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Donato Titolo, Student

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Dr. Mark Coyne, Director of Graduate Studies

USING AN ACTIVE OPTICAL SENSOR TO IMPROVE NITROGEN MANAGEMENT IN CORN PRODUCTION

THESIS

A thesis submitted in partial fulfillment of the
requirements for the degree of Master of Science in the
College of Agriculture
at the University of Kentucky

By

Donato Titolo

Lexington, Kentucky

Director: Dr. John H. Grove, Full Professor of Soil Fertility

Lexington, Kentucky

2012

ABSTRACT OF THESIS

USING AN ACTIVE OPTICAL SENSOR TO IMPROVE NITROGEN MANAGEMENT IN CORN PRODUCTION

Corn nitrogen (N) applications are still done on a field basis in Kentucky, according to previous crop, soil tillage management and soil drainage. Soil tests, as well as plant analysis for N, are not very useful in making N fertilizer rate recommendations for corn. Recommended rates assume that only 1/3 to 2/3 of applied N is recovered, variability largely due to the strong affect of weather on the release of soil N and fertilizer N fate. Many attempts have been made to apply N in a more precise and efficient way. Two experiments were conducted at Spindeltop, the University of Kentucky's experimental farm near Lexington, over two years (2010, 2011), using a commercially available active optical sensor (GreanSeeker™) to compute the normalized difference vegetative index (NDVI), and with this tool/index assess the possibility of early (V4-V6) N deficiency detection, grain yield prediction by NDVI with and without side-dressed N, and determination of the confounding effect of soil background on NDVI measurements. Results indicated that the imposed treatments affected grain yield, leaf N, grain N and grain N removal. Early N deficiency detection was possible with NDVI. The NDVI value tended to saturate in grain yield prediction models. The NDVI was affected by tillage management (residue/soil color background differences), which should be taken into account when using NDVI to predict grain yield. Side-dress N affected NDVI readings taken one week after side-dressing, reducing soil N variability and plant N nutrition. There is room for improvement in the use of this tool in corn N management.

Key words: Nitrogen Management, Corn, NUE, Active Optical Sensors, GreenSeeker™.

Donato Titolo

September 14, 2012

USING AN ACTIVE OPTICAL SENSOR TO IMPROVE NITROGEN MANAGEMENT IN CORN PRODUCTION

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September 14, 2012

This work is dedicated to my family, especially my parents, for all their support and love.

Este trabajo es dedicado a toda mi familia especialmente a mis padres por todo su apoyo y amor.

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CHAPTER 1: INTRODUCTION

Approximately 50% of the world's population relies on nitrogen (N) fertilizer for food production. The world uses today around 83 million metric tons of N, which is about a 100-fold increase over the last 100 years. About 60% of total N fertilizer is used to produce rice, wheat, and maize, which are the world's three major cereals. Projections estimate that 50 to 70% more cereal grain will be required by 2050 to feed 9.3 billion people (Ladha et al., 2005). This will require an increase in the used N of similar magnitude if the efficiency with which N is used by the crop is not improved (Ladha et al., 2005). In the United States, 12.5 million metric tons of N fertilizer are used (USDA, 2012b), and this is more than 15% of the world's N fertilizer use.

In 2011, Kentucky occupied 14th place (583,000 has) in planted corn (Zea mays L.) acreage ranking among the producing states, with 1.56% of the total acreage (37 million has) planted in the US (USDA, 2011). This was a 7.6% acreage increase over that of the previous year.

Almost the total surface planted to corn in Kentucky is N fertilized. A corn crop can produce 30-50 kg grain kg^{-1} N uptake (Satorre et al., 2004). Janssen et al. (1990) found this value to be 49 kg grain kg^{-1} N. If we take a value of, for example, 45 kg grain kg^{-1} N, this would mean that a yield of 10 Mg of grain would require 222 kg of N. The state of Kentucky average yield for 2009-2011 was 9000 kg ha^{-1} (USDA, 2012a), then 200 kgN ha^{-1} would be needed according to our calculations. The USDA (2005) shows that the state of Kentucky used an average nitrogen application rate for corn of 192 kg N ha^{-1} which is very closed to the calculated one. This would mean that the N fertilizer rates recommendations are not accounting for the soil-N supply. This is because the soil-N

supply is highly variable at the field level, which determines that N applications for corn in Kentucky are still done on a field basis, and are adjusted for on the previous crop, soil tillage management and soil drainage (AGR-1, 2009). Soil tests for soil N availability (organic or mineral N) have not proven very useful in Kentucky as a guide for N fertilizer rate recommendations. Recommended N rates take into account that only 35 to 70% of the applied N is recovered (Ladha et al., 2005; Roberts et al., 2012). Along with high variability in N recovery, recovery is also made more unpredictable by the strong effect of seasonal weather on soil biological processes that influence the release of native soil N and the fate of N fertilizer. Also, as stated by Shanahan et al. (2008), a number of factors contribute to reductions in Nitrogen Use Efficiency (NUE), including synchrony between soil N supply and crop N demand, uniformity in the N application, and temporal variation in soil N supply. This makes very difficult to predict, with precision, the N rate needed to maximize corn yield and profit. On top of this, as N recommendations are field-averages, in an attempt to not risk a grain yield loss, it is a normal practice for farmers to apply more than the recommended rate, which implies potential negative impacts due to NO_3^- accumulation in subsurface drainage waters and/or in deep groundwater (Baker and Johnson, 1981; Bakhsh et al., 2001). Also, at present, overuse of N has a larger economic impact due to continuing increase in the N fertilizer price.

Economically optimal N fertilizer rate (EONR) recommendations for corn and other crops can vary substantially within a field. Spatial variability in the soil's capacity to provide N to the crop, which is not addressed by current N management practices, is probably the primary factor controlling variable N fertilizer needs (Scharf and Lory, 2009).

The potential for varying N applications within and among fields is justified by the spatially variable nature of organic carbon and nitrogen mineralization and by N loss potential in agricultural fields (Kitchen et al., 2010). When farmers apply flat rates of N on their fields they are disregarding the fact that the N supply from the soil, crop N uptake, and the N response to N fertilizer are not spatially uniform (Inman et al., 2005). Many attempts have been made to apply N in a more precise and efficient way, attempts which have been either unsuccessful or of limited reach, like the chlorophyll meter or the 'Soil Doctor' (Murdock, 1997). Using the combine yield monitor history to apply variable N rates to corn has also not been successful (Murdock and Howe, 2001). Oklahoma State University has developed a variable rate application system for wheat that has proved to be effective, increasing yield by 314 kg ha^{-1} and lowering N application rates by 11 kg ha^{-1} (Raun, 2002). This technology could be used in corn production. Results from the University of Missouri indicate that variable N rate applications using optical sensors have reduced applied N rates while slightly increasing yields, giving a 11 to 22us$ ha^{-1} positive margin over the farmers' normal practices (Roberts et al., 2010).

In the state of Kentucky, this kind of research in corn has not been done. According to the literature, it is possible to hypothesize that active canopy sensors can help growers increase NUE. In this sense, one question would be how early in corn's lifecycle an N deficiency might be detected, and also how early in the corn's lifecycle grain yield could be predicted using the active optical sensor. Reflectance measurements in both the red (RED) and near-infrared (NIR) wavelengths were taken, and with these measurements the Normalized Difference Vegetative Index (NDVI) was computed and

related to the nutritional status of the crop. Differences in NDVI values previous to growth stage V8 are most desired, because if an N deficiency was already present, an N application at that growth stage occurs after some yield potential has already been lost (Varvel et al., 2007).

CHAPTER 2: LITERATURE REVIEW

2.1. Soil Testing

The usefulness of soil testing to improve N fertilizer rate recommendations has been addressed in previous research. Soil testing for nitrates is currently considered the best option for identifying sites where N fertilization will be ineffective in producing a corn yield response (Khan et al., 2001). Two soil nitrate tests, which differ in the time and depth of sampling, have been developed: i) the pre-plant soil nitrate test (PPNT); and ii) the pre-side-dress soil nitrate test (PSNT).

The use of the PSNT has been limited since the collection of samples is inconvenient, being done during the growing season, and also due to the fact that N fertilization has to be postponed until after testing (Khan et al., 2001). Sample timing appears to be very important, and is based on plant size and the grower's intended side-dressing time (Grove, 1992), adding to the limitations of the technique.

Khan et al. (2001) developed a simple soil N test for Illinois soils, the Illinois Soil Nitrogen Test (ISNT), consisting of an estimation of hydrolyzable amino-sugar N. The test has been successful in detecting sites where there is no corn yield response to N fertilization. Williams et al. (2007) conducted research on North Carolina soils (Ultisols were the dominant soil order) and found the ISNT to be a promising test in developing a soil test based N fertilizer rate recommendation. Sawyer and Barker (2011) conducted research on 14 Iowa soils and found the ISNT was not predictive of corn response to applied N, that it did not reflect corn N uptake or NUE, concluding that the ISNT was not recommended for use in Iowa corn production. Osterhaus et al. (2008) evaluated results from 80 corn-N response experiments in Wisconsin and found that the ISNT was not

related to the optimal N rate and that the test had no ability to separate N responsive from N non-responsive fields.

In drier climates, a PPNT has been shown to be useful in predicting corn yield response to N applications (Grove, 1992; Hauck, 1984). In humid regions, the numerous processes involved in N cycling (mineralization, immobilization, nitrification, denitrification, leaching and plant uptake) limit the usefulness of such tests, causing high spatial and temporal variability in soil nitrate concentrations (Khan et al., 2001).

Grove (1992) stated that the PSNT value was really an "indicator" of the soil's N supplying capacity and recommends the use of the PSNT as an index, just to identify fields where no corn grain yield response to fertilizer N addition was the more likely outcome, using a critical value of about 25 ppm NO_3-N to separate N responsive from N non-responsive fields. Particularly in Kentucky, leaching and denitrification of nitrates during the winter months make soil testing for residual soil nitrate at any time between two summer annual cropping cycles problematic – likely poorly related to the fertilizer N needs of the following crop.

2.2. Tissue Testing

Plant tissue analysis for N is one tool for evaluating N status, offering the advantage, over soil sampling, of including the plant as a factor and causing a better understanding of soil N availability at field scale.

Binford et al. (1992) conducted research to evaluate the total N concentration of small whole corn plants (15 and 30 cm tall) as an indication of N availability, and found that the relationship between N concentration in young plants and fertilizer N applied was not consistent. Furthermore, the N concentration of young plants was a poor predictor of soil nitrate-N concentration and was also a poor predictor of corn grain yield. The N concentration in young plants was greatly influenced by factors having relatively little effect on final grain yield.

Scharf (2001) conducted research to evaluate soil and plant tests for their ability to predict optimum N rates across a number of production fields in Missouri. Soil samples were taken at planting, and again at side-dressing, to a depth of 90 cm. Whole plant tissue samples were taken at side-dressing. Minolta SPAD chlorophyll meter readings were also taken at side-dressing. The author's results agree with those of Binford et al. (1992), where plant tissue N at early growth stages (V4-V5) had only a weak relationship with the optimum N rate. However, at V6 the relationship improved and tissue N became the best predictor of optimum N rate, followed by the SPAD meter. Waiting until V6 to take the tissue sample was critical to the success of the test. Both at-planting and side-dress soil tests for inorganic N exhibited weaker relationships with optimum N rates for corn.

Scharf (2001) concluded that although plant analysis for N was best, a major disadvantage was the turnaround time required in sending samples to the lab and getting results back. This was a serious obstacle for a crop that grows as fast as corn. Another important observation was that the SPAD meter was more convenient technology, providing an immediate result, eliminating the problem with turnaround time, and giving reasonably good predictions of the needed optimal N rate.

2.3. Nitrogen Timing

Nitrogen (N) is an element often used as fertilizer, and suffers many soil transformations. These transformations modify the effectiveness of N fertilizer and affect N use efficiency - the amount of applied N taken up by the crop. Depending on soil conditions, N can be affected by different processes and take different pathways. No other nutrient cycle is as complex as nitrogen's. Physical processes such as leaching, run-off, or volatilization can be important causes of N loss, as can biochemical transformations such as denitrification and nitrification. Biological transformations can immobilize applied N in organic molecules, making that N temporarily unavailable for plants, or can mineralize organic matter, making organic N plant-available. These processes are alternatively affected by organic matter content, temperature, moisture, and as these variables change with landscape position, are highly variable within the field (Ladha et al., 2005). Many of these processes are well understood. However, although the N cycle has been thoroughly studied, new questions arise and new research is needed to provide answers. For some time now, the plant use efficiency of applied nitrogen has been heavily addressed. Agronomists know that in order to increase NUE in corn (Zea mays L.), synchronization between the crop's N need and soil N supply has to improve (Ladha et al., 2005; Shanahan et al., 2008).

Corn is planted at some time in the spring, depending on location. For Kentucky, as the winter is generally wet, a considerable amount of any residual (from the previous season) soil nitrate is lost either through leaching as NO_3^- or denitrification as N_2 (g) or N_2O (g). This is why the soil nitrate test is highly variable within the field and a poor predictor of indigenous soil N supply during corn's growth cycle. An accurate in-season

technique is needed to predict N needs and allow in-season fertilization. In an attempt to improve N synchronization, some farmers are performing side-dress N fertilizer applications before the V6-V7 growth stage. High-clearance applicators allow even for later N application/synchronization.

Usually, less than 15% of the total aboveground N uptake, and about 5% of the total dry matter accumulation, of modern corn hybrids has occurred by the V7 growth stage (Shanahan et al., 2007), though yield potential is being set at this early stage. However, by the silking (VT) growth stage, around 60% of total N uptake has occurred and about 40% of total dry matter has accumulated. Therefore, a considerable amount, around 40%, of the crop's total N uptake occurs during a 30 day period (this amounts to around 60 kg N ha^{-1} of uptake for a yield of 12 Mg ha^{-1}. There are opportunities to improve N synchronization by delaying in-season N applications until sometime between the V7 to VT growth stages, provided that yield potential has not been reduced by an earlier N stress (Holland and Schepers, 2010).

In this sense, and as there was not conclusive evidence of yield loss in dry-land corn production systems with late N fertilizer applications, Scharf et al. (2002) conducted an experiment at 28 locations and over a variety of soils (for the major part silt loam Alfisols), where timing of N fertilizer was the experimental variable. A single application of ammonium nitrate was applied at a rate of 180 kg N ha^{-1} at either: i) planting; ii) V7; iii) V14; or iv) VT. Corn yield responded positively to N fertilizer at the majority of locations. The authors argued that when all 28 trials are considered together there was little evidence of yield reduction with N applications delayed as late as V14. However, when a quadratic plateau model was fitted to the data, the model indicated only a small

risk of yield reduction when N applications were delayed until growth stage V12 to V16. Also, there was little evidence of irreversible yield loss when N applications were delayed as late as V11. The authors stated that climate might affect the relative risk of yield loss with delayed N application, and exemplifies this idea by noting that even in a dry year, for many locations, full yield was attained on water stressed corn by surface applying N as late as July (when N might not be available out of root reach due to positional unavailability). The problem with this study is that many of the locations had been amended with animal manure; many others had soybean as a previous crop, and a number of different tillage systems were combined across the entire experiment. There were two un-manured locations under corn after corn, though both were tilled. These three variables: previous crop; manure management; and tillage management; will affect mineralization rates, soil-N supply and therefore, the level and timing of N deficiency.

Binder et al. (2000) conducted an experiment examining the corn N fertilizer response as affected by the degree of N deficiency and timing of N fertilizer application at one Nebraska location with a silty clay loam (Argiudoll) under double disc tillage. The previous crop was sorghum for the first year and fallow for the second. The authors found that applying side-dress N around V8-V10 was one of the best ways of supplying N to corn due to the crop's physiology. However, they also found that soil N status would affect how late the N application could be delayed without causing a yield reduction. In contrast to Scharf et al. (2002), there was evidence of irreversible yield loss at one of the sites when N was applied on or after V6, which means that N availability must be adequate prior to side-dressing to ensure that maximum yield is obtained. Also, as the level of N deficiency increased (lower N rate at planting), the grain yield response to N

decreased with greater delay in the side-dress N application, meaning that there was a positive interaction between the level of N deficiency and the time of N application on corn yield. As a conclusion they stated that the optimum N application time depends on the degree of N deficiency, which is related to both the available soil N and the crop N demand. This was particularly true in the first year of this research, where the climate (higher precipitation and cooler early season temperatures than average) caused more severe N stress than in the second year. In the first year, for the 0 kgN ha^{-1} N rate, N had to be applied prior to V6 to attain maximum yield, but in the second year N application could wait until V16 without any yield loss.

In the literature there are studies which found a corn yield response to the timing of side-dress N applications and studies where such a response was not found. Among the studies where the response was observed, some found the response earlier, and others later, in the corn growth cycle. Our studies, where we have both short and long term N response studies, under different tillage systems on a silt loam soil, will evaluate how early the N deficiency is detected using an active proximal sensor, given different levels of N deficiency. The overall objective is to correct the N deficiency with little or no yield loss.

2.4. Sensors and NDVI

Tools for diagnosing an optimal N rate are little used by corn producers (Kitchen et al., 2001), producers who often do apply N according to previous crop, soil drainage and soil management. Repeatedly, however, producers also make higher application rates of N fertilizer than needed to ensure production of maximum yield, resulting in unused N moving to ground and surface waters (Scharf et al., 2006). Proximal plant canopy sensors offer another opportunity for corn producers to adjust N management for optimal agronomic outcomes, while minimizing adverse environmental impacts.

Chlorophyll is a pigment located in leaf chloroplasts in the majority of plant species. The pigment absorbs radiation in the visual spectrum and has absorption peaks in the blue (400-530nm) and the red (620-700nm). Carotenoids, another leaf pigment, also have an absorption peak in the blue. This causes sensor manufacturers to set sensors to absorb in the red part of the spectrum – in order to avoid the interference in the blue region of the spectrum.

Chlorophyll leaf content is positively correlated with leaf N, N fertilizer rate, and yield. The chlorophyll meter (SPAD Minolta 502) is an active optical sensor which measures light transmitted through the plant leaf at two different wave lengths, one in the near-infrared (NIR) and one in the red (RED) region of the light spectrum, and computes a value determined by Minolta. The meter is a technology that came to the market as a quick, non-destructive alternative to tissue analysis for the assessment of the N status/nutrition of the plant. Chlorophyll meter readings correlate positively with chlorophyll content (Schepers et al., 1992). Corn research with the meter has focused mainly on separating locations that will respond to N fertilizer from locations that will

not; evaluating the meter as a tool to indicate whether and when N fertigation is needed; and on relationships between instrument readings and either soil or plant N concentrations (Scharf et al., 2006). Calibration of the meter to determine crop N status faces the problem of inherited differences in hybrid characteristics, interacting with N availability, whether from soil or fertilizer (Bullock and Anderson, 1998; Schepers et al., 1992). However, it is possible to normalize the numerical data, for a given hybrid or growth stage, against a high-N nutrition control. This permits comparisons across hybrids, locations and growth stages. As such, application of this technology requires an adequately N-fertilized area within the field that can be used as a reference under local growing conditions (Schepers et al., 1992).

At early corn growth stages (about V7), Bullock and Anderson (1998) found no correlation between chlorophyll meter readings and yield. However, they did find a better correlation between leaf N concentration and yield. On the other hand, at advanced stages (R1 and R4) the meter readings were better related to grain yield than leaf N, though it is important to note that there was not a good yield response to N fertilization in this paper. The meter readings were positively correlated with leaf N, a relationship which followed a distinct pattern across time, the correlations coefficient being greater at early stages and consistently decreasing in value as growth advanced.

Scharf (2001) reported that absolute (rather than relative) chlorophyll meter readings taken at V6 were related to the economically optimal nitrogen rate (EONR) and produced N rate recommendations that were lower than N rates used by producers in the same fields. And although the meter N rate recommendation did not increase profitability as the N tissue test recommendation, the meter recommendation at least maintained

profitability when compared with producer chosen N rates. On the other hand, (Bullock and Anderson, 1998) concluded that absolute chlorophyll meter readings could not be used to make accurate predictions of how much N fertilizer would be needed by a corn crop during the growing season.

With irrigated corn, successful N recommendations using relative chlorophyll meter readings have been developed where irrigation water can be used as an N delivery system, with repeated opportunities for application. By repeatedly checking corn N status with the meter, a fixed low rate of N could be applied whenever meter readings fell below a critical value (Shapiro, 1999; Varvel et al., 1997). The technique consisted on relative meter readings, which implies that an area with a non limiting N rate applied is needed to relativise the readings. Scharf et al. (2006) found that relative chlorophyll meter readings better predicted corn grain yield than absolute meter readings.

In rainfed corn production systems, where the opportunity to make corrective N applications is restricted to one application, there does not seem to be conclusive evidence regarding the relationship between relative meter readings and the amount of N needed by the crop. In contrast to irrigated systems where fix low N rates can be applied repeatedly as needed, in rainfed systems the chlorophyll meter will only be useful in guiding N application rates if the meter can be the basis for that single corrective N rate recommendation (Scharf et al., 2006).

Farmers producing corn in rainfed systems that can’t relay in a central pivot for more immediate N management, need a system that will convert reflectance measurements from vehicle-mounted sensors directly into a N-rate recommendation (Scharf and Lory, 2009).

The hand-held GreenSeeker 505 from Ntech Industries is also an active optical sensor, which, unlike the chlorophyll meter, measures reflected light. The GreenSeeker unit has important advantages over the chlorophyll meter, satellite images and aerial photographs in managing corn N nutrition at a field scale. First, the GreenSeeker is faster and less labor intensive at the farm scale. Second, the instrument does not require full canopy or ultra high resolution as do aerial photographs (Scharf and Lory, 2002; Sripada et al., 2005). Finally, the GreenSeeker is an 'active proximal' sensor, not limited by cloud cover or diurnal variation and emitting the light to be measured upon reflectance back to the sensor. The light is emitted at two different wavelengths, RED_{670nm} and NIR_{780nm}, related mainly with plant color or photosynthetic activity and plant structure or canopy biomass and the capacity to assimilate carbon (Kitchen et al., 2010).

Reflected RED radiation is negatively correlated with canopy photosynthetic activity, whereas the NIR reflectance is positively related to canopy biomass (Knipling, 1970). Nitrogen deficient plants often exhibit higher levels of reflectance in the visible (400 – 700nm) portion of the spectra due to reduced photosynthetic activity, and lower reflectance levels in the NIR (>700nm) region explained by the reduced leaf surface area in the N-stressed plants. Also, leaf tissue is known to reflect more NIR radiation than most soil surfaces (Daughtry et al., 2000).

The GreenSeeker instrument computes the Normalized Difference Vegetative Index (NDVI) as: $(NIR_{780nm} - RED_{670nm}) / (NIR_{780nm} + RED_{670nm})$. The NDVI has been found to be a logarithmic function of the canopy biomass, but after canopy closure the biomass can continue to increase after NDVI reaches a maximum. In other words, the NDVI becomes 'saturated' after canopy closure (Gilabert et al., 1996).

Raun et al. (2002) and Mullen et al. (2003) have shown that the GreenSeeker NDVI value can be used to direct variable rate N applications to wheat and improve fertilizer nitrogen use efficiency (NUE). However, limitations to use of the GreenSeeker during corn's in-season application window (V8 –R1) have been documented by Shanahan et al. (2008). As NDVI becomes saturated at intermediate leaf area index (LAI) values, corn's greater vegetative biomass makes sensor use difficult. Further, there is a sensitivity problem associated with the use of a RED band to assess N status (Gitelson et al., 1996).

Clay et al. (2006), using the Cropscan (Cropscan Inc., Rochester, MN) sensor, conducted a study to determine the influence of water and N stress on corn canopy light reflectance. They evaluated different reflectance indices. They showed that the strength of the relationship between reflectance and N or water stress was growth stage and band dependent. Also, they found that by the V11-VT growth stage canopy closure was completed and added fertilizer increased all the spectral indices while reducing reflectance in all the bands except the NIR. This suggests a low sensitivity in the NIR to N nutrition. A comparison between 3 N fertilizer models showed that at V8-V9 the N fertilizer recommendations based on NDVI were more accurate than the recommendations based on yield or water regime. They found a general trend for N stressed corn canopy reflectance to significantly increase in value, over the whole visible spectral range, between V8 and VT. However, the change in reflectance was larger in the green than in the other bands, and Clay et al. (2006) concluded that green reflectance might be more sensitive to N stress than NIR, while RED reflectance appeared to be more sensitive to water stress, as yield losses due to water stress correlated with reflectance in

the green and red bands and also with NDVI. Ultimately, they found that a green NDVI (GNDVI) index correlated better with corn grain yield than many other indices tested in the study. These results are also supported by the work of (Shanahan et al., 2008).

The transformed soil adjusted vegetative index (TSAVI) was proposed as an alternative index to deal with the problem of the changing influence of soil background influence on the NDVI. However, Shanahan et al. (2001) showed that the TSAVI index was not better than NDVI in detecting corn canopy variation. Green NDVI, which substitutes the RED portion of the NDVI equation with the Green portion of the spectrum, was proposed by Gitelson et al. (1996) to enhance the sensitivity of the NDVI, and was found by Shanahan et al. (2001) to better distinguish corn canopy differences.

Martin et al. (2007) used the GreenSeeker to carry out a study where the progression in NDVI of the growing corn canopy was documented, and the spatial variability of corn growth was evaluated using the coefficient of variation (CV). Corn grain and biomass yields were best correlated with NDVI values taken between V8 and V12. They found that this complementary approach, using both average NDVI value and the CV for that value, as related to corn growth stage, was able to improve yield potential estimation above that with the NDVI value alone.

Solari et al. (2008) used the Chlorophyll meter (CM) and also the Crop Circle (Model ACS-210, Holland Scientific, Inc., Lincoln, NE) sensor with which they evaluated an NDVI value using the green part of the spectrum, the 590nm band, and computed a chlorophyll index (CI) as $CI_{590} = (NIR/VIS_{590}) - 1$. They also examined the question of when, in the corn growth cycle, readings should be taken and which index better predicted grain yield. The results exhibited higher R^2 values when readings were

taken during vegetative growth, suggesting that the presence of the tassel was confounding the relationship between the CM values and sensor NDVI or CI values. The authors stated that this might be due to the reduced ability of the sensor light source to penetrate further than the 5th or 6th leaf into the corn canopy when the reading was taken at a height of around 80cm above the canopy. Also, they found that CI values were more sensitive than GNDVI values in assessing crop N status, and although the two sensor measures were equally sensitive in assessing yield potential, the authors suggested that the CI would have a greater potential for directing spatially variable in-season N applications.

Freeman et al. (2007), using the GreenSeeker, performed by-plant measurements of NDVI and studied the possibility of complementing NDVI readings with plant height information for predicting corn forage yield and forage N uptake, and concluded that the best predictor of corn forage yield and N uptake was NDVI calculated alone at early stages (V7-V9) of plant growth.

Scharf and Lory (2009), using the Cropscan MSR87 multispectral radiometer (Cropscan, Inc., Rochester, MN), conducted a study to calibrate reflectance measurements for prediction of the economically optimal N rate (EONR) for corn in Missouri, where sensing was done at V6 and the N treatments were applied at planting. Many wavelengths were evaluated, as were different sensor orientations. They found that the best orientation of the sensor was facing downwards, with the sensible part of the sensor facing the crop canopy. As did Kitchen et al. (2010), these authors believed that applications of banded or starter fertilizer N may lead to diagnostic errors, since the apparent N availability for the plant early in the season did not represent availability

throughout the season. An interesting comment worth noting is that they observed that the proportion of soil captured in the sensor's field of view undoubtedly influenced reflectance measurements most with the downward orientation, suggesting that this soil interference may actually have aided in diagnosing soil N supply, due to the effects of N on plant size, soil cover, and soil contribution to the measured reflectance values. If true, then the relationship between reflectance and EONR would probably be different according to the color of the soil.

Among the wavelengths evaluated by Scharf and Lory (2009), the different NIR bands had no effect on the R^2 for the relationship, while selection of the band in the visible part of the spectrum significantly influenced the R^2. Simple relationships between NIR and VIS bands were no different than those among the different NDVI indices. The EONR was somewhat better correlated with GNDVI (R^2=0.66) than with NDVI (R^2=0.55). Ultimately, the authors concluded that N savings using their calibrations could be anticipated only when pre-plant N rates were limited – the remaining N need, to be applied after crop establishment, had to be 60 kg N ha^{-1} or more.

Kitchen et al. (2010), using a Crop Circle (Model ACS-210, Holland Scientific, Inc., Lincoln, NE) sensor, conducted a study to evaluate the use of active optical sensors to assess corn N need and derive N fertilizer application rates that would return maximum profit relative to the grower's use of a single application rate at planting. They computed GNDVI, and with that value a sufficiency index (SI), in order to normalize the GNDVI measurements against a GNDVI for a well fertilized area within the field. Doing this also normalized the confounding effects of numerous management (e.g., hybrid) and environmental (e.g., soil and precipitation) factors within the field, focusing sensor

management on the specific N needs of the crop. They found that the sensor recognized differences in crop N status between plots that received no N at planting and plots that received 67 kg N ha^{-1}. They observed that when too much N was applied before sensing there was little or no difference in sensor values between corn from the well N-fertilized reference and those where a response to later N application would be expected. When SI values were around 0.9 the analysis showed that another 50 to 125 kg N ha^{-1} was still needed to maximize profit. They explained this wide range in optimal N rates by noting that the crop was well fed with N at early growth stages, which is what the sensor "sees", although later on, in advanced growth stages, the crop will suffer an N shortage because at V12 crop N uptake is only 30 % of total growing season uptake.

The previous work reflect an important obstacle in using this technology to make an N diagnosis for season-long crop N need using an early-season snapshot of crop N status. At side-dressing, even late side-dressing (V12), the crop has not yet taken even the majority of the total N need, still has a long way to go until physiological maturity, and many weather factors might influence yield between side-dressing and maturity, making the N need prediction difficult. Kitchen et al., (2010) found a weak relationship between optimal yield and SI, but believed that the trend in the dataset could be used, empirically, to derive N application rates. On the other hand, their data suggested that the chlorophyll meter might be more effective in delineating subtle differences in crop N nutrition, as this instrument was able to "see" differences in N nutrition much earlier in the growing season. This was because the ground based sensors, in considerable proportion, "see" the upper leaves of the canopy, whereas the chlorophyll meter was used on the last fully expanded leaf, which is more likely to show N deficiency, if present. Ultimately, they

concluded that understanding N source and fate within fields containing different soils, and in rain-fed environments, is complex. They were not able to offer a solid system of ideas to explain why their results were not consistent – and without a definite pattern.

Dellinger et al. (2008) examined the relationship between EONR and reflectance from a ground-based optical sensor and evaluated the potential for side-dress N recommendations. In this study, the Crop Circle ACS-210 sensor (Holland Scientific, Lincoln, NE) and the GNDVI was used. Their results suggested that the use of the GNDVI would be limited to situations where there little or no N fertilizer was applied at planting. Also, the EONR was significantly correlated with the GNDVI when manure was applied at planting or when fertilizer was not applied at planting. However, as soon as a rate of 56 kg N ha^{-1} was applied at planting, the relationship ceased to exist. They also found that a high N reference area at planting was needed for making side-dress N recommendations, and concluded that with this N enriched area relative GNDVI could be used to successfully develop side-dress N recommendations.

CHARPTER 3: MATERIALS AND METHODS

Two different experiments, the Blevins 23 study and the Green Seeker Corn GSC study, were carried out in the 2010 and 2011 corn production seasons at the University of Kentucky's Spindletop experimental farm near Lexington, in Fayette County, Kentucky. Active optical sensors were used to assess the N status of the crop.

3.1. Blevins 23 Field Trial

This is a long term (41 yr) monoculture corn (Zea maiz L.) N rate by tillage study located on a Maury silt loam (Typic Paleudalf). A cover crop (wheat, Triticum aestivum L.) was seeded after corn harvest in order to capture any residual N. The experimental design was split-strips of tillage by N rate, laid out in four randomized blocks. The main plots consisted of two different tillage management: i) no tillage (NT); and ii) moldboard plow (MP). The subplots consisted of four at planting N rates: i) 0 kgN ha^{-1}; ii) 84 kgN ha^{-1}; iii) 168 kgN ha^{-1}; 336 kgN ha^{-1}. Finally, and considering all combinations of tillage and N rates, there were 8 tillage by N rate treatments, with a plot size of 66 m^2.

The GreenSeeker (Ntech handheld model 505) was used to determine and compute the NDVI values. This active optical sensor emits light at two different wavelengths, one in the red part of the visible spectrum, $RED_{650 \pm 10\ nm}$; and one in the near-infrared part of the spectrum, $NIR_{770 \pm 15\ nm}$. The index was computed as:

$$\text{NDVI} = \frac{NIR - RED}{NIR + RED}$$

Each plot's NDVI value was the average of readings taken over two center rows, and NDVI values were determined at two different heights (0.6 m and 1.2 m above the

canopy). Measurements were taken every two or three days from V4 until the corn plant height was such that continued measurements were not possible, about V13.

The chlorophyll meter (Minolta SPAD 502) readings were taken at the same time as the NDVI readings by the GreenSeeker. The meter is also an active optical sensor, emitting in the $RED_{650\ nm}$ and in the $NIR_{940\ nm}$. Ten SPAD readings were taken randomly in each plot. Each reading was taken on the plant towards the center of the last fully developed leaf, avoiding the central leaf vein.

One question for this field study was how early in the corn life cycle an N deficiency could be found, given that the 0 kg N ha^{-1} control rate in this study has been imposed for 41 years. A second question was whether the differences in soil background reflectance due to the different tillage systems would confound NDVI readings. Ultimately, the objective was to assess the ability of the NDVI to predict corn grain yield, in comparison with the SPAD readings. Ten ear leaf tissue samples per plot were taken randomly at R1 and analyzed for total N (leaf N). Grain yield was determined by hand harvesting 40 foot of 0.9 m row, and weighing all ears. For grain sampling, 5 ears were separated, weighed and saved for moisture and grain N composition analysis.

For 2010, in early May, the cover crop was sprayed with Gramoxone Inteon (paraquat dichloride). In late May, the appropriate plots were moldboard plowed to a depth of 20 cm and secondary tillage was done with a disc to a depth of 15 cm. Two days later, May 27, Pioneer corn hybrid 34F96 was planted at 62000 seed per hectare in 0.9 m rows. On June 4 the N treatments were applied as ammonium nitrate (AN, 34-0-0), broadcasting the N fertilizer by hand. Also, 2576 kg pelletized dolomite (CCE = 95%; 54% passing 100 mesh) per hectare was applied to all plots receiving 336 kg N per

hectare. Pre-emergence weed control was performed on June 5, using 1.2 L ha^{-1} of Dual II Magnum (s-metolachlor); 1.2 kg ha^{-1} of Princep 80 WP; 1.2 kg ha^{-1} of AAtrex 90 DF (atrazine); 2.3 L ha^{-1} of glyphosate; 0.3 L ha^{-1} of 2-4-D and 0.3L ha^{-1} dicamba in 114 L ha^{-1} 0.1% non-ionic surfactant per hectare. The NDVI and SPAD measurements were taken every two or three days from June 16 (V4), until July 15. The NDVI readings were taken at two heights. On July 28 ten ear leaves were collected per plot, and grain samples were taken at harvest on September 24.

For 2011, in early May, the appropriate plots were moldboard plowed to a depth of 20 cm; secondary tillage was done with a disc to a depth of 15 cm. On May 10, the Pioneer corn hybrid 1184HR was planted at 62,000 seed per hectare in 0.9 m rows. Pre-emergent weed control was performed on May 11 using 1.2L ha^{-1} of Dual II Magnum; 2.5 kg ha^{-1} of AAtrex 90 DF; 3.1L ha^{-1} of Gramoxone Inteon and 0.6L ha^{-1} of 2-4-D in 114 dm^3 0.1% non-ionic surfactant per hectare. On May 20 the N treatments were applied as AN (34-0-0), broadcasting the N fertilizer by hand. On June 5, post-emergence weed control was performed using 1.85 kg ha^{-1} of Aatrex 90DF; 0.2L ha^{-1} of Callisto 4EC (mesotrione); 2.3L ha^{-1} Roundup (glyphosate) in 114 L of 1 % crop oil concentrate and 2.5 % urea-ammonium nitrate solution per hectare. The NDVI and SPAD measurements were taken every two or three days from June 3 (V4) until July 5. On July 14 ten ear leaves were collected per plot and grain samples were taken at harvest on October 7.

3.2. GSC Field Trial

The GSC study was located in two different areas within the farm in the two different years. The experimental design was a split strips design randomized in four complete blocks for both years. The main plots consisted of 5 at-planting N rates. The subplots consisted of side-dress N rates sufficient to obtain up to 224 kg N ha^{-1}, in 56 kg N ha^{-1} increments, at each main plot N rate. Finally, and considering all combinations of at-planting N rates and side-dress N rates, there were sixteen fertilizer N treatments: 0 – 0, 0 – 56, 0 – 112, 0 – 168, 0 – 224; 56 – 0, 56 – 56, 56 – 112, 56 – 168; 112 – 0, 112 – 56, 112 – 112; 168 – 0, 168 – 56; 224 – 0; and 280 – 0; where the first number (before the hyphen) is the at-planting N rate and the second number (after the hyphen) is the side-dress N rate. The objective of this study was to evaluate the NDVI response to side-dress N applications and grain yield prediction by NDVI readings.

In this study, five GreenSeeker sensors were mounted on a boom. Because of border effects, the sensors at the boom ends were not used. Of the three sensors used, two were faced directly over the corn crop row and the sensor in the middle faced directly over the plot's middle inter-row. Three NDVI values were computed per plot: i) sensor 53 (right center row) NDVI; ii) sensor 54 (left center row) NDVI and iii) sensor 55 (inter row) NDVI.

In 2010, the GSC study was located on two different soils. The east half of the study was on an Armour (A slope) silt loam (Ultic Hapludalf) and the west half on a Maury (B slope) silt loam (Typic Paleudalf). The area of the study was 4759 m^2 (122 m long and 39 m wide); and each plot's area was 69.7m^2 (15.2m long by 4.6m wide), with a total of 64 plots and 6 corn rows per plot. On May 25, Pioneer hybrid 33N58 was planted

in 0.76m rows. On June 4, at-planting N was applied as banded urea-ammonium nitrate solution (UAN, 32-0-0). On June 24 (V7), the side-dress N treatments were applied as banded UAN. On July 28, at R1, ten ear leaves were collected per plot and analyzed for total N. Two NDVI measurements were made in this year; one on June 23, pre-side-dress (V7); and a second on July 7, post-side-dress (V10).

In 2011, the study was located on a Maury (B slope) silt loam. The area of the study was 3346 m^2 (122 m long and 27.4 m wide), and each plot's area was 46.5m^2 (15.2m long by 3m wide); with a total of 64 plots and 4 corn rows per plot. On May 10, Pioneer hybrid 33F87 was planted at 70,370 seed per hectare in 0.76m rows. On May 20, at-planting N was applied as AN. On June 24 (V8), the side-dress N treatments were applied as AN. On July 14, at R1, ten ear leaves were collected per plot and analyzed for total N. Two NDVI measurements were made in this year; one pre-side-dress on June 17 (V6), and a second on July 1, post-side-dress (V10-V11).

3.3. Plant and Grain Tissue Analysis

A modification of the Kjeldahl acid digestion was performed on both leaf and grain tissue samples in support of total N analysis. A 100 mg dried subsample was put in a 100-mL digestion tube. Five mL of 36 N sulfuric acid, also containing 0.05 g of salicylic acid per mL, were added to each sample. The salicylic acid reacts with any nitrate present, forming nitrosalicylic acid. Half a gram of sodium thiosulfate was added in order to reduce the nitrosalicylic acid to aminosalicylic acid, converting all forms of N in the sample (organic and inorganic) into ammonium N. Ultimately, 1.8 g of potassium sulfate and 3 selenized boiling chips were added and the digestion continued for 2.5

hours. The digests were then analyzed by automated colorimetry using a dual Technicon System II Autoanalyzer. The method utilized was the modification of the Berthelot reaction developed by Chaney and Marbach (1962).

3.4. Statistical Analysis

Statistical Analysis Systems (SAS, 2002) software was used. The General Linear Models (GLM) procedure was used (Dellinger et al., 2008) to partition experimental variance and determine statistically significant treatment differences in grain yield, leaf N, grain N, grain N removal and NDVI values for the GSC field trial and treatment differences in grain yield, leaf N, grain N, grain N removal, SPAD readings and NDVI values for the Blevins 23 field trial. The Least Significant Difference (LSD) was computed so as to perform comparisons between individual treatment means. Considering that the number of replications was not great (only four replications in each study), the *F* tests for ANOVA were considered significant at the 0.10 probability level. However, the LSD value at the 0.05 level of probability was included in the tables for reader benefit. Additionally, in the GSC study, Contrast statements were also used to make comparisons among the following groups of treatments: i) treatments with all N applied at-planting; ii) treatments with all N applied at side-dressing; and iii) treatments where the N was split between at-planting and side-dressing.

Regression analysis was performed, for both experiments, in order to establish relationships between grain yield and other measured variables.

CHAPTER 4: RESULTS AND DISCUSSION

The season had a strong effect on the measured variables. The 2010 corn production year (1 April to 1 October) was drier than 2011, and this impacted crop growth. Daily rainfall, and average daily air and soil temperatures, for the production period, are shown below for 2010 (Figure 1) and 2011 (Figure 2). In 2011 (Figure 2), there was much greater early precipitation at the beginning of the season, and then again at the end, causing the accumulated precipitation to be above 900 mm; whereas in 2010 cumulative precipitation was around 600 mm.

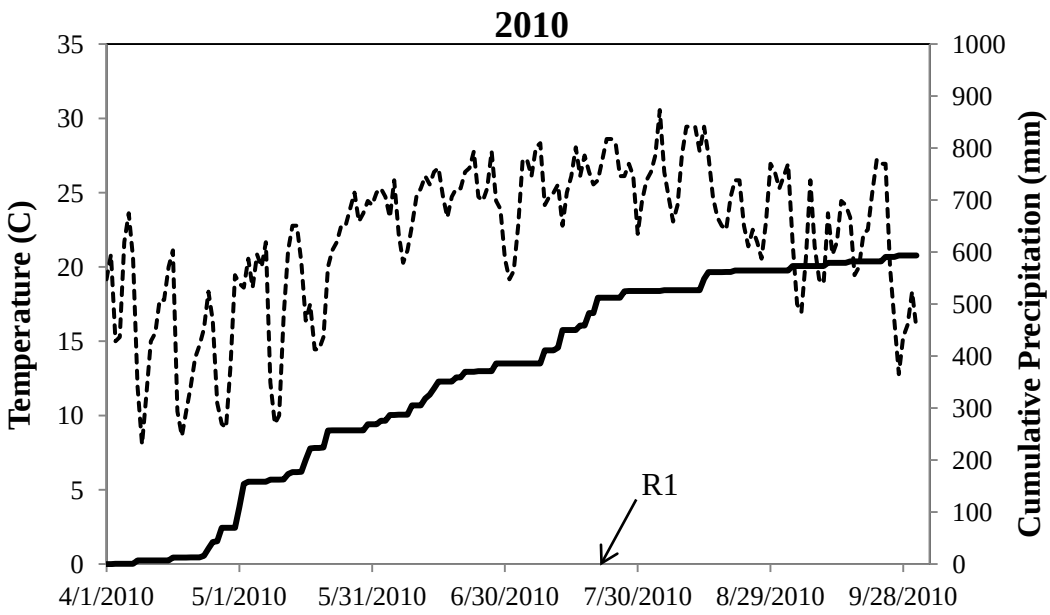

Figure 1. Daily average air temperatures, and the accumulated precipitation, for the 2010 growing season.

Although there was drought in the 2010 season, there was adequate rainfall just prior to the time of tasseling (R1). The air temperature distribution with time was not that different between the two seasons, other than there was a more pronounced drop in air temperature towards the end of the 2011 season.

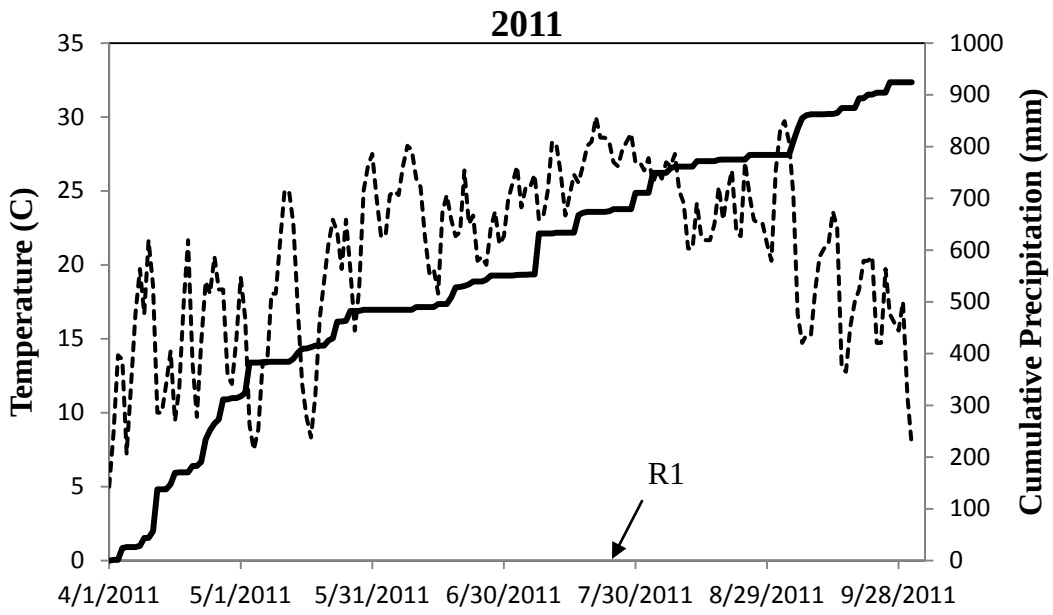

Figure 2. Daily average air temperatures, and the accumulated precipitation, for the 2011 growing season.

4.1. The Blevins 23 Field Trial

4.1.1. 2010 – Traditional Analysis

The 2010 year was dry, and the drought intensified towards the middle-end of the growing season. Nitrogen application treatments significantly affected grain yield, nitrogen concentration in the leaf, nitrogen concentration in the grain and nitrogen removal (Table 1). There were no yield restrictions due to availability of other nutrients, weeds, or disease or insect pressure.

Field observations showed that at the V4-V5 corn's growth stage the N-deficiency was already noticeable to the human eye on the control for both tillage systems. At V5-V6 the deficiency was accentuated. By V7 there was some damage (probably wind) to the new leaves in some plots. At V8-V9 the crop covered the inter rows and the leaves were curled due to severe drought, and N deficiency started to be apparent in the 84 N-rate plots. At V9-V10 the water stress was damaging the crop and seemed more intense in the MP plots. At V13 there were differences in color between the 84 kg N ha^{-1} and the rest of the higher N-rates. Three days later, on July 18 tissue samples were taking at tasseling (R1).

Regardless of the rainfall shortage, 2010 was a good responsive environment for all the measured variables. Yields were not dramatically restricted by water availability, however, the 336 N rate was not able to significantly increase grain yield over the 168 N rate. Significant main effects ($p \leq 0.05$) due to N rate were found in Leaf N and grain N for all the levels of the factor. N removal, as grain yield, didn't presented significant differences between 168 kgN ha^{-1} and 336 kgN ha^{-1}. This means that although N leaf (at

R1) presented better N nutrition for the 336 N rate, it could not be capitalized in significantly higher grain yields.

Significant main effects due to tillage were found only for leaf N ($p \leq 0.1$), though there was a general trend for improved N nutrition and yield with NT soil management. The other variables did not show significant main effect due to tillage; apparently the effects of the drought were too strong for the NT system to make a difference due to its better water use efficiency (WUE). However, there was a positive 560 kg ha^{-1} grain yield difference, across N rates, in favor of the NT system, which might be explained either by better N nutrition or by more available water for crop development, given that the same soil under conservation tillage systems tend to present a better water holding capacity compared with conventional tillage systems (Franzluebbers, 2002; Holland, 2004; Yu et al., 2011).

Table 1. Main effects of N-Rate and Tillage on the measured variables in 2010.

Main Effect of N Rate (across tillage treatments)

	Leaf N (g kg^{-1})	**Grain N (g kg^{-1})**	**Yield (kg ha^{-1})**	**N Removal (kg ha^{-1})**
0	12.7	7.93	3130	21
84	23.2	10.68	8390	76
168	24.9	12.43	10190	107
336	27.1	13.49	10040	114
LSD (0.10)	1.2	0.4	930	10
LSD (0.05)	1.5	0.5	1130	13

Main Effect of Tillage (across N rate treatments)

	Leaf N (g kg^{-1})	**Grain N (g kg^{-1})**	**Yield (kg ha^{-1})**	**N Removal (kg ha^{-1})**
NT	22.5	11.2	8220	83
MP	21.5	11.0	7660	76
LSD (0.1)	0,9	0.3	660	7
LSD (0.05)	1.1	0.4	800	9

Figure 3 shows the grain yield response to N rate for both tillage systems. A quadratic plateau (QP) model was chosen to describe this relationship, maximizing both the goodness of fit (R^2), and better representing the N-rate that maximized yield (Cerrato and Blackmer, 1990). Though this thesis will not discuss N rate recommendations, the choice of the model is of extreme importance (Belanger et al., 2000). In this season, for this study, the N rate that maximized NT yields using a quadratic (dashed black line, Q) model was 80 kgN ha^{-1} higher than that determined by the QP (dashed and dot) model being 150 kgN ha^{-1} for the QP model and 230 kgN ha^{-1} for the Q model, confirming the findings of Cerrato and Blackmer (1990). Also, there is a trend suggesting differences between tillage systems regarding how quickly the yield response rises to reach the

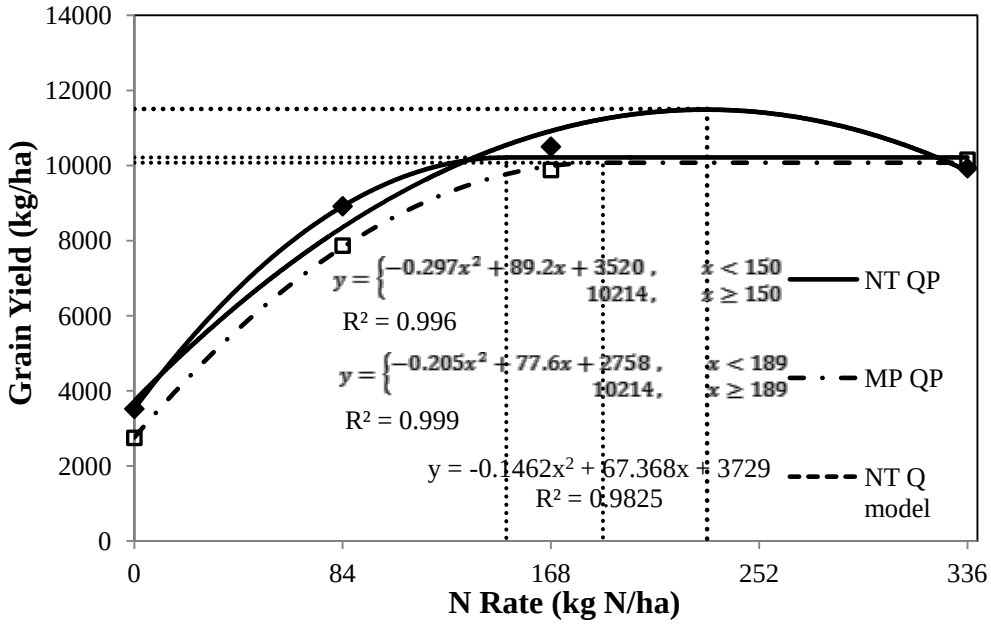

Figure 3. Relationship between applied N and 2010 corn grain yield for two different tillage systems. The quadratic plaeau (QP) regression models are shown, as are the N rates at which yields were maximized, for each tillage system. Also shown a quadratic (Q) model for the NT yield response.

N-rate for maximizing grain yield. Looking at these initial slopes, NT soil management seems to result in a greater NUE (kg grain kg^{-1} N) reaching the plateau at an N rate of 150 kgN ha^{-1} than did MP soil management which reaches the plateau at an N rate of 189 kgN ha^{-1}. Another important difference is that NT starts with greater N availability, greater yield for the control, and remains consistently superior in yield across the lower N rates. The response difference converges/disappears at higher N rates when is restricted by water availability, and, as a consequence, both NT and MP exhibit very similar yields. A particular point is that failing in the proper model choice could not only have serious economic and environmental consequences but could also have consequences in the understanding of the biological processes taking place in the study. Another observation to note was that the variability in the MP data was higher than that for the NT system, suggesting that NT soil management caused a more stable environment.

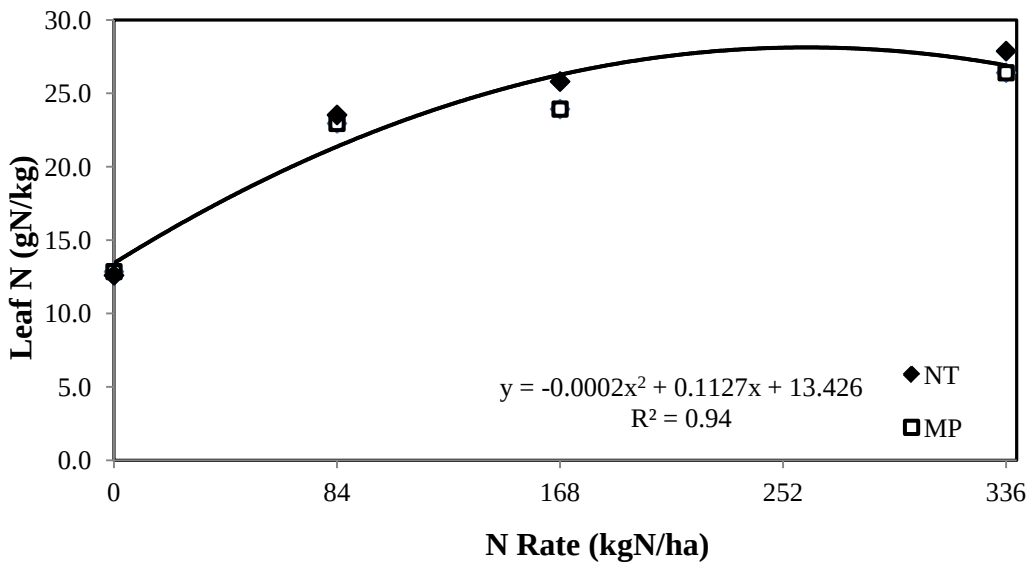

Figure 4. Relationship between the N concentration in the leaf and the N rate for two tillage systems.

Figure 4 illustrates the leaf N response to N-rate. There is a strong correlation between the variables when the relationship is fitted to a Q model. At the 10 % level of significance the differences in leaf N between tillage systems were significant, however, it did not prevented the data to be pulled together into the same model, as it presented a goodness of fit (R^2) equal to 0.94. At the 5% level of significance the differences were not significant. When a relationship between grain yield and leaf-N was explored, a good correlation was found (R2 = 0.95) although the data got clustered around 25 g N kg^{-1}.

4.1.2. 2010 - NDVI and SPAD readings

The evolution of the NDVI readings with time followed a very well defined quadratic pattern. Figure 5 shows that, for both measured heights, the NDVI values for the 0 kg N ha^{-1} control separate from those for the other N rate treatments at a very early growth stage in MP corn, but not for NT corn, where the separation is not as prominent.

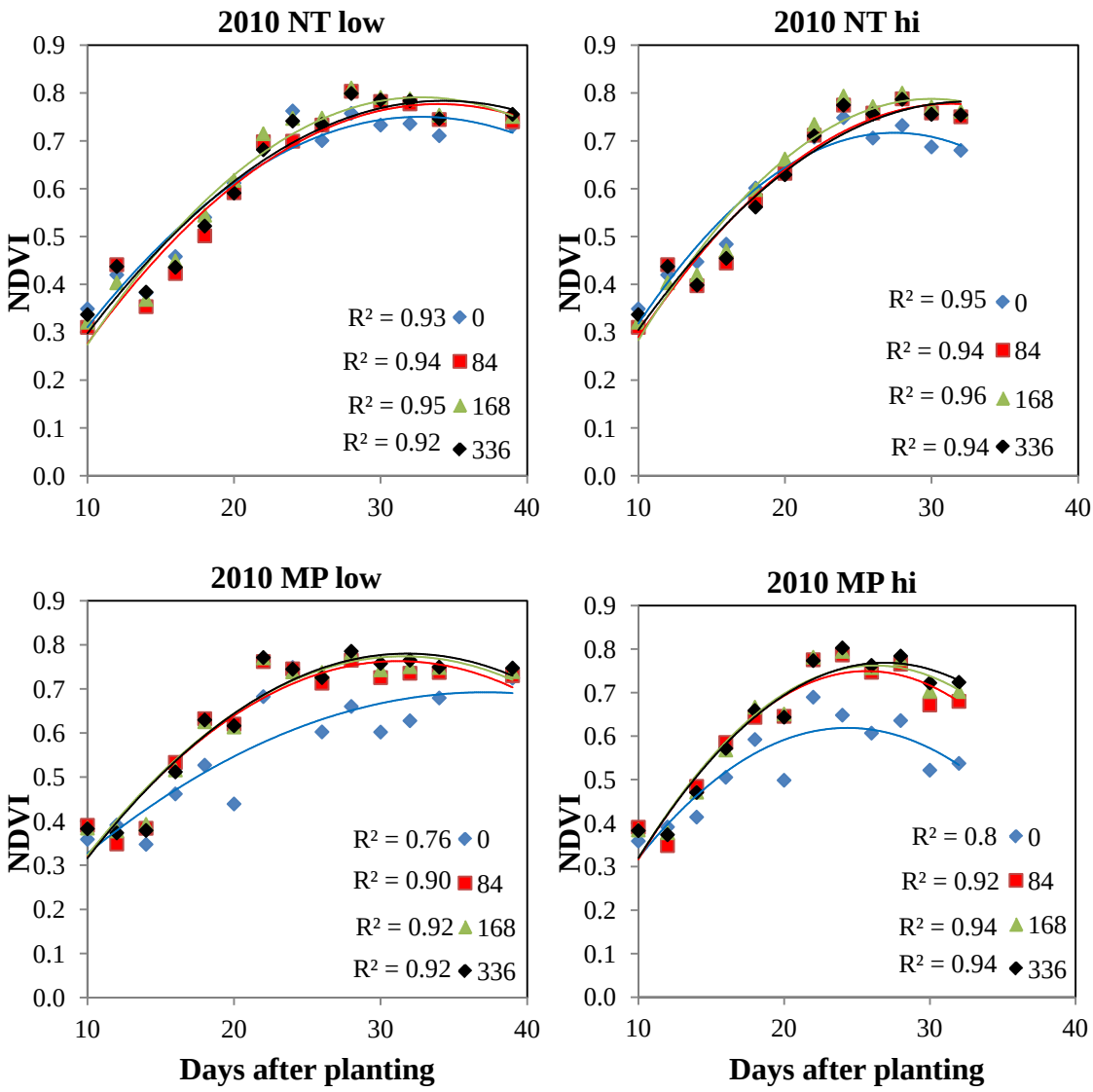

Figure 5. The evolution of corn NDVI values, measured at two heights (low and high), with days after planting, for each N rate, in both tillage systems, in 2010.

This suggests that: i) an earlier N deficiency existed in MP corn than in NT corn, or ii) there were differences in NDVI values between tillage systems due either to N nutrition differences or to soil interference in the index, or to both of these factors.

The evolution in corn leaf SPAD meter readings with time also followed a quadratic pattern (Figure 6). The SPAD worked better than the GreenSeeker in detecting early N deficiency, especially the separation between 0 kg N ha^{-1} and the rest of the N rate treatments (Figure 6). Early measurements show similar values for the 0 kg N ha^{-1} between tillage systems, but higher SPAD values at higher N rates for MP suggesting greater early release of N by MP. The SPAD values also showed, at advanced vegetative growth stages, greater N deficiency at 84 kg N ha^{-1} than at higher N rates, especially in the MP tillage system.

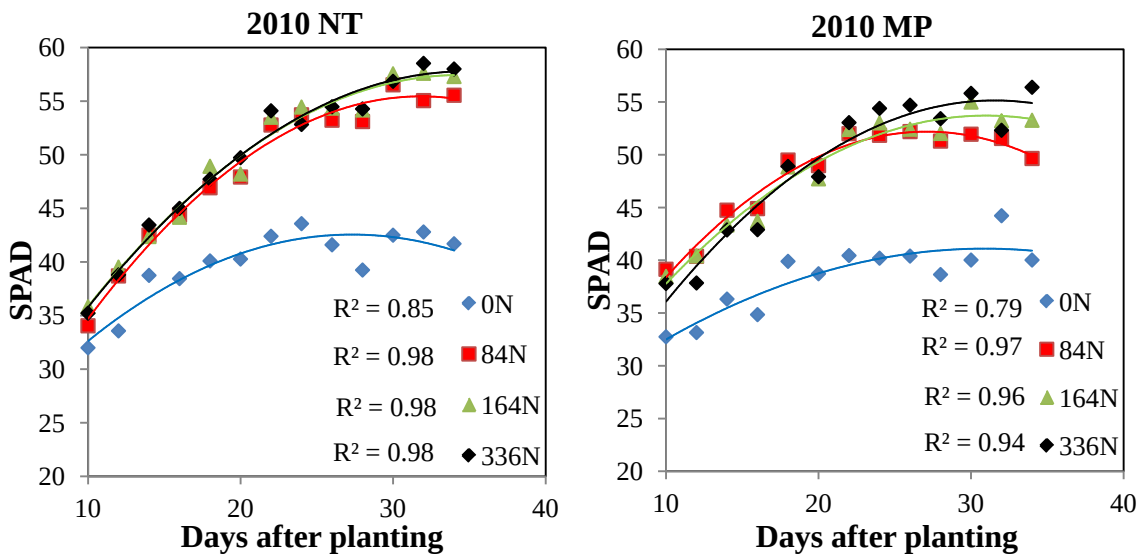

Figure 6. The evolution of corn leaf SPAD meter values with days after planting for each N rate, in both tillage systems, in 2010.

The analysis of variance shed light onto the question of how early these sensors were able to detect N deficiency. This is, essentially, the difference in sensor readings

between the 0 kg N ha^{-1} rate and the rest of the N rate treatments. The GreenSeeker NDVI (lower measurement height) was unable to detect a difference until V6, exhibiting a significant ($p \leq 0.05$) main effect due to N rate at that growth stage, whereas SPAD meter readings indicated a deficiency at V4. As described before, the human eye was able to see the N deficiency at V4 to V5, which is 2 to 4 days after the SPAD and 2 to 4 days before the GreenSeeker. Among the rest of the treatments, the SPAD readings were able to detect differences between the 84 kg N ha^{-1} rate and the higher N rates at V9, whereas the GreenSeeker did not find differences in NDVI values until V13. Field observations indicated that the human eye observed the N deficiency at V8 to V9 on older leaves that were closer to the ground. However, looking downward at the canopy, no color difference separating the 84 kg N ha^{-1} rate from the other N rates could be found by eye – even at V13.

Within each N rate treatment, NDVI readings for each tillage system were plotted against time (days after planting) in Figure 7. Quadratic models were fitted to the data. Figure 7 suggests no differences between tillage systems at any given N rate, but the analysis of variance found a significant main effect of tillage on the NDVI values from the earliest (V4) growth stage, being higher for MP corn until V8, where the relationship switched and NT corn exhibited significantly higher values than did MP corn. At V11, this difference ceased to be significant.

Within each N rate treatment, SPAD meter readings for each tillage system were plotted against time (days after planting) in Figure 8. The meter readings do not suggest any differences due to tillage system for early to middle vegetative growth stages, consistent with analysis of variance. At early growth stages the analysis of variance was

erratic. At V7, significant differences ($P \leq 0.10$) started to be consistent. The NT corn exhibited greater values at every growth stage, especially due to large differences due to tillage at 0 kg N ha^{-1}, suggesting better N nutrition for NT than for MP from V7 onwards.

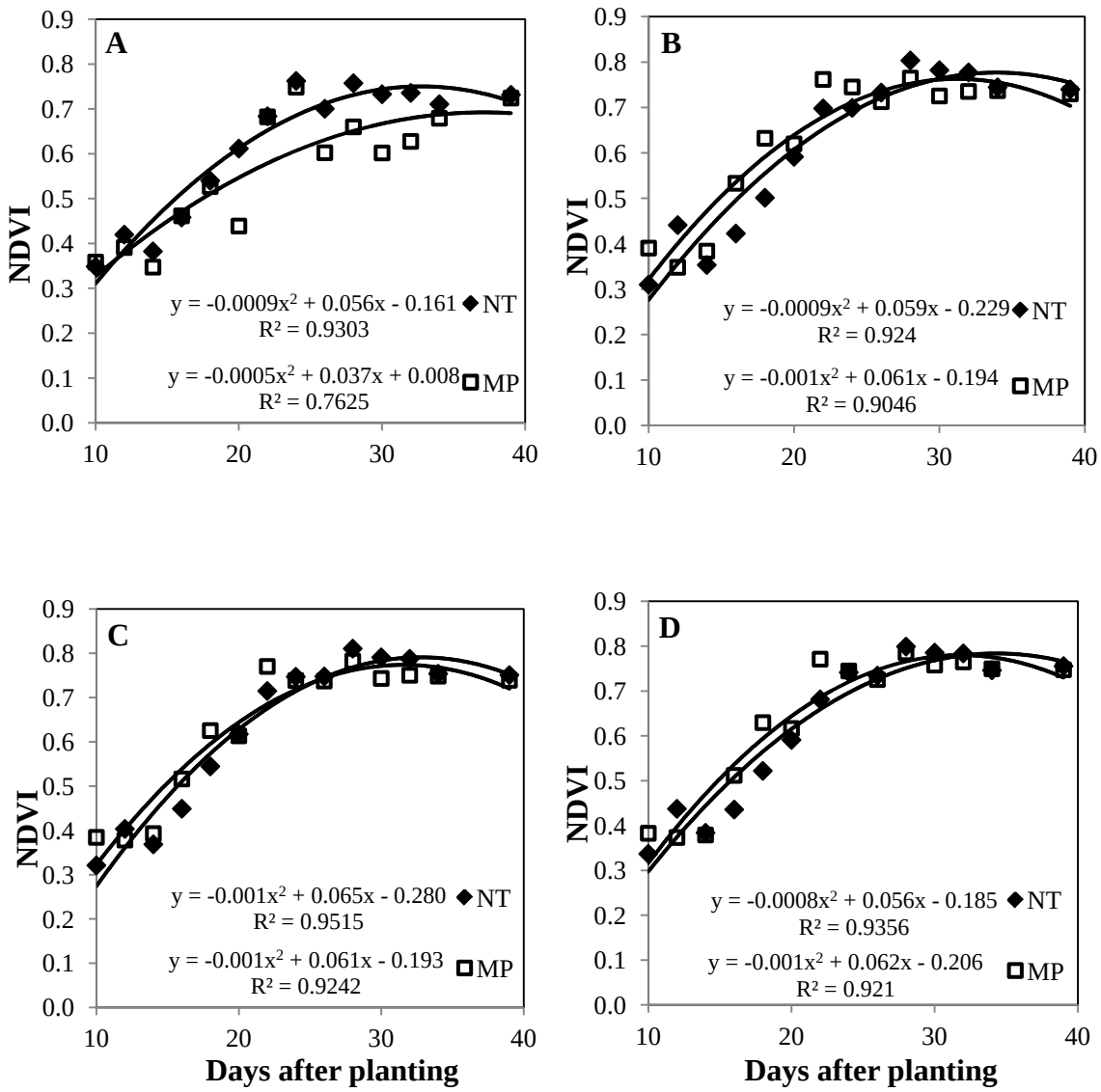

Figure 7. The evolution of 2010 corn NDVI values, measured at the lower of two heights, with days after planting, for each tillage system, at 0 kg N ha^{-1} (A), 84 kg N ha^{-1} (B), 168 kg N ha^{-1} (C), and 336 kg N ha^{-1} (D).

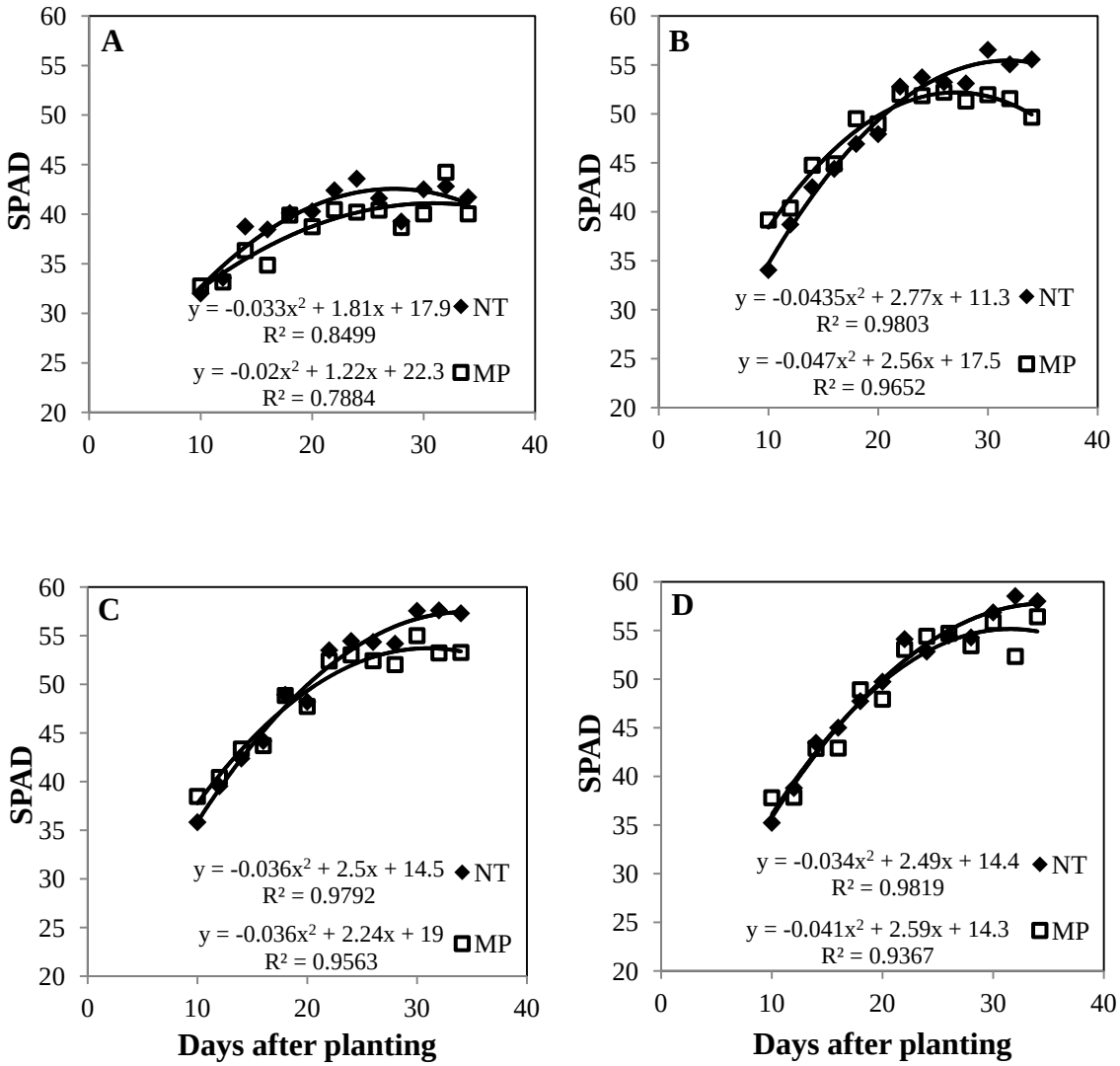

Figure 8. The evolution of 2010 SPAD meter readings, measured at the lower of two heights, with days after planting, for each tillage system, at 0 kg N ha^{-1} (A), 84 kg N ha^{-1} (B), 168 kg N ha^{-1} (C), and 336 kg N ha^{-1} (D).

At early stages (V4-V7) of the corn growth cycle there was no correlation between yield and NDVI for NT corn. On the other hand, MP exhibited better correlations, with an $R^2 = 0.66$ from V6 on (Figure 9). These results suggest that the prediction of yield with early NDVI readings was affected by soil background, and when there was a darker contrast the predictions were better.

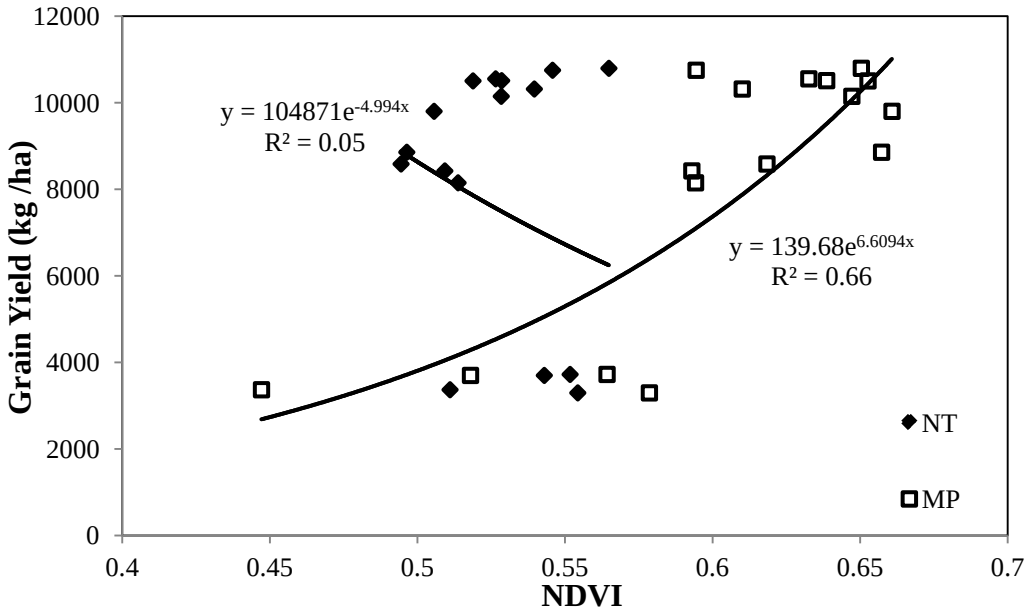

Figure 9. 2010 corn grain yield versus NDVI readings taken at an early growth stage (V6) for the two different tillage systems.

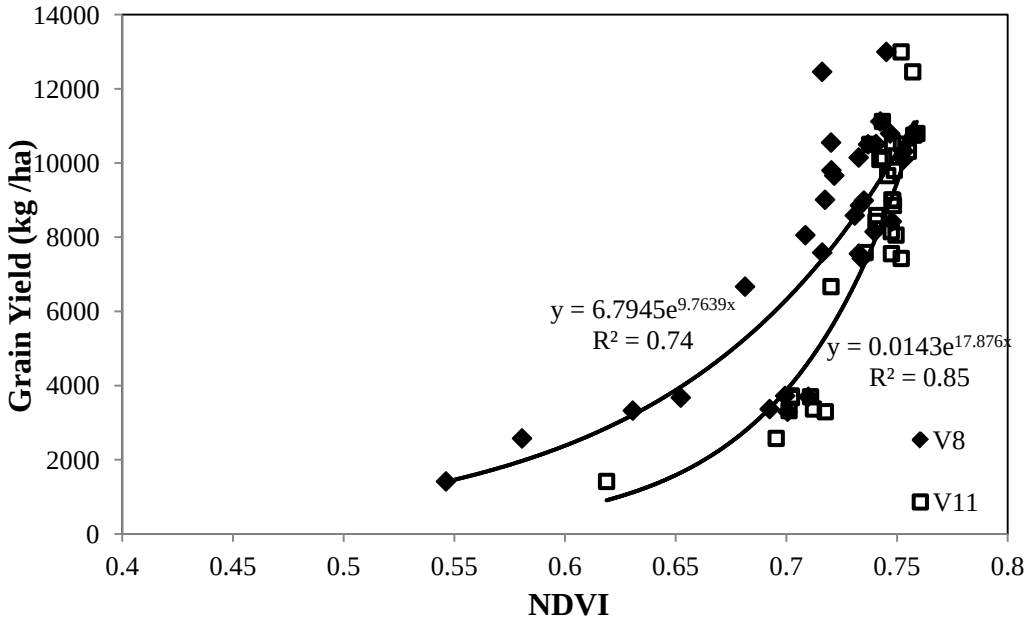

Figure 10. Relationships between the 2010 grain yield and NDVI values determined at V8 and V11 with the data of the two tillage systems pulled together.

Although the resolution was not very good (0.2 NDVI units), the yield versus NDVI relationship was better at growth stage V8, with $R^2 = 0.74$ (and the data for both

tillage systems seeming to follow the same model), than at V6. Figure 10 shows the relationships between grain yield and NDVI determined at two growth stages (V8 and V11) with all 8 treatment means (four N rates by two tillage systems) taken together. This is in accord with the findings of Teal and Tubana (2006). The relationships were only a bit better correlated when yields were relativized, with R^2 values of 0.80 and 0.85 for V8 and V11, respectively.

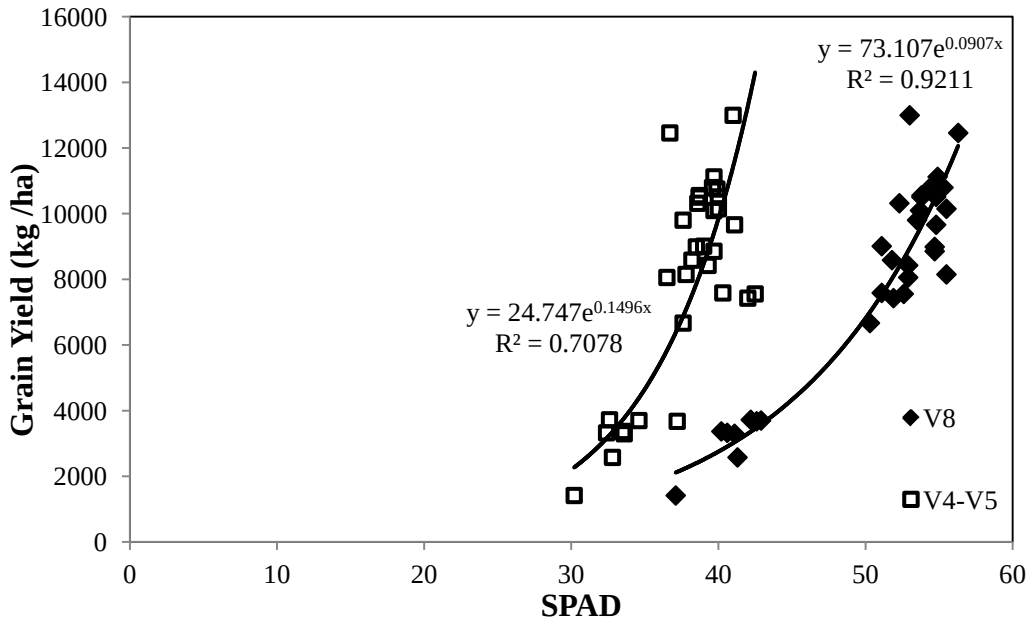

Figure 11. Relationship between the 2010 grain yield and the SPAD meter readings taken at V4-V5 and V8.

The lack of difference due to tillage could be explained by the fact that once the crop covers an important part of the soil area, and then soil color does not interfere as much, causing crop spectral behavior to be similar for both tillage systems (unless differences in N nutrition due to tillage are present). The data for the V11 growth stage supports this hypothesis.

The SPAD meter better predicted corn yields at earlier stages than the GreenSeeker (Figure 11). Exponential models were fitted to these data and R^2 values greater than 0.7 were found from V4-V5 onward (Figure 11).

4.1.3. 2011- Traditional Analysis

The 2011 year was moist, with above-average rainfall. Grain yield potential was exceptionally high, causing the 336 kg N ha^{-1} N rate to produce yields significantly higher than those observed at 168 kg N ha^{-1} (Table 2). There were no restrictions to yield due to nutrient availability (other than N), weeds, or disease and insect pressure.

Field observations found that by V4 the crop's N deficiency was already noticeable. Surprisingly, the NT corn was larger than the MP corn and NT corn color differences were visible, separating the 0 kg N ha^{-1} control from the rest of the N rate treatments. The MP corn also exhibited color differences at this early growth stage, but these plants were not as N-stressed as the NT corn, at the same N rate. At the V5-V6 growth stage, differences in NT corn plant development between the 0 kg N ha^{-1} control and the rest of the N rates intensified. Although MP corn also exhibited differences in plant development, they were not as remarkable as those in NT corn. The MP corn did not show strong N deficiency symptoms at this early growth stage, whereas the lower leaves of NT corn showed N deficiency at 84 kg N ha^{-1}. However, at the V5-V6 growth stage the upper leaf canopy did not yet show color differences separating the 84 kg N ha^{-1} rate from the rest of the N-rates. At V6 even the highest N rate showed N deficiency in lower leaves, but color differences between the 84 kg N ha^{-1} rate and higher N rates were not yet noticeable. The MP corn did not show any N deficiency at V6.

At V8, the MP corn began exhibiting N deficiency. At V9, NT corn at 84 kg N ha^{-1} started to show a little color difference from the higher N rates, but this was not conclusive. At V11 the crop differences (more in size than color) were still small. When looking above the canopy, differences among N rates at 84 kg N ha^{-1} and greater were not

distinguishable. By V13, when walking between the rows, biomass development caused the crop to appear more ‘crowded’ at the higher two N rates, but no color differences were observed.

Table 2. Main effects of N-Rate and Tillage on the measured variables in 2011.

Main Effect of N Rate (across tillage treatments)

	Leaf N ($g\ kg^{-1}$)	**Grain N ($g\ kg^{-1}$)**	**Yield ($kg\ ha^{-1}$)**	**N Removal ($kg\ ha^{-1}$)**
0	18.9	9.1	4270	33
84	21.5	10.0	9480	80
168	27.0	12.5	13210	139
336	28.1	13.7	14170	165
LSD (0.1)	**1.6**	**0.5**	**700**	**7**
LSD (0.05)	**1.9**	**0.6**	**840**	**8**

Main Effect of Tillage (across N rate treatments)

	Leaf N (%)	**Grain N (%)**	**Yield ($kg\ ha^{-1}$)**	**N Removal ($kg\ ha^{-1}$)**
NT	24.7	11.5	10680	110
MP	23.1	11.1	9890	99
LSD (0.1)	**1.1**	**0.3**	**490**	**5**
LSD (0.05)	**1.4**	**0.4**	**600**	**6**

The N rate treatments significantly affected grain yield, N concentration in the leaf, N concentration in the grain and N removal (Table 2). There were significant differences ($p \leq 0.05$) in grain yield among all N rate levels, meaning that in contrast to 2010, in 2011 the highest N rate resulted in higher grain yield. Leaf N was the only variable that seemed to reach a plateau at the highest N rate, implying that up to R1 there was little difference in crop N nutrition between 168 kg N ha^{-1} and 336 kg N ha^{-1}. This caused grain yield prediction with leaf N leaf to be inaccurate. Unlike leaf N, grain N and N removal also exhibited significant differences among all levels of N rate.

There is a significant main effect of tillage for all the measured variables at the 5% level of significance suggesting a better N-nutrition for NT. 2011 was an excellent responsive environment where abundant water availability allowed the different tillage systems to express their yield potential being NT superior to MP.

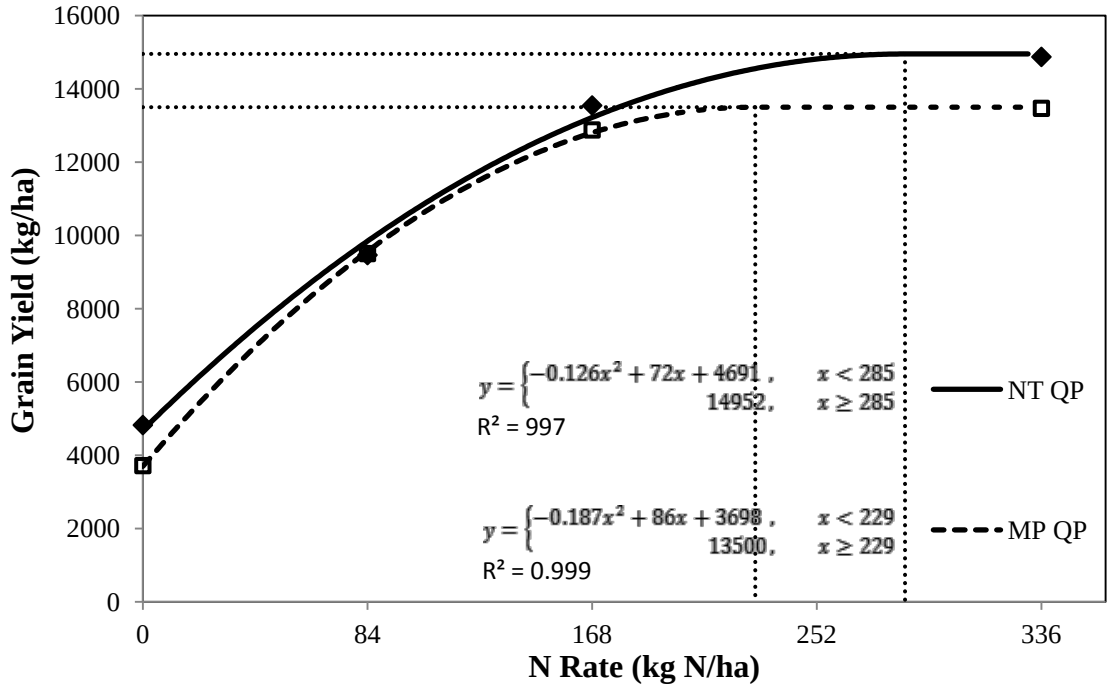

Figure 12. Relationship between applied N and 2011 corn grain yield for two different tillage systems. The QP regression model equations are shown, as are the N Rates at which yields are maximized, for each tillage system.

Figure 12 shows the yield response to N rate, for both tillage systems. Again, QP models best described these relationships. As the figure shows, and looking at the similarity of the slopes, it could be said that there was no NUE difference due to tillage system. Since no symptoms of water stress were observed throughout the season, the greater yield for NT corn at 0 kg N ha^{-1} implies that the NT soil better contributed to the crop's N nutrition. However, the higher yields at the maximum applied N rate suggest that either more N or more water was available for crop development. More water could

have been available at the end of the season given that conservation tillage systems tend to exhibit better plant-available water holding capacity (Franzluebbers, 2002; Holland, 2004; Yu et al., 2011), compared with conventional tillage systems. As fertilizer N was applied at-planting, some nitrate-N could have been washed out of the reach of corn roots under the conventional tillage system (Dinnes et al., 2002). Because of the complexity of the interactions taking place in the soil, perhaps not just one but both of these hypothetical effects impacted corn yield differences due to tillage at 336 kg N ha^{-1}.

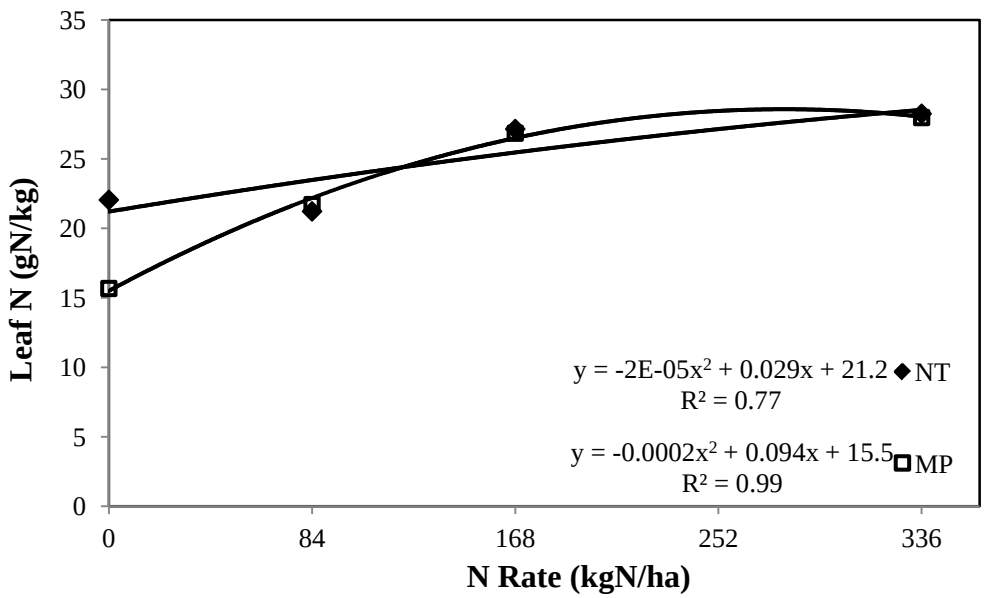

Figure 13. Relationship between leaf N concentration and fertilizer N rate for two tillage systems in 2011.

The 2011 yield data was not as variable as those taken in 2010, but MP corn exhibited a wider range of values than NT corn, confirming the observation made above regarding the higher variability in the 2010 yield data. The fertilizer N rate also affected N nutrition variates in 2011. Figure 13 shows the leaf N response to N rate in 2011.

Unlike 2010, in this year the data followed different models for each tillage system. Surprisingly, NT leaf N was not as responsive to N rate as was MP leaf N.

4.1.4. 2011 NDVI and SPAD readings

The evolution of the NDVI readings with time again, in 2011, followed a very well defined quadratic pattern. Figure 14 shows that, from the earliest measurements, the 0 kg N ha^{-1} control was different from the rest of the N rate treatments. Unlike 2010, NT corn exhibited higher NDVI values than MP corn at the earlier stages in crop

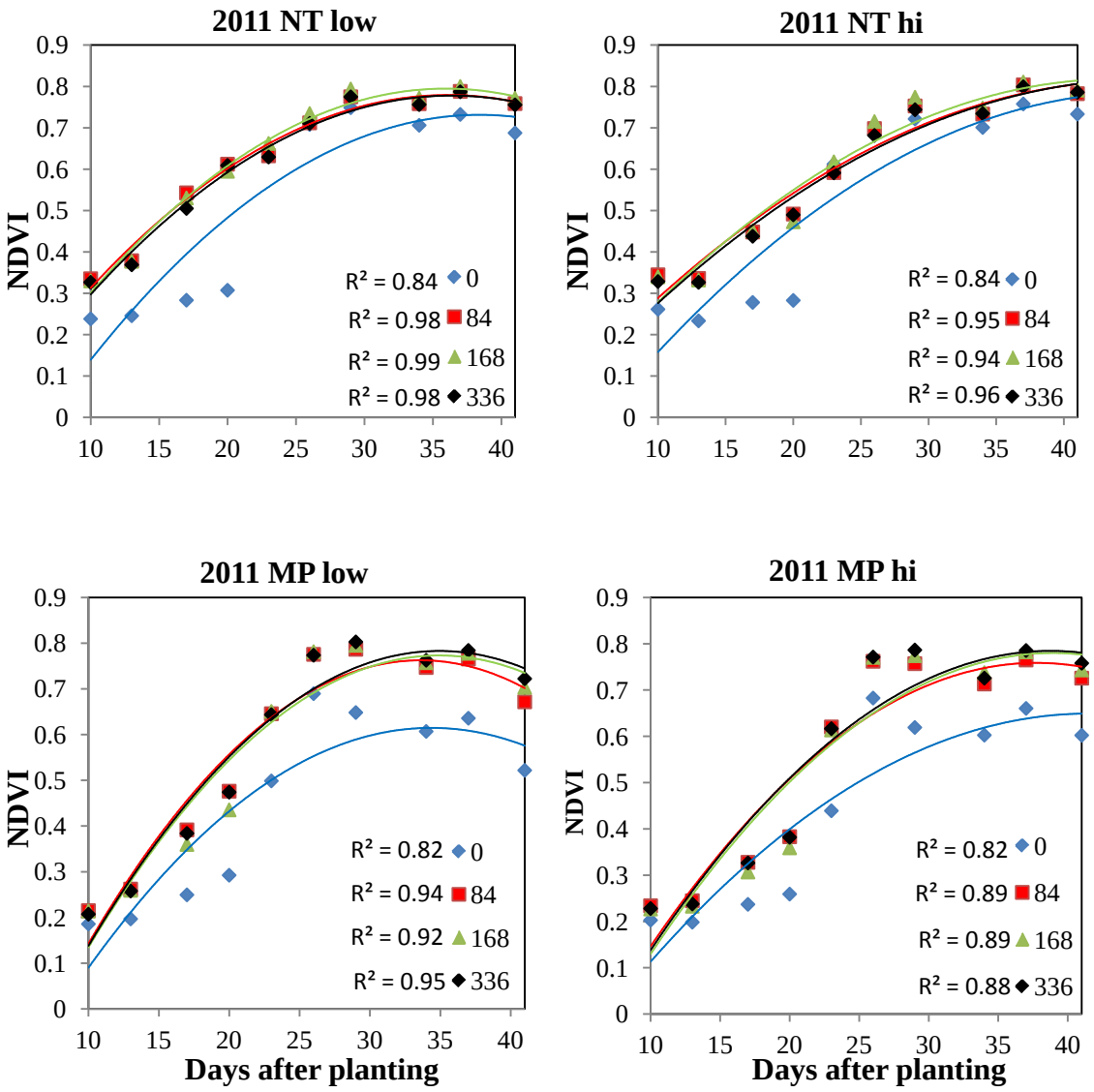

Figure 14. The evolution of corn NDVI values, measured at two heights (low and high), with days after planting, for each N rate, in both tillage systems, in 2011.

development. In this case, the bigger NT plants might have been overcoming the effect of the soil background, which was more dominant in 2010, making MP NDVI values higher that NT NDVI values in the previous year. Generally, the measurements taken at a lower height (0.6 m) above the canopy exhibited better resolution than those taken higher (1.2 m).

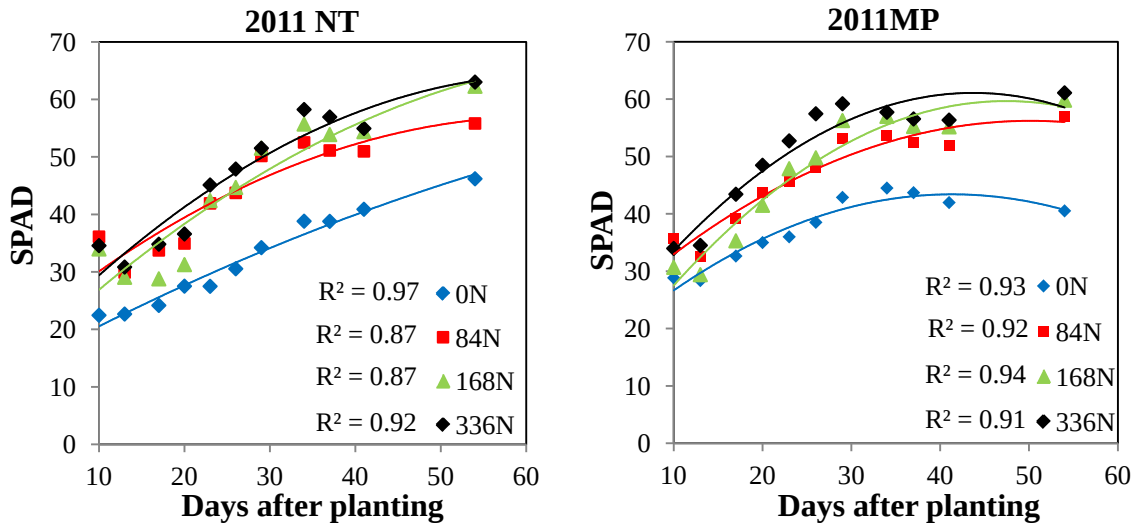

Figure 15. Shows the 2011 evolution in time of the SPAD readings for every N-rate treatment within each tillage system.

Figure 15 shows the temporal evolution of SPAD readings in 2011. Opposite to the NDVI values, the SPAD readings were higher for MP corn than for NT corn, early in the year. This suggests better N nutrition for MP corn at early growth stages. In this case, the higher NDVI values for NT corn were due to bigger plants rather than better N nutrition. The NDVI and SPAD values are providing different kinds of information.

The analysis of variance indicates that the NDVI was able to 'see' the N deficiency ($p \leq 0.05$) in the 0 kg N ha^{-1} control as early as V4, but could not distinguish among the other N rate treatments, even at the most advanced growth stage (V13) where

measurements were taken ($p > 0.10$). The SPAD meter detected the N deficiency at V4, it was able to distinguish the 84 kg N ha^{-1} rate from 336 kg N ha^{-1} N at V7, and 84 kg N ha^{-1} from 168 kg N ha^{-1} at V9 ($p \leq = 0.05$).

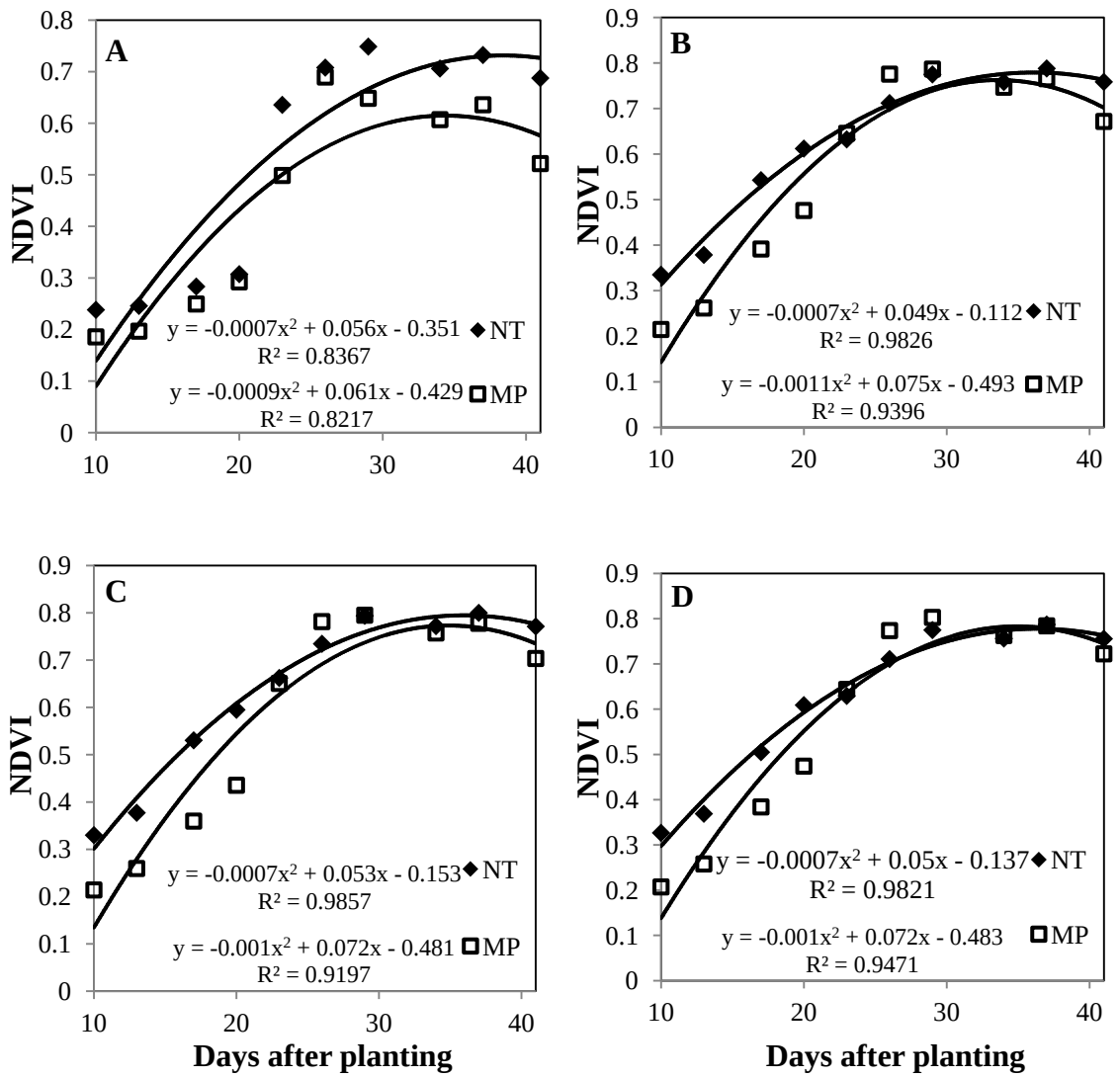

Figure 16. The evolution of 2011 corn NDVI values, measured at the lower of two heights, with days after planting, for each tillage system, at 0 kg N ha-1 (A), 84 kg N ha-1 (B), 168 kg N ha-1 (C), and 336 kg N ha-1 (D).

Figure 16 illustrates the temporal progression of NDVI values for each corn grown in each tillage system, within a given N rate treatment. At 0 kg N ha^{-1}, the MP corn gave lower NDVI values all along the season, with larger differences towards the end. In the other N rate treatments, NDVI values for MP corn were initially lower, but the difference diminished with time.

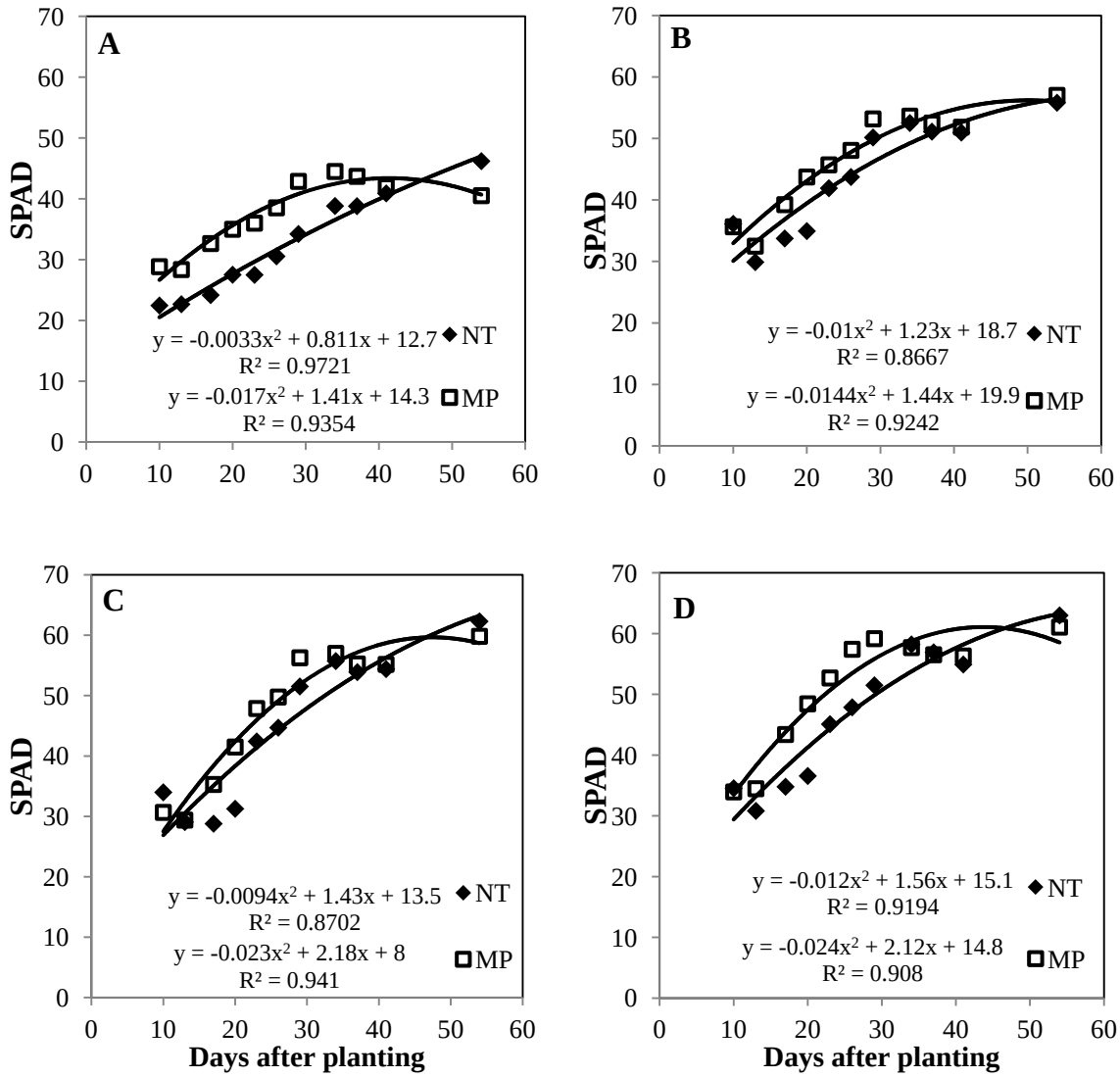

Figure 17. The evolution of 2011 SPAD meter readings with days after planting, for each tillage system, at 0 kg N ha^{-1} (A), 84 kg N ha^{-1} (B), 168 kg N ha^{-1} (C), and 336 kg N ha^{-1} (D).

The season-long SPAD measurements exhibited a different pattern (Figure 17). SPAD values for MP corn were higher most of the time (Figure 17); only at tasseling were NT corn SPAD readings higher. The analysis of variance confirmed the SPAD findings, with MP corn exhibiting significantly higher SPAD values until tasseling, when NT SPAD values became greater. It is interesting to note that right at tasseling NT corn breaks the time-trend and gives significantly higher SPAD readings than MP corn, consistent with leaf N values.

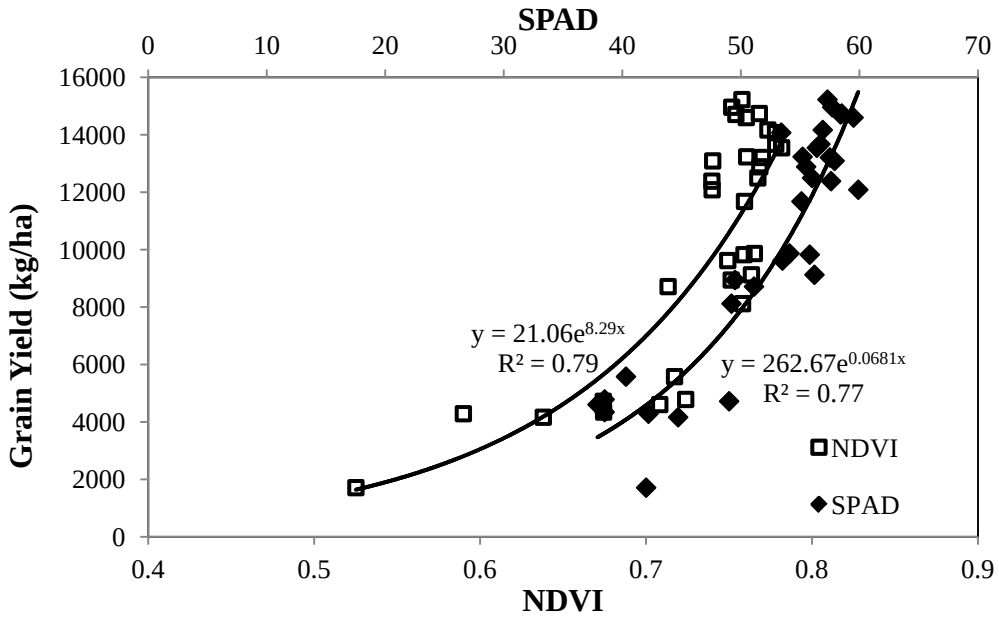

Figure 18. Relationship between the 2011 grain yield and both, SPAD and NDVI readings at V10.

At early (V4 to V8) corn growth stages there was a weak correlation between grain yield and NDVI, in accordance with 2010 results. As such, NDVI at early growth stages was not a good predictor of yield. The typical exponential relationship between NDVI and yield was found at V10, with an $R^2 = 0.79$ (Figure 18). Although side-dress N applications can be made as late as V10 (Scharf et al., 2002), in some situations the crop

would have been N deficient since V4. At V10, the crop would have been N deficient for about 24 days, causing lost yield potential. Varvel et al. (1997) observed lost corn yield potential when N deficiency was present before V8. The SPAD meter was not as good a predictor of 2011 yield as in 2010, with no good relationship until V10 (Figure 18).

Finally, Figure 19 shows a visual comparison between the grain yield prediction equation published by Teal et al. (2006) and the regression equation produced by our data, when data from both tillage systems and both years of measurements at V8 were put together. Giving a closer look at Teal et al. (2006) data, it seems like each of his particular experiments follow a different pattern. However, when put all together, they appear to fit an exponential model (equation shown in Figure 19) with an R2 = 0.77 and with a resolution that is not shown by our data. Although our data also followed an exponential model, it is clear that didn't follow Teal et al. (2006) exponential model.

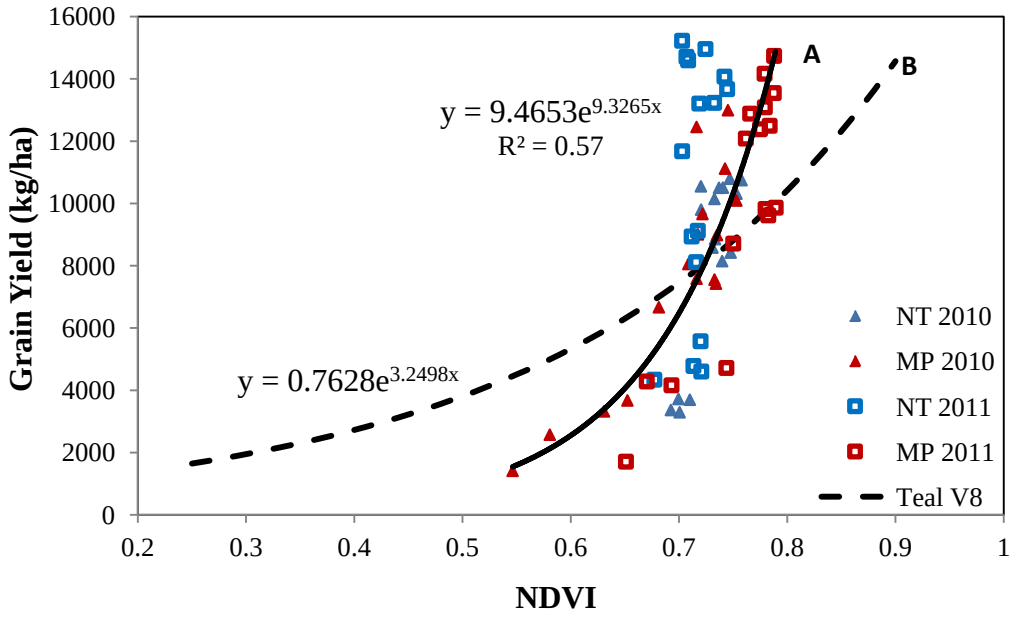

Figure 19. Relationship between grain yield and NDVI. A) For both 2010 and 2011 grain yield from the two tillage systems and NDVI readings taken at V8; B) Teal et al. (2006) regression equation.

4.1.5. Summary

In 2010, the SPAD meter detected the N deficiency as early as V4, but there were no significant differences between tillage systems, indicating no N nutrition differences due to tillage at very early growth stages (with no visible plant size differences either). This suggests that the significant NDVI differences due to tillage found at early growth stages were probably due to differences in soil background interference between the tillage systems. At V7, the SPAD meter detected significant differences due to tillage (being higher for NT). The GreenSeeker detected differences at V8, with NT corn giving significantly higher values, and suggesting that when the background effect was minimized the sensor was capable of detecting N nutrition differences. Grain yield prediction using early growth stage NDVI values was affected by tillage system; there was a good yield prediction for MP corn with V6 NDVI values, but this was not true for NT corn. The SPAD meter readings were better related to yield from the V4-V5 growth stage onwards, suggesting that the ability to ascertain N deficiency early in the growing season made plausible grain yield prediction when N nutrition was limiting.

For 2011, and looking at Figures 12 and 13 with yield versus N rate and leaf N versus N rate, respectively, the results suggest that the NT system provided better N nutrition to the corn crop. There were no water limitations in this season. However, the SPAD meter gave consistently, and significantly, higher readings for MP corn from V4 until R1. This suggests better N nutrition for MP corn until that point (R1), where the situation changed and NT corn exhibited greater SPAD values, consistent with the R1 leaf tissue N concentration data. The NDVI readings were consistently and significantly higher for NT corn from V4 until V13.

Some hypotheses can be made about these results: i) The NDVI values were influenced by the remarkably larger size of the NT corn plants; ii) Difference in interference from different soil tillage backgrounds affected the NDVI; iii) The NDVI 'recognized' better N nutrition in NT corn at advanced growth stages. The author favors the first two hypotheses, but not the third. That the soil background affects NDVI readings is clear from the 2010 study, but the 2010 results found higher NDVI values for MP corn and that was not true in 2011. It is believed that the much larger NT plants overcame the soil background difference, plus the better N nutritional status of MP corn, giving greater NDVI values for NT corn.

This assertion would imply that NDVI is more sensitive to differences in canopy biomass than to differences in canopy color. Once the canopy covered the soil, and even though NDVI tends to 'saturate', the NDVI detects differences between tillage systems. However, it is not likely that the NDVI detected small N nutritional differences due to tillage when NDVI is not able to detect similarly small differences between the 84 kg N ha^{-1} and 336 kg N ha^{-1} N rate treatments.

Yield prediction with early growth stage NDVI was not promising, for either tillage system. The NDVI values developed good relationships with yield around V9 for MP corn and V12 for NT corn, giving the typical exponential relationship in yield versus NDVI, with an acceptable goodness of fit. Teal et al. (2006) exponential equation would not fit the data of these experiments. The data strongly suggests that the model for fitting the data is field and growth stage specific.

In the 2011 season, the SPAD meter was not much better than the GreenSeeker in predicting yield. Good yield prediction could not be found using either sensor until V10,

when both sensors gave acceptable yield predictions (Figure 18). With the SPAD meter, it was important to separate the data according to the tillage system. With this separation, the SPAD meter was able to predict yield at V7, for both tillage systems, while NDVI values were not well related to yield until determined at V10. Being more sensitive to differences in N nutrition, the early SPAD meter readings better predicted grain yield reductions when N nutrition was limiting. However, good yield prediction was delayed relative to when N deficiency was first detected.

Nitrogen in the grain would give us information on how much nitrogen the plants were able to put in the grain. In 2010 there were no significant differences between tillage systems. On the other hand, in 2011, N-Grain was significantly higher for NT. Although MP had better N-nutrition all along the season until R1, it couldn't traduce it into the grain. The data would be suggesting that MP release N faster/earlier in the season whereas NT release it slower and in a more linear fashion offering N to the crop more according to the needs for increasing grain yield.

It could be worth to explore the possibility to work with both technologies in making variable N rate applications. The SPAD meter could be used to detect N deficiency and the GreenSeeker (or another/better sensor) would make variable rate applications of N fertilizer.

4.2. The GSC Field Trial

4.2.1. 2010

In this study weed control and/or nutrient availability (other than N) did not affect grain yield, which was limited in 2010 by water availability, as documented above for the BL23 study. A potassium deficiency was observed early in the season and corrected. Grain yield exhibited a good response to applied N and QP models were fitted to the data, as shown in Figure 20. The responses shown in Figure 20 do not evidence different efficiencies in the use of applied N between all N applied at planting and all N applied side-dress. Although there was no significant difference in grain yield between all N applied at planting and all N applied side-dress at N rates up to 224 kg N ha^{-1}, there was a clear trend suggesting that the all N side-dress treatments could not reach maximum yield.

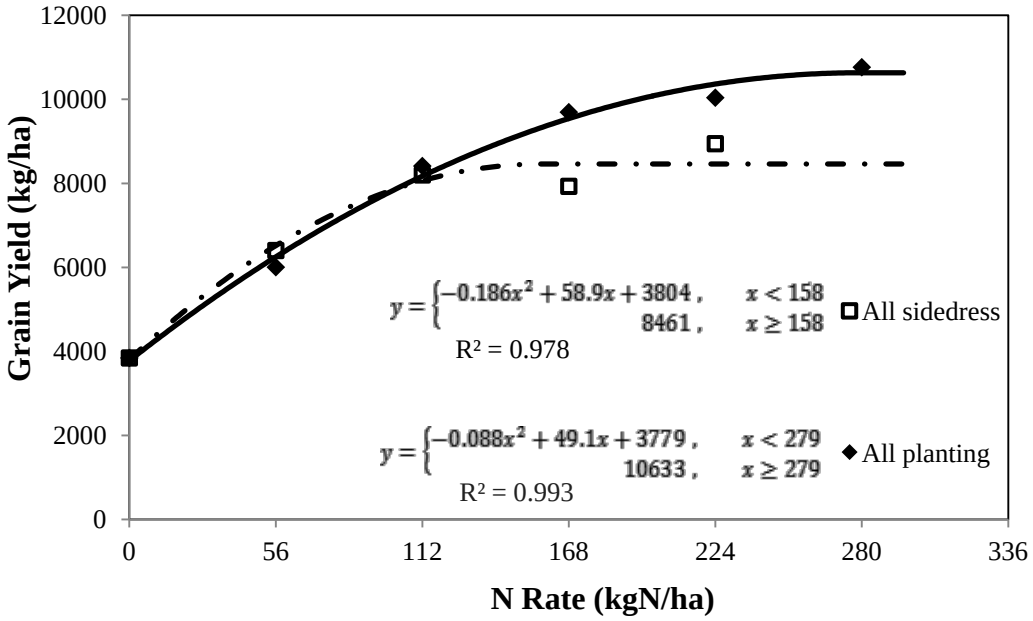

Figure 20. Relationship between applied N and 2010 corn grain yield for two different times of application. The quadratic-plateau (QP) regression models are shown, as are the N rates at which yields were maximized, for each time of application.

This might be due to a loss of yield potential due to waiting until V8 to apply the side-dressed N, delaying N availability for uptake by N starved plants.

In order to explore differences among the alternative strategies for the application of N, orthogonal contrasts were performed between the “all N applied at-planting” treatments (Planting), the “all N applied at side-dress” treatments (side-dress), and split N application treatments (split). Differences between all N at-planting and all N at side-dress treatments are important when evaluating the NDVI response to side-dress N applications (Table 3).

Table 3. Orthogonal contrasts (Pr > F values) between times of N applications in 2010.

	Leaf N	**Grain N**	**Yield**	**N removal**
Sidedress vs. Planting	0.0068	0.2418	0.3138	0.5412
Split vs. Planting	0.2709	0.8330	0.8265	0.8782
Sidedress vs. Split	0.0356	0.1577	0.1482	0.3381
	Pre-side-dress NDVI		**Post-side-dress NDVI**	
	Sen. 54	**Sen. 55**	**Sen. 54**	**Sen. 55**
Sidedress vs. Planting	0.4254	0.9554	0.5277	0.9015
Split vs. Planting	0.9370	0.2526	0.8210	0.9363
Sidedress vs. Split	0.2421	0.6833	0.7410	0.9491

Leaf N values exhibited significant differences due to N application timing; with higher values of leaf N associated with N applied at-planting treatments. It is possible that the observed trend for higher grain yield with N at-planting was related with higher leaf N values (Figure 21). The orthogonal contrasts did not find significant differences in grain yield, suggesting that the different strategies for applying N did not produce statistically significant differences in grain yield, despite the trend in the response data (Figure 20). None of the GreenSeeker sensor measures, taken either pre-sidedress or post-sidedress, gave significantly different NDVI readings due to the N application strategies.

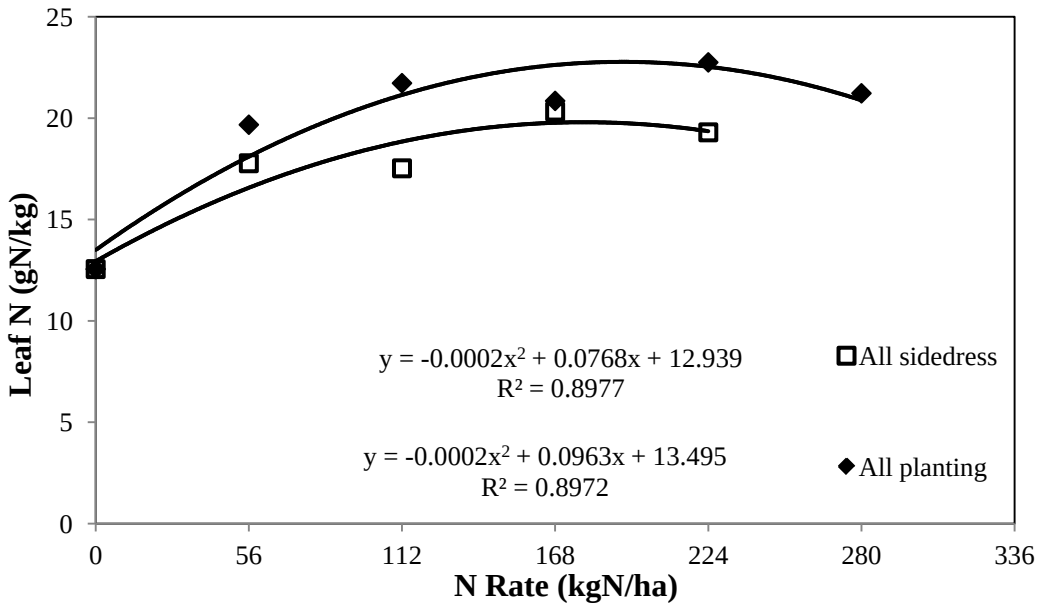

Figure 21. Relationship between leaf N concentration and fertilizer N rate for two N application strategies in 2010.

Nitrogen application treatments did affect grain yield, N concentration in the leaf, and N concentration in the grain (Table 4). Table 4 is organized from the smallest at-planting N rate treatment to the highest at-planting N rate treatment; it summarizes the analysis of variance for the measured variables. The high least significant difference (LSD) values are explained by high coefficients of variation in the data.

Leaf N exhibited consistent significant differences among the side-dress N treatments, as shown among treatments 1 through 5, and also among the N applied at-planting treatments (1,6,10,13,15 and 16). Leaf N reached the highest numerical value at 226 kg N ha^{-1} with all that N applied at-planting. Grain yield exhibited similar behavior, though reaching the maximum numerical value at 280 kg N ha^{-1}.

Table 4. Analysis of variance for the 2010 GSC study.

	N Rate Treatments at-planting – side-dress (kg N ha^{-1})		Leaf N (gN kg^{-1})	Grain N (gN kg^{-1})	Yield (kg ha^{-1})	N removal (kg ha^{-1})
1	0	0	12.7 G	9.2 H	3840 E	30.4 E
2	0	56	17.8 EF	9.3 H	6400 CD	50.2 EF
3	0	112	17.5 F	10.6 DEF	8200 BCD	73.6 CD
4	0	168	20.3 ABCD	11.9 ABC	7930 BCD	80.0 BCD
5	0	224	19.3CDEF	12.0 AB	8940 AB	91.0 ABCD
6	56	0	19.7 DEF	9.5 GH	6000 ED	48.9 EF
7	56	56	21.5 ABC	10.3 EFG	8330 BC	72.6 CD
8	56	112	20.2 ABC	11.1 CDE	9760 AB	91.4 ABCD
9	56	168	21.3 ABC	11.5 ABC	9870 AB	96.9 AB
10	112	0	21.7 ABC	10.1 FGH	8410 BC	70.4 DE
11	112	56	20.8 ABC	11.1 CDE	9820 AB	91.4 ABCD
12	112	112	22.1 AB	12.3 A	9730 AB	101.0 AB
13	168	0	20.8 ABC	11.3 BCD	9690 AB	92.4 ABCD
14	168	56	20.1 BCDE	11.7 ABC	9480 AB	93.6 ABC
15	224	0	22.7 A	11.7 ABC	10040 AB	99.4 AB
16	280	0	21.2 ABC	12.3 A	10760 A	111.9 A
LSD (0.1)			**2.5**	**0.9**	**2210**	**22.2**
LSD (0.05)			**3.0**	**1.1**	**2650**	**26.7**

It was expected that the behavior of both leaf N and grain yield would be reflected in NDVI readings. Table 5 shows the analysis of variance for the 2010 NDVI readings. The readings taken pre-side-dress exhibited few differences due to the treatments, and the sensors were even unable to discern any differences in N nutrition or canopy biomass due to the at-plant N application. The post-side-dress NDVI values were very erratic for the two sensors evaluated (54 and 55), giving strange results: the lowest NDVI value was observed at the highest at-plant N rate and the 0 kg N ha^{-1} control gave high NDVI values (Table 5). This year, the GreenSeeker's NDVI could not even find the N-deficiency. There might be a confounding effect of drought in these measurements.

GreenSeeker's NDVI was not a good predictor of either leaf N or grain yield for none of the two analyzed dates of measurements. No relationship could be established between

any of the two sensors and grain yield or leaf-N. In addition, no relationships could be found when looking at data for “all at planting” only or “all at sidedress” only.

Table 5. Analysis of variance for the 2010 NDVI readings.

	N-Treatments at-planting – side-dress (kg N ha^{-1})		**Pre-side-dress NDVI**		**Post-side-dress NDVI**	
			Sensor54	**Sensor55**	**Sensor54**	**Sensor55**
1	0	0	0.531 A	0.541 ABC	0.717 A	0.553 AB
2	0	56	0.544 A	0.551 ABC	0.723 A	0.557 A
3	0	112	0.522 A	0.501 C	0.642 B	0.472 BC
4	0	168	0.597 A	0.578 AB	0.720 A	0.515 ABC
5	0	224	0.581 A	0.554 ABC	0.717 A	0.556 A
6	56	0	0.519 A	0.562 ABC	0.732 A	0.523 ABC
7	56	56	0.564 A	0.602 A	0.728 A	0.560 A
8	56	112	0.576 A	0.556 ABC	0.721 A	0.543 ABC
9	56	168	0.514 A	0.561 ABC	0.700 AB	0.487 ABC
10	112	0	0.552 A	0.555 ABC	0.704 A	0.552 AB
11	112	56	0.551 A	0.550 ABC	0.691 AB	0.515 ABC
12	112	112	0.524 A	0.569 AB	0.718 A	0.519 ABC
13	168	0	0.539 A	0.544 ABC	0.697 AB	0.492 ABC
14	168	56	0.533 A	0.532 BC	0.715 A	0.535 ABC
15	224	0	0.548 A	0.527 BC	0.716 A	0.546 AB
16	280	0	0.553 A	0.513 BC	0.697 AB	0.463 C
LSD (0.1)			**0.088**	**0.067**	**0.061**	**0.083**
LSD (0.05)			**0.106**	**0.081**	**0.073**	**0.099**

4.2.2. 2011

As in 2010, there were no problems with weeds and nutrient availability. Water availability did not affect yields as in the previous year, allowing grain yield to better express N nutritional limitations. Figure 22 shows the grain yield response to the rate of N application. The QP fitted models suggest a better NUE for the treatments where all N was applied at side-dressing. All N applied at-planting exhibits a higher yield plateau value, in accord with the 2010 results. Also, as in 2010, the difference in yields between N application times, at the same N rate (up to 224 kg N ha^{-1}) was not statistically significant.

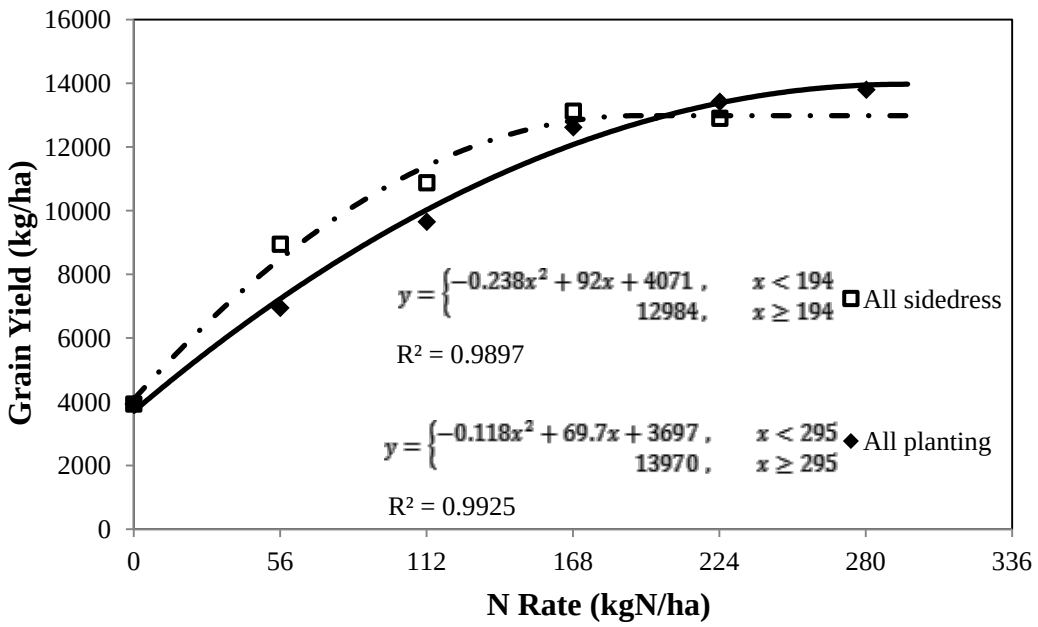

Figure 22. Relationship between applied N and 2011 corn grain yield for two different times of application. The quadratic-plateau (QP) regression models are shown, as are the N rates at which yields were maximized, for each time of application.

The 2011 orthogonal contrasts were performed in order to explore differences among the alternative strategies for the application of N, and are shown in Table 6.

Table 6. Orthogonal contrasts (Pr > F values) between times of N applications in 2011.

	Leaf N	Grain N	Yield	N removal
Sidedress vs Planting	0.0003	0.0039	0.1364	0.0178
Split vs Planting	0.8779	0.4668	0.2580	0.1938
Sidedress vs Split	0.0064	0.0329	0.3506	0.9103

	Pre-side-dress NDVI			Post-side-dress NDVI		
	Sen. 54	**Sen. 53**	**Sen. 55**	**Sen. 54**	**Sen. 53**	**Sen. 55**
Sidedress vs Planting	0.0056	<.0001	<.0001	<.0001	<.0001	<.0001
Split vs Planting	0.3856	0.7063	0.1603	0.3851	0.9636	0.2925
Sidedress vs Split	0.0134	<.0001	0.0007	<.0001	<.0001	<.0001

The table above shows significant differences in leaf N between side-dress and the other two strategies, and no differences between all at-planting and split N applications. No significant differences are shown for grain yield. These results suggest that the N status of the crop at R1 was not going to be reflected in grain yield (although there is a trend similar to the one found in 2010). The NDVI results seem to be more in accord with leaf N than with grain yield. However, this is not the case. While leaf N was consistently higher with all N applied at side-dressing than for all N applied at-planting (Figure 23, Table 7), NDVI values (both pre and post side-dress) gave opposite results (Table 8) with the all N at-planting treatment values being consistently higher than the all N applied at side-dress values. This suggests greater sensitivity of NDVI readings to differences in crop biomass rather than to differences in N nutrition, confirming observations from the BL 23 study discussed above.

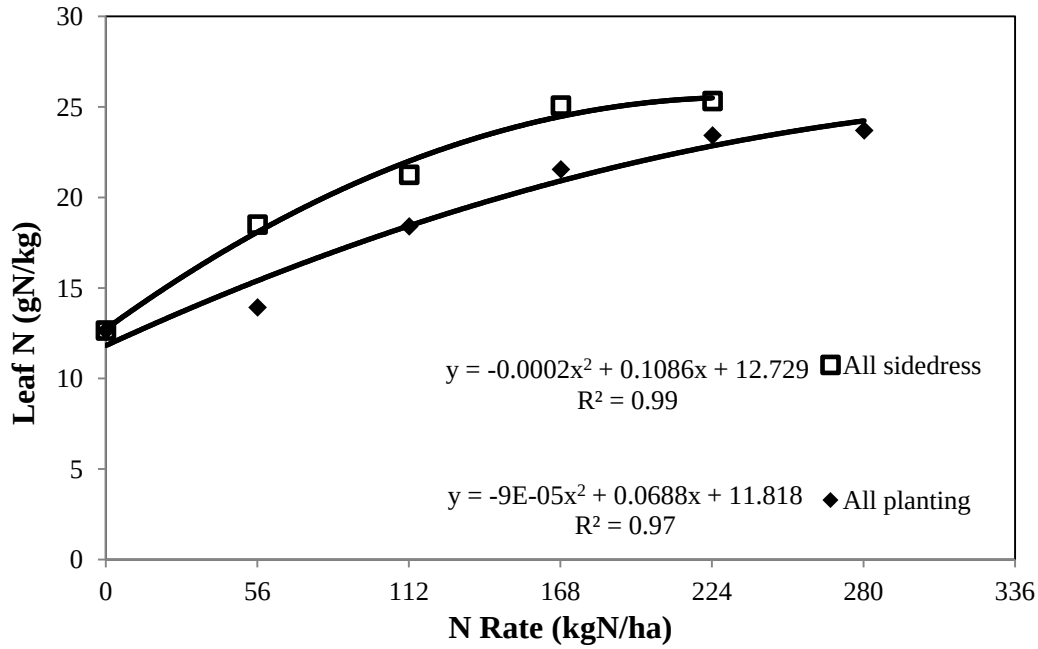

Figure 23. Relationship between leaf N concentration and fertilizer N rate for two N application strategies in 2010.

Table 7. Analysis of variance for the 2011 GSC study.

	N Rate Treatments at-planting – side-dress (kg N ha^{-1})		Leaf N (gN kg^{-1})	Grain N (gN kg^{-1})	Yield (kg ha^{-1})	N removal (kg N ha^{-1})
1	0	0	12.7 G	10.2 EF	3930 F	33.1 G
2	0	56	18.5 F	9.6 F	8950 D	73.3 EF
3	0	112	21.3 DE	11.6 BC	10880 BC	106.2 D
4	0	168	25.1 AB	12.6 A	13120 A	139.1 AB
5	0	224	25.3 A	12.4 A	12900 A	135.6 AB
6	56	0	13.9 G	9.5 F	6950 E	55.3 F
7	56	56	20.0 EF	10.5 DE	12440 AB	110.1 CD
8	56	112	20.4 EF	11.8 ABC	12910 A	128.9 ABC
9	56	168	24.2 ABC	12.3 AB	12840 A	133.3 AB
10	112	0	18.4 F	10.3 EF	9650 CD	83.8 E
11	112	56	22.5 BCDE	11.8 ABC	12880 A	128.2 ABC
12	112	112	21.6 CDE	12.4 A	13880 A	145.3 A
13	168	0	21.6 CED	11.3 CD	12620 AB	120.7 BCD
14	168	56	24 .0 ABCD	11.9 ABC	14020 A	141.2 A
15	224	0	23.4 ABCD	12.2 AB	13420 A	138.0 AB
16	280	0	23.7 ABCD	12.5 A	13800 A	145.4 A
LSD (0.10)			**2.7**	**0.8**	**1780**	**19.2**
LSD (0.05)			**3.3**	**1.0**	**2130**	**23.0**

Table 7 shows the analysis of variance for the measured variables. For grain yield, although the LSD (0.10) was high (1780 kg ha^{-1}), significant differences were found between the treatments. In the table, as in Figure 22, the yield plateaus reached with all N applied at side-dressing and all N applied at–planting are evident. Split N applications seemed to be more efficient, and treatment 7 (112 kg total N ha^{-1}) was not significantly different in yield from that observed with the highest yielding treatments, while exhibiting significantly greater yield than the other 112 kg total N ha^{-1} treatments (treatments 3 and 10). Leaf N also responded to N application, with treatment 5 exhibiting the highest value (Table 7).

As the grain yield response was better in 2011, it was expected that there would be differences in the NDVI readings due to the treatments. Table 8 shows the analysis of variance for the 2011 NDVI measurements. The following comparisons between treatments are of particular interest, and reader attention is drawn to these in an effort to explore agreement between NDVI readings and variables like grain yield or leaf N:

- 1 versus 2,3,4,5: The NDVI was expected to detect N-deficiency, giving a difference between treatment 1 and the rest after side-dressing. However, there are no significant differences among these treatments, for any of the sensors, which suggests that although N nutrition differences might be present, the canopy biomass not generated up to side-dressing at V8 cannot build fast enough so as to cause differences in the NDVI readings at the time the measurements were taken.
- 6 versus 7,8,9: Again, there are differences in N nutrition, as indicated by leaf N values and reflected in grain yield, but NDVI readings were not able to “see”

significant differences among these treatments. It could be said that there was no NDVI reaction to side-dressed N in this case, either.

- 10 versus 11,12: There are significant differences between treatment 10 and the other two, in both leaf N and grain yield. The NDVI exhibited no significant differences.
- 2 versus 6: In this case, side-dress N produced higher leaf N values than the same amount of N applied at-planting, suggesting better N nutrition that was also reflected in grain yield values. In contrast, NDVI readings were higher for treatment 6 (with the N applied at-planting).
- 7 versus 3: There are no differences between these two treatments in either grain yield or leaf N. However, there were significantly higher NDVI values for treatment 7 (which had half the fertilizer N applied at-planting) for both pre- and post side-dress NDVI measurements, supporting the idea of higher sensitivity of NDVI to canopy biomass than N nutrition.
- 7 versus 10: Although there were no significant differences in leaf N, treatment 7 did express a higher leaf N value and a significantly higher grain yield. The NDVI was not different between these treatments.

Table 8. Analysis of variance for the 2011 NDVI readings.

N-Treatments at-planting – side-dress (kg N ha^{-1})			Pre-sidedress NDVI			Post-sidedress NDVI		
			Sen. 54	**Sen. 53**	**Sen. 55**	**Sen. 54**	**Sen. 53**	**Sen. 55**
1	0	0	0.375	0.486	0.261	0.544	0.685	0.326
2	0	56	0.451	0.525	0.237	0.605	0.725	0.373
3	0	112	0.357	0.445	0.207	0.553	0.697	0.307
4	0	168	0.507	0.496	0.268	0.674	0.738	0.458
5	0	224	0.375	0.475	0.206	0.566	0.732	0.339
6	56	0	0.486	0.574	0.311	0.675	0.813	0.520
7	56	56	0.566	0.620	0.337	0.735	0.832	0.577
8	56	112	0.503	0.586	0.318	0.726	0.837	0.534
9	56	168	0.517	0.567	0.281	0.720	0.828	0.560
10	112	0	0.468	0.583	0.272	0.714	0.834	0.547
11	112	56	0.440	0.570	0.285	0.706	0.843	0.562
12	112	112	0.493	0.608	0.276	0.741	0.850	0.600
13	168	0	0.560	0.626	0.352	0.754	0.846	0.611
14	168	56	0.452	0.540	0.298	0.726	0.843	0.566
15	224	0	0.514	0.570	0.330	0.744	0.834	0.604
16	280	0	0.474	0.560	0.304	0.735	0.847	0.623
LSD (0.10)			**0.098**	**0.077**	**0.063**	**0.060**	**0.048**	**0.089**
LSD (0.05)			**0.117**	**0.093**	**0.076**	**0.072**	**0.057**	**0.107**

GreenSeeker NDVI values for 2011 were generally not a good predictor of leaf N or grain yield in this study. As in 2010, when using all the data, there was no relationship between grain yield and NDVI; either for pre- or post-side-dress readings. Considering only the yields with all N applied at side-dressing did not give a relationship, either. However, using only the data for all N applied at-planting, it was possible to find a good correlation between grain yield and the NDVI readings (Figure 24). Exponential models best fitted these relationships, as in the previously discussed study. Sensor 55 (inter row), which was mounted in the inter-row, exhibited higher R^2 and greater resolution. Sensors 53 (over row) and 54 (over row) seemed to exhibit saturated NDVI readings, causing sensor 55 (inter row) to be a better predictor of grain yield. It can be concluded that for grain yield prediction purposes, GreenSeeker sensors should be mounted over the inter-

row when taking readings at advanced growth stages to allow the sensor to better distinguish differences in canopy biomass and avoid saturation of NDVI. However, as there seems to be important differences in the NDVI values between the sensors over the rows (54 and 53) which can be interpreted as a inherited error from the instrument, further study is required to test the hypothesis that at advanced growth stages the best position of the sensor would be in the inter-row.

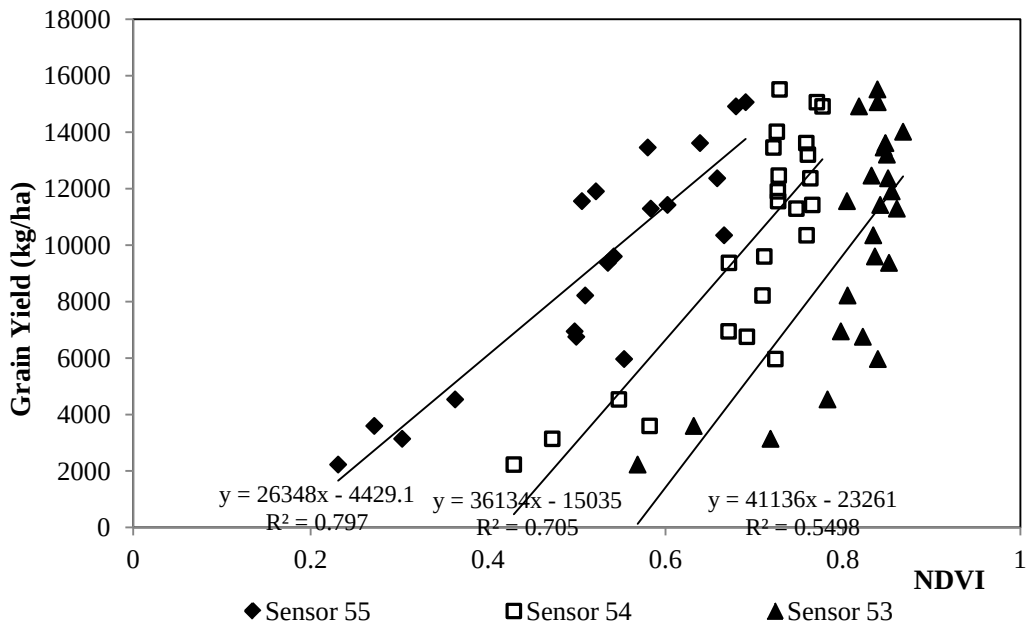

Figure 24. Relationship between 2011 grain yield of pre-plant N treatments and post-side-dress (V10-V11) NDVI readings for the 3 sensors used when no N was applied at side-dress.

4.2.3. Summary

In 2010, there was a significant leaf N and grain yield response to N rate. All N at planting treatments exhibited better N nutrition. However, neither the GreenSeeker sensor 54 (over the row) nor sensor 55 (inter row) found any differences among the treatments. In this case the GreenSeeker was not able to either find N deficiency or produce a response to side-dressing N applications. This means that the GreenSeeker was unable to distinguish canopy biomass or N nutrition among treatments in this year. Sensor 54 (over the row) post-side-dress readings show a reaction relative to the pre-side-dress readings meaning that it was able distinguish between an early growth stage crop and an advance growth stage crop; however, as said before, it could not distinguished among N treatments. Sensor 55 (inter row) was not even able to do that for which the validity of this sensor's data is doubted. The high coefficient of variation in the grain yield data, the drought, and the fact that the blocking didn't help for leaf N this year could have affected the accuracy of the GreenSeeker.

In 2011, the GreenSeeker sensor readings were highly variable. When considering the pre-plant N applied treatments, the pre-side-dressing NDVI values for the 0 kgN ha^{-1} were generally lower than those for the higher N rates; however, the N deficiency is not found consistently. In addition, early NDVI readings were not good predictor of grain yield. The post-side-dressing NDVI readings were able to find N deficiency consistently giving significant differences between the 0 kgN ha^{-1} and the rest of the pre-plant N treatments. Also, if no N was applied at side-dress, post-side-dressing NDVI readings (V10-V11) were able to make a good grain yield prediction. When 56 kgN ha^{-1} or 112 kgN ha^{-1} were side-dressed, although the variability of the NDVI readings got reduced,

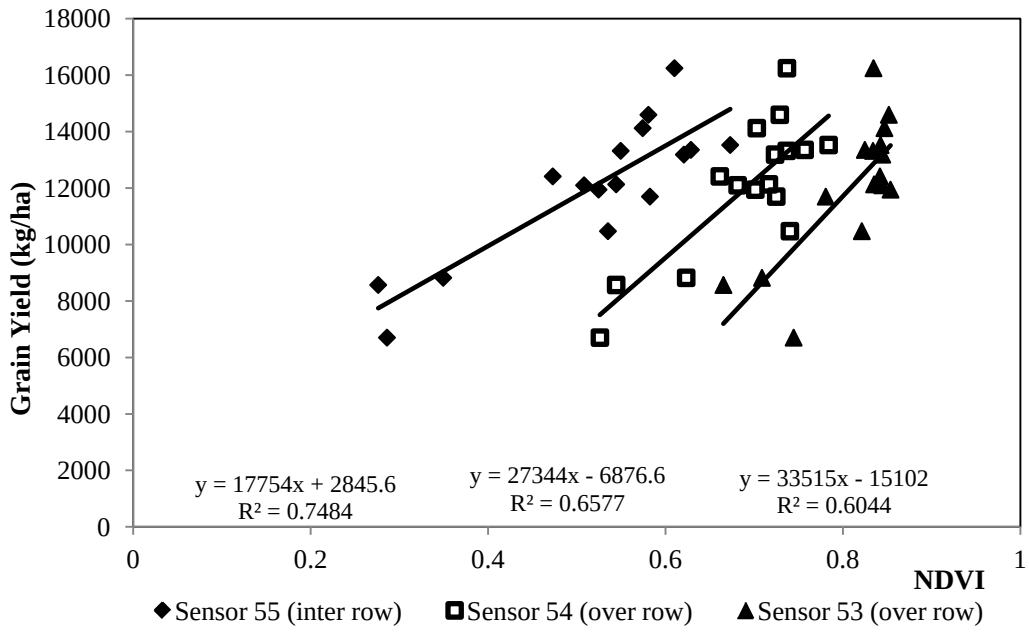

Figure 25. Relationship between 2011 grain yield of pre-plant N treatments and post-side-dress (V10-V11) NDVI readings for the 3 sensors used when 56 kgN ha^{-1} was applied at side-dress.

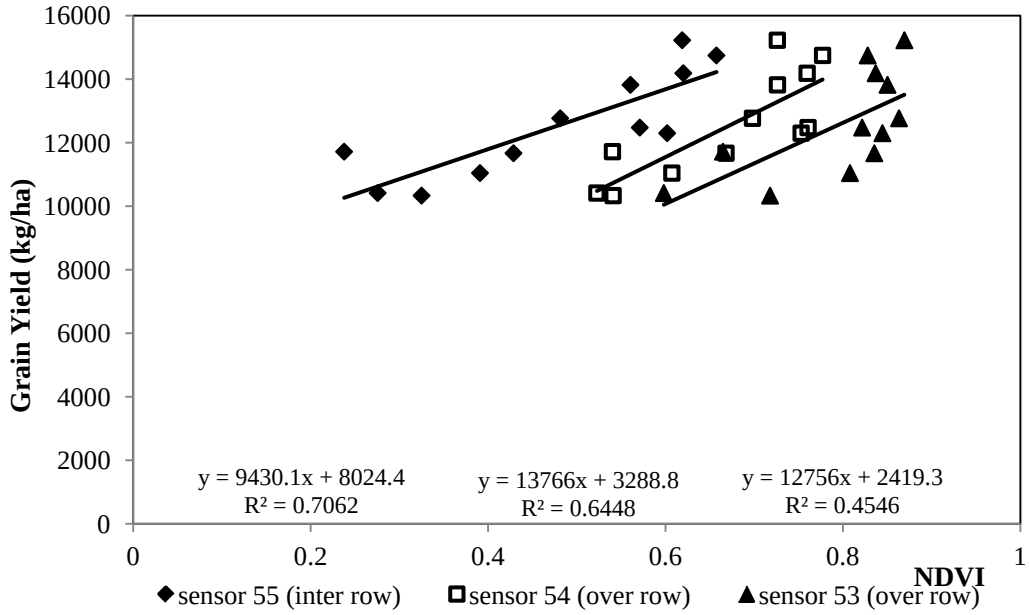

Figure 26. Relationship between 2011 grain yield of pre-plant N treatments and post-side-dress (V10-V11) NDVI readings for the 3 sensors used when 112 kgN ha^{-1} was applied at side-dress.

grain yield was still predicted by the NDVI (Figure 25, Figure 26). I these cases the relationship ceased to clearly exponential and linear models were fitted to the data. It is important to notice a clustering of the data for all the sensors, but most important for the sensors mounted on the rows. Sensor 53 (over the row) and 54 (over the row) NDVI tended to get saturated. The sensor 55 (inter row) didn't seem to get saturated like the others; however, like the rest of the sensors, it didn't present an R^2 as good as in Figure 24.

Finally, as for the Blevins 23 study, the 2011 GSC data was visually compared with the exponential equation for yield prediction by NDVI from Teal et al. (2006). As Figure 19 earlier, Figure 27 shows that our data does not seem to follow Teal's equation.

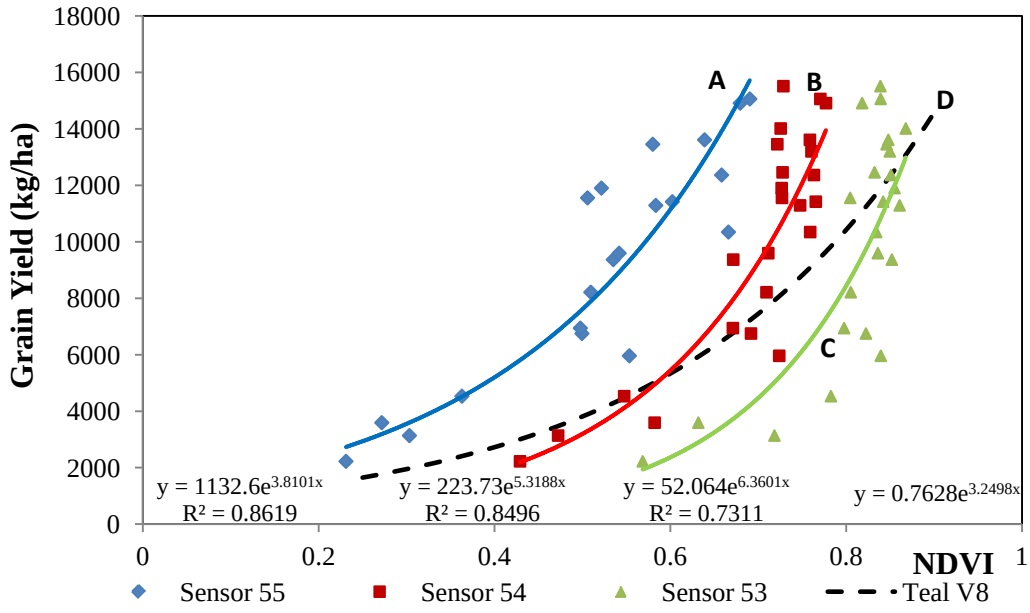

Figure 27. Relationship between 2011 grain yield and NDVI for Senor 55 (Inter row) (A), Sensor 54 (B); Sensor 53 (C) and for Teal et al. (2006) regression equation (D).

As is believed that yield prediction by NDVI is field and growth stage specific, it is important to note that our data was taken at the V10-V11 growth stage while Teal's data

was collected at V8. Teal's data seemed to present a better resolution than ours, particularly for the sensors 54 and 53 (over the row). While the sensors placed on the row seem to be closer to Teal's equation, Senor 55 (inter row) seems to strongly differ from it.

CONCLUSIONS

After two years of research (2010, 2011) significant results can be concluded. Remote sensing technology that employs active NDVI sensors, in this case the GreenSeeker, can detect early N deficiency. Although not as effective as the Chlorophyll meter, the active sensors have the advantage of being able to cover greater sensing area, faster, making it possible to “map” entire fields and creating the opportunity for detection and separation of field areas with N deficiency.

Early Grain yield prediction with the active NDVI sensors was not possible with these data. Further research with small N rate increments like 15 kg N ha^{-1} would be necessary to better understand the N deficiency resolution at which young corn plants would provide information regarding their N nutritional need.

Background reflectance differences due to levels of crop residues resulting from different soil tillage management choices significantly affected NDVI readings at early growth stages. There was an important contrast between the seasons; the 2010 year was a dry season and the 2011 was a wet season, causing the difference in background reflectance due to tillage to be inconsistent for the two seasons. After V8, soil interference was reduced, and it was possible to fit the yield versus NDVI data to a single model, across both tillage treatments. However, special care has to be taken when combining NDVI data from different sensors, because these might differ in their calibration. This happened in the GSC field trial. There were consistent differences between sensors 54 and 53, both mounted over the row. It was difficult to confirm with enough confidence that NDVI values from a sensor mounted over the inter-row (sensor 55) better predicted grain yield when determined after V8. The observation that a sensor

over the inter-row was more sensitive to changes in canopy biomass because of better contrast with the soil background, and was, therefore, a better predictor of grain yield, should be studied further.

The literature indicates that NDVI measurements should be taken at a height not greater than 0.8 m. Although the NDVI measurements taken low (0.6m) in this study were preferred because they were not as erratic as the ones taken high (1.2 m), the height at which the NDVI measurements were taken did not have a big impact on the behavior of the readings. A slightly better relationship was found between the low height NDVI measurements and the SPAD readings, but this was not always the case.

Side-dressing the N did not significantly affect maximum grain yield, but threre was a trend suggesting that if an N deficiency was present very early in the growing cycle, then some yield potential could be lost. In 2010, leaf N reacted differently than in 2011, suggesting this parameter would be dependent on the season. The impact of side-dressing N on the NDVI readings taken a week from side-dressing was noticeable in 2011. This crop response to side-dressing N a week from its application might be due to N nutrition rather than canopy biomass. This suggests that NDVI would also be sensitive to N nutrition at the V10-11 growth stage. Further research would be necessary to assess the usefulness of this kind of response in leading to an N recommendation.

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VITA

Donato Titolo was born on February 10th, 1979 in Santa Rosa, La Pampa, Argentina. He was raised in an environment where cattle grazing was the main agricultural activity. In 2002, Donato was accepted as an undergraduate at the National University of La Pampa in order to pursue an Agriculture Engineer degree, which was completed in 2007. In his years as an undergraduate, he had wonderful experiences. He participated of the student government as press secretary and was elected student counselor for the university government.

After completing his undergraduate degree, he traveled around the world for one year and did temporary work for nine months in New Zealand, all related to agricultural activities. Upon returning to Argentina, he worked for one year at a “La Sombra S.A.” a 4000 has. farm located south of Cordoba province, where the main activities were grain crop production and cattle grazing. He entered the Graduate School of the University of Kentucky in December 2009 and was appointed to a Graduate Research Assistantship under the supervision of Dr. John H. Grove. In June 2012, Donato accepted employment with Dow Agrosciences as a Field Research Biologist.