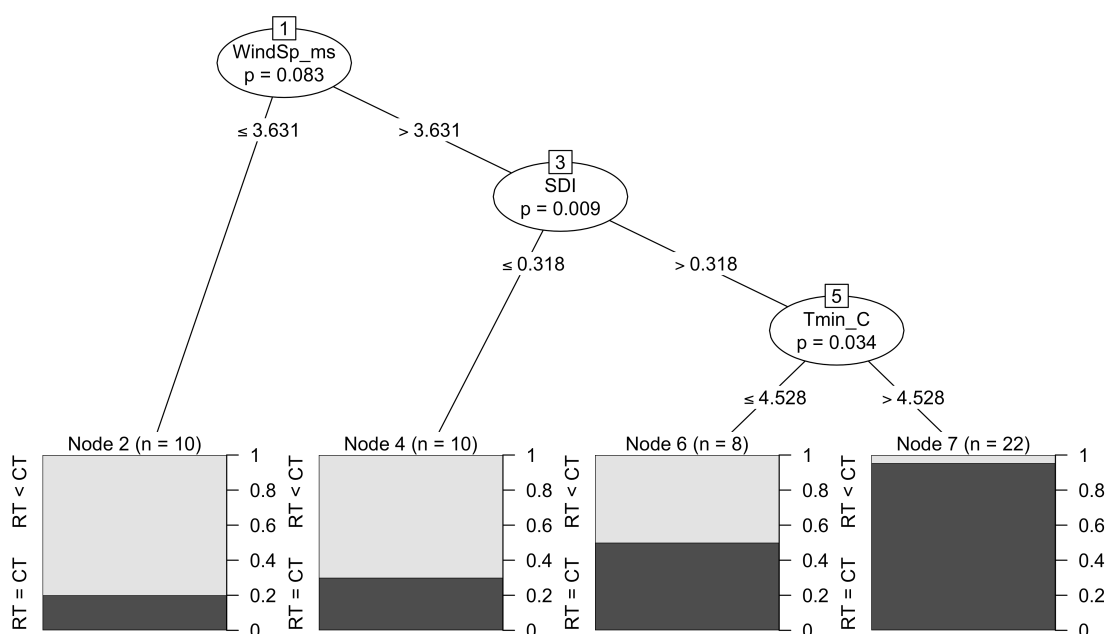


Figure 3.17. Conditional inference tree for the response of tillage (negative or neutral based on $\text{Yield}_{\text{reducedtillage}} - \text{Yield}_{\text{conventionaltillage}}$ from the $\text{Year} \times \text{Tillage}$ interaction) across year over two weeks after fertilizer application. Each terminal node contains the ratio of cases in each yield response category. Note: n = number of observations, WindSp_ms is the average wind speed in m s^{-1} , SDI is the precipitation Shannon diversity index, and Tmin_C is the average air minimum temperature in $^\circ\text{C}$.

The probabilities of observing a tillage-related yield response to WindSp , SDI , and Tmin based on the $\text{Year} \times \text{Tillage}$ interaction are shown on Figure 3.18. In general, negative yield

responses to RT compared to CT were most probable when WindSp was intermediate (4.3 m s^{-1}) and precipitation was highly concentrated (SDI=0), regardless of T_{min}. At higher WindSp, negative yield responses to RT became less probable as both T_{min} and SDI increased, except for SDI=0 when RT < CT increased with T_{min}. The probability of observing no tillage effect on yield was greatest when WindSp was intermediate (4.3 m s^{-1}) and precipitation was highly distributed (SDI=0.76), with a minor effect of T_{min}.

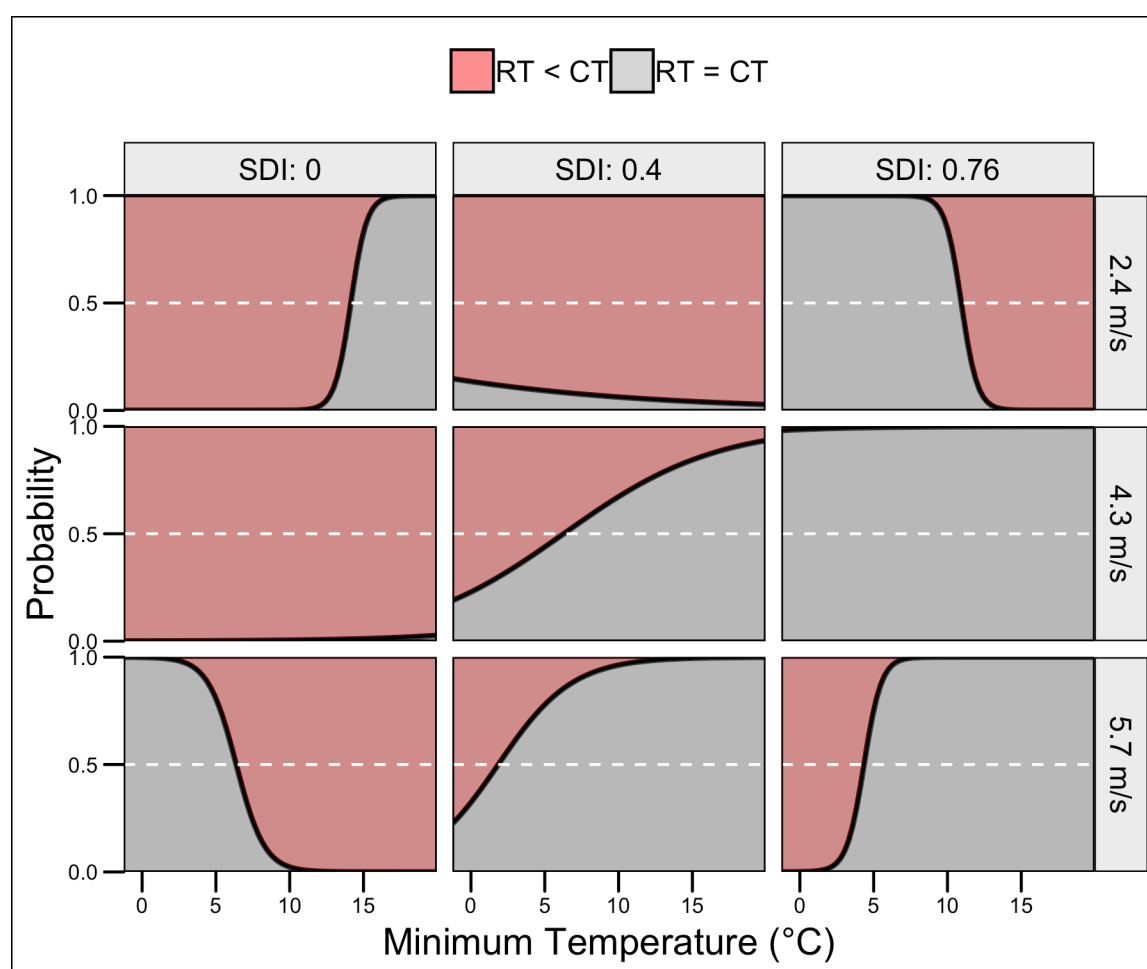


Figure 3.18. Logistic regression for the response of Tillage (negative or neutral based on $\text{Yield}_{\text{reducedtillage}} - \text{Yield}_{\text{conventionaltillage}}$ from the Year \times Tillage interaction) as impacted by precipitation Shannon diversity index (SDI), wind speed and average air minimum temperature over two weeks after fertilizer application.

Year × Nrate

Grain yield was significantly affected by N rate over the years (Figure 3.19). In general, the three N rates followed a similar pattern in time, producing more in high-yielding years and less in low-yielding years, differing mostly by the magnitude of the yield response. Overall, 300N produced the largest grain yields over time, but only significantly higher than the 150N in nine out of 28 years. Furthermore, only in one occasion the 75N yielded similarly to 300N.

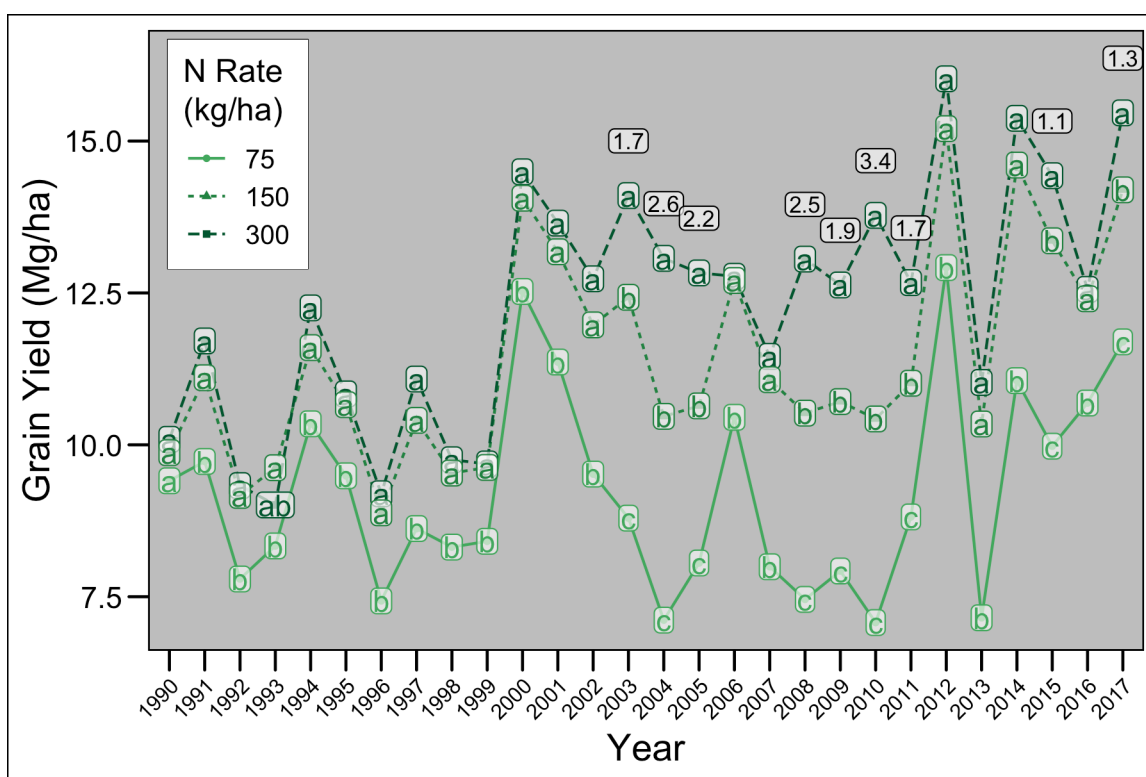


Figure 3.19. Mean ($n=16$) corn grain yield as affected by N rate (75, 150 and 300 kg N ha⁻¹) for period two (1990 through 2017). Means within a given year followed by a common letter are not significantly different at $\alpha=0.05$. Numbers in the plot area represent Δ yield calculated as Yield_{300N} – Yield_{150N}, in Mg ha⁻¹.

Based on the yield responses observed for the Year × Nrate interaction (Figure 3.19), years were classified as 300N > 150N (2003, 2004, 2005, 2008, 2009, 2010, 2011, 2015, 2017),

and 300N = 150N (all others) based on $\text{Yield}_{300\text{N}} - \text{Yield}_{150\text{N}}$. The summarizing week-window interval that best predicted the yield response from this interaction was two weeks after fertilizer application. Summarized weather variables, application timing and year-yield group (high-, medium- and low-yielding years) were fit to a CIT. The most important variables in explaining yield response were year-yield potential, average air maximum temperature and SDI (Figure 3.20), with a predictive accuracy of 84% on training data. Positive yield response from applying 300N over 150N was observed with the highest frequency under medium-yielding years when SDI was above 0.39. This response was also observed under low- and high-yielding years when air maximum temperature was below 18°C, however at a lower frequency. No response from applying 300N over 150N was observed at the highest frequency under high- and low-yielding years when maximum temperature was above 18°C, followed by medium-yielding years when SDI was below 0.39.

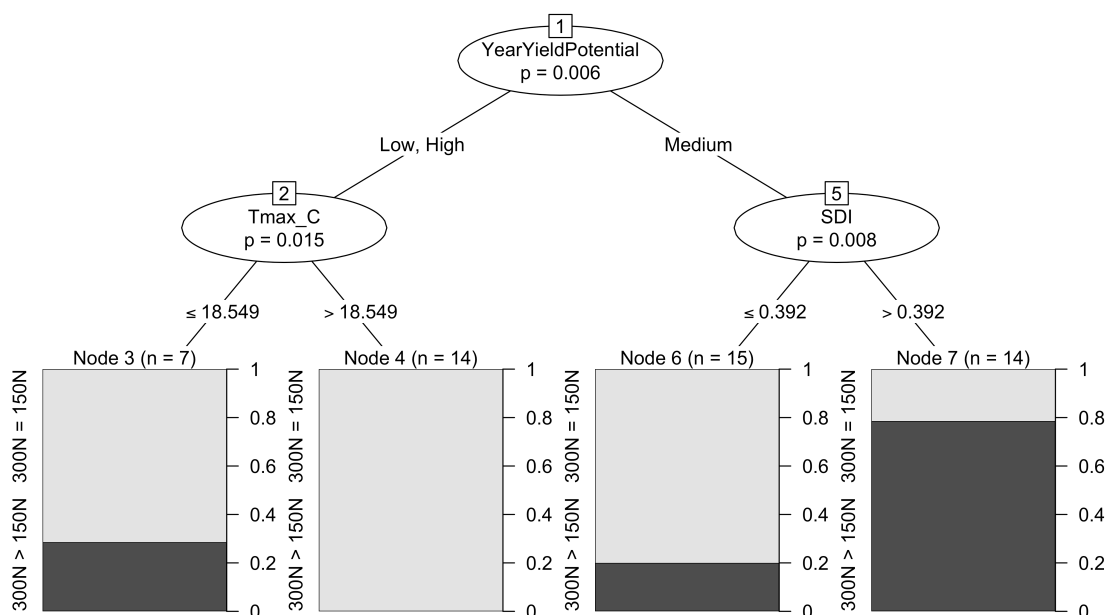


Figure 3.20. Conditional inference tree for the response of Nrate (positive or neutral based on $\text{Yield}_{300\text{N}} - \text{Yield}_{150\text{N}}$ from the Year \times Nrate interaction) across year over two weeks after fertilizer application. Each terminal node contains the ratio of cases in each yield response category. Note: n= number of observations, YearYieldPotential is the year-yield potential grouping (High from 12.6 to 14.7 Mg ha⁻¹, Medium from 10 to 12 Mg ha⁻¹ and Low from 8.5 to 9.8 Mg ha⁻¹), Tmax_C is the average air maximum temperature in °C, and SDI is the precipitation Shannon diversity index.

The probabilities of observing different yield response groups based on the Year \times Nrate interaction as affected by year-yield potential, average air maximum temperature and SDI are shown on Figure 3.21. The probability of observing higher grain yield under 300N compared to 150N was highest under medium- and high-yielding years at high SDI and maximum temperature values. However, higher maximum temperature levels only increased the probability of a positive response (300N > 150N) if accompanied by more-distributed rainfall (SDI=0.76). Under more-concentrated rainfall cases (SDI<0.4), increasing maximum temperature decreased the probability of higher yield at 300N compared to 150N. The probability of observing a neutral

yield response ($300N = 150N$) was highest and the only predicted outcome under low-yielding years.

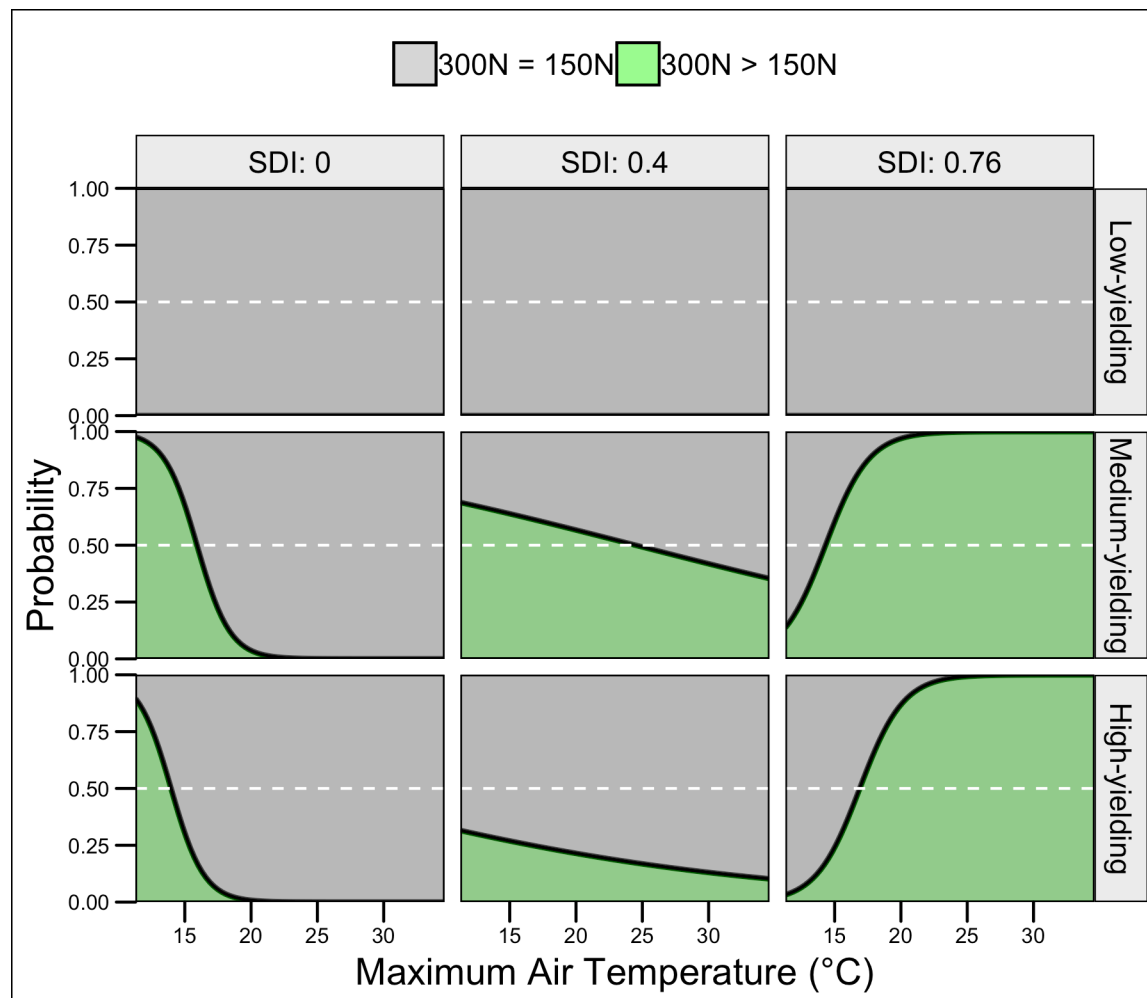


Figure 3.21. Logistic regression for the response of Nrate (neutral or positive based on $Yield_{300N} - Yield_{150N}$ from the Year \times Nrate interaction) as impacted by precipitation Shannon diversity index (SDI) and year-yield potential over two weeks after fertilizer application.

Tillage \times Nserve and Nrate \times Tillage

Grain yield was affected by the two-way interactions of Tillage \times Nserve (n=336) and Nrate \times Tillage (n=224) (Table 3.4). Using Nserve had no yield effect under CT, and CT yields were greater than RT yields. The lowest yield was under RT without Nserve and adding Nserve improved RT yields, but not enough to reach mean CT yields. Grain yield was highest at 300N, regardless of tillage type. For lower N rates (75N, 150N), however, yields were higher under CT compared to RT.

Table 3.3. Mean corn grain yield from Tillage vs. Nserve (n=336, averaged over levels of N rate and N timing) and Tillage vs. N rate (n=224, averaged over levels of Nserve and N timing) interactions. Means followed by a common uppercase and lowercase letter are not significantly different for the Tillage vs. Nserve and Tillage vs. N rate interactions, respectively, at $\alpha = 0.05$.

	CT	RT
	---- Yield (Mg ha ⁻¹) ----	
Nserve		
W	11.18 A	10.87 B
W/O	11.21 A	10.71 C
N Rate (kg ha ⁻¹)		
75	9.45 d	9.01 e
150	11.77 b	11.07 c
300	12.36 a	12.29 a

Discussion

Soil Properties

Soil properties were affected over time by N rate, tillage type and Nserve. In 2000, 14 years after experiment initiation, soil pH, K and NO₃⁻ in the 0-20 cm depth were the main soil features impacted by N rate. Lower soil pH values and higher residual nitrate values were observed at higher N rates. Applying ammonium-containing fertilizer increases soil acidification by stimulating nitrification, which produces H⁺. For example, Flowers and O'Callaghan (1983) reported on a pH decrease of 1 unit after complete nitrification of 250 ppm of NH₄⁺-N. Greater

nitrification of NH_4^+ into NO_3^- also increases the risk of NO_3^- leaching which decreases NO_3^- availability for plant uptake, further promotes soil acidification because plant roots neutralize soil acidity when NO_3^- is taken up (Bolan et al., 1991).

In 2006, 20 years after experiment initiation, soil pH, POM, OM, K, and P were affected by different treatment factors at different depths. Soil pH was mostly affected by N rate up until a depth of 61 cm, and N rate and tillage at the 0-15 cm depth, with pH lowering as both N rate and tillage increased. Other long-term tillage studies have found CT to both increase (Dick, 1983; Hickman, 2002) and decrease (Ismail et al., 1994) soil pH at the surface layers compared to no-tillage practices. Ismail et al. (1994) attributed lower soil surface pH under CT compared to NT due the lack of soil mixing following lime application six years prior to soil sampling. In our study, no record of lime application occurred to the best of our knowledge, but the reason for lower surface soil pH with CT could be attributed to higher rates of mineralization compared to RT.

Soil POM is expected to increase as N rate increases to levels that optimize grain production (Liebig et al., 2002; Brown et al., 2014), and as tillage decreases (Hussain et al., 1999). That is because optimum levels of N input increase plant biomass production, and lack of tillage reduces residue incorporation and decomposition rate. In our study, this trend was partially confirmed, with higher POM observed under the highest N rate plus RT at the 0-15 cm depth. However, given that the lowest observed POM at this depth was under the highest N rate plus CT, it appears that tillage played a stronger effect than N rate. At intermediate depths, POM was only affected by Nserve and tillage, with the use of Nserve decreasing POM, and CT having higher POM than RT. While the positive effect of CT on POM at depth is expected due to residue incorporation, the reason for a negative effect of Nserve on POM is unclear.

Grain Yield

Year-to-year weather variability was the main driver of corn yield differences over the 28-yr study period. Different studies have attempted to understand the main N management factors driving corn response (Wolt, 2004; Kyveryga et al., 2010; Tao et al., 2018). While these reviews are important for understanding yield responses over larger geographic regions, they fail to quantify the complex soil, weather, and management interactions inherent to each individual study and across them. In the current report, we focus on the primary effects of weather variability and management on grain yield and assume that changes in soil properties over time have a secondary feedback role in yield variability.

Year, N timing, and Nserve Interaction

The use of Nserve with PP fertilization improved yield more frequently than when applied SD. The longer the elapsed time between N application and plant N demand the higher chances for N to be lost by processes such as leaching and denitrification. Therefore, PP applications are more prone to losses and thus are more responsive to protecting N by the use of Nserve than if applied at SD. In contrast to our findings, Burzaco et al. (2014) found no significant yield response to either NI or application timing when UAN was applied with and without nitrapyrin in three corn studies in Indiana. Despite the lack of yield response, Burzaco et al. (2014) observed a significant increase in plant N uptake when N was applied at V6 as compared to at-planting. These authors also reported an increase in N recovery efficiency when UAN was applied at-planting with NI, but no effect of NI when UAN was applied side-dress. Studies evaluating the use of NI applied at different timings normally evaluate fall vs. spring applications, and planting vs. side-dress comparisons are scarce. Randall and Vetsch (2005) evaluated AA application to corn both in the fall and spring in six years, with and without an NI,

and found that adding NI in the fall had a larger positive effect on yield compared to untreated fertilizer than when applied in the spring. On a survey over 920 corn fields from four US Midwest states under multiple different N management practices, Tao et al. (2018) found that spring-applied and SD-applied anhydrous ammonia did not significantly differ in promoting higher end-of-season cornstalk nitrate test (CSNT), a measure of nitrogen use efficiency. The same study also reported on the lack of significance of NI in changing the odds ratio of a cornstalk to test in a higher CSNT category (i.e. more efficient).

The use of Nserve decreased yields compared to fertilizer alone in 2012 and 2001 for PP and SD, respectively. Although 2012 was one of the driest and hottest years in this dataset with cumulative precipitation inputs of only 48 mm in the first three weeks after PP fertilizer application, irrigation successfully alleviated the severe drought conditions such that 2012 was the highest yielding year in this long-term study. The negative effect of Nserve in 2012 may have reflected restricted N mobility, especially as NO_3^- , to the roots in a year when plant N demand was high and low soil moisture early in the season limited N movement. While NO_3^- is highly mobile and moves towards the roots mainly by mass flow, NH_4^+ is involved in reactions with clay mineral lattices and moves towards the roots mainly by diffusion (Forde and Clarkson, 1999), which has been estimated to be from 50- to 500-fold slower than mass-flow-driven NO_3^- (Forde and Clarkson, 1999).

Weather analysis identified the highest proportion of positive yield response to Nserve under low- to medium-yielding years when average minimum temperature was below 6°C. This scenario is likely conducive to lower plant N demand early in the season and prolonged fertilizer N protection due to temperature-driven slower Nserve degradation (Keeney, 1980; Hoefl, 1984). On the other hand, negative effects of Nserve on yield were predicted to happen under high-

yielding years, likely related to limiting accessibility of N in the mobile nitrate form, and especially as this was the case in 2012 when drier conditions early in the season were observed. Furthermore, the probability of a negative yield effect from Nserve in high-yielding years increases as minimum temperature increases, likely a result of faster plant growth and its associated higher N demand coupled with soil drying. Medium-yielding years were predicted to have a positive yield effect from Nserve under lower air minimum temperatures, and this probability decreases in place of neutral yield response as temperature increases, likely as a result of Nserve degradation (Keeney, 1980; Hoefl, 1984).

Year, N Rate, and N Timing Interaction

Applying fertilizer at SD increased corn grain yield 47% of the years compared to PP under at least one N rate. The most frequent yield benefits from SD over PP application occurred at the lowest N rate, and benefits decreased in frequency as N rate increased. Side-dressing at the lowest N rate likely resulted in positive yields by better matching N availability with crop N demand. Side-dressing also likely avoided greater N risk loss over time compared to pre-plant N, particularly for this low N rate. The yield benefits of delaying fertilizer application to occur during the growing season (instead of before it as PP) was reflected in the higher fraction of PP < SD years under high solar radiation in medium- to high-yielding years, especially when cumulative rainfall over 13 weeks after fertilizer application was above 211 mm. Under these conditions, high solar radiation promoted more plant growth and development in yield-responsive years. In turn, the accompanied demand for N likely could not have been met with PP as higher precipitation likely increased N losses during this window of time, giving SD a yield advantage.

A negative effect of SD compared to PP was also observed in 21% of the years. In those years when $PP > SD$, this effect happened with the highest frequency at the highest N rate. The highest proportion of this scenario happened under lower solar radiation conditions, but had a decreased probability of being observed as both solar radiation and cumulative precipitation increased. Therefore, PP application is more likely to yield greater than SD in low-yielding years, when N demand is lower, and under lower cumulative precipitation, when N loss potential is also decreased.

Year, N Rate, and Nserve Interaction

The use of Nserve at different N rates produced variable yield responses over the years. It is expected that the use of a NI will reflect in higher grain yield in case a response to N fertilizer exists and N loss pressure is high to the point of limiting N availability to crops (Hergert and Wiese, 1980). However, the possibility of an NI to negatively impact grain yield also exists and has been reported in various occasions (Blackmer and Sanchez, 1988; Cerrato and Blackmer, 1990; Sassman et al., 2018). In this study, the use of Nserve produced higher, lower and equal yields at different N rates compared to fertilizer alone over the years.

Positive yield response from NI use was observed in at least one N rate in 32% of the years. This is in agreement with the 10-40% frequency of positive yield response from NI summarized by Hergert and Wiese (1980) for irrigated corn in fine textured soils in Kansas and Nebraska. However, the frequency of positive yield response to NI found in the present study is well below the 75% frequency found by Wolt (2004) in a summary of 436 mean comparisons, mostly from field corn studies in the U.S. Midwest states. The highest frequency of greater yield from using Nserve was observed under higher cumulative precipitation and CHU in medium- to

high-yielding years. Under this scenario, the overall higher year-yield potential was met by both higher thermal units to drive growth and development, and higher precipitation. Higher precipitation volume can often lead to increased chances for N losses, which in this case were avoided by both higher plant N demand and the use of Nserve.

The highest frequency of a negative yield effect of Nserve was observed in years with a cumulative precipitation below 142 mm over 10 weeks after fertilizer application. Dry conditions early in the growing season and before irrigation likely decreased nutrient mobility from the bulk soil towards the rhizosphere. By keeping N in the NH_4^+ form, it is possible that the use of Nserve limited N supply to the roots in such dry conditions. Studies demonstrating a decrease in yield and/or N uptake from NI use normally attribute this effect to i) drier soil conditions causing N positional unavailability (Hoefl, 1984; Sassman et al., 2018), ii) NI-induced N immobilization (Ferguson et al., 1991, 2003), and iii) adverse effects of NI on plant growth (Blackmer and Sanchez, 1988). Hoefl (1984) reported a yield decrease from the use of AA at 67 kg N ha^{-1} with NI in a dry year, and attributed this to positional unavailability since roots were likely extracting water from deeper soil layers whereas N was positioned on layers closer to the dry surface. Sassman et al. (2018) observed the use of NI to decrease corn grain yield in two out of three years, and increase agronomic and economic optimum N rate in one year when urea-ammonium nitrate was applied at multiple N rates. The authors suggested that the negative impact of NI could be due to the high efficacy of the inhibitor, thus maintaining more N as NH_4^+ in a small soil volume, decreasing the chances of fertilizer interception by roots. Ferguson et al. (1991) observed a decrease in inorganic N in NI-treated AA injection bands in three years of field corn studies, and suggested that this was due to NI-induced temporary N immobilization. Blackmer and Sanchez (1988) observed that most of the site-year-rate data points that increased corn leaf,

stover, and grain N concentration yet produced grain yields below plateau levels were NI-treated, and attributed this to a negative effect of the inhibitor on plant growth.

Tillage Interactions

Tillage type was part of two-way interactions with Year, Nrate, and Nserve. Tillage had a variable impact on grain yield over the years, where yields were either not affected by tillage, or greater under CT than RT but never vice-versa. Similarly, Drury et al. (2012) observed higher corn yields under CT vs. NT in two out of three years in Ontario, Canada. Tao et al. (2018) also found that CSNT, an end-of-season proxy for corn N sufficiency, was more likely to be higher under CT than no-till (NT), likely due to greater N availability after tillage disturbance. Ismail et al. (1994) found that CT outyielded NT in the first 12 years of a 20-year continuous corn study, but that NT outyielded CT with greater frequency later in the study (7 years). Similar temporal trends were found in our study, where CT outyielded RT at a greater frequency early in the study compared to later in the study (50%, 50%, 13% for 1990-1999, 2000-2009, 2010-2017, respectively). Therefore, although the frequency of CT yield benefit over RT has decreased in the last eight years, this relationship has not shifted (i.e. CT has not yielded less than RT) over the years as observed by Ismail et al. (1984).

When averaged over years, using Nserve in CT had no effect on grain yield whereas it increased yield in RT compared to untreated fertilizer. When averaged over years, CT and RT produced similarly only at the 300N fertilizer rate, with CT producing more than RT at the 150 and 75N rates. Both of these interactions demonstrate that less N was available to the crop under RT than CT, either because more N was lost (e.g. as NO_3^- leaching and/or denitrification) in RT

or because N was less available in RT compared to CT (e.g. due to greater immobilization of fertilizer-N in RT and/or greater mineralization of soil organic matter in CT).

Yield increases under CT compared to RT occurred with highest frequency in years with lower average wind speed during the two-week window after fertilizer application. The relationship with wind speed did not meet our expectation that tillage-related yield responses would be more sensitive to weather variables that directly impacted soil water and temperature status (e.g. precipitation, air temperature). Given that wind speed is independent of tillage practice, it is possible that its integrated effects on other soil thermal and hydrological properties derived from tillage type were more explanatory of yield differences. For example, low speed winds will cause less soil water evaporation, which in turn maintains a lower soil temperature especially under high-residue conditions found in continuous corn RT. Stanley and Smith (1956) observed in a laboratory incubation study that soil moisture from the top five cm layer decreased from 23% to 7% after supplying heat and air movement for 48 hours, and that when no heat or wind was supplied only 2% of soil moisture was lost. Similarly, Greb (1966) suggested that the presence of residue decreased soil water evaporation because the physical presence of residue reduced both soil temperature and the gradient of wind speed on the soil surface. Sauer et al. (1996) noted a decrease in soil moisture evaporation as surface cover increased, with a reduction in evaporation of 41 and 43% on two different soils. Ussiri and Lal (2009) found lower soil temperatures from June to October and higher soil gravimetric water content during the growing season under NT vs. CT, attributed differences to the residue cover in NT decreasing evaporation and enhancing soil water retention through increases in SOM. These conditions can have an impact on both corn early season growth and development (Hatfield et al., 2001) and N mineralization, ultimately impacting grain yield.

The second most frequent weather condition causing CT to yield higher than RT was under higher wind speeds accompanied by a lower SDI. This scenario reinforces the hypothesis of damp RT conditions as the cause for lower yields, as more concentrated rainfall would likely cause a similar effect on soil moisture and temperature, even at higher levels of wind speed and evaporative pressure. The condition most likely to be observed under years where RT produced similarly to CT was at higher wind speeds, higher SDI and higher air minimum temperature. These conditions favor soil evaporation in both tillage systems and especially under RT to the point of soil temperature not being a limiting factor for early-season plant growth and N mineralization.

Year and N Rate Interaction

Nitrogen rate had a variable impact on yield over the years. In all years, at least the first N increment (i.e. 75N) was able to produce greater yield than 0N, indicating N responsiveness throughout the study duration (data not shown). In only one occasion 75N produced similarly to 300N. Given that 150N is close to the average rate used in the region (i.e. 160 kg N ha⁻¹) and that 300N would be excessive and likely represent a non-limiting N condition, these two rates were compared more closely for their effect on yield.

The 300N treatment produced more than 150N in 32% of the years, indicating that 150N may have been yield-limiting possibly due to higher N loss pressure, higher crop N demand, or both. The highest frequency of observing a positive yield response from 300N over 150N happened under medium-yielding years at high SDI values over 13 weeks after fertilizer application. These conditions suggest that more evenly distributed rainfall benefited a response to higher N rate. However, since this scenario happened mostly in medium-yielding years, it is possible that other factors were then limiting to yield (e.g. other nutrients, stress timing, etc.).

Nonetheless, results from the logistic regression model indicate that not only medium- but also high-yielding years were more likely to respond to 300N, especially as maximum temperature and SDI increase. Higher temperatures coupled with evenly distributed precipitation likely promoted enhanced crop growth and development and crop N demand, which in turn was met by higher N application rates.

Averaged over years, CT produced more than NT at 75N and 150N, and produced equally to RT at 300N. Based on yield and weather results, it is likely that i) N mineralization under CT was higher, being able to supply more N and thus improve grain yield at a yield-limiting N rate, ii) continuous corn RT immobilized a portion of the applied N fertilizer, making less of it available for crop uptake and thus causing lower grain yield at yield-limiting N rate, or iii) a combination of both conditions. However, neither mineralization nor immobilization were measured, and these yield results do not agree with what is reported for long-term tillage studies. Kitur et al. (1984) reported higher soil N immobilization in NT compared to CT at a low N fertilizer rate only (84 kg N ha^{-1}), but no net effect on grain yield was found in three years. Rice et al. (1986) reported that N mineralization from a 16-yr tillage study on corn was similar between NT and CT, but that mineralization differences were observed prior to the 10th yr. The authors suggested that lower N availability in NT compared to CT was transient, and that it becomes unimportant after a new OM steady state is reached (about after 10 yrs).

Conclusions

Soil characteristics and yearly weather interact with N fertilizer management practices to impact grain yield response over time in irrigated corn. The management practices that had the greatest magnitude of response in grain yield were Nrate > tillage > N timing > Nserve. For the conditions of this study, the use of Nserve would be recommended when applied pre-plant only.

When averaged over application timing, Nserve produced yield responses ranging from negative to positive, with the former happening mostly in drier-than-normal early springs. When considering different timing options alone, side-dress application around V4 should be recommended over pre-plant applications. In this study, CT either produced as much or more grain yield than RT. It is not clear why RT did not produce greater yields than CT, especially as the study progressed past 10 yrs. The choice of tillage should not only consider yield, but also other benefits that RT may present over CT (e.g. erosion control; decreased fuel, labor and equipment cost; weed suppression, water retention).

This study elucidated how weather variables interacted with different N management practices to impact yield response in irrigated corn. The specific weather variables most important in explaining yield responses from different N management practices varied. The most important weather variables in explaining different yield responses over time were related to year- yield potential, air temperature, precipitation volume and distribution, solar radiation and wind speed. These relationships were only able to be assessed given the many years of both yield and weather data. Long-term studies are important in allowing the assessment of complex interactions between management and weather that may not be evident in short-term studies.

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Chapter 4 - Active and Passive Sensors are Comparable for Variable Rate Nitrogen Determination and Accuracy in Irrigated Corn

Introduction

Nitrogen (N) is often the most limiting nutrient to crop productivity. To maximize yield potential, fertilizer N is supplemented to non-legume (i.e. non N-fixing) crops such as corn (*Zea mays* L.). The annual fertilizer input to U.S. corn crops alone is substantial, with ~46% of all N applied to crops in 2010 (USDA-NASS, 2017). The large quantities of fertilizer applied for crop growth can pose environmental risks once applied to the field. Nitrogen transformations are dynamic, and losses resulting from these processes can be significant especially when soil N supply is much greater than the demand by the crop (Cameron et al., 2013). Because conditions for both crop N demand and environmental N losses vary spatially and temporally, applying a single fertilizer rate to an entire field that varies in landscape characteristics can create areas of under- and over- fertilization (Mamo et al., 2003; Scharf et al., 2005). To better match fertilizer application rate with crop N demand and landscape characteristics, the use and development of crop canopy sensors for assessing crop N status and applying N variably has been of major research interest.

Both active and passive crop canopy sensors can be used for variable rate N (VRN) management. Active sensors emit their own modulated light. Because of that, sensing performance is theoretically independent of atmospheric conditions, such as cloud cover and time of day. Moreover, active sensors have been used “on-the-go”, capable of assessing crop N status and directing VRN application on the same pass. Because they emit their own light source, active sensors require a certain proximity to their target, and thus are mostly limited to ground-

based platforms (e.g. tractor, application implement boom) and low-flying (0.5 to 1.5 m above canopy) unmanned aerial systems (Krienke et al., 2015).

Passive sensors rely on sunlight as the energy source and thus may be limited by atmospheric conditions like time of day and cloud cover (de Souza et al., 2010). Furthermore, the use of passive sensors to generate VRN application is a two-step process, where first the field is imaged, and only after data correction and processing can a prescription map be generated and fed into a variable rate applicator software. Historically, passive sensors have been mostly employed in agriculture via satellite or aircraft. Recently, unmanned-aerial systems (UAS) have become a popular platform for carrying passive sensors both in research and commercially.

Recently, many studies have compared how different active crop canopy sensors can be used for VRN (Barker and Sawyer, 2010; Shaver et al., 2011, 2014; Li et al., 2014), but fewer have compared active vs. passive sensors (Erdle et al., 2011). There is strong interest in passive sensors because these are the most common sensors used from airplanes and, more recently, on unmanned aerial systems (UAS). With the rapidly growing UAS market, there will be an increasing opportunity to use passive sensors for quantitative decision-making in agriculture, including N management.

Various vegetation indices (VIs) have been developed for assessing different vegetation parameters (Mulla, 2013). Two of the most common VIs calculated from bands found in both active and passive sensors are the normalized difference vegetation index (NDVI) and the normalized difference red-edge (NDRE). Reflectance on the spectral region of red (600-700 nm), which is used to calculate NDVI, saturates when leaf area index (LAI) values are > 2 (Gitelson et al., 1996; Viña et al., 2011) and at chlorophyll concentrations as low as 3-5 micrograms cm^{-2} (Gitelson and Merzlyak, 1997). The saturation of the red band renders it insensitive in

differentiating N deficient and sufficient crop condition (Blackmer and Schepers, 1994; Holland and Schepers, 2013). One solution is to use a different index that does not contain the red band, such as NDRE that replaces the red band with the red-edge band (700-800 nm) in its formula. For example, Gitelson et al. (2003) demonstrated that reflectance in the green and red-edge bands was significantly more sensitive to increasing chlorophyll levels than those in the red and blue (400-600 nm) bands.

Different algorithms have been developed to translate sensor-measured plant nutrient deficiency status into an N rate recommendation (Raun et al., 2005; Teal et al., 2006; Holland and Schepers, 2010; Scharf et al., 2011). In the algorithm developed by Holland and Schepers (2010), input variables include optimum N rate, management zone scalar, different sources of N credits (i.e. previous crop, organic matter, water nitrate, manure application, fertilizer applied prior to sensing), a sufficiency index (SI) and a delta SI (DSI). The SI is the ratio of the vegetation index (VI) values from a the field area receiving VRN to the VI of a high N reference strip, or N-rich strip (NRS) (Biggs et al., 2002). The NRS represents the N rate for maximum crop growth under no N limitation. By normalizing sensor data from unknown parts of the field to the NRS, sensor-derived comparisons can be made across different hybrids, planting dates, fields, and sampling dates (Blackmer and Schepers, 1995). Establishing an NRS, however, can be inconvenient and even restricted in commercial sensor-derived N applications (Holland and Schepers, 2013). The high N inputs used for an NRS could also induce crop sulfur deficiencies (Franzen et al., 2016), which sensors are unable to differentiate from N deficiency effects on crop canopies.

Given these limitations, Holland and Schepers (2013) proposed the use of a virtual reference (VR), defined as the 95th cumulative percentile of a histogram from a given VI data

collected over parts of the field demonstrating different crop N status levels. Although the implementation of the VR concept has been introduced as a solution for large-scale commercial field applications, it is unclear how VRN recommendations are impacted by choice of reference (e.g. NRS vs. VR) to calculate SI and, subsequently, DSI. The delta SI (DSI) is the difference in SI between the reference and an N-unfertilized area [i.e. $1 - SI(0)$], and these values impact the accuracy of sensor-based N rate recommendation (Holland and Schepers, 2010). Although DSI can change according to sensor type, the specific bands utilized to calculate the VI used for VRN, and crop stage (Holland and Schepers, 2010), the major challenge to calculating DSI is that commercial production fields are unlikely to have any non-fertilized areas. As a result, a default DSI value of 0.3 [i.e. $SI(0)=0.7$] has been proposed (Holland and Schepers, 2010).

Sensor-based VRN application can be an important tool to adjust N rates while maintaining grain yield levels, thus enhancing the efficiency with which the crop uses fertilizer. One way to assess the agreement of the sensor-based N recommendation to the optimum N rate required by the crop is to compare the recommendation to the end-of-season calculated economic optimum N rate (EONR). The EONR is the N rate that economically optimizes grain yield production, and after which the return on investment decreases with increasing N rates.

The hypotheses that were tested were that i) NRS and VR create similar reference VI values; ii) DSI varies depending on site, sensor type, and VI; iii) active and passive sensors generate similar VRN; iv) NDRE-based VRN is higher than NDVI-based VRN; and v) sensor-generated VRN, when summed to the pre-plant N rate, approximates the field EONR. The objectives of this research were to i) assess the agreement between the NRS vs. VR values for each individual site-year (SY)-sensor-VI combination; ii) assess if and how DSI varies over different SYs, sensor types and VIs, and inform the variable rate algorithm on proper DSI term

selection; iii) compare active and passive crop canopy sensors' recommended side-dress N rate derived from different VIs; and iv) assess side-dress N rate recommendation accuracy of different sensor and VI types compared to the EONR in irrigated corn.

Material and Methods

Site Description and Field Experimental Design

This study is comprised of eight site-years (SYs), conducted from 2015 through 2018 on different soil types and with a range of N fertilizer rates applied at pre-plant (Table 4.1). The studies were located either on-farm (Central City and Hastings, NE) or at the University of Nebraska-Lincoln's South Central Agricultural Laboratory (SCAL) near Clay Center, NE. The soils were classified as Novina sandy loam (coarse-loamy, mixed, superactive, mesic Fluvaquentic Haplustolls) at SY1; Crete silt loam (fine, smectitic, mesic Pachic Udertic Argiustolls) at SYs 2, 3, 5, 8; Hastings silt loam (fine, smectitic, mesic Udic Argiustolls) at SYs 4, 6; and Cass fine sandy loam (Coarse-loamy, mixed, superactive, mesic Fluventic Haplustolls) at SY 7. Corn was planted in 0.76-m spacing at 79,800 to 82,000 plants ha⁻¹. Each plot was 15-20 m long and comprised four rows. For all site-years, the experiment was single factor (treatment = N rate) in a randomized complete-block design with four blocks (Table 4.1). The N source utilized varied among SYs and included urea-ammonium nitrate (UAN), urea (U), or anhydrous ammonia (AA). Fertilizer was either surface applied (UAN, U) or injected (AA) prior to corn planting. Fertilizer N rate was calculated based on the University of Nebraska-Lincoln N recommendation algorithm for corn (Shapiro et al., 2008).

Table 4.1. Description of each site-year (SY) study related to site characteristics, N management and passive sensor utilized.

Site-year	Site	Year	Soil [†]	N Source	N rates	UNL [‡]	AONR [§]	EONR	Sensor
						kg N ha ⁻¹			
SY1	Central City	2015	SL	UAN	0, 65, 96, 130, 161	161	-	-	Tetracam
SY2	SCAL	2015	SiL	UAN	0, 65, 96, 130, 161	161	0	0	Tetracam
SY3	SCAL	2015	SiL	AA	0, 94, 126, 157	157	84	30	Tetracam
SY4	SCAL	2016	SiL	UAN, U	0, 108, 161, 173, 215	215	160	108	RedEdge
SY5	SCAL	2017	SiL	AA	0, 77, 163, 233, 309	233	293	235	Sequoia
SY6	SCAL	2017	SiL	UAN	0, 45, 90, 133, 178, 268	178	173	160	Sequoia
SY7	Hastings	2018	SiL	UAN	0, 98, 146, 194, 388	194	149	133	Sequoia
SY8	SCAL	2018	SiL	AA	0, 72, 152, 217, 289	217	288	210	RedEdge

[†]SL = sandy loam; SiL = silt loam

[‡]UNL = optimum N-rate for corn recommended by UNL algorithm

[§]AONR and EONR of “-” or 0 indicate no crop response to N added at any rate compared to no fertilizer added.

Sensor Description and Sensor Data Processing

Crop reflectance data was acquired using four different sensors: RapidScan (handheld, active) and Tetracam, MicaSense RedEdge or Parrot Sequoia (unmanned aerial system-mounted, passive). On each SY, a specific passive sensor was utilized (Table 4.1), and for all SYs RapidScan was used as the active sensor. Crop canopies were sensed during the V12 stage for each SY, with data used to calculate simulated side-dress N rates that would be recommended for management systems using split-N applications (described below). To maintain equivalent field conditions among sensor types, active and passive sensing were completed on the same day within a SY, and sensing was limited to sunny days to maximize passive sensor performance.

The RapidScan CS-45 (Holland Scientific, Lincoln, NE, USA) is an active handheld sensor equipped with a modulated light source and three photodetector measurement channels at 670, 730 and 780 nm (Table 4.2). RapidScan was oriented on the nadir position and measurements taken at ~0.6 meters directly over the corn row. The two central rows of each plot were scanned individually. Values generated for each row were averaged to create one value per wavelength per plot. The passive multispectral sensors used were Tetracam MCA6 Mini

(Tetracam Inc., Chatsworth, CA, USA), MicaSense RedEdge (MicaSense Inc., Seattle, WA, USA) or Parrot Sequoia (Parrot Inc., San Francisco, CA, USA) (Table 4.2).

Table 4.2. Sensor information on band, wavelength center and full width at half maximum (FWHM, in parenthesis).

Sensor name	Sensor type	Blue	Green	Red	Red Edge	NIR
		Wavelength Center and FWHM (nm)				
RapidScan	Active	-	-	670	730	780
Tetracam	Passive	-	530 (10)	670 (10)	760 (10)	800 (10)
RedEdge	Passive	475 (20)	560 (20)	668 (10)	717 (10)	840 (40)
Sequoia	Passive	-	550 (40)	660 (40)	735 (10)	790 (40)

Each passive sensor was mounted on a UAS and flown to an altitude of 70 to 120 m. Imaging scenes were acquired with overlapping regions over the entire study area. A downwelling radiation sensor on the UAS was used for radiometric correction. Corrections were performed in PixelWrench II (Tetracam Inc., Chatsworth, CA) when Tetracam was the passive sensor, and in Atlas (MicaSense Inc., Seattle, WA, USA) and Pix4D (Pix4D S.A., Lausanne, Switzerland) when MicaSense RedEdge and Parrot Sequoia were the passive sensors, respectively. Following image radiometric and geometric adjustment, the remaining image processing steps were performed in R Statistical Software (R Core Team, 2017). Unsupervised classification and image reclassification were used to exclude soil pixels from plant pixels.

Calculation of Vegetation Index, Sufficiency Index, and N Rate Recommendations

VIs were calculated for the entire field and averaged within each plot. NDVI and NDRE were derived from the reflectance data of the red and near-infrared (NIR) bands and red-edge (RE) and NIR bands, respectively. The RE band from the passive sensor Tetracam had a wavelength center positioned too close to the NIR band (760 and 800 nm, respectively), resulting in NDRE values that were unrelated to pre-plant applied N rates. Due to this, NDRE values from

the SYs where Tetracam was the passive sensor (i.e. SY1, SY2, and SY3) were removed from the dataset.

Two different methods were tested to set the reference VI values used for SI calculated from active and passive sensors used for each SY. The first reference method, the NRS, was calculated as the VI from the treatment receiving the highest N rate in each treatment block. The second reference method, the VR, was calculated as the 95th percentile of the histogram for each treatment block across all N rates. For both reference types, SI values were calculated for all other N treatments by block so as to retain n=4 per treatment combination per site-year (below).

To calculate an SI, the VI of a treatment was divided by that of the VR derived for each SY-sensor-VI-block combination. Then, the SI was used as an input in the simplified algorithm developed by Holland and Schepers (2010) for side-dress N rate determination (Eq. [4.1]):

$$N_{app} = (N_{opt} - N_{PreFert}) \cdot \sqrt{\frac{(1-SI)}{\Delta SI}}, \text{ where} \quad [4.1]$$

N_{app} = calculated recommended side-dress N rate, in kg ha⁻¹

N_{opt} = optimum N rate calculated using the UNL nitrogen fertilizer algorithm for corn, in kg ha⁻¹

$N_{PreFert}$ = pre-plant applied N rate, in kg ha⁻¹

SI = sufficiency index

$\Delta SI = DSI$ = difference between 1 and SI(0), which is the SI for when pre-plant applied N rate=0. Allowed to vary for each SY-sensor-VI combination.

Delta SI was calculated for each SY-sensor-VI combination as 1 – SI(0), where SI(0) represents the SI value for the 0N check plot. Prior to side-dress N rate calculation, the SI and DSI datasets were filtered to meet the VRN algorithm constraints of DSI >0 and 0 ≤ SI ≤ 0. A

total of five data points were removed at this step, resulting from blocks where the 0N VI value was greater than the VR value such that $SI_{0N} > 1$ and $DSI < 0$ for a given SY-sensor-VI-block-treatment. It is important to make the distinction that pre-plant N rates were actual applied treatments, whereas recommended side-dress N rates (hereafter referred to as side-dress N rate) were only simulated (not applied) for all treatments using data collected at the V12 corn growth stage.

The effectiveness of the sensor-based side-dress recommendation rate was assessed by calculating the total N fertilizer input (pre-plant plus simulated side-dress) for each treatment, then comparing total simulated inputs to the economic optimum N rate (EONR). First, crop N-responsiveness was determined for each SY as a linear contrast of grain yield from 0N vs. grain yield from added N rates. When contrasts were significant ($\alpha = 0.05$), grain yield was considered responsive to N fertilization and AONR and EONR could be calculated. Otherwise, no values for EONR could be calculated for corn with no N response. EONR was calculated by regressing grain yield data against N rate using linear, linear-plateau, quadratic, and quadratic-plateau models, then selecting the model with the lowest Akaike information criterion. EONR was then determined from the linear and quadratic terms derived from the selected model (Scharf et al., 2005) and by assuming a corn price of \$134.8 Mg⁻¹ grain and fertilizer price of \$0.93 kg⁻¹ N.

Statistical Tests

All statistical analyses were conducted in R (R Development Core Team, 2017). First, to evaluate reference type effect on reference VI (response variable), reference VI data were tested using a four-way mixed effect ANOVA with fixed main and interaction treatments of SY (1 through 8); reference type (NRS, VR); sensor type (active, passive); and VI (NDVI, NDRE), and the random effect of block nested in SY. Because all main and interaction effects of reference

type were not significant (see Results), all SI and DSI values were recalculated using the VR approach only. Second, to evaluate treatment effects on DSI, the fixed main and interaction effects of SY, sensor type, and VI on DSI were calculated using a three-way mixed effect ANOVA with the random effect of block nested in SY. Finally, the resulting side-dress N rate recommendations were analyzed for each SY. For SYs 1-3, side-dress N rates were assessed with a three-way mixed effect model ANOVA with block as random effect and the main and interaction fixed effects of pre-plant N rate, sensor type (active, passive), and VI (NDVI, NDRE), except for passive sensor NDRE data because Tetracam data was excluded. For SYs 4-8, side-dress N rates were assessed with a three-way mixed effect model ANOVA with block as random effect and the main and interaction effects of pre-plant N rate, sensor type (active, passive), and VI (NDVI, NDRE). For all ANOVAs, treatment means were compared using Fisher's Least Significant Difference test for significant treatment effects. Significance for model terms and mean comparisons was set at $\alpha = 0.05$. Data were visually assessed by constructing fitted vs. residual, residual quantile-quantile and residual histogram plots, and met all assumptions of homogeneity and heteroscedacity.

Results

Reference Comparison

Reference VI values varied over SYs, sensor types and VIs ($p < 0.001$), but was not affected by the main or interaction effects of reference type (VR = NRS) (Figure 4.1). All SI and DSI values hereafter were based on the VR approach in order to retain the highest N rate treatment in the VRN calculation (e.g. otherwise excluded in the NRS approach).

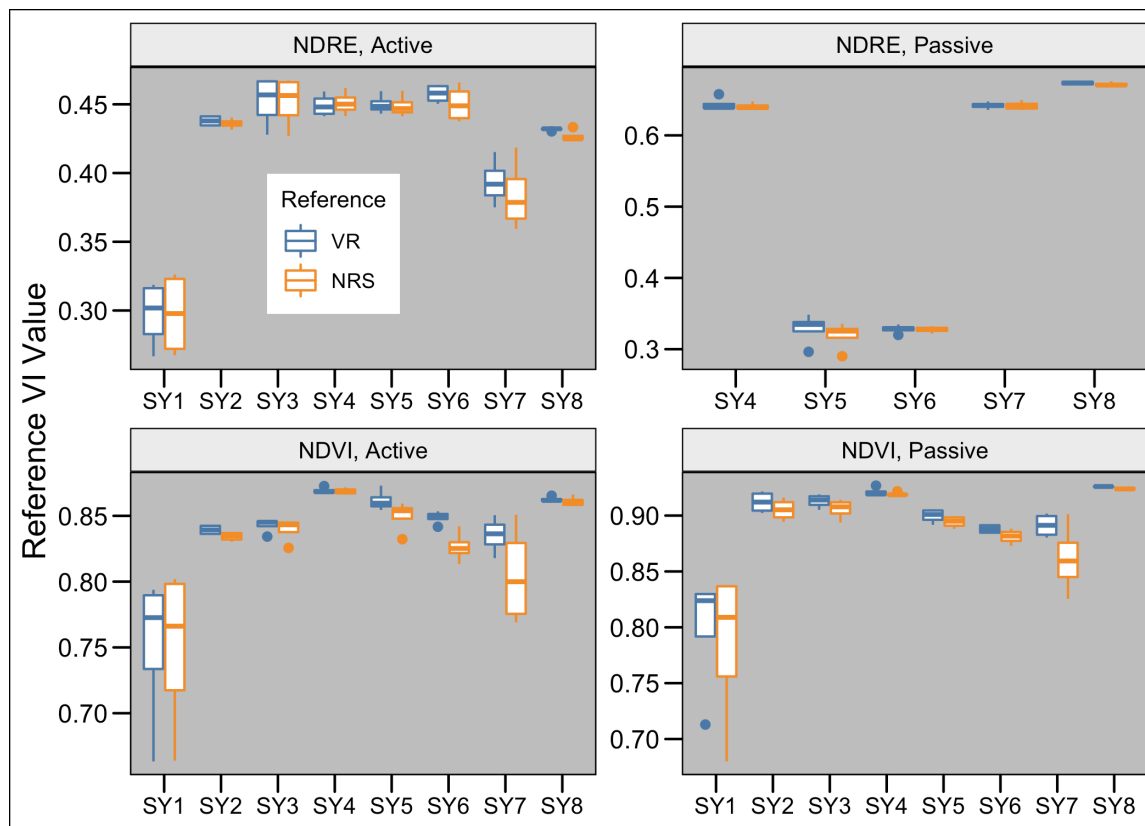


Figure 4.1. Boxplots (n=4) of reference vegetation index value for different reference types [virtual reference (VR) and N-rich strip (NRS)], sensor types (active vs. passive), and vegetation indices (NDRE and NDVI) at each site-year (SY).

Delta SI

Delta SI (DSI) was significantly affected by the 3-way interaction of SY \times sensor type \times VI ($p < 0.001$), and ranged from 0.001 to 0.23 (Table 4.3). DSI averaged within SYs ranged from 0.118 to 0.013, while DSI averaged over sensor-VI ranged from 0.027 to 0.107. Given the significance of different variables, DSI was calculated for each SY-sensor-VI combination and allowed to vary accordingly when calculating side-dress N rate.

Table 4.3. Delta SI for site-years (SY) 1 through 8 as affected by sensor type and vegetation index (VI). Means within a given SY followed by a common lowercase letter are not significantly different at $\alpha=0.05$.

Sensor	VI	SY1	SY2	SY3	SY4	SY5	SY6	SY7	SY8	Mean
Active	NDRE	0.12 a	0.03 a	0.021 a	0.122 a	0.157 b	0.145 a	0.095 a	0.067 a	0.095
Passive	NDRE	NA	NA	NA	0.089 a	0.229 a	0.155 a	0.024 b	0.036 ab	0.107
Active	NDVI	0.078 b	0.009 a	0.007 a	0.04 b	0.052 c	0.085 b	0.021 b	0.007 bc	0.037
Passive	NDVI	0.087 ab	0.005 a	0.01 a	0.03 b	0.032 c	0.038 c	0.016 b	0.001 c	0.027
Mean		0.095	0.014	0.013	0.07	0.118	0.106	0.039	0.028	

Side-Dress N Rate as Affected by Pre-Plant N Rate, Sensor, and VI

Recommended side-dress N rates based on both NDRE and NDVI from active and passive sensors decreased as pre-plant N rates increased at all SYs (Figure 4.2). For SYs 1 through 3 (no passive NDRE data), and SYs 4 through 6, side-dress N recommendations did not differ between sensor type or VI methods. For SYs 7 and 8, a significant three-way interaction between pre-plant N rate, sensor, and VI-type reflected that passive sensors resulted in both lower or higher side-dress recommendations, depending on VI-type and intermediate pre-plant rates. For SY7, side-dress N rate at pre-plant N rate=98 kg N ha⁻¹ from passive-NDRE (89 kg N ha⁻¹) was higher than other sensor-VI combinations, with the lowest side-dress N rate from active-NDVI (16 kg N ha⁻¹). At pre-plant N rate=146 kg N ha⁻¹, however, side-dress N rate was highest when derived from passive-NDRE (32 kg N ha⁻¹) and different only from the lowest side-dress N rate (passive-NDVI, 8 kg N ha⁻¹). For SY8, side-dress N rate at pre-plant N rate=72 kg N ha⁻¹ from active-NDVI (124 kg N ha⁻¹) was the highest and different only from the lowest side-dress N rate (passive-NDVI, 61 kg N ha⁻¹). For SY8, side-dress N rate at pre-plant N rate=172 kg N ha⁻¹ from passive-NDVI (97 kg N ha⁻¹) was higher than other sensor-VI combinations, with the lowest side-dress N rate observed from both active- and passive-NDRE, and active-NDVI (22, 22, and 46 kg N ha⁻¹, respectively).

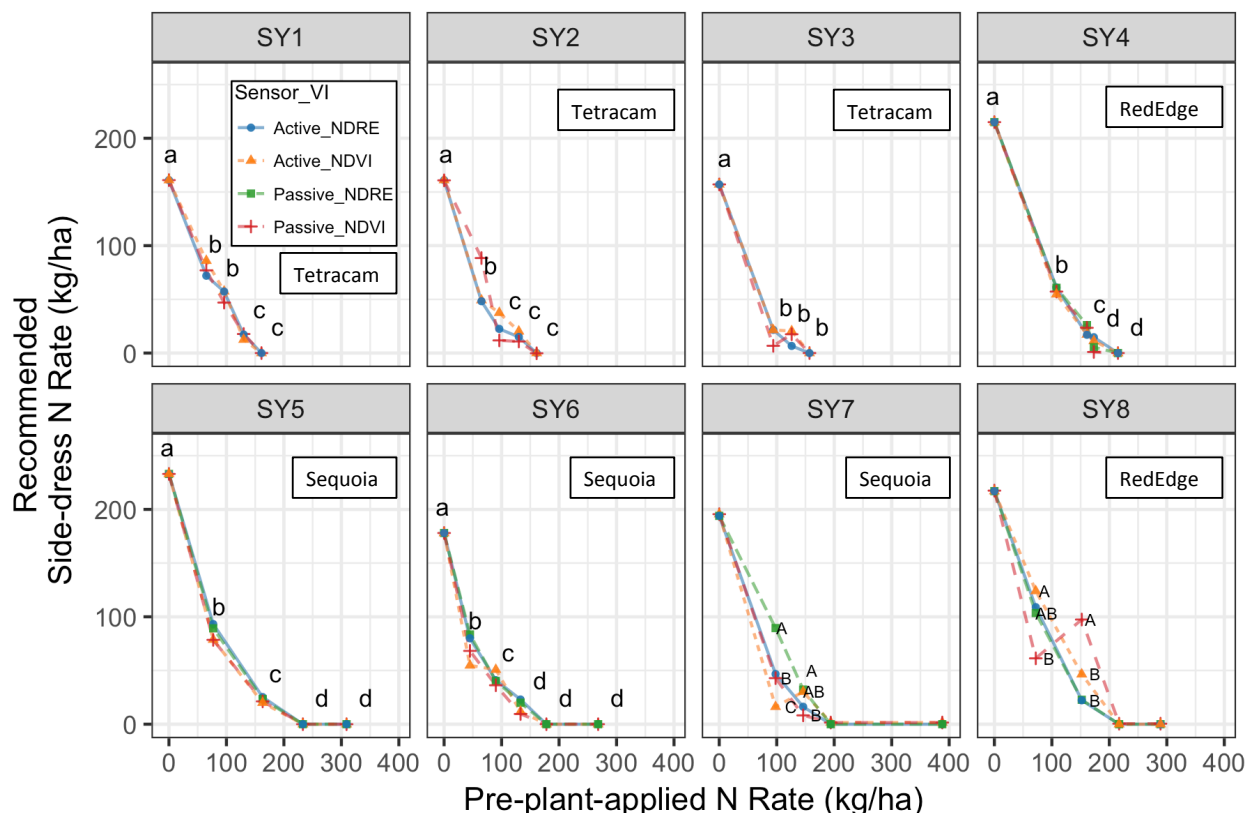


Figure 4.2. Recommended side-dress N rate derived from both sensor types (active and passive) and vegetation indices (NDRE and NDVI) as a function of pre-plant-applied N rate at different site-years (SY). Side-dress N rate means within a given SY and over multiple pre-plant N rates followed by a common lowercase letter are not significantly different at $\alpha=0.05$. Side-dress N rate means within a given SY and pre-plant N rate followed by a common uppercase letter are not significantly different at $\alpha=0.05$. Text box in each panel is the passive sensor used at that SY.

N-responsiveness and EONR

Response to N application varied among SYs, where all years showed positive crop N response to fertilizer except for SYs 1 and 2 (Table 4.1). For SY1, N deficiency at time of sensing was severe and affecting all pre-plant N rates such that EONR was not estimable. For SY2, yield showed no response to pre-plant N rate compared to the already high-yielding 0N check (13.7 Mg ha^{-1}), so EONR was set to zero. For SYs 3 through 8, the best-fit model between

grain yield and pre-plant-applied N rate was either quadratic or quadratic plateau. For those SYs, EONR varied from 30 to 235 kg N ha⁻¹ (Table 4.1).

VRN Accuracy

For each SY, the accuracy of VRN recommendations was assessed by how well a side-dress N rate plus pre-plant N rate (i.e. total annual fertilizer input) approximated EONR (Figure 4.4). A side-dress N rate recommendation was considered accurate if it met two criteria: i) under N-deficient conditions, the total annual N input was $EONR \pm 10 \text{ kg N ha}^{-1}$, and ii) under N-sufficient conditions, the side-dress N rate recommendation was near or at zero.

Given that side-dress N rate at SYs 1 through 6 was not affected by either sensor type or VI, side-dress N rate accuracy was assessed for each pre-plant N rate (Figure 4.3). At SY1, N deficiency at time of sensing was severe to the point of affecting all pre-plant N rates. Thus, EONR was not estimable due to the overall low grain yields regardless of pre-plant N rate. At SY2, no pre-plant N rate was able to improve yield compared to the already high-yielding 0N check, and thus EONR was set to zero. Although a lack of grain yield differences among pre-plant N rates was observed, all sensors and VIs inaccurately recommended side-dress N rate at SY 2. For SYs 3 and 4, all recommended side-dress N rates were greatly above EONR, with the highest discrepancy for the 0N checks. For SY5, side-dress N rates were accurate for the 0N check but too low at 77N and 163N. The recommendation to add no side-dress N was accurate at 233N and 309N, which equaled or exceeded EONR. Similarly for SY6, side-dress N rates were accurate for 0N and pre-plant rates >133N, but too low for 45N and 90N rates.

Side-dress N rate at SYs 7 and 8 was affected by both sensor type and VI, and thus side-dress N rate was calculated for all four combinations (Figure 4.4). At SY7, all sensor x VI combinations generated side-dress N rates that surpassed EONR when pre-plant N rate=0, while

only active-NDRE side-dress N rate was able to match EONR at the other pre-plant N rates. At SY8, all sensor x VI combinations generated side-dress N rates that matched EONR when pre-plant N rate=0, while active-NDVI was the best combination for creating side-dress N rates that approached EONR at other pre-plant N rates while concurrently recommending no side-dress N rate when pre-plant N rate > EONR.

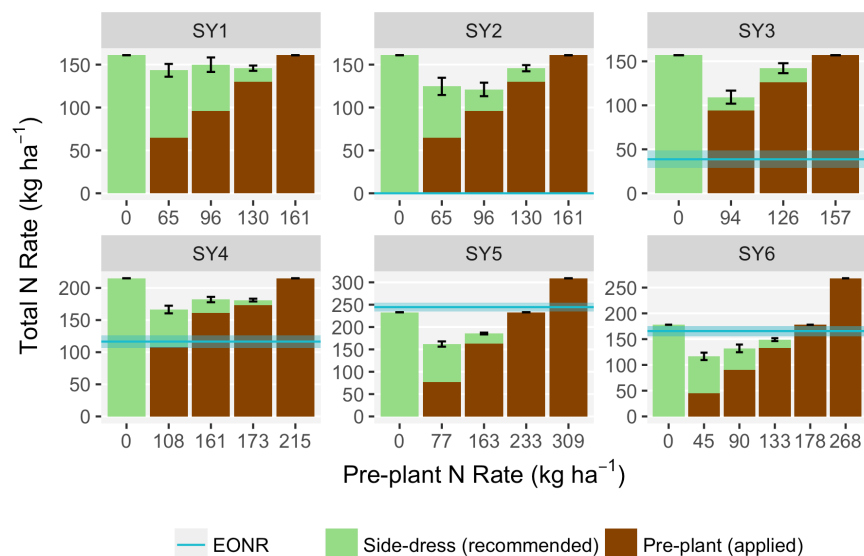


Figure 4.3. Recommended side-dress N rate averaged over vegetation indices (NDRE, NDVI) and sensor types (active, passive) at V12 growth stage for site-years (SY) 1 through 6 using the Holland-Schepers algorithm. Black bars represent standard error of the mean side-dress N rate. Light blue horizontal line represents SY-specific EONR, with shaded light blue band representing EONR ± 10 kg N ha⁻¹.

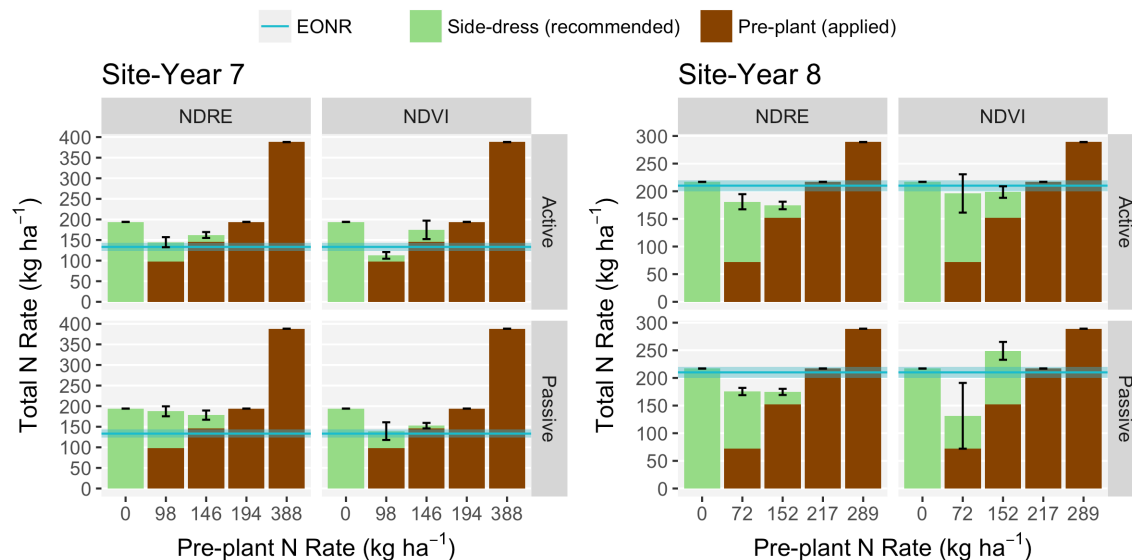


Figure 4.4. Recommended side-dress N rate for each combination between vegetation indices (NDRE, NDVI) and sensor types (active, passive) at V12 growth stage for site-years (SY) 7 and 8 using the Holland-Schepers algorithm. Black bars represent standard error of the mean side-dress N rate. Light blue horizontal line represents SY-specific EONR, with shaded light blue band representing $EONR \pm 10 \text{ kg N ha}^{-1}$.

Discussion

The selection of reference type, sensor type, VI, and DSI can impact the accuracy of in-season sensor-based N application rates. The negligible differences ($\sim 0.6\%$) between reference VI values calculated with NRS or VR suggests that NRS can be replaced by VR without loss of information under these conditions. Different combinations of SY, sensor type, and VI are not expected to have similar reference VI values due to inherent differences among SYs (sensing time, planting date, soil type, weather conditions until sensing date, level of N stress), sensor types, and between VIs (absolute NDVI values are higher than NDRE due to differential response of R and RE to increased biomass and chlorophyll content). Vegetation index values could differ between NRS and VR, however, when i) the VR area does not include N-sufficient patches; ii) the VR area includes many patches of bare soil (i.e. poor stand establishment); iii) the

VR area is sensed before sufficient canopy cover; and/or iv) the NRS exhibits high-N-induced sulfur deficiency as decreased crop vigor (Franzen et al., 2016). These conditions pose a greater problem when on-the-go active sensors are utilized because VR area sizes are limited unless continuously updated.

In-season side-dress recommendations are sensitive to DSI, and we found that DSI was sensitive to sensor type, VI calculation, and SY-specific crop and N management characteristics. Overall, however, DSI based on NDRE was higher than NDVI, and NDVI-based DSI never exceed that calculated from NDRE. Smaller DSI values from NDVI likely occurred because the red band reflectance used to calculate NDVI saturates at LAI values >3 (Viña et al., 2011) and chlorophyll concentrations $> 3 \mu\text{g cm}^{-2}$ (Gitelson et al., 1996), above which NDVI loses sensitivity in differentiating between varied crop N status levels. NDRE, however, continues to differentiate because it uses the RE band instead of the red band (Viña et al., 2011), and could show larger DSI values. In contrast to our findings, Holland and Schepers (2010) suggested that RE-based DSI are normally smaller than those from red bands. Other authors attribute differences between NDVI and NDRE in active sensors to the mathematical function itself, because reflectance from red and NIR bands move in opposite directions as crop biomass increases whereas reflectance from RE and NIR bands move in the same direction (Bean et al., 2018).

Side-dress N rate will only vary when $0 < \text{pre-plant N rate} < N_{\text{opt}}$. In cases where pre-plant N rate = 0, side-dress N rate = N_{opt} because 0N is used to set SI(0) and to calculate DSI. Under these conditions, the square root component of the algorithm becomes 1. When pre-plant N rate $> N_{\text{opt}}$, side-dress N rate is set to zero since otherwise side-dress N rate would be calculated as a negative number. Overall, we found that crops were N-responsive in 75% of SYs

(6 of 8), and that partially or fully accurate side-dress N rates were recommended for 67% of N-responsive years (4 of 6). The accuracy to side-dress N rates were based on whether pre-plant plus side-dress rates approximated EONR ($\pm 10 \text{ kg N ha}^{-1}$) under N deficient conditions, or if side-dress recommendations were near or at 0 under N-sufficient conditions. Furthermore, in 75% of SYs, side-dress N recommendation rates were similar regardless of sensor type nor VI calculation method, varying only as a function of pre-plant N rate.

At SY1, grain yield was not responsive to N fertilizer, likely due to high N deficiency in all treatment plots that resulted from N leaching through coarse-textured soils under high water inputs (315 mm of rainfall plus 318 mm of irrigation water from June to August). In contrast, side-dress N rates for SY2 were set to 0 because non N-responsive yield was due to overall high-yielding conditions. At SYs 2 and 3, side-dress N rate was well above EONR, evidenced from the lack of differences in active sensor data acquired throughout the growing season.

At SY4, NDRE SI varied more over time. The increase in NDRE SI for pre-plant N rate=0 from V12 to R5 indicates that considerable N mineralization from SOM occurred during this period. Since side-dress N rate was based on sensor data collected at V12, no combination between sensor and VI could have accounted for atypical N mineralization that occurred after sensing. When comparing rainfall and air temperature data after sensing among SYs conducted at SCAL over different years (data not shown), the only noticeable difference is the average mean air temperature in September. At SYs 2 and 3 (both conducted in 2015 at SCAL), average mean air temperature in September was 25.7°C , compared to 19.2 , 19.6 , and 19°C at SYs 4-5, 6, and 8, respectively, which were the other SYs conducted at SCAL over different years. This significantly higher temperature in September at SYs 2 and 3 could have led to increased N

mineralization and explain, in part, the hypothesis of enhanced N mineralization in those two occasions.

At SYs 7 and 8, side-dress N rate was affected by pre-plant applied N rate, sensor, and VI. Passive-NDVI side-dress N rate performance at SY8 was an example of a desirable outcome when using sensor-derived VRN, because it was able to generate a high enough side-dress N rate that was within $EONR \pm 10 \text{ kg N ha}^{-1}$ under N-deficient conditions, and a low enough side-dress N rate that did not surpass EONR under N-sufficient conditions.

Bean et al. (2018) evaluated the performance of different VRN algorithms in recommending a side-dress N rate that was aligned with end-of-season calculated EONR for 49 sites. The authors observed that the Holland-Schepers algorithm performed better when NDRE was used instead of NDVI, although this algorithm was developed in such a way to be sensor-VI independent. However, Bean et al. (2018) used a constant DSI value of 0.3 when assessing the algorithm performance. In our study, DSI varied considerably and the use of a SY-sensor-VI specific DSI considerably improved the side-dress N rate agreement between different sensors and VIs.

While active sensor data quality can be impacted by the amount of non-plant pixels sensed, passive sensors data are more prone to variability related to multiple sources and decisions taken in the imagery acquisition and processing steps. For instance, factors that could have impacted passive sensor data variability are time of the day when sensing was performed, cloud cover, quality of downwelling radiation acquisition for reflectance correction, proper geolocation of multiple bands, proper non-plant pixel identification and removal, etc. Although these conditions and decisions were controlled to our best capacity, normal variation could have created somewhat different outcomes. Under these studies, the passive sensor was flown between

10:30 to 16:00 local time, with most flights between ± 2 h from solar noon under sky conditions varying from clear to some scattered clouds not casting shadow on the study area. Furthermore, the same statistical procedures were utilized for non-plant pixel identification and separation before band reflectance was averaged within each plot.

It has been demonstrated that NDVI loses sensitivity in differentiating crop N status at higher biomass and chlorophyll levels (Gitelson et al., 1996; Viña et al., 2011). These conditions were observed in our irrigated corn studies at the V12 growth stage, as evidenced by VI data (not shown). However, the implementation of a variable DSI for each SY-sensor-VI combination was able to create side-dress N rates that were comparable regardless of VI and sensor used in most cases. This finding is key because i) it demonstrates that sensors having only red and NIR bands can still generate reliable information to derive side-dress N rates even when VI data demonstrates sensitivity loss, and ii) it reinforces the need to use site-sensor-VI specific DSI rather than a default value. One limitation of using a variable DSI is that its calculation involves collecting reflectance data from an area not fertilized with N. Thus, the use of variable DSI in sensor-based VRN of normal production fields where non-fertilized areas are not desirable may not be practical.

It is important to note that EONR was calculated based on pre-plant N rate treatments only, and that EONR based on side-dress N application could have been different. A U.S. Midwest multi-state, multi-year corn N management study comprising a total of 49 sites with N rates varying from 0 to 315 kg N ha⁻¹, applied both all at pre-plant or split into 45 kg N ha⁻¹ pre-plant plus different side-dress N rates, observed a wide range in EONR from both pre-plant and split-applied fertilizer (from 0 to 315 kg N ha⁻¹), with average pre-plant and split application EONR of 169 and 159 kg N ha⁻¹, respectively (Kitchen et al., 2017). This is an indication that

EONR based on pre-plant N rates may be a reliable proxy to what side-dress EONR could have been had it been evaluated.

Conclusions

The performance of different crop canopy sensors in assessing in-season corn N status and recommending a side-dress N rate that matches EONR has been shown. The main findings of our work were: i) no difference was found in reference VI when comparing NRS and VR; ii) observed DSI \ll default DSI of 0.3 in Holland-Schepers algorithm; and iii) different passive and active sensors can be used to effectively recommend in-season N rates, though efficacy can be dependent on VI selection in some years.

The Holland-Schepers algorithm was utilized to translate sensor information into an N recommendation. To implement the algorithm, a virtual reference approach was utilized when calculating SI, based on preliminary evaluation that found NRS and VR approaches resulted in the same SI under conditions of this study. Delta SI significantly varied depending on the SY, sensor type and VI. Once DSI was allowed to vary accordingly, side-dress N rates from different sensors and VIs were comparable. Therefore, DSI should be calculated using information specific to the sensor and VI being employed at the site that is receiving VRN. Different sensor types and VIs have the potential to similarly assess corn N stress and create a side-dress N rate in agreement with EONR when proper algorithm inputs are selected and when no significant N-stressing and yield-reducing event happens after the time of sensing.

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Chapter 5 - Summary and Generalized Recommendations for Nebraska

Corn Production

Introduction

On a state level, Nebraska corn producers have continuously improved their nitrogen (N) fertilizer use efficiency (NUE) through increasing grain production at near-constant average N rates over the past 60 years. However, corn production NUE under certain groundwater management areas has stagnated in the past three decades, possibly due to current N fertilizer management practices having achieved their maximum potential efficiency. In order to further improve NUE, next-generation management practices such as fertigation, the use of stabilized fertilizers, and in-season sensor-based N management have been proposed (Ferguson, 2015).

These tools can be categorized as protective and reactive approaches. Protective approaches include the use of stabilized fertilizers in order to keep N from being lost given conducive weather conditions exist. These conditions could be no rainfall following urea-based fertilizers application, or excessive rainfall after N application leading to leaching. By protecting fertilizer from weather-driven environmental losses, protective approaches may maintain more N available for the crop and avoid a nutritional deficiency that can lead to yield penalty.

Reactive approaches include the use of in-season sensor-based assessment of crop vigor and the formulation of an N rate to mitigate a stress when present. Reactive approaches work by allowing weather conditions to affect N dynamics and crop nutrition to the point of a mild nutritional stress, and then work to correct it. Because a reactive approach is normally used during the mid-vegetative growth stages in corn, it reduces the need to predict and account for early-season weather-driven losses before they happen. With that, the uncertainty of how much

N will be needed to finish the season is also decreased, and N rates can be fine-tuned to better match the optimum required for production.

The benefits of using sensors to assess spatial variability in crop vigor and vary N rate accordingly are often confounded with the fact that it is also split-applied (Colaço and Bramley, 2018). Studies evaluating both the sensor-based and the timing aspects of in-season N management separately have demonstrated that the use of sensors can improve NUE and grain yield compared to fixed-rate split application, but that split application alone comprised most of the improvement over pre-plant-only applied N fertilizer (Colaço and Bramley, 2018).

The objectives of this chapter were: i) to summarize recent evaluations of the use of stabilized fertilizers including urease inhibitors (UI) and nitrification inhibitors (NI), and the use of crop canopy sensors for in-season crop vigor assessment and fertilizer recommendation; and ii) to generalize these research findings to other corn growing regions of Nebraska based on county-level weather and soils data. To extrapolate sensor-based N management, it was hypothesized that county-level soil texture class variability was related to within and between fields soil texture variability.

Materials and Methods

In order to generalize recent research findings about the effect of next-generation N management practices on corn production to the entire state, various spatial data layers were downloaded, processed, and analyzed. Those included county-level corn planted and harvested area in 2018 (USDA NASS, 2019); state-level monthly cumulative precipitation normal (1981-2010); and state-level STATSGO soil surface texture data (USDA Geospatial Data Gateway, 2019).

The first processing step was to select only counties that reported corn production data for the 2018 growing season. The filtered county layer was utilized to subset the state-level monthly precipitation and soil texture layers, and to extract a mean value for each county. Whenever a county comprised multiple values for a given spatial layer, the area extent of each different value within the county was derived, and a weighted mean value per county was calculated. For the precipitation layers, county-level data was extracted for the months of February through April. Thereafter, county-level precipitation layers were categorized according to different response thresholds (explained below).

To extrapolate UI use to Nebraska, the main variable considered was cumulative precipitation following fertilizer application, assuming 20-mm precipitation volume to be the threshold between high and low potential for ammonia loss for surface-applied ammonium-containing fertilizer. Since the precipitation data layers were the average cumulative precipitation in a given month, the specific timing of fertilizer application could not be derived. To grossly overcome this limitation, a threshold of 50 mm cumulative rainfall was considered for each of the months March, April, and May, assuming that N surface application to corn would mostly happen within this period. Thereafter, counties were classified as <50 mm and >50 mm for each of these months, assuming <50 mm to be under higher risk of ammonia loss and most probably responsive to UI.

To extrapolate NI use to Nebraska, the main variable considered was cumulative precipitation in two-month intervals (February and March, March and April, and April and May). The interval of two months (~eight weeks) was chosen because, according to results observed in Chapter 3 of this document, 10 weeks after fertilizer application was the summarizing window that best described the response of corn yield to NI use (negative, neutral, and positive NI effect).

The monthly average cumulative precipitation of the two months considered for a given interval were summed, and a threshold of 142 mm was used to classify counties as <142 mm (higher chance of negative NI effect on yield) and >142 mm (higher chance of positive NI effect on yield).

To extrapolate the use of sensor-based N management to Nebraska, the main variable considered was soil surface textural class variability within a county. Because the extrapolation was aimed at the county level, it was hypothesized that greater variability of soil texture in a county would also be reflected in greater variability within and among fields in that county, and be most benefitted from sensor-based variable rate N management.

In order to calculate surface soil texture variability at the county level, the number of different soil texture classes and their relative area to the county total area was calculated using STATSGO surface soil texture data. Thereafter, counties were classified as having low, medium, and high surface soil texture variability when the soil texture class with the higher proportion in the county represented >70%, between 50% and 70%, and <50% of the county total area.

Results and Discussion

County Selection

Of the total 93 counties in the state, only 72 reported corn production data in 2018 (Figure 5.1) and were selected for subsequent analysis. The largest and smallest harvested corn area in 2018 was recorded in Perkins (95,142 ha) and Garfield (4,569 ha) counties, respectively.

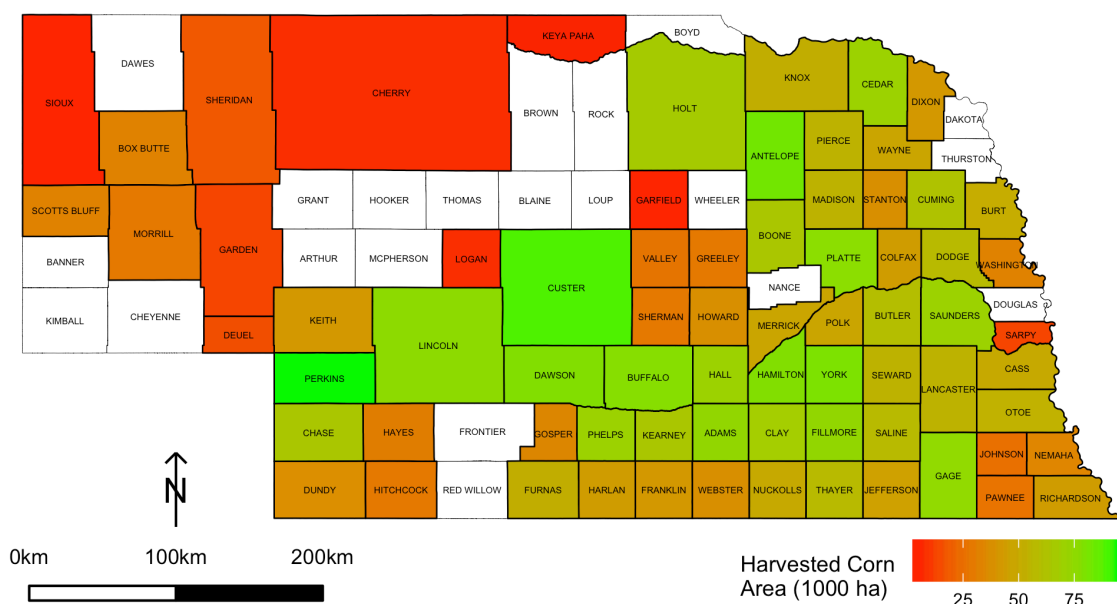


Figure 5.1. Nebraska state and county boundaries, including county names, depicting the harvested corn area (1000 ha) for the 2018 growing season based on USDA reported data.

Urease Inhibitor – Summary and Extrapolation to Nebraska

The UI study was aimed at i) comparing different UI, including a new commercial formulation, and UI+NI products on how they affect sealed-chamber measured NH_3 volatilization losses from surface-applied urea-ammonium nitrate; and ii) assessing the impact of different UI and UI+NI products on corn growing season vigor and grain yield. This study was comprised of five site-years (SY), conducted from 2014 through 2017 on different soil types (silt loam, loamy sand) and with a range of pre-plant-applied N rates (0 to 268 kg N ha⁻¹).

Over all SYs, volatilization losses ranged from 0 to 26 $\text{NH}_3\text{-N}$ ha⁻¹. The use of UI was able to decrease NH_3 losses from 4 to 16 kg $\text{NH}_3\text{-N}$ ha⁻¹, which represented a reduction in loss of 21-62%. The use of UI+NI increased NH_3 losses compared to UI only at the loamy sand site, possibly due to lower cation exchange capacity and H^+ buffering capacity, and was not different

from UI in two other silt loam sites. Three of the five SYs responded to N fertilization, with agronomic optimum N rate ranging from 160 to 208 kg N ha⁻¹. However, corn grain yield was not affected by UIs in any of the sites.

Based on our research results, review of the literature, and availability of public data, the average normal (1981-2010) cumulative precipitation for the months of February through May was summarized for the selected counties (Figure 5.2) and used as a proxy for the probability of response to UI use in Nebraska (Figures 5.3 through 5.5). Cumulative precipitation varied in space and time, with smallest and largest volumes of 0 to 23 mm in February, 0 and 51 mm in March, 25 and 76 mm in April, and 51 and 124 mm in May, respectively.

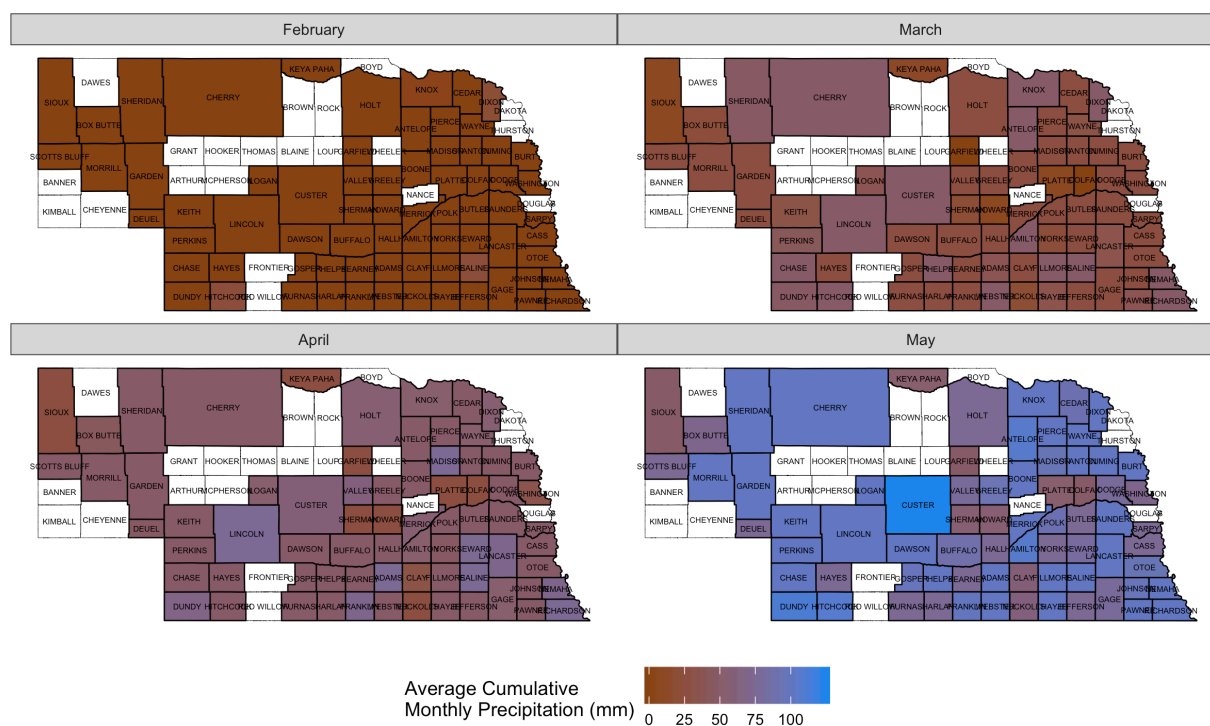


Figure 5.2. Nebraska state and county boundaries, including county names, depicting the average normal (1981-2010) cumulative precipitation, in mm, for the months of February through May.

For surface applications of urea-containing N fertilizer during the month of April, 13 of the total 72 counties were classified as <50 mm and thus considered susceptible to N losses in the form of ammonia volatilization (Figure 5.4). These counties would have a higher probability of decreasing ammonia volatilization by using a UI and therefore protecting final grain yield.

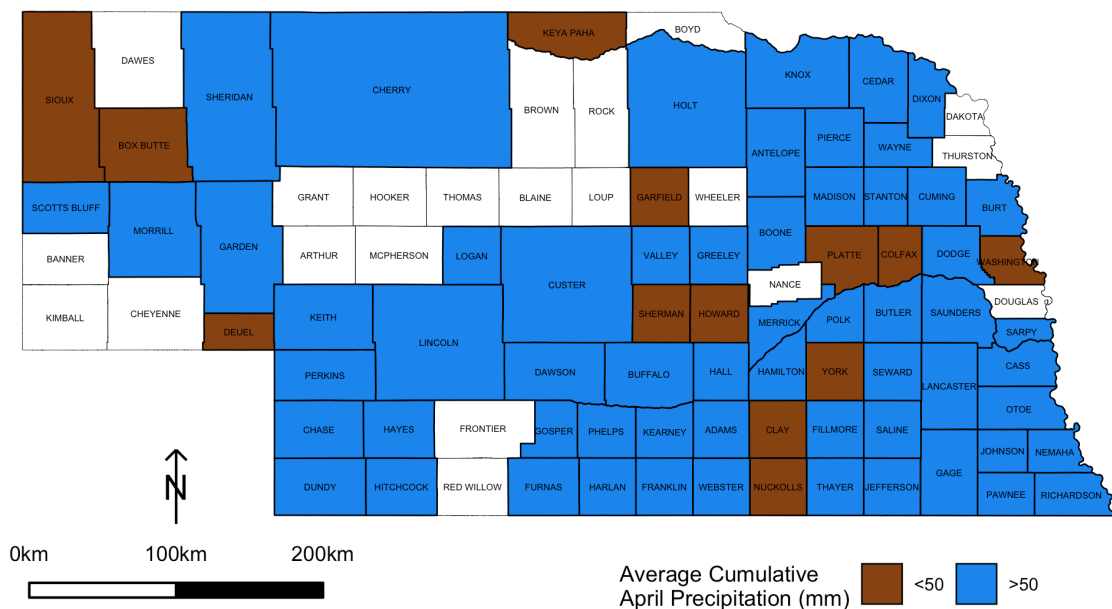


Figure 5.4. Nebraska state and county boundaries, including county names, depicting the average normal (1981-2010) cumulative precipitation for the month of April as <50 mm and >50 mm.

For surface applications of urea-containing N fertilizer during the month of May, none of the total 72 counties were classified as <50 mm and thus considered susceptible to N losses in the form of ammonia volatilization (Figure 5.5). Therefore, the risk of ammonia loss and probability of benefiting from UI use when surface-applying urea-containing N fertilizer is greatest for March applications, intermediate for April applications and very limited for May applications for different counties in Nebraska.

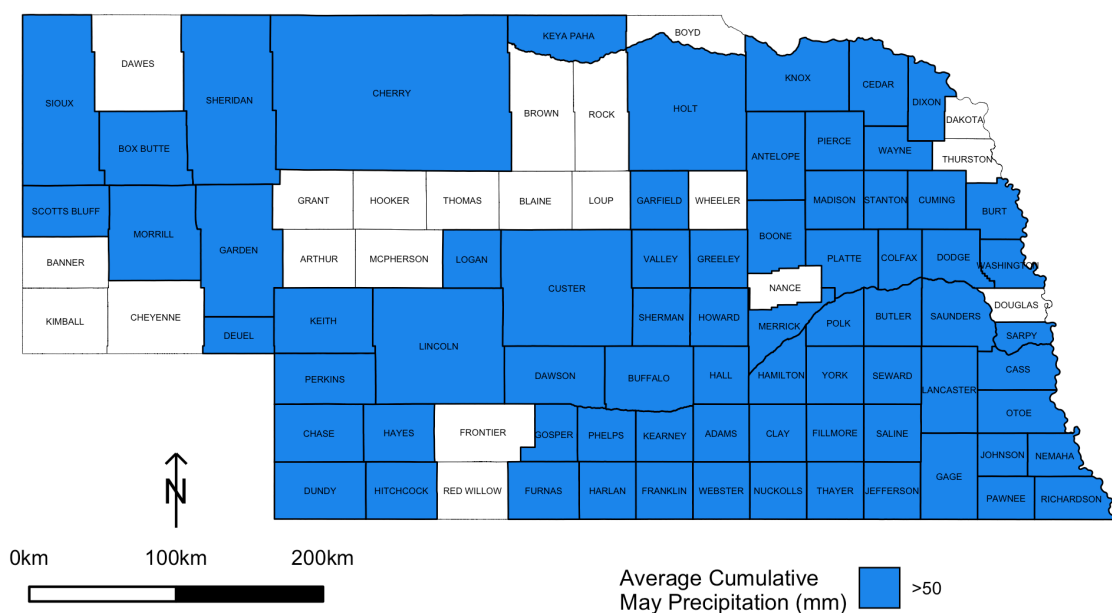


Figure 5.5. Nebraska state and county boundaries, including county names, depicting the average normal (1981-2010) cumulative precipitation for the month of May as <50 mm and >50 mm.

One limitation of the UI extrapolation is the fact that rainfall timing in relation to N application timing could not be explicitly considered. Also, this extrapolation does not account for the effect of temperature on urea hydrolysis. Urea hydrolysis will be slower at cooler temperatures. Thus, urea applied in March may remain in the urea form longer than with warmer temperatures in May, and the likelihood of rainfall incorporating urea before much volatilization occurs is higher. Nonetheless, this extrapolation provides a general idea of where and when in Nebraska conditions could be more conducive to ammonia volatilization considering the lack of rainfall as the main driving factor.

Nitrification Inhibitor – Summary and Extrapolation to Nebraska

The NI study was aimed at i) understanding the long-term effects of N rate (0, 75, 150 and 300 kg N ha⁻¹), tillage [conventional (CT) vs. reduced (RT)], N application timing (pre-plant and side-dress) and the use of a NI (Nserve, with vs. without) on irrigated corn grain yields, and ii) assessing weather patterns responsible for different yield responses over time. The study has been continuously conducted near Clay Center, NE on a silt loam for over 28 years.

All treatment factors impacted corn grain yield over time, in the order of N rate > Tillage > N Timing > NI. The most important weather variables in explaining different yield responses over time were related to year-yield potential, air temperature and precipitation. NI applied at pre-plant increased yield over fertilizer alone more often than when applied side-dress (32 vs. 5% of the years), especially under medium to low-yielding years under lower mean air temperature (<6°C) three weeks after fertilizer application (AFA). Side-dress fertilizer application increased yield over pre-plant in 47% of the years, which occurred mostly during medium- and high-yielding years under dry conditions (cumulative precipitation over 13 weeks AFA <210 mm).

The use of NI at different N rates created positive (32% of years), neutral (54% of years) and negative (14% of years) impact on yield as compared to fertilizer alone. Most of the years with a negative impact of NI on yield happened under dry conditions (cumulative precipitation over 10 weeks AFA < 141 mm), whereas most of the years with a positive impact of NI happened in wetter (>142 mm) and hotter (average corn heat units >15) conditions under medium- and high-yielding years. CT yielded higher than RT 40% of the years, and in no occasion RT yielded higher than CT. Weather conditions most conducive to CT yielding higher

were higher wind speeds ($> 3.6 \text{ m s}^{-1}$), more evenly distributed rainfall and higher mean air temperature ($>4.5^\circ\text{C}$) over two weeks AFA.

Overall, the use of NI had the least impact on yield over time compared to other N fertilizer practices, and promoted positive, neutral, and negative yield outcomes. The use of NI was most beneficial when applied pre-plant followed by wet and hot weather conditions early in the season. The use of NI had the most negative impact on yield under dry conditions early in the season, and thus cannot be considered a risk-free practice.

Based on our research results, review of the literature, and availability of public data, the average normal (1981-2010) cumulative precipitation for the periods of February to March, March to April, and April to May were summarized, assuming fertilizer application in the first month of each period, for the selected counties and used as a proxy for the probability of response to NI use in Nebraska (Figures 5.6 through 5.8). Cumulative precipitation varied in space and time, with all counties receiving $<142 \text{ mm}$ rainfall in the period of February-March (Figure 5.6) and March-April (Figure 5.7), and thus being at higher risk of NI negatively impacting corn yield if fertilizer was applied in early February and early March. The period of April-May had 29 and 43 counties receiving <142 and $>142 \text{ mm}$, respectively. In this case, a larger number of counties would have a higher probability of NI use to benefit grain yield if fertilizer was applied in early April.

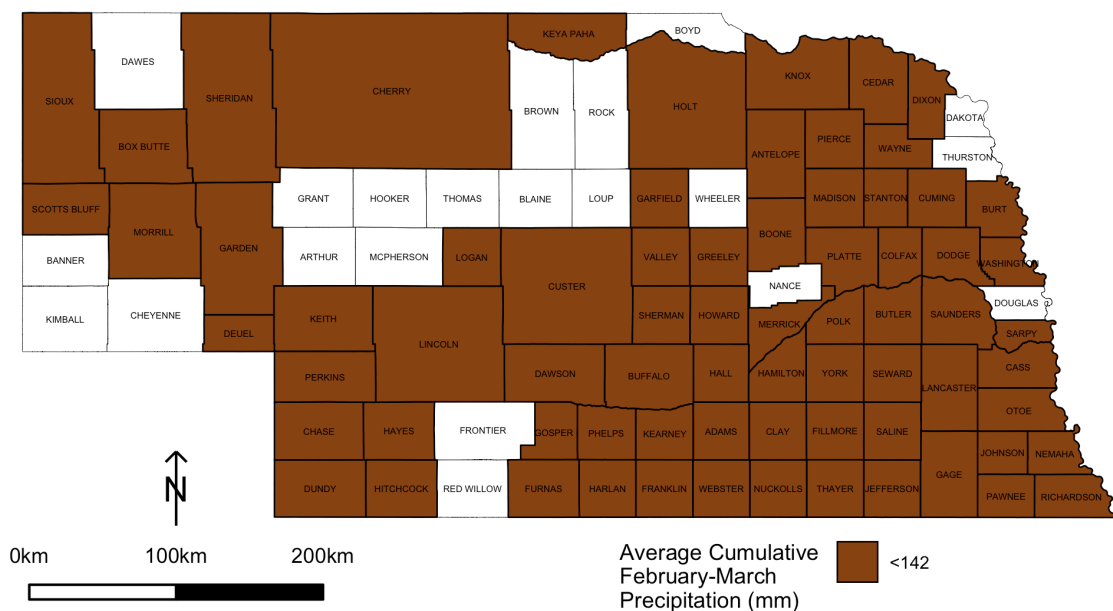


Figure 5.6. Nebraska state and county boundaries, including county names, depicting the average normal (1981-2010) cumulative precipitation for the period of February-March as <142 mm and >142 mm.

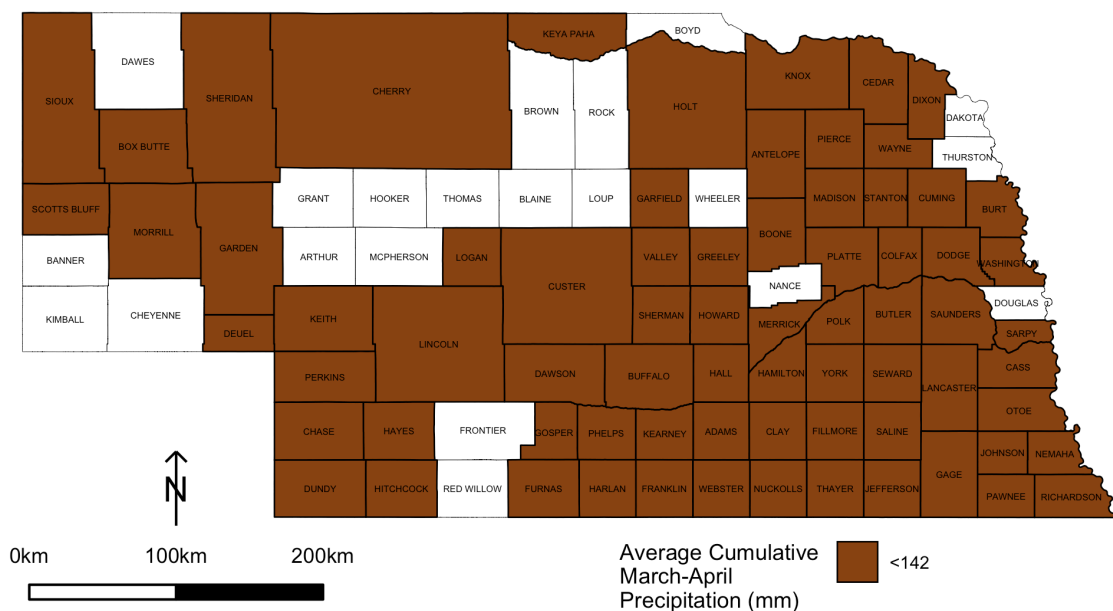


Figure 5.7. Nebraska state and county boundaries, including county names, depicting the average normal (1981-2010) cumulative precipitation for the period of March-April as <142 mm and >142 mm.

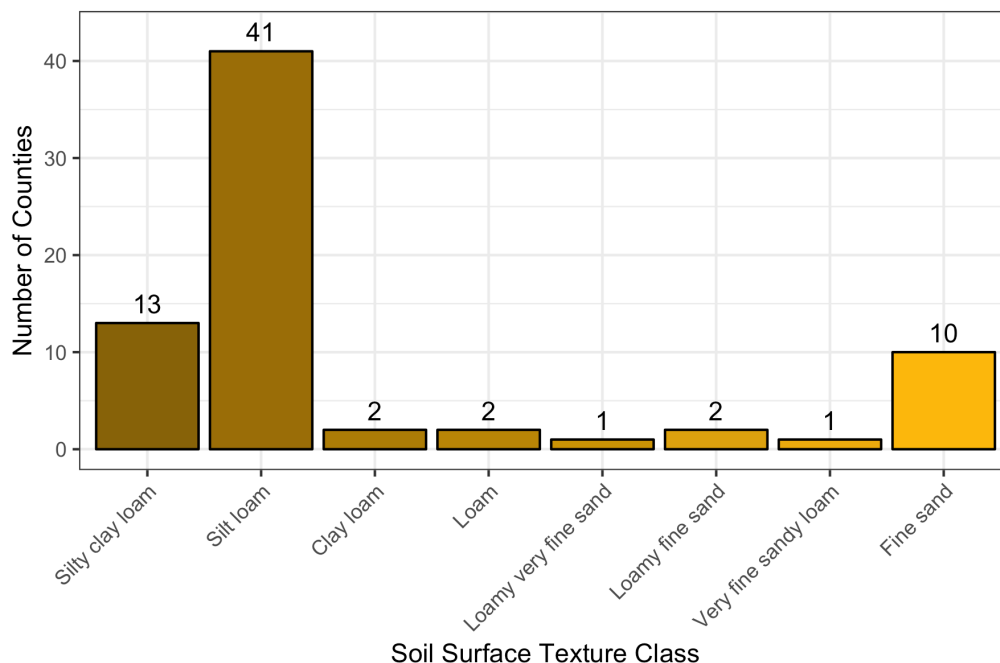


Figure 5.9. Number of counties with a given predominant surface soil texture class.

In-Season Sensor-Based Variable Nitrogen Rate – Summary and Extrapolation to Nebraska

The sensor-based N management study was aimed at i) comparing active and passive crop canopy sensors' recommended side-dress N rate derived from different vegetation indices (VI); and ii) assessing recommended side-dress N rate recommendation accuracy of different sensor and VI types compared to the economic optimal N rate (EONR) in irrigated corn. This study was comprised of eight site-years (SYs), conducted from 2015 through 2018 on different soil types (silt loam, loam, and sandy loam) and with a range of pre-plant-applied N rates (0 to 390 kg N ha⁻¹). Crop reflectance data was acquired using four different sensors: RapidScan (handheld, active) and Tetracam, MicaSense RedEdge or Parrot Sequoia (unmanned aerial system-mounted, passive). Sensors were utilized at the V12 growth stage. For all sensors,

normalized difference vegetation index (NDVI) and normalized difference red-edge (NDRE) were calculated.

Recommended side-dress N rate based on both NDRE and NDVI was affected by pre-plant N rate at all SYs, and further affected by sensor type at SYs 7 and 8. Overall, side-dress N rate varied from 0 to 233 kg N ha⁻¹ and decreased as pre-plant N rate increased for all SYs. Six out of eight SYs were responsive to pre-plant applied N, of which four performed partially or fully satisfactorily in creating a side-dress N rate that when summed to the pre-plant N rate was within EONR \pm 10 kg N ha⁻¹ of that SY. Different sensor types and VIs have the potential to similarly assess corn N stress and create a side-dress N rate in agreement with EONR when proper algorithm inputs are selected and when no significant N-stressing and yield-reducing event happens after the time of sensing.

Because the main objective of this study was to compare N variable rate recommendation from different sensors, it did not generate results that could be directly used for state-level extrapolation of the suitability of this technology. Therefore, the likelihood of a positive response from sensor-based variable rate N management was extrapolated to the state of Nebraska based on the number and extent of different soil textural classes within the selected counties (Figure 5.10).

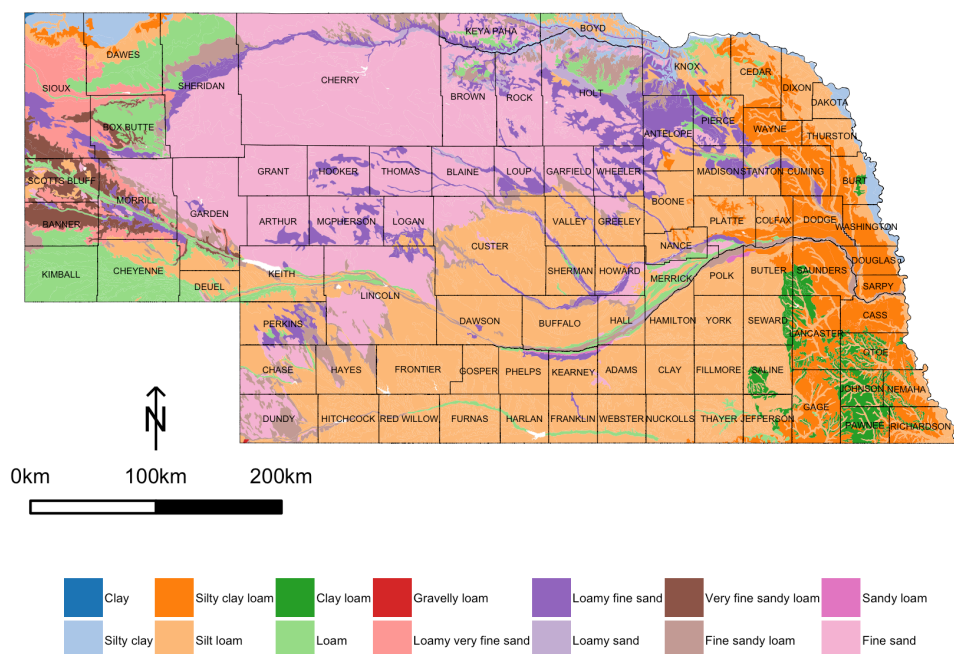


Figure 5.10. Nebraska state and county boundaries, including county names, depicting soil surface texture classes based on STATSGO data.

The hypothesis of using soil texture variability was that greater variability of soil texture in a county would also be reflected in greater variability within and between fields in that county, and be most benefitted from sensor-based variable rate N management. Selected counties were classified as low, medium, and high variability in soil textural class when the major soil texture class in that county represented $>70\%$, between 50% and 70% , and $<50\%$ of the total county area, respectively.

Of the total 72 counties, 32, 22, and 18 were classified as low, medium, and high soil textural class variability, respectively (Figure 5.11). With that, approximately 56% of the selected counties in Nebraska could potentially benefit from utilizing in-season crop canopy

sensors to not only assess the effects of spatial variability in corn, but also to variably apply N to compensate for this variability.

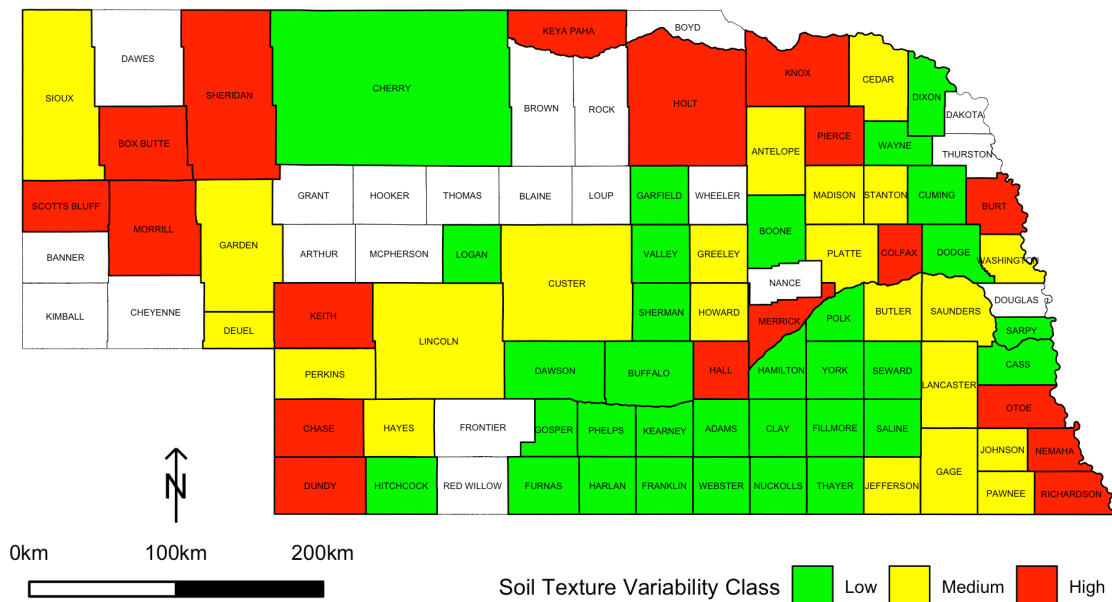


Figure 5.11. Nebraska state and county boundaries, including county names, depicting soil texture variability class of low (>70%), medium (between 50% and 70%), and high (<50%) according to the soil texture class with highest proportion in the county.

County-level Suitability of the Three Next-Generation Strategies

The suitability of using each individual next-generation N management strategy has been demonstrated above. To better understand which management options a producer may have in a given county, a summary containing all three strategies was created (Figure 5.12). The use of UI was classified as “recommended” when either cumulative precipitation in March or April was <50 mm; the use of NI was classified as “recommended” when the cumulative precipitation during the period between April and May was >142 mm; and the use of sensor-based in-season

N variable rate application was classified as “recommended” when the soil texture class variability was classified as either medium or high.

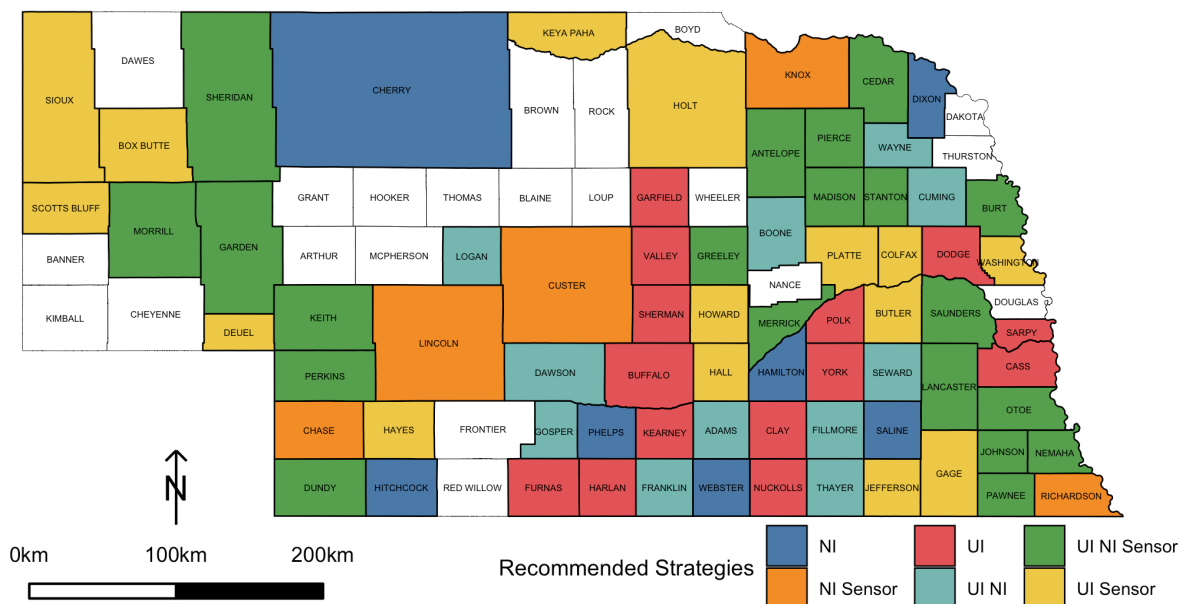


Figure 5.12. Nebraska state and county boundaries, including county names, depicting recommended next-generation N management strategies. UI = urease inhibitor, NI = nitrification inhibitor.

Overall, all selected counties could benefit from the use of at least one next-generation N management strategy, based on the assumptions of this work. The number of counties with a higher probability of positive response from the use of NI alone, UI alone, UI/NI, NI/Sensor, UI/Sensor, and UI/NI/Sensor was 7, 14, 11, 5, 15, and 20, respectively. The fact that all selected counties were considered suitable for at least one next generation strategy demonstrates how N fertilizer can be at risk in the entire state if not properly managed.

Counties identified as suitable for the use of more than one technology (e.g. UI/NI) are at risk of N loss via different pathways and could benefit from managing N by implementing more than one strategy. However, this may become unfeasible given the added cost of different

technologies. Other aspects of N fertilizer management such as N source, placement, and timing should be considered to identify the most critical N loss conditions in a given field and guide the decision of adopting a technology that better protects N loss and grain yield. For example, producers that inject N fertilizer into the soil will not benefit from using a UI, even if the field is within a county classified as “UI/NI”. In this case, these producers could consider the adoption of NI only, which best suits their specific N management conditions.

The results of this extrapolation should be validated using information from previous studies evaluating the effect of these technologies in different growing conditions of Nebraska. Furthermore, once validated, this extrapolation could aid in the targeted selection of regions with a high probability of response for future studies including the use of NI, UI, and sensor-based management.

Conclusions

The use of next-generation protective and reactive approaches to N management have been demonstrated for Central Nebraska. Overall, the use of a UI decreased N losses as NH_3 , but this was not translated into higher yields when compared to untreated fertilizer. Nonetheless, loss-saved fertilizer may have been incorporated into soil organic matter and become available in following growing seasons. The use of an NI created negative, neutral, and positive yield responses compared to untreated fertilizer depending on weather conditions. The use of sensor-based in-season N management was able to recommend an N rate that partially or fully matched crop demand in four of six N-responsive SYs.

The lack of consistent response of stabilized fertilizers on yield has been attributed to many factors. Those include i) lack of yield response to N application (i.e. N was not the limiting

factor); ii) large contribution of N from soil organic matter mineralization or other sources including excessively high N rates; iii) conditions not conducive to loss; iv) N positional unavailability in relation to root active uptake region; v) negative effect of inhibitor on crop growth.

Stabilized fertilizers have been utilized as “insurance” against weather uncertainty. While UIs either have shown positive or no effect on yield depending on the conditions stated above, NIs cannot be considered risk-free since under certain conditions they can also negatively impact yield. Future studies should focus on the probability of different yield responses from the use of stabilized fertilizers based on past weather to generate probable scenarios for a current growing season.

The conditions under which both protective and reactive approaches have failed in these studies were commonly related to weather. The lack of yield response from UI was attributed to either low NH_3 losses and/or N from other sources such as soil organic matter mineralization, both of which are governed by weather. The effect of NI on both loss and yield was weather-driven, with higher chance of decreasing losses and having a positive yield response under wet years. In-season sensor-based N management failed when increased soil mineralization likely happened after sensing.

The extrapolation of these technologies at the county level in the state of Nebraska was demonstrated. Overall, all counties where corn is produced had a high probability of benefiting from the implementation of at least one of the three technologies. This demonstrates that N fertilizer can be at risk of different loss pathways in different regions of the state, and that the use of next-generation N management technologies can aid in managing these losses.

Once validated, this extrapolation could be used to inform the placement of future research field trials evaluating the effect of UI, NI and sensor-based in season N management. Furthermore, future N management approaches would likely benefit from explicitly incorporating weather information into their adoption decision and implementation, rather than be expected to indirectly account for weather uncertainty.

References

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