AERIAL IMAGERY AND OTHER NON-INVASIVE APPROACHES TO DETECT NITROGEN AND WATER STRESS IN A POTATO CROP

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ABSTRACT

Post-emergence nitrogen (N) fertilizer is typically split applied to irrigated potato (*Solanum tuberosum* L.) in Minnesota in order to minimize the likelihood of nitrate leaching and to best match N availability to crop demands. Petiole nitrate-nitrogen (NO_3-N) concentration is often used as a diagnostic test to determine the rate and timing of split applications, but determining spatial variability within a field using this approach is difficult. Canopy-level spectral measurements, such as hyperspectral and multispectral imagery, have the potential to be a reliable tool for making in-season N management decisions for precision agriculture applications. A two year field study was conducted on a loamy sand soil to evaluate the effects of variety, N treatment, and water stress on: (i) tuber yield, tuber quality, and plant N uptake characteristics; (ii) total N and NO_3-N concentrations in petiole, leaflet, and whole leaf tissue samples throughout the season; and (iii) the ability of spectral data, especially from aerial hyperspectral and multispectral imagery, to predict leaf N concentration and tuber yield. The study included two potato varieties (Russet Burbank and Alpine Russet), two irrigation regimes (unstressed and stressed), and five N treatments categorized by three N rates (34 kg N ha^{-1}, 180 kg N ha^{-1}, and 270 kg N ha^{-1}) in which the 270 kg N ha^{-1} rate had post-emergence N either split applied or applied early in the season. In addition, one of the 270 kg N ha^{-1} rates with post-emergence N split applied included a soil surfactant application. Reflectance measurements were made using both ground- and aerial-based platforms using several sensors: SPAD-502 chlorophyll meter, Cropscan MSR16R multispectral radiometer, AISA-Eagle hyperspectral camera, and Redlake MS4100 multispectral camera.

Insufficient supplemental water during critical growth stages was found to negatively affect tuber yield, tuber quality, and plant N uptake, but surprisingly had a positive overall effect on frying quality. Because of its high yields, higher proportion of tubers >170g, higher N uptake, and similar processing qualities compared with Russet Burbank (RB), the Alpine Russet (AR) variety is attractive from both an economic and environmental perspective. For all N treatments except the 34 N early treatment, N use efficiency and N uptake efficiency values were less than 50 g g^{-1} and 50%, respectively. This indicates a relatively low efficiency of N use during 2010 and 2011 attributed to heavy leaching events during these years. Within the same N rates, N uptake efficiency was higher when post-emergence N was split applied, which is likely the result of less N being lost due to leaching. The use of the soil surfactant had a minimal effect on the parameters measured in this experiment.

Tissue samples analyzed for NO_3-N were very responsive to N fertilizer applications that occurred within about 7 days of the sampling date, and are therefore a good indicator of current plant N uptake. Alternatively, tissue samples analyzed for total N were more stable over sample dates, and appear to be a better indicator of overall plant N uptake at the time of sampling. In general, ground-based spectral data could predict tuber yield better than any of the N-based tissue sampling procedures; their coefficient of determination (r^2) values ranged from 0.40-0.85 on all measurement dates throughout the 2010 and 2011 seasons. Applying the nitrogen sufficiency index (NSI) to plant measurements and spectral indices/models made them mostly insensitive to the effects of variety; this is an effective way to normalize data based on local growing conditions and cultural practices.

The best predictor of N stress was determined to be the partial least squares regression model using derivative reflectance as input for its independent variables (r^2 of 0.79 for RB and 0.77 for AR). However, the best technique for determining N stress level for variable rate application of N fertilizer using the NSI was determined to be MTCI (MERIS Terrestrial Chlorophyll Index) because the combination of its good relationship with leaf N concentration and high accuracy. The inherent variability of a spectral index should be considered before determining the N sufficiency threshold for determining the rate and timing of post-emergence N fertilizer applications. GRVI (Green Ratio Vegetation Index) normalized by an NSI that used the recommended rate and timing from the research plots as a reference could detect areas of the commercial field that were most unsuitable for supplemental N fertilizer applications on different measurement dates. On 56 and 79 DAE in 2011, most of the commercial field was above the GRVI NSI over-sufficiency threshold level of 120% (the mean pixel values were greater than 127% for both varieties).

Because of differences in potato variety, growth stage, sensors, or other local conditions, reference areas are needed in order to make accurate recommendations. The results from this study suggest that diagnostic criteria based on biomass and nutrient concentration (e.g., canopy-level spectral reflectance data) were best suited to determine overall crop N status for determination of in-season N fertilizer recommendations.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

GENERAL INTRODUCTION

Minnesota ranks 7th in U.S. potato (*Solanum tuberosum* L.) production with about 18,000 ha of irrigated potato grown in 2010 (NASS, 2010). The sandy loam and loamy sand soils used for irrigated potato production in Minnesota are typically low in organic matter and cation exchange capacity, and therefore, have a relatively low reserve of soil nutrients. The low inherent soil fertility and the large nutrient requirement result in a high rate of fertilizer inputs (Dean, 1994). Because of the high nitrogen (N) fertilizer rates and high hydraulic conductivity soils used for irrigated potato, there is a high potential for nitrate-nitrogen (NO_3-N) leaching. In addition, potato plants are relatively shallow rooted compared with other field and vegetable crops; this feature results in below average nutrient uptake and poor nutrient use efficiency, which can ultimately lead to higher amounts of nitrate leaching (Lynch et al., 2012; Rosen and Bierman, 2008; Zebarth and Rosen, 2007; Zvomuya et al., 2003).

Potato yield and quality are highly dependent on an adequate supply of nitrogen (N) fertilizer. However, high rates of N fertilizer and rainfall/irrigation on coarse-textured soils have been shown to increase the risk of N loss from the root zone (Errebhi et al., 1998). This results in an economic loss to the grower and can degrade groundwater quality via elevated groundwater nitrate concentrations. Based on previous experiments on coarse-textured soils in Minnesota, only about a third to a half of applied N is recovered in years of moderate to heavy leaching (Errebhi et al., 1998; Waddell et al., 2000). Plant N uptake in the presence of an abundant supply of N (i.e., N uptake capacity) can vary by potato variety (Zebarth et al., 2004). Increased plant N uptake over the growing season can reduce the quantity of N lost from the root zone, thereby minimizing financial losses. Therefore, from both an economic and environmental perspective, it is desirable to grow potato varieties that have the ability to optimize the use of added N (Errebhi et al., 1999).

Potato plants are also very sensitive to water stress (Bailey, 2000; Gregory and Simmonds, 1992; Phene and Sanders, 1976). When leaf stomates close, stomatal conductance decreases to the extent necessary to prevent leaf water potential from falling below critical levels, which could ultimately impede growth and reduce tuber yield (Bailey, 2000; Gregory and Simmonds, 1992; Phene and Sanders, 1976; Wright and

Stark, 1990). Water stress can also affect tuber quality characteristics such as specific gravity, incidence of misshapen tubers, and susceptibility of bruising (Ojala et al., 1990; Wright and Stark, 1990).

A logical strategy for determining an appropriate rate and timing of split applied N fertilizer is to make adjustments based on in-season plant monitoring. Since this approach uses plant measurements during the growing season, it is able to account for the effect of seasonal weather conditions on crop N availability (Meisinger et al., 2008). Currently, a best management practice for potato production in Minnesota is to base the rate and timing of post-emergence N fertilizer applications on petiole NO_3-N concentrations (Rosen and Bierman, 2008) and is being implemented by many commercial growers. The limitation of this procedure is that it is difficult to account for within-field spatial variability. The opportunity exists to use remote sensing to predict crop biophysical parameters that depend on crop N uptake such as tissue N or NO_3-N concentration and/or leaf chlorophyll concentration in order to account for within-field spatial variability for variable rate application of fertilizer. Spectral imagery is a powerful research tool that can provide information necessary to determine the best techniques to monitor crop N status over a variety of conditions and growth stages.

In general, there are a number of factors that limit the use of remote sensing from being used commercially for monitoring in-season plant status for potato production. First, spectral data processing techniques can be time consuming, and there is some uncertainty in the ability to accurately detect deficiencies for supplemental N to be applied in a timely manner, especially while accounting for within-field spatial variability. Little is known about the effect of crop growth stage or potato variety on the ability of spectral data to be used to adjust in-season applications. A better understanding of the response of spectral data and processing techniques, especially their behavior under varying conditions (i.e., growth stage, variety, yield potential), can improve the feasibility of this approach to allow its use at a commercial scale.

NOTE: This field study was designed and carried out in collaboration with a group in Israel in which field experiments were completed at both locations. The overall objective of the entire study is to fuse remotely sensed data and imagery in the visible/near-infrared and thermal spectrum to distinguish between water and nitrogen status in a potato crop. The contents of this thesis include both irrigation and nitrogen fertilizer treatments, but data analysis was primarily done using only visible/near-infrared imagery. The group in Israel is working more with the thermal data, and the completion of this overall objective is still in progress.

RESEARCH OBJECTIVES

The overall objectives of this research were to:

1) Evaluate the effects of variety (Russet Burbank and Alpine Russet), nitrogen (N) treatment, and water stress on: (i) tuber yield and quality characteristics, (ii) nitrogen use indices, (iii) residual inorganic soil N levels, and (iv) reducing sugar concentrations and how they relate to potato chip color after frying.
2) Evaluate how total N and nitrate-nitrogen concentrations for petiole, leaflet, and leaf samples change throughout the season for Russet Burbank and Alpine Russet varieties.
3) Determine which tissue samples/N analysis techniques and growth stages can best predict Grade A tuber yield.
4) Examine the ability of chlorophyll meter readings and narrowband spectral indices from Cropscan to predict leaf N concentrations, leaf area index, and Grade A tuber yield at different growth stages throughout the season.
5) Evaluate the ability of chlorophyll meter readings, narrowband and broadband indices, and partial least squares regression models to predict leaf N concentration in Russet Burbank and Alpine Russet varieties.
6) Evaluate the ability of chlorophyll meter readings and spectral indices/models to classify nitrogen sufficiency index stress levels into pre-determined stress classes via an accuracy assessment.
7) Evaluate the differences in variability within and across experimental treatments for petiole nitrate-nitrogen and leaf N concentration, chlorophyll meter readings, and spectral indices/models after being normalized by a nitrogen sufficiency index.
8) Evaluate the potential for using canopy-level hyperspectral reflectance as an indicator of potato N status in two varieties subjected to various N treatments and irrigation regimes.
9) Determine how nitrogen sufficiency index values can vary at different growth stages because of differences in the nitrogen sufficiency index reference used
10) Identify whether estimates of N stress can be made in a commercial field without using a reference N strip.

11) Investigate how the variability within a commercial potato field changes with spatial resolution at different growth stages.

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Chapter 1.

LITERATURE REVIEW

1. History and importance of potato production

Wild species of the potato (*Solanum tuberosum* L.) are native to South America, Central America, and possibly parts of North America; however, they are thought to have been first cultivated as a food source 2,500-10,000 years ago in the region of the Andes of what is now known as south Peru and west Bolivia (Hawkes, 1992; Salaman, 1985; Burton, 1989). During the Spaniard's exploration of South America in the mid-1500s, they exploited the potato and returned prosperous to Spain by selling chuñus (i.e., potatoes dried in the sun) for human consumption (Burton, 1989). Although chuñus were brought to Europe in the 1550s, it was not until about the 1570s when the potato was finally introduced into Europe as a domesticated crop (Burton, 1989; Hawkes, 1992). Shortly after the introduction into Europe, the domesticated potato spread throughout the rest of the discovered world relatively quickly (Hawkes, 1992). Although potatoes may have been native to parts of North America, they were not domesticated in what is now the northern continental United States until European settlers discovered America (Burton, 1989; Dean, 1994).

The potato is a very important food source, both nationally and globally. Among all other non-cereal food crops, potatoes rank third in the United States after sugar beet and sugar cane with over 18 million Mg produced in 2010; worldwide, potatoes rank first among all other non-cereal food crops with over 324 million Mg produced in 2010 (FAOSTAT, 2010; NASS, 2010). Minnesota is the seventh largest potato producing state in the U.S. with just over 0.77 million Mg produced in 2010 (NASS, 2010). Raw Russet potatoes contain over 18% carbohydrates, over 2.1% protein, low levels of fat, and high levels of many vitamins and minerals, which make them an excellent source of energy when consumed as food by humans (Dean, 1994; NND, 2011).

2. Economic and environmental implications of nitrogen in agricultural systems

2.1. Nitrogen cycle

Within agricultural soils, nitrogen (N) is continuously being transformed between different organic and inorganic forms. The rates of these N transformations depend on biological processes being carried out by organisms within the system (Vos and MacKerron, 2000). It is important to understand the cycling between the different forms of N because most agricultural crops require substantial quantities of inorganic N for adequate growth. The inorganic compounds of N that are generally taken up by plants include the ammonium ion (NH_4^+) and the nitrate ion (NO_3^-). The net positive charge present on the NH_4^+, allows it to adsorb to soil exchange sites if the soil has a sufficiently high exchange capacity (Tisdale et al., 1985). Conversely, the net negative charge on the NO_3^- permits it to move freely within the soil/water system (Tisdale et al., 1985). Because nitrate-N (NO_3-N) is mobile and is generally present in higher concentrations in soils than ammonium-N (NH_4-N), it is often the dominant source of N for plant uptake (Tisdale et al., 1985).

Organic forms of N occur naturally in soil systems as compounds of proteins, amino acids, microbial cell-wall polymers, amino sugars, nucleic acids, and a variety of vitamins, antibiotics, and metabolic intermediates (Sylvia et al., 2005). Ammonification (often referred to in a more general sense as mineralization) is the biological transformation of these organic forms of N to ammonium. Ammonification occurs when heterotrophic microorganisms consume organic material for their own energy and/or carbon demands. If the catabolic metabolism of the microorganisms on the organic material exceeds anabolic processes, there will be excess ammonium which is then released as an NH_4^+ (Myrold and Bottomley, 2008). Immobilization is the conversion of NH_4^+ to an organic form of N. Immobilization occurs when microorganisms take up the NH_4^+ to satisfy their N needs, in which case it is ultimately incorporated into the organic biomass – this consequently results in the temporary unavailability of N to plants (Myrold and Bottomley, 2008; Sylvia et al., 2005).

Under aerobic conditions (typical in most agricultural soils), NH_4^+ can be oxidized and ultimately converted to NO_3 in a process called nitrification. Nitrification is a two-step process in which the NH_4^+ is first oxidized and converted to a nitrite ion (NO_2^-) by ammonia oxidizing bacteria such as *Nitrosomonas* or *Nitrosospira*; the NO_2^- is then oxidized further and converted to a NO_3^- by nitrite oxidizing bacteria such as *Nitrobacter*. Because of the susceptibility of NO_3-N to be lost from the root zone, it is desirable to manage agricultural soils to reduce nitrification in an attempt to reduce groundwater nitrate pollution (Norton, 2008). Under anaerobic conditions (usually because of soil saturation), nitrate can be reduced and ultimately converted to gaseous forms of N through anaerobic decomposition in a process called denitrification (Tisdale et al., 1985). Due to the limited availability of oxygen under anaerobic conditions, the NO_3^- is used as an alternate electron acceptor for the metabolism of facultative aerobic bacteria such as *Pseudomonos*, *Bacillus*, and *Paracoccus* (Tisdale et al., 1985). During this process, the NO_3^- is reduced to nitric oxide (NO), then to nitrous oxide (N_2O), and finally to elemental N (N_2); since both NO and N_2O are powerful greenhouse gases, it is desirable to manage agricultural soils to limit incomplete denitrification in an attempt to reduce greenhouse gas emissions (Coyne, 2008; Sylvia et al., 2005). N use efficiency (NUE) is also reduced during nitrification and denitrification since both of these processes ultimately result in a permanent loss of N from the root zone (Coyne, 2008; Norton, 2008). In addition to soil aeration and the presence of the appropriate microorganisms, nitrification and denitrification rates depend largely on soil NH_4-N/NO_3-N concentrations, temperature, pH, and carbon availability (Sahrawat, 2008; Sylvia et al., 2005).

2.2. Nitrogen deficiencies

Optimum yields and maximum profits will not be achieved if producers do not supply sufficient levels of N to their crop throughout the growing season. Organic compounds in plants such as proteins (enzymes), amino acids, nucleic acids, and nucleotides (among others) are essential to carry out important life processes involved in plant growth and metabolism (Haynes, 1986; Troeh and Thompson, 2005; Vos and MacKerron, 2000). Many organic compounds in plants, especially proteins, are particularly rich in N; without adequate supplies of N to the plant during growth, breakdown and remobilization of protein N may occur, limiting their development (Haynes, 1986; Troeh and Thompson, 2005; Vos and MacKerron, 2000). This causes symptoms of general yellowing of the leaves and overall stunted growth, and if severe enough, will lead to accelerated maturity and reduced yields in agricultural crops (Dean, 1994; Tisdale et al., 1985; Troeh and Thompson, 2005; Vos and MacKerron, 2000).

Following uptake, N is easily transported throughout the plant either in the form of the NO_3^- or amino acids (Haynes, 1986). If a plant is experiencing N deficiency, the N that has already been taken up will translocate from the older growth to the newer growth in an attempt to supply the actively growing regions with N (Bucher and Kossmann, 2007). Because of this, the yellowing of the leaves is most apparent in the older growth; yellowing begins along the veins at the leaf tips, and progresses inward along the leaf midribs (Troeh and Thompson, 2005). To optimize yields and maximize profitability, it is very important to supply plants with an adequate supply of N, especially during critical growth stages (i.e., growth stages when crop N uptake is highest; (Burton, 1989)).

2.3. Excessive nitrogen

The manufacture of synthetic N from the Haber-Bosch process has resulted in relatively cheap and a seemingly limitless source of fertilizer N. This, coupled with the lack of an environmental value for most production land and the surrounding water resources, has enabled crop producers to be very flexible with the quantity of fertilizer N to apply to their crops (Raun and Schepers, 2008); some producers apply above-optimal rates in an attempt to eliminate any chance of N deficiency. If the release of soluble N by microbial activity plus applied N fertilizer exceeds the rate of use by growing plants, excessive available soil N will exist. Nitrogen supply to plants that is above the optimum rate can lead to undesirable increases in the shoot/root ratio that may negatively impact the acquisition of water and other nutrients, especially when nitrate is supplied as the N source (Lynch et al., 2012; Marschner et al., 1986). Furthermore, high N uptake by agricultural crops can cause rapid, dark green vegetative production, which may ultimately result in structural weakness, increased vulnerability to pests, delayed maturity, reduced yields, and/or reduced crop quality (Addiscott et al., 1991; Rosen and Bierman, 2008; Troeh and Thompson, 2005). These negative effects are most pronounced if the crop is not provided with an adequate supply of other essential nutrients that have structural and metabolic functions in plant development (e.g., phosphorus or potassium) (Troeh and Thompson, 2005).

Agriculture is responsible for a vast amount of total nonpoint source N pollution in the United States, primarily because much of the N from fertilizer applications is not taken up by the crop (Carpenter et al., 1998). All soil inorganic N is susceptible to loss via leaching or volatilization if it is not immediately taken up by plants. In addition to these environmental ramifications, inorganic N can also be temporarily lost via immobilization. Any N that escapes the crop rooting zone, or that is not in a plant available form during the time of uptake, is a financial loss to the producer since the lost N cannot provide a yield return from the investment of N as a purchased input (Rosen and Bierman, 2008).

Nitrate-N leaching from agriculture systems has been known to degrade ground and surface water resources, resulting in eutrophication and non-potable water supplies (Vollenweider, 1982). Because of the nitrification process, excess N in agricultural soils tends to accumulate in the form of nitrate (Troeh and Thompson, 2005). Nitrate is soluble in soil and largely moves with soil water, and will therefore likely leach into the groundwater during any rainfall or irrigation event in which drainage occurs (Tisdale et al., 1985). Coarse textured soils with low organic matter are especially vulnerable to nitrate leaching because of their drainage capabilities (Rehm et al., 2008, revised). Experiments on coarse textured soils in MN showed that the quantity of NO_3-N leached in a potato crop increased linearly as the proportion of N applied at plating increased (Errebhi et al., 1998b).

Surface waters can also be contaminated because of nitrate leaching, especially in regions where subsurface tile-drainage systems are used to increase crop productivity in soils where high water tables are common (Mulla and Strock, 2008). Tile-drainage systems are pathways for leached nitrate to be transported to surface waters. In one study, NO_3-N concentrations as high as 10 mg L^{-1} were measured in downstream water bodies while tile drains were flowing, but very low concentrations were measured when tile drains went dry (Fenelon and Moore, 1998). This indicates that the high NO_3-N concentrations in the downstream water bodies were caused by nitrate losses from tile drains. Nitrate losses from tile drains depend on a variety of factors, including:

precipitation, mineralization, cropping system, tillage, and rate and timing of N application (Randall and Mulla, 2001).

There are potential human health risks if nitrate contaminated groundwater is used as a source of drinking water. Infants below the age of six months who consume water containing levels of NO_3-N exceeding the maximum contaminant level (MCL) could develop symptoms of shortness of breath and methemoglobinemia (blue-baby syndrome), which could lead to serious illness and potential death (EPA, 2009). The current MCL for NO_3-N concentrations in drinking water as set by the U.S. Environmental Protection Agency is 10 mg L^{-1} (EPA, 2009). However, of over 10,000 drinking water wells sampled in a nationwide study conducted by the USGS National Water-Quality Assessment Program, nearly 14% exceeded the MCL for NO_3-N (Burkart and Stoner, 2008).

Because of denitrification (and to a smaller extent, nitrification), nitrate can be converted to gaseous forms of N (primarily as N_2O and N_2), which cannot be taken up by plants and are easily volatilized (Tisdale et al., 1985). Atmospheric N_2O concentrations are rising at a rate of over 0.2% yr^{-1}, primarily because of enhanced microbial production in expanding fertilized agricultural lands (Forster and Ramaswamy, 2007). Another, more direct loss of gaseous N is through non-biological volatilization of free ammonia (NH_3) which often occurs in agriculture when urea and NH_3 based fertilizers are applied; the greatest susceptibility of gaseous N loss occurs when urea and NH_3 based fertilizers are applied to high pH, sandy soils (Tisdale et al., 1985).

3. Managing irrigated potato production in Minnesota

3.1. Irrigated potato production

In Minnesota, processing potatoes are typically planted from mid-April to the beginning of May, depending on weather conditions. Russet Burbank, the most popular processing variety in the U.S. and Minnesota, has a mid-season maturity class of 111-120 days after planting (Whitworth et al., 2011). Alpine Russet is a relatively new variety that is being grown in Minnesota; it has a late-season maturity class of 121-130 days after planting, but has similar yields to Russet Burbank, has superior processing quality after long-term storage, and is more resistant to water-stress induced tuber defects (Whitworth et al., 2011).

Shortly after emergence (when the plants are 5-7.5 cm high), a hill is typically formed over the plants (Dean, 1994). Hilling promotes root and tuber development, helps to control weeds, helps to moderate soil moisture from excessive rainfall or irrigation, and protects tubers from sun damage and possible late-season frosts (Scherer et al., 1999). Mechanical weed control can be done efficiently during early growth stages, but to control diseases, weeds, and/or insects throughout the growing season, chemical inputs are necessary (Engel et al., 2012). The major insect pest of the potato is the Colorado potato beetle; in many areas of the Midwest, it has built up resistance to many insecticides. For producers to overcome this resistance, they should rotate between insecticide products that have different Insecticide Resistance Action Committee Group Numbers (Engel et al., 2012). In early- to mid-September, vines of non-processing varieties are typically killed using non-translocated herbicides, followed by mechanical harvest 1-2 weeks later (Dean, 1994). Killing the vines prior to harvest is advantageous because it allows the periderm to develop (decreases susceptibility to scuffing of the skin during harvest and transport), and controls the spread of diseases (Dean, 1994).

There are four major growth stages associated with the development of the potato (Ojala et al., 1990; Wright and Stark, 1990). Stage I includes sprout and early vegetative development from planting to tuber initiation, and lasts between 15 and 30 days; these are sometimes considered to be two separate growth stages (Zebarth and Rosen, 2007). Crop N uptake at the end of this stage is only about 20% of the seasonal total, so high rates of fertilizer N before this stage should be avoided (Zebarth and Rosen, 2007). During stage II (tuber initiation, or "tuberization") tubers are formed at the tips of stolons – this stage lasts about 10-14 days. Although this stage is short in relation to the others, about 50% of the seasonal total N is taken up by the crop during this time (Zebarth and Rosen, 2007). During stage III (tuber enlargement, or "bulking"), the tubers gain most of their mass; this stage can last from 50 to over 120 days, depending on variety, length of growing season, and presence of pathogens. The remainder of N is mostly taken up within the first half of this stage (Zebarth and Rosen, 2007). Stage IV is most easily characterized by the commencement of plant senescence and loss of leaves. During this stage, the tuber skins mature and tuber dry weight increases due to the translocation of nutrients from the shoots and roots to the tubers (Ojala et al., 1990; Wright and Stark, 1990; Zebarth and Rosen, 2007).

Due to coarse textured soils in central MN, it is essential to supply supplemental water to a potato crop throughout the growing season in order to optimize tuber yield and quality. Sufficient levels of soil water are reflective of high plant nutrient uptake rates, which are strongly correlated with high tuber yields (Wright and Stark, 1990). Because even moderately compacted soil will impede root growth, potato plants will often form relatively shallow and inefficient rooting systems in regard to other field and vegetable crops (Bailey, 2000). The roots of irrigated Russet Burbank potatoes grown in a loamy sand soil were found to be restricted to the upper 30 cm, with less than 15% of the roots below this depth (Lesczynski and Tanner, 1976). This shallow root system leads to poor water uptake efficiency (Bailey, 2000). To combat this problem, scheduled irrigation supplements must be applied in order to maintain soil water potential throughout the growing season, so potato plants can consistently take up sufficient levels of water. Potato plants themselves, are very sensitive to water stress (Bailey, 2000; Gregory and Simmonds, 1992; Phene and Sanders, 1976). Leaf stomates will close with only mild soil water deficits; this will decrease potato stomatal conductance to the extent necessary to prevent leaf water potential from falling below critical levels, which could ultimately impede growth and reduce tuber yield (Bailey, 2000; Gregory and Simmonds, 1992; Phene and Sanders, 1976; Wright and Stark, 1990).

Water stress can also affect tuber quality. Although tuber initiation only lasts 10-14 days, introduction of a water deficit during this period generally has the greatest effect on tuber quality over all other stages (Wright and Stark, 1990). Water stress during this period can result in reduced specific gravity and an increased incidence of secondary growth, pointed ends, growth cracks, and knobby tubers, and can also increase susceptibility of tuber bruising and internal diseases (Ojala et al., 1990; Wright and Stark, 1990). Any moisture deficits following tuber initiation can result in tubers with “common scab disease” (Bailey, 2000). Sugar end potatoes, which ultimately result in a very dark appearance of the high sugar tissue of fried potato products at the tuber ends, can also result from moisture deficits (Eldredge et al., 1996; Shock et al., 1992; Shock et al., 1993). Introduction of a water stress during tuber bulking negatively affects tuber quality, although it is not as severe as when a stress is introduced during tuber initiation (Miller and Martin, 1987). Any water stress during this stage can reduce tuber specific gravity because of the tuber bulking period being shortened (Miller and Martin, 1987; Wright and Stark, 1990). Drought tolerance may vary by variety, and the drought tolerance of a specific variety can vary depending on the timing of the water limitation (Vos and Haverkort, 2007).

Excessive irrigation can also negatively affect a potato crop. Excess irrigation has been shown to increase the incidence of brown center and hollow heart (Wright and Stark, 1990), as well as total and marketable tuber yields (Stark et al., 1993). From an environmental perspective, excess irrigation can result in increased drainage and saturated soil conditions, which can result in an increased susceptibility of nitrate leaching and denitrification, respectively. Producers should follow irrigation best management practices to maintain soil moisture throughout the growing season (Shock et al., 2007).

Well-drained, coarse textured soils (soils with high oxygen diffusion rates) with soil matric potentials that are maintained to be nearly constant are ideal for potato production (Phene and Sanders, 1976). Unfortunately, having constant soil moisture over an entire growing season is extremely rare under natural conditions on the coarse textured soils in central MN. To accommodate this, producers rely on irrigation. The sandy loam and loamy sand soils used for potato production in Minnesota are typically low in organic matter and cation exchange capacity (CEC), and therefore, have a relatively low reserve of soil nutrients. The low inherent soil fertility, coupled with the large nutrient requirement for potato production (i.e., nitrogen and phosphorus) results in high fertilizer inputs (Dean, 1994). Potato plants are relatively shallow rooted in regard to other field and vegetable crops; this physiological feature results in below average nutrient uptake and poor NUE, which ultimately leads to higher rates of nitrate leaching (Lynch et al., 2012; Rosen and Bierman, 2008; Zvomuya et al., 2003).

There is a high susceptibility for N losses (especially via nitrate leaching) in irrigated potato production, primarily because of unpredictable rainfall and the interaction among the factors listed in the previous paragraph (i.e., well-drained, low CEC soils, the use of irrigation, and the high nutrient requirement and low NUE of potatoes). Based on previous experiments on coarse textured soils in Minnesota, only about a third to a half of applied N is recovered in years of moderate to heavy leaching (Errebhi et al., 1998b; Waddell et al., 2000). Groundwater nitrate concentrations have been found to be significantly higher in irrigated than non-irrigated cropland (Burkart and Stoner, 2008). Out of 370 water samples from Minnesota drinking water wells, 6% were found to exceed the 10 mg L^{-1} MCL for NO_3-N (Lewandowski et al., 2008).

3.2. Minimizing nitrogen losses

The goal of N management in agriculture is to provide the crop with a sufficient quantity of available N throughout the growing season to optimize yields, while minimizing the quantity of N lost from the root zone. This is a very difficult task to achieve because there are usually many unknown factors involved. It is especially difficult for irrigated potato because of the increased susceptibility of nitrate losses compared to other crops (as described in previous sections).

There are many strategies that can be used to manage fertilizer N in attempting to optimize potato plant uptake while minimizing losses. The first focus for producers is N fertilizer application rate. Assuming there will be no loss throughout the season, the optimum rate depends on yield goal (which varies depending on variety, harvest date, and general management practices), N present in irrigation water, and residual soil N that is available or will become available during the course of the growing season (Meisinger et al., 2008; Rosen and Bierman, 2008). The problem with relying solely on N rate is that it does not help to minimize losses; any N lost from the system is N that cannot be taken up by the crop, and will therefore likely be the cause of a plant N deficiency. Some producers may apply above-optimum rates as a way to account for N that may be lost; although this may help to maximize crop uptake, it does not help to minimize N losses.

A second N fertilizer management strategy intended to increase fertilizer NUE/minimize losses is application timing (i.e., split N fertilizer applications). This strategy is used in conjunction with application rate. Matching the timing and rate of fertilizer N with the N needs of the crop during different growth stages will help to optimize crop uptake while minimizing N losses (Canter, 1997; Errebhi et al., 1998b). Increased NUE and yields have been shown to result from split applications of N fertilizer during tuber initiation and bulking (Errebhi et al., 1998b; Westermann et al., 1988). The producer must be sure to apply the full N rate in a timely manner, however. Excessive late-season N uptake can promote late-season vegetative growth, resulting in poorer net development, delayed tuber maturity, and poor skin quality (Ojala et al., 1990). Late season N is also less likely to be taken up by the crop, making it vulnerable to nitrate leaching (Rosen and Bierman, 2008; Vos, 1999). Split applications can be adjusted over a period of at least 60 days after emergence (Vos, 1999), and the majority of total N should be applied before tuber bulking (Ojala et al., 1990).

Controlled release N fertilizers and nitrification inhibitors are intended to provide the crop with a supply of available soil N at a rate that matches the physiological demands of the crop. Controlled release N fertilizers typically consist of urea coated with a polymer. The purpose of the polymer coating is to meter the supply of N to better match N availability to the needs of the crop (release is temperature dependent) (Randall et al., 2008). Experiments in Minnesota showed that tuber yields and NO_3-N leaching obtained with controlled release fertilizers were similar to those obtained with split N applications (Wilson et al., 2009; Wilson et al., 2010). Nitrification inhibitors are chemicals that are nontoxic to plants, but are toxic to nitrifying bacteria (*Nitrosomonas*). When added to the soil, the nitrification inhibitor temporarily blocks or slows the nitrification process by inhibiting the growth or activity of nitrifying bacteria so they cannot convert NH_4^+ to NO_3^- (Randall et al., 2008; Tisdale et al., 1985). A recent experiment where nitrification inhibitors were used with potato showed that success was dependent on the form of N fertilizer – yields were reduced when the inhibitor was used with a pure ammonium fertilizer, but yields were found to increase in some cases with a mixed ammonium/nitrate fertilizer (Kelling et al., 2011).

Best management practices for irrigated potato on coarse textured soils in Minnesota recommend a minimum of three split N applications if fertigation not is available; applications should occur at planting, emergence, and post-emergence (Rosen and Bierman, 2008). To more precisely meet the needs of the crop, *post-emergence* applications can be split; this can be done very easy if fertigation is available. The challenge here lies in the ability of the producer to estimate the appropriate rates and timing of split N applications so that fertilizer N best matches crop demands. This is because crop uptake rate and soil N transformations/losses depend on the interaction of many complicated (and sometimes unpredictable) factors throughout the growing season, including: fertilizer source, soil fertility, soil type/CEC, and weather conditions.

4. In-season monitoring of crop nitrogen status

A logical strategy for determining the exact rate and timing of post-emergence N applications is to make adjustments based on in-season soil or plant monitoring. This approach uses soil or plant measurements after the growing season has begun, so it is able to account for the effect of seasonal weather conditions on crop N availability (Meisinger et al., 2008). This section will evaluate several strategies that can be used to monitor in-season soil or crop N status. When evaluating strategies used to prescribe rates and timing of in-season N, the prediction accuracy is of primary concern; in addition to accuracy, however, other aspects such as precision, sensitivity and specificity to N (defined by Goffart et al. [2008]), feasibility (i.e., ease, expense, and time lag between sampling and obtaining results), and spatial scale should also be considered.

4.1. Soil-based monitoring

Soil sampling is perhaps the most accurate strategy to determine plant available N since it is a direct analysis of NH_4-N and NO_3-N in the soil (Meisinger et al., 2008). The soil NO_3-N content is a good representation of the net balance between nitrate production processes and nitrate loss processes (Meisinger et al., 2008). Only 1-3% of the total N in agricultural soils is present in the available form; much of the remainder is present in the organic form and is only available to the crop following mineralization (Goh and Haynes, 1986). Mineralized N is very difficult to predict since it depends on the interaction of many environmental factors (Goh and Haynes, 1986). Furthermore, coarse textured soils have a high vulnerability to nitrate leaching, so in order to accurately prescribe accurate rates and timing of post-emergence N applications, soil samples should be collected weekly (Dean, 1994). Subsamples should be collected to a depth of 30 cm (depth of the root zone) from representative areas in the field (Dean, 1994).

Spatial variability of soil fertility, especially NO_3-N, is recognized as a major problem with the soil analysis approach (Goh and Haynes, 1986). To account for this, it would be ideal to analyze subsamples separately in order to determine the spatial variability (Dean, 1994); however, this can be very costly and can prolong the time before soil results are received. Overall, soil sampling is accurate, but it is time consuming, expensive, and does not precisely account for within-field spatial variability. Since soil testing is a good method to get an initial approximation of overall soil fertility, perhaps the most useful approach would be to implement a combination of soil testing and a plant-based approach for within-season N monitoring (Raun and Schepers, 2008).

4.2. Plant-based monitoring

In many cases, it may be a more feasible approach to base post-emergence N applications on in-season *plant* measurements. Plant measurements can be a good indicator of crop N status, as long as they are able to detect N deficiency at its earliest stages – this requires a precise, time efficient approach (Gianquinto and Bona, 2000). Before discussion of the approaches, the mobilization and storage processes of N following plant uptake will be introduced in an attempt to explain why plant-based analysis can be an accurate basis for prescribing post-emergence N fertilizer.

Immediately following ammonium absorption by plant roots, it is assimilated to amino acids and either remains in the roots to be stored, or is transported toward the shoots; only small amounts of ammonium are translocated to the xylem (Haynes, 1986). Once nitrate is absorbed, it may be either reduced and synthesized into amino acids while being stored in the root tissue, or may be transported across the root and deposited in the xylem for movement toward the shoots (it may not necessarily change forms; (Haynes, 1986)). Portions of both nitrate and amino acids can be transported to the stem and petiole cells; the bulk of this is transported further into the leaflets where it is mostly reduced to ammonium and stored (Haynes, 1986). In other words, the stems and petioles are the plant organs that transport N that has been recently taken up, and the leaflets are the plant organs that store N. This means the leaflets are a good indicator of *current* plant N status, whereas the stems and petioles are good indicators of *future* plant N status.

There are many plant-based approaches for detecting the N status in a potato crop. Some are based primarily on measurements and analysis from the leaflets, while others are based primarily on measurements and analysis from the petioles. Some approaches are well-developed and are currently being implemented commercially, while others require further research before they can be efficiently used at a large scale. The remainder of this section will focus on the discussion of several invasive and noninvasive approaches for estimating in-season crop N status. Invasive approaches result in the destruction of plant structure before measurement, whereas noninvasive approaches do not (Haase et al., 2000).

4.2.1. Leaf tissue nutrient concentration

Since it is a direct measurement, N determination by dry matter leaf tissue is perhaps the most accurate strategy for determining plant N status. There are several well documented, accepted procedures for this approach, each of which may be sensitive to different forms of N (Haase et al., 2000). Some procedures are described in detail by Haase et al. (2000) and include digestion (measures organic N and some forms of inorganic N, depending on acid and catalyst used), combustion (measures total N), spectroscopy (measures organic and inorganic N), and chromatography (measures NO_3-N). Conductimetric procedures (measures NO_3-N and NH_4-N), such as those described by Carlson et al. (1990) are also widely accepted. All of these procedures are carried out in a laboratory setting, so leaf tissue samples must be collected prior to analysis. The position of the leaf on the plant can influence nutritional diagnosis, so it is important to remain consistent when sampling (Gianquinto and Bona, 2000). Some procedures require samples to be dried and then ground finely (<1 mm) to allow intimate mixing prior to analysis.

The current best management practice for potato production in Minnesota is to base the rate and timing of post-emergence N fertilizer applications on petiole NO_3-N concentrations determined from tissue dry matter (Rosen and Bierman, 2008); this approach is currently being implemented by many commercial growers. It is based on the concept that the petiole is the transport organ of N (in the form of NO_3^- and amino acids) from the roots to the leaflets, and is a good indicator or NO_3 uptake by the plant. Wu et al. (2007) found that petiole NO_3-N concentrations were closely reflected in N treatment variations created by split applied N. Overall, N determination by dry matter leaf tissue provides very accurate results, and the interpretations are straightforward (Haase et al., 2000). However, this approach is generally expensive (depending on procedure used), time consuming, and requires trained operators and specialized equipment (Haase et al., 2000). Also, this approach does not account for spatial variability.

4.2.2. Petiole sap nitrate concentration

The petiole sap nitrate concentration (PSNC) is a method to quickly and directly measure petiole sap nitrate concentration in the field. Petiole samples must be collected from a consistent position on the plants as NO_3-N concentration in petiole sap increases with depth into the canopy (MacKerron et al., 1995). Following collection of leaf petiole samples, PSNC is measured using one of two types of procedures – one uses a nitrate specific electrode, the other uses nitrate test strips followed by a reflectometer reading (Goffart et al., 2008). PSNC has been found to be significantly related to dry petiole NO_3-N concentration and plant N uptake (Errebhi et al., 1998a; MacKerron et al., 1995), and increases with increasing fertilizer supply (MacKerron et al., 1995). High PSNC variability has been reported relative to N uptake (MacKerron et al., 1995; Westcott et al., 1993), and caution should be made when evaluating crop N status using this approach.

Although this approach is quick, easy, and cheap, reports have suggested that the change in PSNC over the growing season depends on external factors other than N (e.g., soil conditions or weather) (Goffart et al., 2008; MacKerron et al., 1995). Also, this approach does not account for spatial variability.

4.2.3. Relative chlorophyll

The SPAD-502 chlorophyll meter (Minolta Camera Co., Ltd, Osaka, Japan) is a widely used instrument for detecting relative chlorophyll. Its output is based on measurements of light transmittance at two wavelengths: 650 nm and 940 nm. The 650 nm wavelength is in a region of peak chlorophyll absorbance, and the 940 nm wavelength is in a region of very low chlorophyll absorbance and serves as a reference to compensate for leaf thickness and moisture content (Gianquinto et al., 2004; Schröder et al., 2000). The measured values at each wavelength are converted to digital signals, and then output as a ratio (i.e., chlorophyll reading). This chlorophyll reading provides a good indication of the relative amount of chlorophyll present in the plant leaves (Gianquinto et al., 2004; Vos and Bom, 1993). Most plant N is stored in chloroplasts and chlorophyll proteins in plant leaves, so leaf chlorophyll is closely related to plant N (Gates et al., 1965; Vos and Bom, 1993; Wood et al., 1992); therefore, relative chlorophyll measurements can be used to indirectly determine crop N status. For a potato crop, Wu et al. (2007) found that relative chlorophyll values were closely related to N treatment variations created by split applied N.

Chlorophyll meters should be used with caution, however. Since its output is a relative measurement, chlorophyll readings can vary depending on soil type, light intensity, crop growth stage, and variety (Gianquinto et al., 2004). Also, environmental or stress factors (e.g., water content, nutrient deficiency besides N, and pests/diseases) can confound output values (Gianquinto et al., 2004; Ladha et al., 2005; Schepers et al., 1996), so normalization to a non-limiting N reference area within the same field is required for each use (Blackmer et al., 1993; Ladha et al., 2005; Schepers et al., 1992). Chlorophyll readings have a poor response to N fertilizer rate throughout the growing season as compared to PSNC measurements, especially during early growth stages (Gianquinto et al., 2004). In general, chlorophyll readings for agricultural crops reach a plateau at moderate N supply (Tremblay et al., 2011); this is because a very small amount of excess

N is actually converted into chlorophyll under conditions of excessive N uptake, which results in chlorophyll readings being poor predictors of N sufficiency under these conditions (Schröder et al., 2000; Wood et al., 1992).

Overall, chlorophyll readings can be a good way to determine crop N status. The initial cost of the meter can be moderately high, but operating costs are inexpensive. This is a noninvasive approach, so measurements are easy and can be made relatively quickly (certainly quicker than measurements of PSNC); this allows for same-day fertilizer recommendations (Gianquinto et al., 2004). A major limitation to this approach is its poor sensitivity and/or specificity to N under certain circumstances. The user must be aware of the potential variability, and must have knowledge of its calibration requirements. Furthermore, this approach does not account for spatial variability, unless many individual measurements are taken. Even if many measurements are taken, it is still difficult to make objective N fertilizer decisions for variable rate application, and it is even more difficult to implement.

4.2.4. Remote sensing of the crop canopy

4.2.4.1. Overview of remote sensing

The most sophisticated approach for monitoring crop N status is through the use of remote sensing. Remote sensing is most simply defined as the acquisition of information about an object without making physical contact (Rees, 2001). This can be done quantitatively by the use of passive and active sensors: passive sensors detect naturally occurring radiation, and active sensors emit their own source of radiation (Rees, 2001). Specialization of sensors allows for the acquisition of information over a range of spectral wavelengths – due to the absorption of radiation by the earth’s atmosphere at some wavelengths, useful wavelengths are limited to the visible and infrared parts of the electromagnetic spectrum (from about 300 nm to 12,000 nm; (Rees, 2001)). Spectral reflectance (i.e., reflectance as a function of wavelength) is a measurement that can be acquired about a target object from these sensors (Rees, 2001). Measurements of reflectance are output as the ratio between energy reflected by the target and total energy incident on the target (Hansen and Schjoerring, 2003). Crop canopy spectral reflectance measurements can provide the user with useful information about structural and physiological characteristics of the crop (Peñuelas et al., 1994; Scotford and Miller, 2005), including relative measures of leaf chlorophyll content. Chlorophyll content has an influence on crop canopy reflectance, specifically in the visible and NIR wavelengths of the spectrum (Diker and Bausch, 2003; Jongschaap and Booij, 2004; Solari et al., 2008). Because leaf chlorophyll content is also closely related to plant N status (Gates et al., 1965; Vos and Bom, 1993; Wood et al., 1992), canopy reflectance measurements can be useful for monitoring crop N status. Scotford and Miller (2005) provide additional information behind the theory of spectral reflectance techniques.

4.2.4.2. Comparison to other approaches

Similar to the approaches previously discussed, the use of remote sensing to monitor in-season crop N status can provide the producer with information to make accurate decisions regarding the optimum rate and timing of post-emergence N fertilizer applications. However, it is not practical to collect spatial data using the other approaches, and using them for whole-field management is problematic. A major advantage of using remote sensing over the other approaches is that remote sensing data can be associated with spatial data much easier, which gives it the ability to account for within-field spatial variability. A primary reason for this is that physical contact is not required, so it is relatively easy to take measurements over large areas (e.g., an entire field) in a short amount of time, and in some cases, instantaneously. The methods by which spatial data are acquired vary depending on the type of sensor and the type of sensing platform as discussed in sections below.

The recent improvement in geographic information systems (GIS), global positioning systems (GPS), and variable-rate N applicators for precision agriculture has made it possible for producers to apply N at sub-meter accuracy (Meisinger et al., 2008). These tools, together with spatial information acquired from remotely sensed measurements, allow for the optimum *placement* of post-emergence N fertilizer. Optimum placement is an important consideration when attempting to account for the within-field spatial variability in precision agriculture.

Currently, the translation of information collected via remote sensing to N fertilizer recommendations has been a primary limiting factor for its use commercially. Calibration of measurements with plant N status in the attempt to objectively prescribe a recommended N fertilizer rate has been difficult because of wide variations in leaf chlorophyll estimation due to external factors other than plant N concentration (Samborski et al., 2009). Currently, feasibility is another limiting factor. Satellite and aerial data are very useful because they contain spatial data; however, data acquisition is not as flexible for satellite and aerial sensors compared to other approaches, and data quality is largely dependent on weather conditions. Also, the acquisition of image data is costly, and the time lag before results are obtained can be on the order of days to weeks (Deguise et al., 1998). However, because of the high spatial and spectral capabilities of image data, it is much more useful from a research perspective. Equipment-mounted sensors can be configured to process data in real-time in order to make immediate N fertilizer recommendations. For irrigated potato crops, there is opportunity to mount sensors to center-pivot irrigation equipment to do just this. Perhaps the development and fine-tuning of spectral data processing techniques for specific crop and temporal conditions may make this the most feasible approach for commercial application in the future.

4.2.4.3. Types of sensors

There are three main types of sensors (i.e., radiometers, spectrometers, and digital imagers), and three different sensing platforms (i.e., ground, aerial, and space) in which spectral reflectance measurements are typically obtained for agricultural purposes – each is explained in some detail by Scotford and Miller (2005). Radiometers and spectrometers typically have ground-based platforms since they are only able to obtain point measurements; because sampling area depends on the height above the target area, it would not be useful to take point measurements from aerial or satellite platforms. Alternatively, it is common for digital imaging sensors to be used with any of the three sensing platforms, but especially from aerial and satellite platforms. Spectral imagery contains two spatial dimensions (*x*- and *y*-axes), with a third dimension (*z*-axis) holding the spectral data for each pixel (Lucas et al., 2004), whereas radiometers and spectrometers only obtain direct measurements of spectral data; the use of GPS and GIS does, however, make it possible to associate spatial data to the spectral data obtained from radiometers or spectrometers. Radiometers and spectrometers can also be mounted to field equipment (e.g., tractors, applicators, irrigation sprinklers) to provide real-time measurements in order to account for spatial variability (Lammel et al., 2002). Whether spectral data are acquired from ground-based, aerial, or satellite sensing platforms, quantitative techniques can be applied in order to estimate the N status of the crop (Nellis et al., 2009).

As described by Scotford and Miller (2005), radiometers can only obtain spectral data from one wavelength; however, they can use either natural or artificial light as their illumination source (defined as a passive or active sensor, respectively), whereas spectrometers and imaging sensors must rely on natural light as an illumination source. Since passive sensors use naturally occurring radiation as their energy source, less than perfect atmospheric and environmental conditions can negatively impact data quality. Spectrometers and imaging sensors are much more robust in their ability to obtain spectral data, however. Depending on their configuration, they can obtain multispectral or hyperspectral data. *Multispectral* sensors gather data in a few discrete wavelength intervals (i.e., bands), usually with relatively broad bandwidths (often termed “broadband” data); alternatively, *hyperspectral* sensors gather data in many narrow contiguous wavelength intervals (often termed “narrowband” data (Varshney and Arora, 2004)). The bandwidth, spectral resolution, and spatial resolution (for imagery) of the data can vary, depending on the sensor configuration. To some extent, multispectral and hyperspectral data can be analyzed using similar processing techniques; however, there are a tremendous amount of opportunities for the interpretation and analysis of hyperspectral data that do not exist with multispectral data, especially for identifying and modeling plant biophysical parameters such as N stress (Aspinall et al., 2002).

4.2.4.4. Spectral reflectance measurements for monitoring crop nitrogen status

The use of spectral reflectance data for monitoring crop N status is effective primarily because the spectral characteristics of green-leaved vegetation change as leaf chlorophyll content changes. As leaf chlorophyll increases, leaf reflectance generally decreases in the visible region (i.e., 400-670 nm) and increases in the near-infrared (NIR) wavelengths [i.e., ~670-1000 nm; (Goffart et al., 2008; Hansen and Schjoerring, 2003; Rees, 2001)]. Reflectance is low in the red wavelengths as a consequence of absorption by chlorophyll, and high in the near-infrared wavelengths as a result of scattering in the mesophyll, and thus, reflectance in these parts of the spectrum are strongly correlated with the amount of photosynthetically active radiation (PAR; (Rees, 2001)). In addition to the optical properties of leaves, N deficiencies affect canopy architecture and leaf area (Moran et al., 2004), which can also affect the spectral characteristics of a crop at the canopy level.

Leaf area index (LAI) is used as an indicator of overall crop growth and productivity, which is strongly related to leaf chlorophyll content and crop N status (Moran et al., 1995; Ray et al., 2006). LAI has a direct relationship to the canopy reflectance of a crop (Haboudane et al., 2004; Ray et al., 2006; Wang and Rosen, 2004) and the development of different remote sensing techniques to improve its estimation over large areas has been of interest (Haboudane et al., 2004). It is important to point out, however, that spectral measurements at the canopy level are influenced by the combined response of variations in environmental properties other than LAI, such as leaf chlorophyll content, canopy shadows, background soil reflectance, and solar zenith angle (Haboudane et al., 2002). Therefore, for canopy level spectral reflectance data to be a good indicator of LAI, it should be relatively insensitive to effects of all of these environmental properties except LAI (Haboudane et al., 2004). Similarly, if leaf chlorophyll is of primary interest, spectral data should be insensitive to all properties except leaf chlorophyll. Haboudane et al. (2002) developed a spectral index that takes the ratio of an index which is sensitive to chlorophyll concentration (TCARI; Haboudane et al., 2002) to one that minimizes soil background effects (OSAVI; (Rondeaux et al., 1996)). There are many other published indices (original and revised) that were developed for similar reasoning (Daughtry et al., 2000; Haboudane et al., 2004; Haboudane et al., 2008; Qi et al., 1994; Wu et al., 2008).

There are hundreds of possible methods for predicting crop biophysical properties using spectral reflectance data. A combination of spectral transformations (e.g., indices, derivatives, continuum removal, band depth, etc.) and statistical techniques (e.g., ordinary least square regression, multiple linear regression, partial least squares regression, artificial neural networks, etc.) can be applied to spectral data to quantitatively compare crop N stress in field studies (Stroppiana et al., 2012). Many of these techniques have been tested on numerous field and vegetable crops, some of which are summarized below. There are many published reports that describe the techniques and discuss their usefulness for predicting N status under the specific conditions tested. This is an ever-emerging area of research as most studies have been published within the last 15-20 years.

4.2.4.4.1. Regression analysis

One of the most basic technique to determine the usefulness of reflectance measurements as a predictor of another plant measurement (i.e., N concentration of dry matter leaf tissue) is to correlate the reflectance spectra to the plant measurement. The coefficient of determination (r^2) is a quantitative way to assess the relationship between the reflectance and the plant measurement. Miao et al. (2009) used linear regression to compare the relationship between canopy reflectance and chlorophyll meter readings from a corn crop. They found that 73-88% of chlorophyll meter variability could be explained by reflectance from the best performing single narrow band. Cohen et al. (2010) found that reflectance data were better correlated with leaf N concentration than petiole NO_3-N concentration.

4.2.4.4.2. Spectral indices

The combination of data from two or more spectral bands by ratioing, differencing, ratioing differences, or applying other algebraic functions to form what are commonly referred to as spectral indices, is a simple technique commonly used for prediction of biophysical properties (Jackson and Huete, 1991). In general, the aim of spectral indices is to enhance the signal of vegetation, while minimizing the effects of solar irradiance and soil background (Jackson and Huete, 1991; Rees, 2001). There are hundreds of published spectral indices, and each was developed for testing a particular biophysical parameter of interest (e.g., chlorophyll, LAI, N content, soil background). Hatfield et al. (2004) summarized the development of indices and discussed their application to crop canopies; Le Maire et al. (2004) summarized a comprehensive set of broadband and narrowband indices that can be effectively used for detecting chlorophyll at different scales. R^2 values can be determined to quantitatively asses the relationship between a specific index and a plant biophysical parameter; this is a useful technique to assess the ability of that index for determination of crop N status.

4.2.4.4.3. Reflectance derivative

The reflectance derivative is a type of band transformation that is most useful when applied to hyperspectral data; it can be analyzed by individual band or by the shape of the reflectance derivative curve as a function of wavelength (Filella and Peñuelas, 1994). When plotted as a function of wavelength, the peak of the reflectance derivative curve (which occurs at the red-edge inflection point for green vegetation) shifts to longer wavelengths and increases in amplitude with higher leaf chlorophyll content/N concentration (Filella and Peñuelas, 1994; Le Maire et al., 2004). Derivative reflectance (particularly the second derivative) has also been shown to be useful in reducing the effect of the soil background (Ray et al., 2006).

4.2.4.4.4. Classification

Another technique for determination of crop N status is through image classification. The ENVI software (Exelis, Inc., USA) provides many supervised and unsupervised classification techniques, as well as more sophisticated classification algorithms (e.g., spectral angle mapper [SAM], linear spectral unmixing [LSU], among others). Goel et al. (2003) investigated the effectiveness of *maximum likelihood, minimum distance, Mahalanobis distance, parallelepiped, and binary coding* supervised classification techniques, as well as SAM and LSU for detecting N status and weed infestations in corn. Based on an accuracy assessment, very poor results were achieved when all combinations of the three N levels and three weed control measures were classified at once. When only N was considered, classification accuracy improved dramatically; of all classification techniques, SAM performed the best (66% accuracy). Classification of spectral data into predefined classes is a good way to evaluate their use for variable-rate application decisions at a commercial scale (Cohen et al., 2010).

4.2.4.4.5. Predictive models using data compression

Some techniques, such as partial least squares regression (PLS) or principle component regression, compress hyperspectral data by reducing the large number of measured collinear spectral variables to a few non-correlated principal components (Hansen and Schjoerring, 2003). These techniques are able to account for the majority of the variability that exists in a hyperspectral dataset using only a fraction of the information provided from the dataset, and they are based on the statistical analysis of wavelengths from a wide spectrum, in contrast to simple regression for raw spectral data or indices (Cohen et al., 2010). Ehsani et al. (1999) briefly review the steps involved in the PLS algorithm. Alchanatis et al. (2005) were able to predict N status at the leaf scale in corn from ground-based imagery using PLS models constructed from the first derivative of the spectral data. They found that prediction of N status was much better during a later growth stage (i.e., VT). Cohen et al. (2010) used similar methods for predicting N status in potato leaf tissue (petiole NO_3-N and leaf total N). The PLS model resulted in R^2 values of 0.82 and 0.95 for petiole NO_3-N and leaf N concentration, respectively.

5. Conclusions

Potato yield and quality in Minnesota are highly dependent on N fertilizer and supplemental water via irrigation. Limited or excess quantities of either N fertilizer or water to a potato crop can result in reduced tuber yield and/or quality. Furthermore, excess crop available N and/or water poses an increased vulnerability to nitrate leaching, especially in the coarse textured soils typically used for potato production in Minnesota. Data obtained via remote sensing can be effectively used to monitor in-season plant nitrogen status. This information can be used to adjust rates and timing of within-season N fertilizer applications to ultimately maintain production while minimizing losses.

However, there are a number of factors that limit this approach from being used commercially for monitoring in-season plant status for potato production. First, spectral data processing techniques can be time consuming, and there is some uncertainty in the ability to accurately detect deficiencies for supplemental N to be applied in a timely manner, especially while accounting for within-field spatial variability. Little is known about the effect of crop growth stage or potato variety on the ability of spectral data to be used to adjust in-season applications. A better understanding of the response of spectral data and processing techniques, especially their behavior under varying conditions (i.e., growth stage, variety, yield potential), can improve the feasibility of this approach to allow its use at a commercial scale.

Spectral imagery is a very powerful research tool because it contains spatial data in addition to spectral data. Perhaps the role of spectral imagery in agriculture should be to be used as a tool to *discover* and *develop* optimum spectral processing techniques for monitoring in-season plant status. Once appropriate indices and processing techniques are developed, automated, equipment mounted sensors could implement these “best” techniques, so in-season N monitoring can be more feasible for producers on a commercial scale.

6. References

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Chapter 2.

IRRIGATION AND NITROGEN MANAGEMENT EFFECTS ON POTATO NITROGEN USE INDICES AND TUBER YIELD AND QUALITY

Chapter Summary

In order for a potato (*Solanum tuberosum* L.) variety to be economically viable for processing, it must produce high yields and have good quality attributes. Later maturing processing varieties require high rates of nitrogen (N) fertilizer to optimize yields, and this, coupled with the high risk of nitrate-nitrogen (NO_3-N) leaching in irrigated potato production, makes it both economically and environmentally important to have varieties that possess good N use efficiencies. A two year field study was conducted on a loamy sand soil to evaluate the effects of variety, N treatment, and water stress on: (i) tuber yield and quality characteristics, (ii) N use indices, and (iii) residual soil NO_3-N levels. The study included two irrigation regimes (unstressed and stressed), five N treatments categorized by three N rates (34 kg N ha^{-1}, 180 kg N ha^{-1}, and 270 kg N ha^{-1}) in which the 270 kg N ha^{-1} rate had post-emergence N either split applied or applied early in the season (also, the 270 kg N ha^{-1} rate with post-emergence N split applied included a soil surfactant application as one of its treatments), and two potato varieties (Russet Burbank and Alpine Russet). The use of the soil surfactant had a minimal effect on the parameters measured in this experiment. Insufficient supplemental water during critical growth stages was found to negatively affect tuber yield, tuber quality, and plant N uptake, but surprisingly had a positive overall effect on chip color. Under the conditions of this study, water stress did not significantly affect nitrogen use efficiency (NUE; plant dry matter per unit available N supply), N utilization efficiency (NUtE; plant dry matter per plant N uptake), or N uptake efficiency (NUpE; plant N uptake per unit available N supply), even though it did have a negative effect on plant N uptake and plant dry matter (DM) production. These N use indices, as well as N harvest index (NHI; tuber N uptake per plant N uptake), decreased with increasing N rate. For all N treatments except the 34 N early treatment, NUE and NUpE values were less than 50 g g^{-1} and 50%, respectively. This indicates a relatively low efficiency of N use during 2010 and 2011 and is attributed to heavy leaching events during these years. Within the same N rates, NUpE was higher when post-emergence N was split applied, which is likely the result of less N being lost due to leaching. NUtE and NHI were higher for the Russet Burbank variety, whereas NUpE was higher for the Alpine Russet variety. Alpine Russet tuber yields were comparable to or greater than Russet Burbank, but had a specific gravity that was slightly lower than Russet Burbank. Overall, neither variety was found to be better than the other with regard to reducing sugar concentrations and frying quality measurements. Because of its high yields, higher proportion of tubers >170g, higher N uptake, and similar processing qualities compared with Russet Burbank, the Alpine Russet variety is attractive from both an economic and environmental perspective.

Keywords: Nitrogen uptake, nitrogen use efficiency, water stress, reducing sugars, fry quality, Russet Burbank, Alpine Russet

1. Introduction

Potato (*Solanum tuberosum* L.) yield and quality are highly dependent on an adequate supply of nitrogen (N) and water. However, high rates of N fertilizer and rainfall/irrigation on coarse-textured soils have been shown to increase the risk of N loss from the root zone (Errebhi et al., 1998). This results in an economic loss to the grower and can degrade groundwater quality via elevated groundwater nitrate concentrations. Based on previous experiments on coarse-textured soils in Minnesota, only about a third to a half of applied N is recovered in years of moderate to heavy leaching (Errebhi et al., 1998; Waddell et al., 2000). Plant N uptake in the presence of an abundant supply of N (i.e., N uptake capacity) can vary by potato variety (Zebarth et al., 2004). Increased plant N uptake over the growing season can reduce the quantity of N lost from the root zone, thereby minimizing financial losses. Therefore, from both an economic and environmental perspective, it is desirable to grow potato varieties that have the ability to optimize the use of added N (Errebhi et al., 1999).

Potato plants are very sensitive to water stress (Bailey, 2000; Gregory and Simmonds, 1992; Phene and Sanders, 1976). Leaf stomates will close with only mild soil water deficits; this will decrease potato stomatal conductance to the extent necessary to prevent leaf water potential from falling below critical levels, which could ultimately impede growth and reduce tuber yield (Bailey, 2000; Gregory and Simmonds, 1992; Phene and Sanders, 1976; Wright and Stark, 1990). Water stress can also affect tuber quality. The introduction of a water deficit during tuber initiation generally has the greatest effect on tuber quality over all other stages (Wright and Stark, 1990). Water stress during this period can result in reduced specific gravity and an increased incidence of secondary growth, pointed ends, growth cracks, and knobby tubers, and can also increase susceptibility of tuber bruising and internal diseases (Ojala et al., 1990; Wright and Stark, 1990). Any moisture deficits following tuber initiation can result in tubers with “common scab disease” (Bailey, 2000). High sugar concentrations in potato ends, which ultimately result in a very dark appearance of the high sugar tissue of fried potato products at the tuber ends, can also result from moisture deficits (Eldredge et al., 1996; Shock et al., 1992; Shock et al., 1993). Introduction of a water stress during tuber bulking negatively affects tuber quality, although it is not as severe as when a stress is introduced during tuber initiation (Miller and Martin, 1987). Specifically, water stress during tuber bulking can shorten the tuber bulking period, which can ultimately reduce tuber specific gravity (Miller and Martin, 1987; Wright and Stark, 1990). Drought tolerance may vary by variety, and the drought tolerance of a specific variety can vary depending on the timing of the water limitation (Vos and Haverkort, 2007).

Excessive irrigation can also negatively affect a potato crop. Excess irrigation has been shown to increase the incidence of brown center and hollow heart (Wright and Stark, 1990), as well as total and marketable tuber yields (Stark et al., 1993). From an environmental perspective, excess irrigation can result in increased drainage and saturated soil conditions, which can increase the potential for nitrate leaching and denitrification, respectively.

In order for a potato variety to be economically viable, it must produce high yields, but must also have good tuber quality characteristics. Potato chip color after frying is a common index of chipping quality, and is important for processing varieties such as Russet Burbank and Alpine Russet. A light brown chip color is preferred by consumers (Smith, 1955). Sugars are critical in determining the processing quality of potatoes because frying at high temperatures results in a typical Maillard reaction between the reducing sugars (e.g., glucose and fructose) and free amino acids present in the tuber (Marquez and Anon, 1986; Shallenberger et al., 1959). Completion of the Maillard reaction results in a dark brown potato chip color (Gray and Hughes, 1978); the reducing sugars are the limiting factor in this reaction (Marquez & Anon, 1986), so the concentration of reducing sugars can be a good indicator of how well a potato crop is suited for chipping (Sowokinos, 1973). However, in some situations (especially in high N potatoes), fry color may differ for similar sugar concentrations (Roe et al., 1990). Roe et al. (1990) concluded that amino acids may play a synergistic role in the Maillard reaction for high N potatoes.

Sucrose concentration actually has very little direct contribution to the Maillard reaction, but sucrose concentration at harvest can be a good indicator of the resulting sugar accumulation (Sowokinos, 1973). Sowokinos (1973) found that poor processing potato varieties contained sucrose concentrations two to three times as high as acceptable varieties. Increasing N fertilizer rates have been shown to decrease the concentration of reducing sugars in tubers (Hughes, 1986; Roe et al., 1990).

Alpine Russet is a later maturing processing variety that has similar yields and superior processing quality after storage to Russet Burbank (Whitworth et al., 2011). However, since it is a relatively new variety, research comparing the effects of N and irrigation stress on N uptake and tuber frying quality after storage is limited, particularly under Midwest growing conditions. Therefore, the objective of this study was to evaluate the effects of variety (Russet Burbank and Alpine Russet), N treatment, and water stress on: (i) tuber yield and quality characteristics, (ii) N use indices, (iii) residual inorganic soil N levels, and (iv) reducing sugar concentrations and how they relate to potato chip color after frying. This research is part of a larger study to use hyperspectral and thermal imagery to detect nitrogen and water stress in potato (Nigon et al., 2012).

2. Materials and methods

2.1. Study site

Field experiments were conducted over two years (2010-2011) at the University of Minnesota Sand Plain Research Farm (45°23’N, 95°53’W) near Becker, MN. The soil at this location is classified as an excessively drained Hubbard loamy sand (sandy, mixed, frigid Typic Hapludoll) comprised of 82% sand, 10% silt, and 8% clay. The available water holding capacity in the upper 150 cm of soil is 10 cm. Experiments were set up at different areas of the research farm each year; the previous crop in both years was non-irrigated cereal rye (*Secale cereal* L.).

To determine pre-plant soil inorganic N, soil samples from the upper 60 cm were collected from each whole plot on 23 March 2010 and 6 April 2011 in order to determine pre-plant soil inorganic N. After harvest, soil samples from the upper 60 cm were collected from each treatment plot to determine residual soil NO_3-N. All samples were air-dried, ground to pass a 2-mm sieve, and analyzed for KCl extractable nitrate-N (NO_3-N) using a Wescan N analyzer (Carlson et al., 1990). Pre-plant soil samples were also analyzed for KCl extractable ammonium-N (NH_4-N). Pre-plant soil inorganic total N (NH_4-N + NO_3-N) was 15 kg ha^{-1} in 2010 and 20 kg ha^{-1} in 2011. Additional pre-plant soil properties are shown in Table 2-1.

2.2. Experimental design

The field study was set up as a randomized complete block design with a split-split plot restriction on randomization replicated four times. The whole plot treatment was irrigation rate, (i.e., unstressed and stressed treatments). Irrigation was applied with an overhead sprinkler system. A water balance method was used to schedule irrigation applications for the unstressed treatment (Wright, 2002). The stressed plots were irrigated at a rate in which cumulative water (rainfall + irrigation) equaled approximately 89% and 81% of the unstressed plots in 2010 and 2011, respectively, and was calculated from total water applied between emergence and vine kill. Daily rainfall totals and daily irrigation and cumulative water for both the unstressed and stressed irrigation regimes are presented in Fig. 2-1 for 2010 and Fig. 2-2 for 2011. Timing of irrigation was variable between years and depended on weather conditions. On average, approximately four days elapsed between irrigation applications.

The subplot treatment included a low, medium, and high N rate (i.e., 34, 180, and 270 kg N ha^{-1}) with variable timing of post-emergence N applications for the high rate, for a total of five N treatments (i.e., 34 N early, 180 N split, 270 N split, 270 N split + surfactant (s), and 270 N early; Table 2-2). The 270 N split and 270 N split + s had the same rate and timing of N application, but a soil surfactant (IrrigAid Gold) was applied to 270 N split + s at a rate of 10 L ha^{-1} on top of the hill just prior to emergence to investigate the effects of the surfactant on N uptake under different irrigation regimes. On average, about two weeks passed between split applications of post-emergence N for the 180 N split, 270 N split, and 270 N split + s treatments. In both years, the planting and emergence N source was mono-ammonium phosphate and urea, respectively. Post-emergence N was split applied four times by hand as a 1:1 mixture of urea and ammonium nitrate in 2010 and five times by spray boom and tractor as 28% urea-ammonium nitrate solution in 2011. All post-emergence N was irrigated in immediately following application.

The sub-subplot treatment consisted of two potato varieties, (i.e., Russet Burbank and Alpine Russet). Russet Burbank (RB), the most widely grown processing potato variety in the U.S. and in Minnesota, has a mid-season maturity class of 111-120 days after planting (Whitworth et al., 2011). Alpine Russet (AR) has a late-season maturity class of 121-130 days after planting, and is a relatively new variety grown in Minnesota. Compared to RB, it reportedly has similar yields, superior processing quality after long-term storage, lower N fertilizer requirements to obtain similar yields, and greater resistance to water-stress induced tuber defects (Whitworth et al., 2011).

In both years, whole "B" seed and cut "A" seed were used for the RB and AR varieties, respectively. Seed was hand planted in furrows with 90 cm row spacing and approximately 30 cm spacing between seed pieces within rows. Each plot consisted of seven 13.7 m rows. Planting dates were 16 April 2010 and 29 April 2011 and plant emergence occurred on 15 May 2010 and 24 May 2011. Two days after plant emergence in each year, emergence fertilizer was applied and rows were mechanically hilled. Chemicals were applied as needed during the season for the control of pests, disease, and weeds according to standard practices in the region (Engel et al., 2012).

2.3. Field measurements and data sampling

Average daily temperature and daily rainfall and irrigation totals were measured throughout the growing season for both years (Figs. 2-1 and 2-2). Average daily soil temperature was also measured in 2011 (5TM sensor, Decagon Devices, Pullman, WA) for both the unstressed and stressed irrigation regimes (Fig. 2-2).

To estimate N supplied by precipitation and irrigation, water samples were collected throughout the growing season. Concentrations of NO_3-N and NH_4-N from the samples were determined using a Wescan N analyzer. Following reduction of NO_3-N to NH_4-N using granular zinc, NO_3-N was determined as the difference between NH_4-N + NO_3-N and NH_4-N (Carlson, 1986; Carlson et al., 1990). For the unstressed irrigation treatment, total N supplied by rainfall + irrigation was 53 and 41 kg N ha^{-1} in 2010 and 2011, respectively, and for the stressed irrigation treatment, total N supplied by rainfall + irrigation was 41 and 29 kg N ha^{-1} in 2010 and 2011, respectively.

Vines were mechanically killed (via flail mower) on 10 Sept. 2010 (118 days after emergence [DAE]) and 14 Sept. 2011 (113 DAE). Immediately prior to mowing, a 3 m length of vine samples was collected by hand from both the third and fourth rows from the alley. Vine samples were weighed (fresh weight), and then a subsample was collected from each plot to be analyzed for percent dry matter and vine N concentration. Tubers were mechanically harvested from the third and fourth rows from the alley in each treatment plot approximately 1-2 weeks after the vines were killed. Tubers were sorted into weight classes for total and graded yield. Grade A yield was determined by subtracting undersized tuber yields (<85 g) from the total yield. The proportion of misshapen tubers was calculated using all tubers >85 g in size. A sample of twenty-five representative tubers from 85-392 g were collected from each plot to measure incidence of hollow heart, specific gravity (calculated using the ratio between the weight in air and weight in water; Dean, 1994), percent dry matter, and tuber N concentration.

Vine subsamples and tuber samples were weighed, oven-dried at 60° C for at least 72 hours, and weighed again to determine percent dry matter. Dry matter yield (total yield × percent dry matter) was then calculated for both vines and tubers; the sum of dry matter vine yield and dry matter tuber yield make up what is hereafter referred to as plant dry matter. The oven-dried vine and tuber samples were ground with a Wiley mill to pass a 2-mm sieve, and were analyzed for total N concentration using a combustion analyzer (Elementar Vario EL III, Elementar Americas Inc., Mt. Laurel, NJ) following the methods of Horneck and Miller (1997). Nitrogen content in the vines and tubers was then calculated by multiplying the dry matter yield and total N concentration; the sum of vine N content and tuber N content make up what is hereafter referred to as plant N uptake.

A sample of 8-10 tubers in the 170-283 g size class was also collected from each plot to measure sucrose concentration, glucose concentration, and light reflectance, a measure of chip color. Tuber sugars were analyzed with a YSI 2700, Select, Industrial Analyzer (Yellow Springs Instrument Co., Inc., Yellow Springs, OH, USA) following the methods of Sowokinos et al. (2000). Light reflectance measurements were made with a HunterLab D-25/DP-9000® (Hunter Associates Laboratory, Inc., Reston, VA, USA) standardized with black glass and C2-39360 calibration tiles. All measurements were taken on the stem and bud ends of the tubers at harvest and following 6 months of storage at 7.8° C.

2.4. Nitrogen efficiency indices

Nitrogen efficiency indices help to explain how crop available N is recovered and utilized by the potato crop. Nitrogen utilization efficiency (NUtE; Eq. (2-1)) refers to the efficiency of a crop in utilizing the N that has been taken up for plant dry matter production (Bock,1984; Zebarth et al., 2004). Nitrogen uptake efficiency (NUpE; Eq. (2-2)) refers to the efficiency of a crop in recovering plant-available N (Bock,1984; Zebarth et al., 2004). Nitrogen use efficiency (NUE; Eq. (2-3)) refers to the efficiency of a crop in producing plant dry matter per quantity of plant-available N (Zebarth et al., 2004). Tuber fertilizer efficiency (TFE; Eq. (2-4)) refers to the efficiency of a crop in utilizing applied N fertilizer for dry matter tuber production. Nitrogen harvest index (NHI; Eq. (2-5)) refers to the efficiency of the crop in transporting N to the tubers after it has been taken up by the crop (Zebarth et al., 2004; Zvomuya et al., 2002).

$$NUtE = \frac{DM_{plant}}{N_{uptake}} \qquad \text{Eq. (2-1)}$$

$$NUpE = \frac{N_{uptake}}{N_{available}} \qquad \text{Eq. (2-2)}$$

$$NUE = \frac{DM_{plant}}{N_{available}} \qquad \text{Eq. (2-3)}$$

$$TFE = \frac{DM_{tuber}}{N_{fertilizer}} \qquad \text{Eq. (2-4)}$$

$$NHI = \frac{T_{uptake}}{N_{uptake}} \qquad \text{Eq. (2-5)}$$

where *NUtE* is nitrogen utilization efficiency (g g^{-1}), *NUpE* is nitrogen uptake efficiency (%), *NUE* is nitrogen use efficiency (g g^{-1}), *TFE* is tuber fertilizer efficiency g g^{-1}), *NHI* is nitrogen harvest index (%), DM_{plant} is plant (vine + tubers) dry matter, N_{uptake} is plant nitrogen uptake (vine + tuber), $N_{available}$ is plant-available nitrogen, $N_{fertilizer}$ is rate of N fertilizer, and T_{uptake} is tuber nitrogen uptake.

Plant-available N was calculated as the sum of: soil inorganic N at planting; N in the seed tubers (estimated to be 6 kg N ha^{-1}); N fertilizer applied throughout the season; N provided to the crop via irrigation + rainfall; and estimated net N mineralization. Net N mineralization was calculated using a mass balance approach (Eq. (2-6)) according to Errebhi et al. (1998) using only measurements from the 34 N early treatment. Net N mineralization was calculated separately for all Russet Burbank treatment plots within the 34 early N treatment (i.e., within years and irrigation regimes). The average value (calculated to be 83 kg N ha^{-1}) was used to represent net N mineralization for all treatment plots.

$$N_{mineralization} = [(U + L + RS) - (IS + S + F + W)] \qquad \text{Eq. (2-6)}$$

where *U* is plant nitrogen uptake (vine + tuber), *L* is nitrogen that became unavailable due to leaching as described below (~91 kg N ha^{-1}), *RS* is residual soil inorganic nitrogen in the soil after harvest, *IS* is initial soil inorganic nitrogen in the soil at planting, *S* is nitrogen in the seed tubers (~6 kg N ha^{-1}), *F* is nitrogen supplied to the crop as applied fertilizer, and *W* is nitrogen supplied to the crop via irrigation and rainfall; all terms are expressed in kg N ha^{-1}.

Using this mass balance approach, it was assumed that: (i) the difference between N remaining in the roots and N lost due to volatilization and denitrification was negligible, (ii) there was no enhancement of mineralization over the 34 N early treatment caused by an increasing rate of N fertilizer applications, and (iii) there was no significant immobilization of applied N fertilizer. Although there are several assumptions that go into the calculation of plant-available N for use in NUpE and NUE equations, this method provides a more realistic estimate than would be obtained using the quantity of N fertilizer alone (Zebarth et al., 2004) as has been frequently used (Bock, 1984; Errebhi et al., 1999; Zvomuya et al., 2002).

2.4.1. Nitrate leaching estimation

Leaching can be described using the general water budget equation (Errebhi et al., 1998), where change in water storage = inputs – outputs, or:

$$\varDelta S = (P + I) - (ET + D) \tag{7}$$

where $\varDelta S$ is change in soil water storage, P is precipitation, I is irrigation, ET is evapotranspiration, and D is drainage/percolation; all terms are expressed in mm^3 water mm^{-2} soil day^{-1}. It was assumed that soil-water storage on a daily time step did not exceed the available water holding capacity of the soil profile. Daily ET was estimated using the Penman-Monteith equation as described by Venterea et al. (2011). Temperature, wind speed, relative humidity, and net radiation were measured from an on-site weather station in which values were recorded at 10-min intervals. Saturation vapor pressure and actual vapor pressure were estimated using appropriate equations given by Allen et al. (1998). Because ET values using this method are for a grass reference crop, values were multiplied by a crop coefficient based on the stage of growth in the potato crop to give daily ET estimates for potato using equations in Stegman et al. (1977) and Allen et al. (1998).

To determine N lost due to leaching, water samples from below the rooting zone were collected at a depth of 1.2 m using suction cup lysimeters according to the methods of Venterea et al. (2011). Water samples were collected only from the Russet Burbank treatment plots within the 34 early N treatment on 22 dates in 2010 and 28 dates in 2011. Sample collection beginning within three weeks after planting, and terminating within two weeks after residual soil N was collected. Daily rates of nitrate leaching were calculated according to the methods of Errebhi et al. (1998).

2.5. Statistical analysis

Data from the study were analyzed using PROC MIXED (SAS Institute, 2008) with irrigation regimes, N treatments, and variety considered as fixed variables and years and replications (nested within years) considered as random variables. For the main effects and interactions of the fixed variables, pairwise comparisons of the least square means were made using the *lsmeans / pdiff* option of the MODEL statement of the MIXED procedure of SAS (SAS Institute, 2008 ; α = 0.05). The PDMIX800 macro (Saxton, 1998) was used to place treatment means into letter groupings based on the pairwise comparisons. The interactions between the fixed effects and years were assessed by year-specific inference using best linear unbiased predictors (BLUPs) as described by Littell et al. (2006). Since random variables cannot be tested in the *lsmeans* statement, the *estimate* statement was used to make pairwise comparisons between treatment means for the interactions of the random variables (α = 0.05).

3. Results and discussion

3.1. Weather

In 2010, 32 and 22 cm of irrigation were applied to the unstressed and stressed treatment plots, respectively. After rainfall (80 cm), total cumulative water at the end of the growing season was 112 and 102 cm for the unstressed and stressed treatment plots, respectively (Fig. 2-1). In 2011, 26 and 13 cm of irrigation were applied to the unstressed and stressed treatment plots, respectively. After rainfall (64 cm), total cumulative water at the end of the growing season was 90 and 77 cm for the unstressed and stressed treatment plots, respectively (Fig. 2-2). Through the growing season (April through September), the 30-year average rainfall and temperature values at a weather station near Becker, MN are 55.0 cm and 16.5° C, respectively (1971-2000; Midwest Regional Climate Center). Overall, 2010 and 2011 were much wetter than the average growing season. Average daily and monthly air temperatures for the 2010 and 2011 growing seasons are compared with the monthly 30-year average in Figs. 2-1 and 2-2. The average temperatures over the entire growing season were 17.5° C in 2010 (1.0 °C above average) and 16.3° C in 2011 (0.2 °C below average). In 2011, an increase in soil temperature for the stressed plots compared to the unstressed plots was observed towards the end of the season (Fig. 2-2; deviations from the unstressed soil temperatures began at the end of July). This was likely due to an earlier senescence of the stressed plots.

3.2. Tuber yield and quality

There was a significant effect of year on tuber yields (Table 2-3). Higher total and marketable yields were produced in 2010 than in 2011, and the proportion of tubers greater than 170 grams was also higher in 2010 (Table 2-4). The difference between years was likely due to temperature conditions that affected tuber bulking. Higher daytime and nighttime temperatures during tuber initiation and bulking can expedite crop senescence and ultimately reduce tuber yield (Gregory, 1956; Midmore, 1990). High temperatures can also affect the number of small tubers per plant (Haverkort, 1990), and this negatively impacts both Grade A tuber yield the proportion of tubers >170 g. A major reason for yield reduction at higher temperatures is because the photosynthetic efficiency of potato plants is substantially reduced in high temperature environments (Prange et al., 1990). The average nighttime minimum temperature throughout the month of July (important tuber bulking period) was higher in 2011 than 2010. From 2010 to 2011, an increase of 1.2° C during the first half of July and an increase of 3.0° C during the second half of July was measured (precise data not presented). Also, the stressed irrigation regime was slightly more stressed in 2011 than in 2010. Relative to the unstressed irrigation regime, cumulative water for the stressed regime was 89% in 2010 and 81% in 2011, although the year × irrigation interaction was not significant for any yield measurement.

The unstressed irrigation regime resulted in higher total and Grade A tuber yields, and also a higher proportion of tubers > 170 g than the stressed regime (Table 2-4). The main effect of variety did not affect total tuber yield, but Grade A tuber yield and the proportion of tubers >170 g was higher in AR than RB. The proportion of tubers >170 g was higher in AR than RB for each irrigation regime, but there was a significant irrigation × variety interaction because there was a greater decrease from the unstressed to the stressed regime for AR compared with RB (data not presented).

All N treatments were higher than the 34 N early control for total and Grade A tuber yields, as well as the proportion of tubers >170 g (Table 2-4). The N treatments that had split applications of post-emergence N had higher total tuber yields than the 270 N early treatment. The proportion of tubers >170 g was higher for the N treatments with the 270 kg N ha^{-1} rates than for the 180 N split treatment, regardless of N fertilizer timing. Grade A tuber yield was higher in AR than in RB for every N treatment except 270 N early (in which RB and AR were statistically similar), and this resulted in a significant N treatment × variety interaction (data not presented). The soil surfactant did not affect any of the tuber yield measurements, and it had a minimal effect (if any) on the tuber quality, N uptake and efficiency, and fry quality measurements evaluated in this study.

The main effect of N, the N treatment × variety interaction, the N treatment × variety × year interaction and the irrigation × variety × year interaction were significant for the proportion of misshapen tubers (Table 2-3). There were many factors that caused these significant three-way interactions (Fig. 2-3). The most apparent observation was that AR had a significantly higher proportion of misshapen tubers than RB for the 34 N early treatment in both years (for the 34 N early treatment, the average proportion of misshapen tubers was 55% for AR, but was only 16% for RB). For all other N treatments, the proportion of misshapen tubers between varieties was similar in 2010, but in 2011, AR had a higher proportion of misshapen tubers than RB for all N treatments except 270 N split (Fig. 2-3a). In addition, RB had a significantly higher proportion of misshapen tubers in 2010 than 2011 for the treatments with 270 kg N ha^{-1} split. For AR, there were not a higher proportion of misshapen tubers in 2010 compared with 2011 for any of the N treatments. In fact, the 34 N early treatment actually had a lower proportion of misshapen tubers for AR in 2010. The irrigation × variety × year interaction was also significant (Table 2-3). In 2010, the proportion of misshapen tubers was the same between varieties in the unstressed irrigation, but was higher for RB in the stressed irrigation; in 2011, the proportion of misshapen tubers was also the same between varieties in the unstressed irrigation, but the proportion of misshapen tubers in the stressed irrigation was actually lower for RB (Fig. 2-3b). In 2010, water stress began at the beginning of July and continued through about mid-August, whereas in 2011, water stress did not begin until about mid-July and continued through the beginning of September. This is perhaps evidence that the timing of water stress affects the varieties differently. Also, the proportion of misshapen tubers was the same between varieties and years in the unstressed irrigation; while the proportion of misshapen tubers in the stressed irrigation was also similar between years for AR, the proportion was significantly higher in 2010 than 2011 for RB. These complicated interactions with year illustrate: (i) that the proportion of misshapen tubers averaged over irrigation regimes were higher in AR than RB for the 34 N early treatment and (ii) that the proportion of misshapen tubers averaged over N treatments were variable with variety, irrigation regime, and year. Although Whitworth et al. (2010) did not consider differences in water or N stress, they concluded that AR culls, consisting of both misshapen and rotten tubers, were lower than RB at their reported locations.

The incidence of hollow heart was significantly affected by irrigation and N treatment, but year was not significantly different (Table 2-3). Averaged over all other variables (i.e., for the main effects), there was a higher incidence of hollow heart in the unstressed irrigation regime, the N treatments with the 270 kg N ha^{-1} rate, and the RB variety (Table 2-4). However, there was a significant effect for the irrigation × N treatment × variety three-way interaction. For both irrigation regimes and both varieties, there was a 0% incidence of hollow heart for the 34 N early treatment. The incidence of hollow heart was very rare for the AR variety. The only circumstances in which it appeared was in the unstressed irrigation regime for the 270 N split treatment, but the incidence was very low and was not statistically different from the other N treatments (data not presented). For the RB variety and stressed irrigation regime, the only N treatment that had a significantly higher incidence of hollow heart than the other N treatments was the 270 N early treatment. However, for the unstressed irrigation regime, the only N treatments with a significantly higher incidence of hollow heart than the other N treatments were the 270 N split treatments. Previous research has shown zero percent incidence of hollow heart for the AR variety (Whitworth et al., 2011). For RB, hollow heart is typically most serious in larger tubers (Beattie, 1989). This is consistent with data from our study since the treatments with the higher N and irrigation rates had a higher proportion of tubers greater than 170 g (Table 2-4), and these were the treatments with the highest incidence of hollow heart.

Specific gravity was higher in 2011 than 2010, and was higher for RB than for AR (Table 2-4). Neither irrigation nor N treatments significantly affected specific gravity. Cumulative water supplied to the crop was very similar between irrigation treatments and years until the very end of June (Figs. 2-1 and 2-2); substantial differences did not appear until the beginning of August in each year. This indicates that any differences in specific gravity due to environmental conditions through the end of July were likely due to temperature differences between years; previous research has shown a reduction in specific gravity due to high soil (Epstein, 1966) and air temperatures (Van den Berg et al., 1990; Wilson et al., 2009). Specific gravity was not significantly different between RB and AR varieties over all trial locations evaluated by Whitworth et al. (2011). However, within locations and in contrast to our results, they reported a significantly higher specific gravity for AR than RB at two of nine locations (when the crops were late harvested only).

3.3. Crop nitrogen uptake and nitrogen use indices

There was a significant effect of year, irrigation, N treatment, and variety on plant N uptake, and none of the interactions were significant (Table 2-3). Averaged over all other variables (i.e., for the main effects), plant N uptake was significantly higher in 2010, for the unstressed irrigation regime, and for the AR variety (Table 2-4). Increased plant N uptake for the unstressed irrigation regime was observed in previous research (Dalla Costa et al., 1997). This increased N uptake can be attributed to a higher quantity of N being supplied to the unstressed irrigation regime via irrigation water; increased N uptake could also be due to an increase in root biomass. Plant N uptake was significantly higher for all other N treatments than for the 34 N early treatment, and plant N uptake was highest for the 270 N split treatments. Plant N uptake for the 180 N split and 270 N early treatments was similar to each other and lower than the 270 N split treatments. The increase in plant N uptake with increasing N fertilizer rate for the treatments with split applications of post-emergence N suggests that plant N uptake is determined primarily by the rate of N fertilizer. The significantly lower plant N uptake for 270 N early compared with the 270 N split treatments indicates that plant N uptake is also affected by the timing of N fertilizer. Under the conditions of this study, this difference was likely due to NO_3-N leaching with excessive rainfall events in June and July each year (Figs. 2-1 and 2-2). Nitrate-nitrogen (NO_3-N) leaching results in a reduction of plant available N, and therefore can decrease plant N uptake. Increased NO_3-N leaching rates have been shown to result from early applications of post-emergence N fertilizer compared with split applications, especially during heavier leaching years (Errebhi et al., 1998; Westermann et al., 1988).

Plant dry matter was significantly affected by year and N treatment, but irrigation and variety did not have a significant effect (Table 2-3). Averaged over all other variables (i.e., for the main effects), plant dry matter was significantly higher in 2010 than in 2011 (Table 2-4). Plant dry matter was lowest for the 34 N early treatment and was highest for the three N treatments with the 270 kg N ha^{-1} fertilizer rate. An increase in plant dry matter with increasing N fertilizer rate was also observed by Zebarth et al. (2004).

Nitrogen use efficiency (NUE) was significantly affected by year and N treatment, but not by irrigation and variety (Table 2-3). Averaged over all other variables (i.e., for the main effects), NUE was significantly higher in 2010 than in 2011 (Table 2-4). NUE decreased by a significant amount for each step in N rate (i.e., NUE was highest for the 34 N early, next highest for 180 N split, and lowest for the treatments with the 270 kg N ha^{-1} rates). Previous research has shown a decrease in NUE (Zebarth et al., 2004) with increasing N fertilizer rate. Neither the soil surfactant nor the timing of the treatments with the 270 kg N ha^{-1} rate resulted in a significant difference in NUE. The NUE estimates in this study were lower than values previously reported, especially for the lowest N rates (Zebarth et al., 2004). Because leaching was not considered, actual NUE was overestimated by Zebarth et al. (2004).

None of the interactions were significant for nitrogen utilization efficiency (NUtE), nitrogen uptake efficiency (NUpE), or tuber fertilizer efficiency (TFE), but the main effects of year, N treatment, and variety were significant for each (Table 2-3). Similar to NUE values, NUtE, NUpE, and TFE values were higher in 2010 than in 2011.

NUtE was significantly lower for 180 and 270 N treatments than for the 34 N early treatment, and was lowest for the 270 N split treatments (Table 2-4). It is important to note, however, that high NUtEs are not valid if N uptake is very low because there is a genetic limit to how much biomass a plant can produce with low N uptake (Errebhi et al., 1999). The means separation among N treatments for NUtE showed the exact opposite trend with what was observed for plant N uptake (i.e., NUtE decreased and plant N uptake increased with increasing N fertilizer rate). Even though DM also increased with N rate, it did not increase to the same extent as plant N uptake. This illustrates that at least some of the differences in NUtE across N treatments was apparently due to the variation in plant N concentration across N treatments. NUpE and TFE also decreased as N fertilizer rate increased; the statistical relationships among N treatments for NUpE and TFE were similar. The 270 N early treatment had a lower NUpE and TFE than the 270 N split treatments, which was likely due to NO_3-N leaching. Compared with the 270 N split treatments, the higher NUtE for 270 N early was apparently due to the lower quantity of plant N uptake for 270 N early, as indicated by the similar response of plant dry matter among these treatments. The general decrease in NUtE and NUpE with increasing N fertilizer rate in this study is consistent with results from other studies (Errebhi et al., 1999; Kleinkopf et al., 1981; Zebarth et al., 2004, Zvomuya et al., 2002).

NUtE was higher for RB, but NUpE was higher for AR, and this can be attributed to lower plant N uptake for RB. The higher TFE for RB when compared to AR was due to higher tuber dry matter yield for RB. Although plant N uptake and total tuber yield were significantly higher for the unstressed irrigation, NUtE, NUpE, and TFE for the main effect of irrigation were not significant. The lack of significance for NUtE can be attributed in part to the similar trends between the numerators and denominators with the different irrigation treatments (i.e., if the numerator increased with decreasing water stress, so did the denominator, ultimately resulting in no significant change in the N index). The lack of significance for NUpE can be attributed in part to plants in the unstressed irrigation having a greater quantity of available N provided via irrigation. In contrast to NUpE, the fertilizer uptake efficiency (FUpE; plant N uptake per unit of applied fertilizer) was significantly higher for the unstressed irrigation (data not presented). This could be in part due to more prolific root growth for the unstressed irrigation; however, because the unstressed irrigation treatment also provided more available N to the crop than the stressed irrigation treatment, there is a confounding effect that makes it difficult to identify the exact cause for the higher FUpE. Therefore, for this study, NUpE is a more useful term than FUpE because NUpE takes into consideration all forms of available N, not just N available from applied fertilizer (Zebarth et al., 2004).

Nitrogen harvest index (NHI) was significantly affected by N treatment, irrigation, variety, and the irrigation × variety interaction (Table 2-3). NHI was higher in RB than in AR for the unstressed irrigation regime, but not for the stressed irrigation regime (in which case RB and AR were statistically similar), and this resulted in a significant irrigation × variety interaction (data not presented). NHI was not affected by timing of post-emergence N fertilizer for the treatments with the 270 kg N ha^{-1} rate, but NHI did decrease with increasing N fertilizer rate (Table 2-4). This indicates that relative to the total amount of N taken up by the crop, less N is translocated to the tubers as N rate increases.

3.4. Residual soil nitrate

None of the interactions for residual soil NO_3-N were significant ($\alpha=0.05$), so the mean values of the main effects only are presented (Table 2-5). Residual soil NO_3-N in the top 60 cm was significantly higher in 2011 than 2010. Irrigation and variety did not significantly affect residual NO_3-N, but N treatment did. The 34 N early treatment had a significantly lower NO_3-N concentration than any of the other treatments, and the 270 N split + s treatment had a significantly higher NO_3-N concentration than the 180 N split treatment; however, the 270 N split + s treatment had a statistically similar NO_3-N concentration as the other treatments with the 270 kg N ha^{-1} rates. The similarity between the early applied and split applied 270 kg N ha^{-1} rates was also observed in previous research on irrigated potato (Wilson et al., 2010), even though the quantity of NO_3-N leaching for these N treatments were likely quite different between studies.

3.5. Tuber frying quality before and after storage

The analysis of variance for tuber sucrose and glucose concentration is presented in Table 2-6. Sucrose concentration was higher in 2010 than 2011 in the stem end after storage, as well as in the bud end both at harvest and after storage (Table 2-7). The differences between years could be due to environmental conditions since tuber sugar concentration can change depending on environmental conditions such as temperature or soil moisture (Kumar et al., 2004). The unstressed irrigation regime resulted in significantly lower sucrose concentrations for all sucrose measurements except for in the bud end after storage (in which case mean values for sucrose were statistically similar). Sucrose concentration was highest for the 34 N early treatment in both the bud and stem ends, but only at harvest. The sucrose concentration was lower in RB than in AR for all N treatments; however, in the bud end in RB, there was not a significant difference among N treatments and this triggered a significant N treatment × variety interaction (data not presented). In AR, the sucrose concentration for 270 N split + s was significantly lower than 180 N split and 270 N early. The soil surfactant did not affect sucrose concentration at equivalent rate and timing of N fertilizer. In the bud end after storage, there was a significant irrigation × variety interaction; this was triggered because sucrose concentration was similar over irrigation regimes in RB, but the stressed irrigation increased sucrose concentration in AR.

Glucose concentration was higher in 2010 than 2011 in the stem and bud ends after storage (Table 2-7). At harvest, glucose concentration in the stem end triggered an irrigation × year interaction because glucose concentration for the unstressed irrigation regime was higher in 2011, but glucose concentration for the stressed irrigation regime was higher in 2010. The glucose concentration in the stem end after storage was higher for the unstressed irrigation regime than for the stressed, and there was no significant difference between irrigation regimes for bud end glucose concentration either at harvest or after storage. In this study, 2010 stem end glucose concentration at harvest, both stem and bud end sucrose concentrations at harvest, and stem end sucrose concentration after storage were similar to the findings of Shock et al. (1993). However, 2011 stem end glucose concentration at harvest and stem end glucose concentration after storage (over both years) is in conflict with those results. Shock et al. (1993) found elevated levels of total reducing sugars at harvest, particularly in the stem ends of RB potatoes subjected to water stress. The differing results between these experiments are unclear, but it could potentially be due to the severity and/or timing of the water stress. Glucose concentration decreased with higher N fertilizer rates in both the stem and bud ends of the tuber. This is contrary to results presented by Westermann et al. (1994); they found that the reducing sugars decreased with higher N fertilizer rates in the stem end, but actually increased with increasing N rates in the bud end. Glucose concentration was highest for the 34 N early treatment in both the bud and stem ends at harvest and after storage. Glucose concentration for the 270 N split and 270 N split + s treatments were always lower than for 180 N split, and were either lower or statistically similar to 270 N early. Averaged over all other variables (i.e., for the main effects), glucose concentration in the stem end was lower in AR than RB, but glucose concentration in the bud end was lower in RB. Overall, the glucose concentration in both varieties was higher in this study than that reported by Whitworth et al. (2011); however, they found substantial variation in percent reducing sugars among locations evaluated in their study.

The analysis of variance for light reflectance (AGT score) after frying is presented in Table 2-6. Light reflectance was higher (lighter chip color) in 2011 than 2010 in the stem end at harvest and after storage, and in the bud end after storage (Table 2-7). Light reflectance was also higher in the stressed irrigation regime in the stem end after storage and in the bud end at harvest and after storage. However, for these three measurements, there was a significant year × irrigation interaction because light reflectance was not significantly different between irrigation regimes in 2010, but light reflectance in the unstressed irrigation regime was lower than in the stressed irrigation regime in 2011. The 2011 data are contrary to what was observed in previous research. Shock et al. (1993) found that water stress in late June or throughout July resulted in light reflectance readings that were significantly lower than the unstressed control, although this was observed in only one of two years. They observed that water stress in late July and August resulted in light reflectance readings that were not significantly different from the unstressed control, similar to results of 2010 data from the current study.

Bud and stem end light reflectance at harvest and after storage was significantly lower in the 34 N early treatment than in the other N treatments. For 270 N split and 270 N split + s, bud end chip color after storage was higher than the 180 N split treatment. Stem end light reflectance at harvest was higher in AR, and bud end light reflectance after storage was higher in RB. For stem end light reflectance after storage, there was a significant N treatment × variety interaction because 180 N split, 270 N split, and 270 N early were similar between varieties, but light reflectance was higher in RB for 34 N early and lower in RB for 270 N split + s (data not presented). Overall, AR had a more uniform light reflectance due to a smaller difference in reducing sugars between the bud and stem ends and this is consistent with results reported by Whitworth et al. (2011).

In general, AR contained more sucrose in both ends of the tuber and more glucose in the bud end, and RB contained more glucose in the stem end, resulting in lower light reflectance in the stem end at harvest in RB, and lower light reflectance in the bud end after storage in AR. It was difficult to identify the effect of irrigation on tuber sugar concentration and its relationship to chip color. In general, stem end sucrose was higher in the stressed irrigation regime, but stem end glucose was higher in the unstressed irrigation regime; this ultimately resulted in higher stem end light reflectance for the unstressed irrigation regime, but only after storage. There were many irrigation interactions for light reflectance as well, which further complicated its relationship with sugar concentration. It was also difficult to identify the effect of N treatment on tuber sugar concentration and its relationship to light reflectance because of differences in responses between sugar concentration and chip color. For example, among N treatments, bud end sucrose after storage was similar, but light reflectance was different. However, nitrogen stress that occurred with the 34 N treatment clearly resulted in the highest tuber sucrose and glucose levels and the lowest light reflectance. Perhaps more N rates would make it easier to distinguish the precise relationship between sugar concentration and light reflectance.

4. Conclusions

This two year study has shown that insufficient supplemental water during critical growth stages negatively affected tuber yield/quality and plant N uptake, but surprisingly had a positive overall effect on chip color. The late season timing of water stress may have been responsible for the chip color results. The 270 kg N ha^{-1} rates with split applications of post-emergence N had a positive effect on tuber yield, size, fresh quality, plant N uptake, and dry matter production, but resulted in lower efficiency ratings according to the N use indices when compared with lower N rates. The use of the soil surfactant had a minimal effect on the parameters measured in this experiment. The NUE and NUpE values were relatively low in the two years of this study, which is likely the result of 2010 and 2011 being high leaching years. Within similar N rates, NUpE was higher when post-emergence N was split applied in 4 or 5 applications than when it was applied in one early application. The higher NUpE is likely the result of less N being lost due to leaching. Over years, NUE was not affected by irrigation or variety, and it decreased with increasing N rate. Low NUpE for the treatments with high N rates was reflected by the higher residual soil NO_3-N levels for the higher N rate treatments. Alpine Russet had tuber yields that were comparable or greater than Russet Burbank, but had a specific gravity that was slightly lower than Russet Burbank. The two varieties had similar overall reducing sugar concentrations and frying quality measurements. Alpine Russet produced higher Grade A yields, a higher proportion of tubers >170 g, and had higher NUpE than RB. Overall, Alpine Russet is a processing variety that is attractive from both an economic and environmental perspective.

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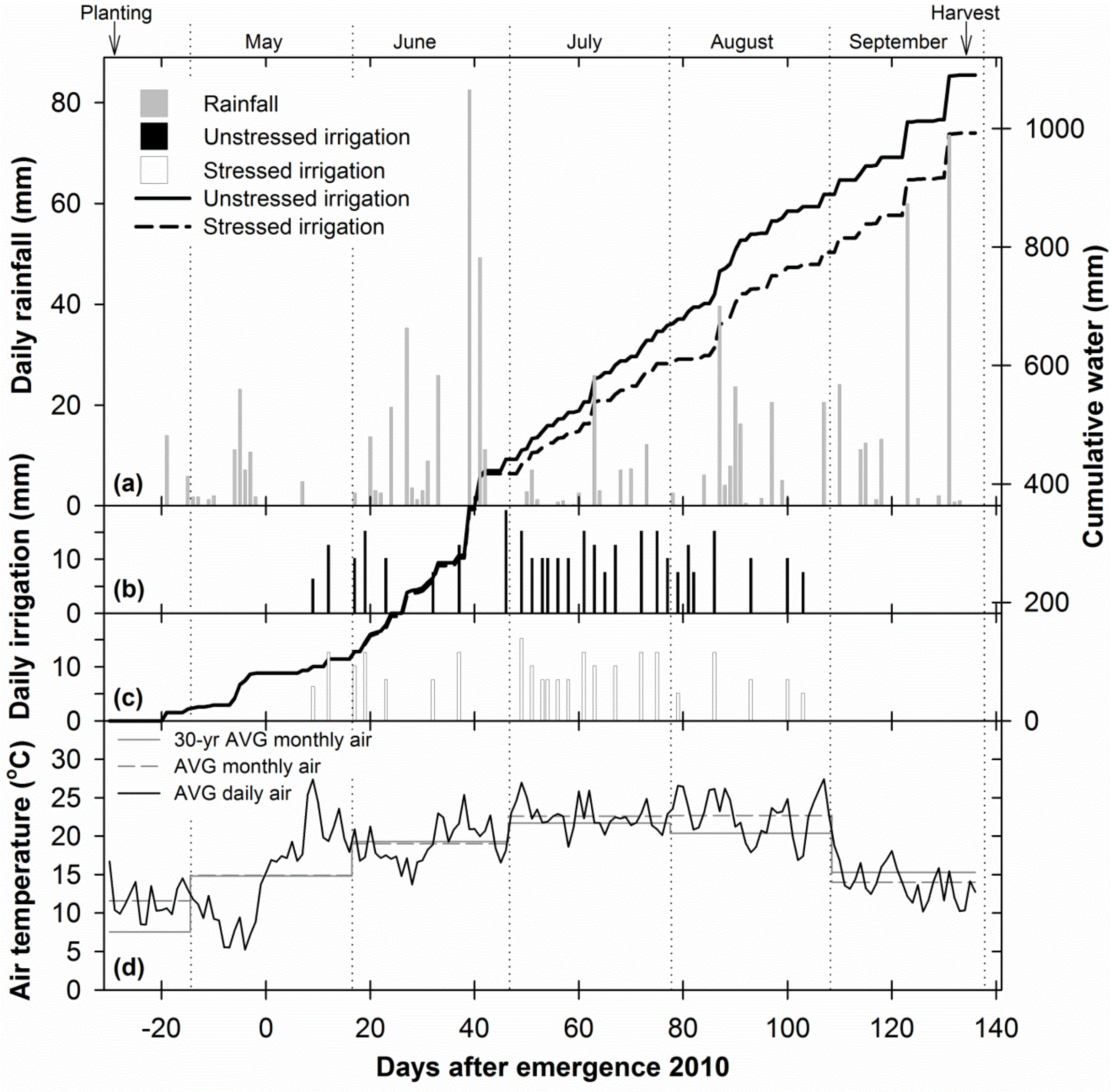

Fig. 2-1. Daily rainfall (a), unstressed irrigation (b), stressed irrigation (c), cumulative water (a-c), and air temperature (d) throughout the 2010 growing season.

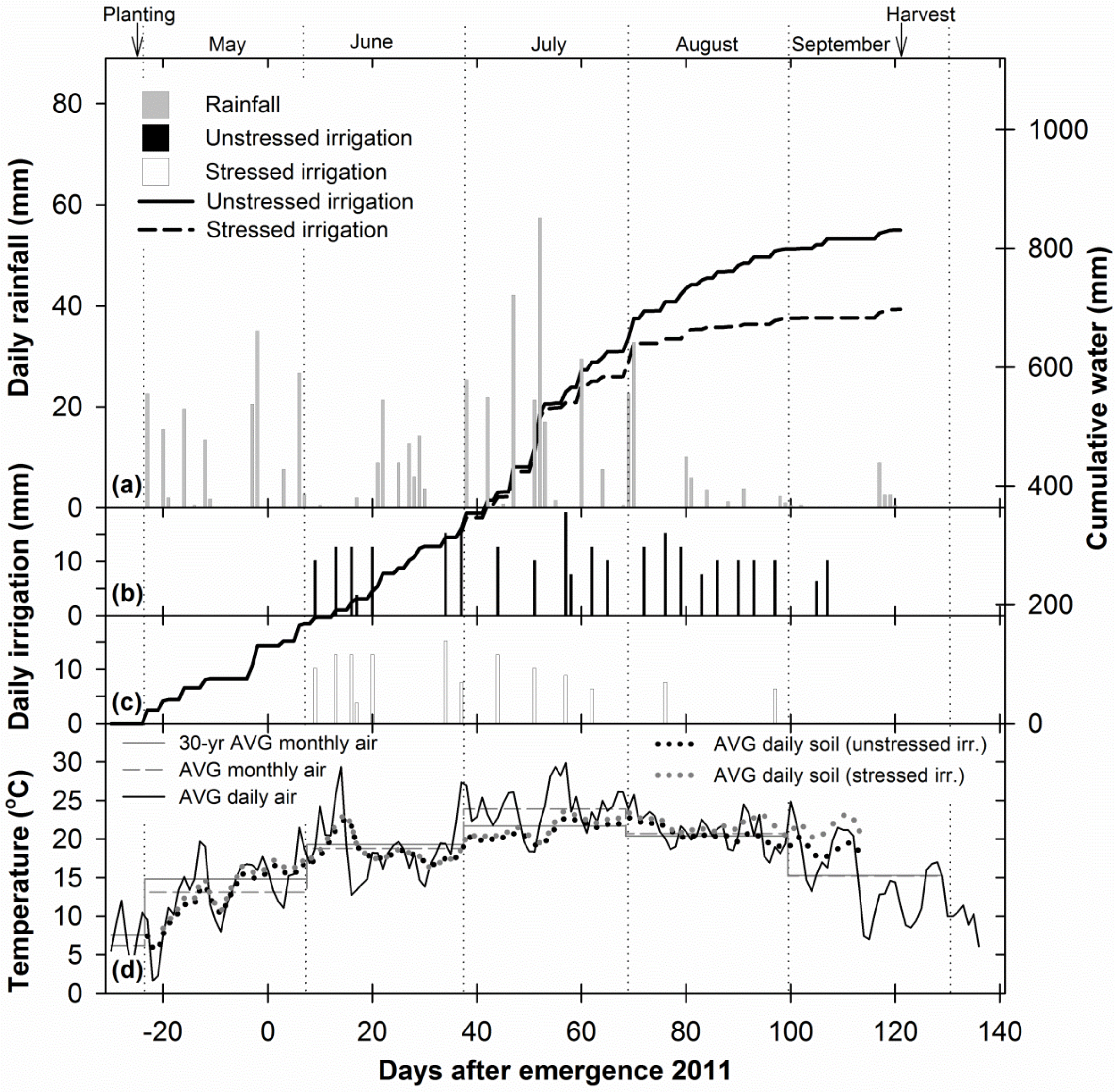

Fig. 2-2. Daily rainfall (a), unstressed irrigation (b), stressed irrigation (c), cumulative water (a-c), and air and soil temperatures (d) throughout the 2011 growing season.

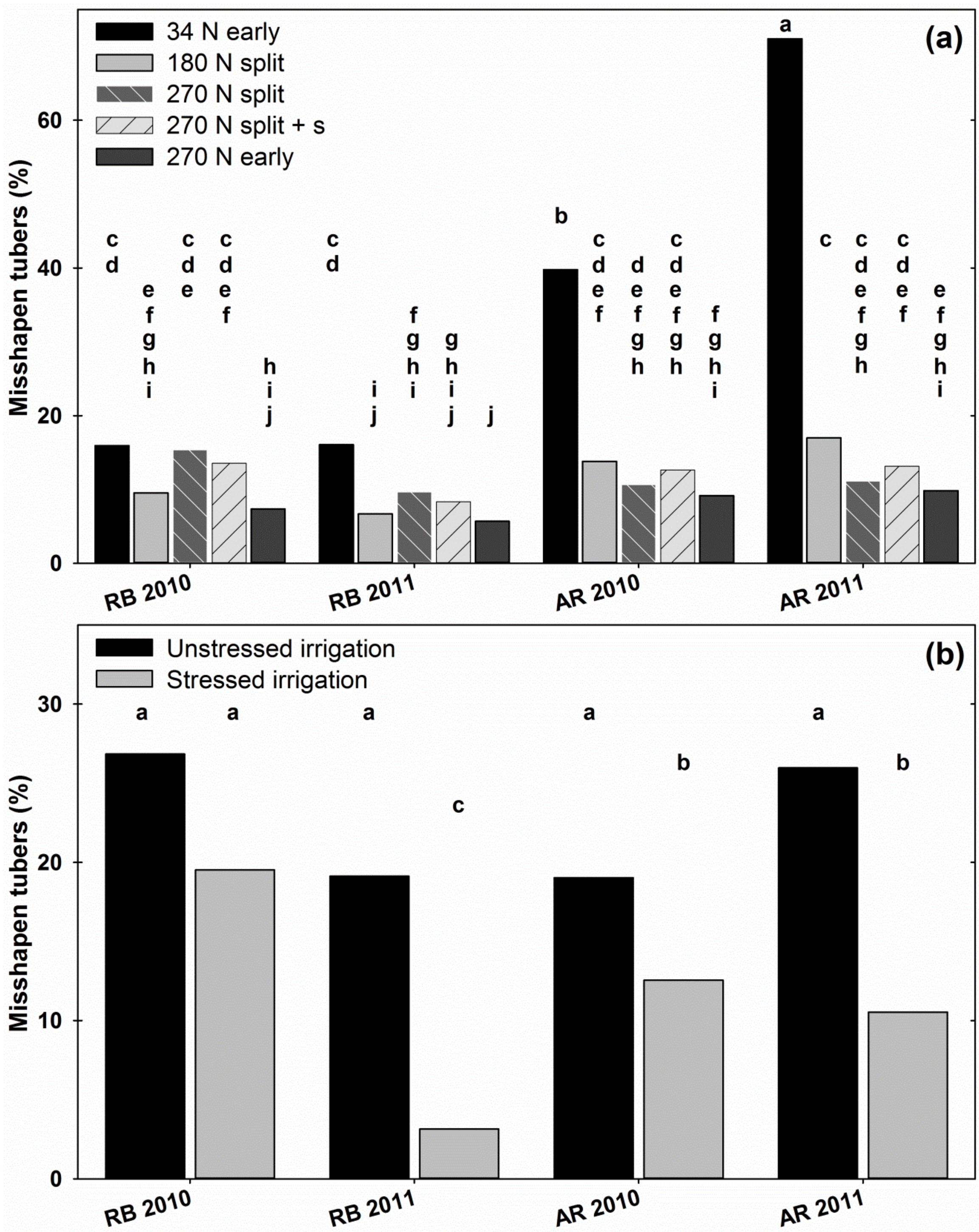

Fig. 2-3. Mean values for the proportion of misshapen tubers for the N treatment × variety × year interaction **(a)** and the irrigation × variety × year interaction **(b)**. Values with the same letter are not significantly different (α = 0.05). RB = Russet Burbank, AR = Alpine Russet.

Table 2-1. Selected pre-plant soil chemical properties of site soils. Samples were collected before planting each year from the upper 15 cm, except for inorganic N samples, which were collected from the upper 60 cm. Means with standard errors in parentheses are shown (*n*=4).

Soil Property[†]	2010	2011
Water-pH	6.8 (0.06)	6.2 (0.03)
Organic Matter (%)	1.5 (0.11)	2.0 (0.09)
	--------- mg kg^{-1}	---------
Bray-P	30 (2.7)	44 (2.6)
K[‡]	101 (9.1)	109 (7.0)
Ca[‡]	847 (44)	875 (25)
Mg[‡]	166 (9.1)	146 (7.5)
Zn[§]	0.9 (0.12)	1.5 (0.06)
Cu[§]	0.4 (0.05)	0.7 (0.07)
Fe[§]	15 (1.2)	38 (2.2)
Mn[§]	4.0 (0.29)	11 (0.58)
SO_4-S	5.7 (1.4)	3.3 (0.25)
NO_3-N	1.6 (0.10)	0.9 (0.06)
NH_4-N	0.1 (0.07)	1.3 (0.10)

[†] Methods of analysis from Brown (1998).

[‡] Extracted with ammonium acetate.

[§] Extracted with diethylenetriaminepentaacetic acid (DTPA).

Table 2-2. Rate and timing of nitrogen (N) fertilizer treatments for 2010 and 2011.

Nitrogen fertilizer treatment	Timing of application					Total N
	planting	emergence		post-emergence		
		2010	2011	2010	2011	
	---------------------------		kg N ha^{-1}		---------------------------	
34 N early	34	0	0	0	0	34
180 N split	34	78	90	17 × 4[‡]	11.2 × 5[‡]	180
270 N split	34	124	124	28 × 4[‡]	22.4 × 5[‡]	270
270 N split + s[†]	34	124	124	28 × 4[‡]	22.4 × 5[‡]	270
270 N early	34	124	124	112	112	270

[†] A soil surfactant (IrrigAid Gold) was used in this treatment, and was applied at a rate of 10 L ha^{-1}.

[‡] On average, about two weeks passed between split applications of post-emergence N.

Table 2-3. Analysis of variance for potato tuber yields, fresh quality, nitrogen (N) use indices, plant N uptake, and plant dry matter (DM).

Effect	Total yield	Grade A yield	Tubers >170g	Tubers Misshapen	Specific gravity	Hollow heart	NUE	NUtE	TFE	NUpE	NHI	Plant N uptake	Plant DM
Year [Y]	***[†]	***	***	-	***	-	**	***	**	*	-	**	*
Irrigation [I]	**	*	**	-	-	*	-	-	-	-	*	*	-
N treatment [N]	***	***	***	***	-	***	***	***	***	***	***	***	***
Variety [V]	-	***	***	-	**	***	-	***	**	***	***	***	-
I × N	-	-	-	-	-	**	-	-	-	-	-	-	-
I × V	-	-	*	-	-	-	-	-	-	-	**	-	-
I × Y	-	-	-	-	-	-	-	-	-	-	-	-	-
N × V	-	*	-	*	-	***	-	-	-	-	-	-	-
N × Y	-	-	-	-	-	-	-	-	-	-	-	-	-
V × Y	-	-	-	-	-	-	-	-	-	-	-	-	-
I × N × V	-	-	-	-	-	*	-	-	-	-	-	-	-
I × N × Y	-	-	-	-	-	-	-	-	-	-	-	-	-
I × V × Y	-	-	-	**	-	-	-	-	-	-	-	-	-
N × V × Y	-	-	-	***	-	-	-	-	-	-	-	-	-

[†] ***, **, and * are significant 0.001, 0.01, and 0.05, respectively; - is nonsignificant.

Table 2-4. Main effects of year, irrigation, nitrogen (N) treatment, and variety on potato tuber yields, fresh quality, nitrogen (N) use indices, plant N uptake, and plant dry matter (DM).

Sources of Variation	Total Yield	Grade A yield†	Tubers >170 g†	Misshapen Tubers†	Hollow heart†	Specific gravity	NUE	NUtE	TFE	NUpE	NHI†	Plant N uptake	Plant DM
Year	--- ton ha^{-1}	---	-------------	%	-------------		----------	g g^{-1}	----------	------- %	-------	kg ha^{-1}	ton ha^{-1}
2010	58.5a‡	52.1a	55.4a	22.4a	1.4 a	1.074 b	40.0a	81.1a	95.2a	50.2 a	87.4a	172 a	14.5a
2011	49.5 b	40.3 b	37.1 b	22.1a	1.2 a	1.078 a	34.0 b	79.0 b	77.2 b	43.4 b	90.3a	146 b	12.3 b
Irrigation													
Unstressed	57.1a	49.8a	50.8a	28.5a	2.0 a	1.076 a	38.0a	79.7a	90.0a	48.4 a	87.9 b	167 a	14.1a
Stressed	50.9 b	42.7 b	41.7 b	16.0a	0.7 b	1.076 a	36.1a	80.5a	82.5a	45.3 a	89.8a	150 b	12.8a
N treatment§													
34 N early	43.9 c	33.6 c	27.3 c	40.5a	0.0 b	1.074 a	51.9a	97.4a	248 a	55.6 a	92.5a	98 c	10.1 c
180 N split	56.2ab	48.6ab	47.5 b	17.1ab	0.3 b	1.076 a	38.9 b	81.9 b	62.1 b	48.2 b	89.8 b	155 b	13.8 b
270 N split	57.4a	50.4a	52.5a	21.2 b	2.4 a	1.077 a	31.8 c	72.3 c	42.6 c	44.7 c	87.8 c	184 a	14.5ab
270 N split + s	57.7a	50.5a	51.6a	20.9 b	2.4 a	1.076 a	32.2 c	70.3 c	43.2 c	46.5 c	87.7 c	192 a	14.6a
270 N early	54.8 b	48.1 b	52.4a	11.4 c	1.5 a	1.077 a	30.9 c	79.1 b	40.7 d	39.6 d	86.7 c	163 b	14.0ab
Variety													
Russet Burbank	53.6a	43.2 b	34.9 b	21.9a	2.6 a	1.078 a	37.3a	85.2a	86.9a	44.4 b	89.9a	152 b	13.6a
Alpine Russet	54.4a	49.3a	57.5a	22.6a	0.1 b	1.074 b	36.8a	75.0 b	85.6 b	49.3 a	87.9 b	166 a	13.2a

†Response variables that had at least one significant interaction; refer to Table 2-3 and text for explanation.

‡Means followed by the same letter are not significantly different (α=0.05).

§N rates are expressed in kg N ha^{-1}; refer to Table 2-2 for detailed rates and timing.

Table 2-5. Main effects of year, irrigation, nitrogen (N) treatment, and variety on residual (post-harvest) soil nitrate-nitrogen (NO_3-N; 0-60 cm).

Sources of Variation	Residual soil NO_3-N
Year	-- mg kg^{-1} --
2010	2.1 b[†]
2011	3.9 a
Irrigation	
Unstressed	2.8 a
Stressed	3.2 a
N Treatment[‡]	
34 N early	2.2 c
180 N split	2.8 b
270 N split	3.2 ab
270 N split + s	3.5 a
270 N early	3.1 ab
Variety	
Russet Burbank	3.2 a
Alpine Russet	2.8 a

[†] Means followed by the same letter are not significantly different (α=0.05).

[‡] N rates are expressed in kg N ha^{-1}; refer to Table 2-2 for detailed rates and timing.

Table 2-6. Analysis of variance for sucrose concentration, glucose concentration, and AGT score in tubers for both the stem and bud ends at zero months after harvest (T0) and after six months of storage (T6).

Effect	Sucrose				Glucose				Light reflectance after frying			
	Stem end		Bud end		Stem end		Bud end		Stem end		Bud end	
	T0	T6	T0	T6	T0	T6	T0	T6	T0	T6	T0	T6
Year [Y]	-[†]	***	**	***	-	***	-	***	***	***	-	**
Irrigation [I]	**	*	*	-	*	*	-	-	-	**	*	*
N treatment [N]	***	-	**	-	***	***	***	***	*	***	***	***
Variety [V]	***	***	***	***	***	**	***	***	***	-	-	***
I × N	-	-	-	-	-	-	-	-	-	-	-	-
I × V	-	-	-	*	-	-	-	-	-	-	-	-
I × Y	-	-	-	-	*	-	-	-	-	***	*	**
N × V	-	-	***	-	-	-	-	-	-	*	-	-
N × Y	-	-	-	-	-	-	-	-	-	-	-	-
V × Y	-	-	-	-	-	-	-	-	-	-	-	-
I × N × V	-	-	-	-	-	-	-	-	-	-	-	-
I × N × Y	-	-	-	-	-	-	-	-	-	-	-	-
I × V × Y	-	-	-	-	-	-	-	-	-	-	-	-
N × V × Y	-	-	-	-	-	-	-	-	-	-	-	-

[†] ***, **, and * are significant 0.001, 0.01, and 0.05, respectively; - is nonsignificant.

Table 2-7. Main effects of year, irrigation, nitrogen (N) treatment, and variety on sucrose concentration, glucose concentration, and AGT score in tubers for both the stem and bud ends at zero months after harvest (T0) and after six months of storage (T6).

Sources of Variation	Sucrose				Glucose				Light reflectance after frying			
	Stem end		Bud end		Stem end		Bud end		Stem end		Bud end	
	T0	T6	T0[†]	T6[†]	T0[†]	T6	T0	T6	T0	T6[†]	T0[†]	T6[†]
Year	——————————— mg g^{-1} ———————————											
2010	1.52 a[‡]	0.93 a	2.6 2a	1.69 a	2.99 a	5.23 a	0.37 a	0.95 a	47.5b	36.5b	54.9a	43.1b
2011	1.24 a	0.36 b	2.06 b	0.85 b	3.10 a	3.59 b	0.48 a	0.74 b	49.9a	46.6a	53.3a	49.8a
Irrigation												
Unstressed	1.10 b	0.57 b	2.23 b	1.19 a	3.36a	4.93 a	0.44 a	0.92 a	47.9a	40.7b	53.2b	45.6b
Stressed	1.66 a	0.72 a	2.45 a	1.35 a	2.74 b	3.89 b	0.41 a	0.78 a	49.4a	42.4a	55.0a	47.3a
N Treatment[§]												
34 N early	1.82 a	0.63 a	2.90 a	1.25 a	4.31 a	5.89 a	0.7 0a	1.31 a	45.8b	38.9b	50.1b	43.5 c
180 N split	1.47 b	0.6 1a	2.31 b	1.25 a	2.97 b	4.43 b	0.44 b	0.93 b	48.4a	41.8a	54.9a	45.5b
270 N split	1.19 bc	0.66 a	2.20 b	1.27 a	2.51 c	3.78 cd	0.31 cd	0.59 c	49.8a	42.3a	55.5a	47.9a
270 N split + s	1.11 c	0.67 a	1.99 b	1.34 a	2.65 c	3.72 d	0.27 d	0.64 c	49.7a	42.4a	55.6a	48.5a
270 N early	1.30 bc	0.65 a	2.30 b	1.24 a	2.79 bc	4.21 bc	0.41 bc	0.77 bc	49.5a	42.3a	54.4a	46.9ab
Variety												
Russet Burbank	1.09 b	0.34 b	1.75 b	1.12 b	3.73 a	4.66 a	0.35 b	0.47 b	47.1b	41.7a	53.8a	48.2a
Alpine Russet	1.66 a	0.95 a	2.93 a	1.4 2a	2.36 b	4.16 b	0.50 a	1.22 a	50.2a	41.4a	54.4a	44.7b

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[†] Response variables that had at least one significant interaction; refer to Table 2-6 and text for explanation.

[‡] Means followed by the same letter are not significantly different (α=0.05).

[§] N rates are expressed in kg N ha^{-1}; refer to Table 2-2 for detailed rates and timing.

Chapter 3.

PLANT-BASED APPROACHES FOR TRACKING THE NITROGEN STATUS OF TWO POTATO VARIETIES THROUGHOUT THE SEASON

Chapter Summary

Petiole nitrate-nitrogen (NO_3-N) concentration is often used as a diagnostic test in potato (*Solanum tuberosum* L.) to determine the rate and timing of in-season nitrogen (N) fertilizer applications. Non-invasive sensing methods are also being investigated as a means to detect crop N stress; however, few studies have compared the relative abilities of these two approaches. A two year field study was conducted on a loamy sand soil to: (i) evaluate how total N and NO_3-N concentrations for petiole, leaflet, and whole leaf samples change throughout the season for Russet Burbank and Alpine Russet varieties, (ii) determine which plant tissues/N analysis techniques and growth stages can best predict Grade A tuber yield, and (iii) examine the ability of chlorophyll meter readings and remotely sensed narrowband spectral indices to predict leaf N concentrations, LAI, and Grade A tuber yield at different growth stages. Five N treatments were tested with varying rates and timing of N fertilizer. Total N and NO_3-N concentrations were measured in petiole, leaflet, and whole leaf tissue samples on several dates each year. Chlorophyll meter readings and three narrowband spectral indices (NDVI, NDI2, and SR8; from Cropscan) were also measured and their relationship with leaf N, LAI, and Grade A tuber yield were determined using linear regression analysis. The main effect of variety was statistically significant (α=0.05) for all measurements on most sample dates. Tissue samples analyzed for NO_3-N were very responsive to N fertilizer applications that occurred within about 7 days of the sampling date, and are therefore a good indicator of current plant N uptake. Alternatively, tissue samples analyzed for total N were more stable over sample dates, and appear to be a better indicator of past plant N uptake. These measurements have traditionally been used to predict current crop N needs; however, inaccurate sufficiency recommendations could lead to over- or under-application of N fertilizer. According to a linear regression analysis, the non-invasive measurements (particularly NDVI and NDI2 narrowband spectral indices) had a better relationship with Grade A tuber yield than any of the tissue samples throughout the season. The r^2 values for NDVI and NDI2 gradually increased as the season progressed; they ranged from 0.40-0.65 on the first sample date in each year, and ranged from 0.55-0.85 by the last sample date in each year. These results suggest that information such as biomass or leaf area index, as well as plant nutrient concentration, may be needed to accurately determine overall in-season crop N status for determination of in-season N fertilizer recommendations.

Keywords: Petiole, leaflet, leaf, nitrogen, nitrate-nitrogen, chlorophyll meter, leaf area index, NDVI, yield

1. Introduction

Minnesota ranks 7th in U.S. potato (*Solanum tuberosum* L.) production with about 18,000 ha of irrigated potato grown in 2010 (NASS, 2010). The sandy loam and loamy sand soils used for irrigated potato production in Minnesota are typically low in organic matter and cation exchange capacity, and therefore, have a relatively low reserve of soil nutrients. The low inherent soil fertility and the large nutrient requirement result in a high rate of fertilizer inputs (Dean, 1994). Because of the high nitrogen (N) fertilizer rates and high hydraulic conductivity soils used for irrigated potato, there is a high potential for nitrate-nitrogen (NO_3-N) leaching. In addition, potato plants are relatively shallow rooted compared with other field and vegetable crops; this feature results in below average nutrient uptake and poor nutrient use efficiency, which can ultimately lead to higher amounts of nitrate leaching (Lynch et al., 2012; Rosen and Bierman, 2008; Zebarth and Rosen, 2007; Zvomuya et al., 2003).

Groundwater nitrate concentrations have been found to be significantly higher in irrigated than non-irrigated cropland (Burkart and Stoner, 2008). Based on previous experiments on coarse textured soils in Minnesota, only about a third to a half of applied N is recovered by the potato crop in years of moderate to heavy leaching (Errebhi et al., 1998; Waddell et al., 2000). The goal of N management for irrigated potato production is to estimate the appropriate rate and timing of split applications of N fertilizer so that N availability matches crop N demands throughout the growing season; this will optimize tuber yields while minimizing environmental losses of N. However, managing N for potato is a challenge because crop N uptake rates and soil N transformations/losses depend on the interaction of many complicated (and sometimes unpredictable) factors throughout the growing season, including: fertilizer source, soil fertility, soil type/CEC, and weather conditions.

One strategy used to maximize N use efficiency is to minimize the potential for N to be lost from the root zone by split applying post-emergence N fertilizer. Matching the timing and rate of N fertilizer with the N needs of the crop at different growth stages will help to optimize crop uptake while minimizing N losses (Zebarth and Rosen, 2007; Canter, 1997; Errebhi et al., 1998). Increased NUE and yields have been shown to result from split applications of N fertilizer during tuber initiation and bulking, especially during leaching years (Errebhi et al., 1998; Westermann et al., 1988). The use of fertigation provides a convenient method to split apply post-emergence N fertilizer, but the challenge is to accurately estimate the appropriate rate and timing of split applications so that N supply best matches crop N demands.

A logical strategy for determining an appropriate rate and timing of split applied N fertilizer is to make adjustments based on in-season plant monitoring. Since this approach uses plant measurements during the growing season, it is able to account for the effect of seasonal weather conditions on crop N availability (Meisinger et al., 2008). Currently, a best management practice for potato production in Minnesota is to base the rate and timing of post-emergence N fertilizer applications on petiole NO_3-N concentrations (Rosen and Bierman, 2008) and is being implemented by many commercial growers. Petiole nitrate analysis is based on the concept that the petiole is the transport organ of N from the roots and stems to the leaflets (in the form of NO_3^- and amino acids), and is therefore a good indicator of current NO_3-N uptake by the plant. Wu et al. (2007) found that petiole NO_3-N concentrations were closely related to N treatment variations created by split applications of N fertilizer.

The opportunity exists to use non-invasive sensing methods (e.g., chlorophyll meter readings or spectral reflectance measurements) to predict crop biophysical parameters in order to account for within-field spatial variability for variable rate application of fertilizer. However, chlorophyll meter readings are measured from the potato leaflets alone, and canopy-level reflectance measurements depend very little on petioles since the leaflets account for almost all of the total leaf area at a canopy-scale (Goffart et al., 2008; Jongschaap and Booij, 2004). In order to use non-invasive measurements for in-season N fertilizer recommendations, it is important to determine the relationships between total N and NO_3-N concentrations for petiole, leaflet, and whole leaf tissue samples. This is because non-invasive sensing methods depend entirely or primarily on leaflet measurements and the current best management practice relies on petiole NO_3-N concentrations.

Research comparing total N concentration to NO_3-N concentration of tissue samples is limited for RB and has not been published for AR. The objectives of this study were: (i) to evaluate how total N and NO_3-N concentrations for petiole, leaflet, and whole leaf samples change throughout the season for Russet Burbank and Alpine Russet varieties, (ii) to determine which tissue samples/N analysis techniques and growth stages can best predict Grade A tuber yield, and (iii) to examine the ability of chlorophyll meter readings and narrowband spectral indices from Cropscan to predict leaf N concentrations, LAI, and Grade A tuber yield at different growth stages throughout the season.

2. Materials and methods

2.1. Study site

Field experiments were conducted over two years (2010-2011) at the University of Minnesota Sand Plain Research Farm (45°23'N, 95°53'W) near Becker, MN. The soil at this location is classified as an excessively drained Hubbard loamy sand (sandy, mixed, frigid Typic Hapludoll) comprised of 82% sand, 10% silt, and 8% clay in the top 15 cm. The available water holding capacity in the upper 150 cm of soil is 10 cm. Representative soil samples in the top 15 cm were collected in the spring before planting for routine soil tests (Brown, 1998); averaged over both years, pH, percent organic matter, Bray-P, and extractable K were 6.5, 1.8%, 37 mg kg^{-1}, and 105 mg kg^{-1}, respectively. The previous crop in both years was non-irrigated cereal rye (*Secale cereal* L.).

To determine pre-plant soil inorganic N, soil samples from the upper 60 cm were collected from each whole plot on 23 March 2010 and 6 April 2011. The samples were air-dried, ground to pass a 2-mm sieve, and analyzed for KCl extractable NO_3-N and NH_4-N using a Wescan N analyzer (Carlson et al., 1990). Pre-plant soil inorganic N (NH_4-N + NO_3-N) was 15 kg ha^{-1} in 2010 and 20 kg ha^{-1} in 2011.

2.2. Experimental design

This field study was set up as a randomized complete block design with a split-split plot restriction on randomization replicated four times. The whole plot treatment was irrigation rate, (i.e., unstressed and stressed). To evaluate the hypotheses in this particular study, only data from the unstressed plots were used. Therefore, the data in this experiment were analyzed using a randomized complete block design with a split plot restriction on randomization. Irrigation was applied with an overhead sprinkler system, and a water balance method was used to schedule the irrigation rate (Wright, 2002). Timing of irrigation was variable between years and depended on weather conditions. On average, approximately four days elapsed between irrigation applications.

The subplot treatment included a low, medium, and high N rate (i.e., 34, 180, and 270 kg N ha^{-1}) with variable timing of post-emergence N applications for the high rate, for a total of five N treatments (i.e., 34 N early, 180 N split, 270 N split, 270 N split + surfactant (s), and 270 N early; Table 3-1). On average, about two weeks passed between split applications of post-emergence N for the 180 N split, 270 N split, and 270 N split + s treatments. The 270 N split and 270 N split + s treatments had the same rate and timing of N application, but a soil surfactant (IrrigAid Gold) was applied to 270 N split + s at a rate of 10 L ha^{-1} to investigate the effects of the surfactant on soil moisture and N uptake under different irrigation regimes. The effects of the soil surfactant were not an objective of this particular study, and it actually had little or no statistical significance on the parameters measured. Therefore, the data from the 270 N split + s treatment were included in the dataset for this study. Because treatments 180 N split, 270 N split, and 270 N split + s had split applications of post-emergence N fertilizer, actual N applied at the time of data acquisition varied (Table 3-2). All N applications were completed by the last sample date in each year. In both years, the planting and emergence N source was mono-ammonium phosphate and urea, respectively. Post-emergence N was split applied four times by hand as a 1:1 mixture of urea and ammonium nitrate in 2010 and five times by spray boom and tractor as 28% urea-ammonium nitrate solution in 2011. All post-emergence N was irrigated in immediately following application.

The sub-subplot treatment consisted of two potato varieties, (i.e., Russet Burbank and Alpine Russet). Russet Burbank (RB), the most widely grown processing potato variety in the U.S. and in Minnesota, has a mid-season maturity class of 111-120 days after planting (Whitworth et al., 2011). Alpine Russet (AR) has a late-season maturity class of 121-130 days after planting, and is a relatively new variety grown in Minnesota.

Compared to RB, it has similar yields, superior processing quality after long-term storage, and greater resistance to water-stress induced tuber defects (Whitworth et al., 2011).

In both years, whole "B" seed and cut "A" seed were used for the RB and AR varieties, respectively. Seed was hand planted in furrows with 90 cm row spacing and approximately 30 cm spacing between seed pieces within rows. Each plot consisted of seven 13.7 m rows. Planting dates were 16 April 2010 and 29 April 2011 and plant emergence occurred on 15 May 2010 and 24 May 2011. Two days after plant emergence in each year, emergence fertilizer was applied and rows were mechanically hilled. Chemicals were applied as needed during the season for the control of pests, disease, and weeds according to standard practices in the region (Engel et al., 2012).

To estimate N supplied by precipitation and irrigation, water samples were collected. The concentrations of NO_3-N and NH_4-N from the samples were determined using a Wescan N analyzer. Following reduction of NO_3-N to NH_4-N using granular zinc, NO_3-N was determined as the difference between NH_4-N + NO_3-N and NH_4-N (Carlson, 1986; Carlson et al., 1990). Total N supplied by rainfall + irrigation was 53 and 41 kg N ha^{-1} in 2010 and 2011, respectively.

2.3. Plant sampling and field measurements

In-season field measurements were collected on four dates in 2010 and five dates in 2011 (Table 3-2), and included: tissue samples (petioles and leaflets); chlorophyll meter readings (Minolta SPAD-502, Spectrum Technologies, Plainfield, IL); and multispectral reflectance (MSR16R Cropscan, Cropscan, Inc., Rochester, MN, Serial no. 249).

The fourth leaf from the apex of the shoot was sampled from twenty plants in the fifth row from the alley in each treatment plot. Relative chlorophyll was immediately measured at a central point on the terminal leaflet between the midrib and leaf margin. The 20 measurements from each plot were averaged to represent a single value for each treatment plot. Immediately following relative chlorophyll measurements on each leaf, leaflets were stripped from the petiole and both the leaflets and the petioles were separately saved for analysis.

Leaflet and petiole samples were oven-dried at 60°C, each tissue part was weighed for dry matter yield, and then each was ground separately with a Wiley mill to pass a 2-mm sieve. Total N concentration in ground tissue samples was determined with a combustion analyzer (Elementar Vario EL III, Elementar Americas Inc., Mt. Laurel, NJ) following the methods of Horneck and Miller (1997). Following extraction with water, NO_3-N concentration was determined using a Wescan analyzer after reduction to NH_4 using granular zinc following the methods of Carlson (1986) and Carlson et al. (1990). To obtain total N and NO_3-N concentrations for the whole leaf, petiole and leaflet concentrations were first used to calculate N and NO_3-N content on a dry weight basis by multiplying dry matter by N or NO_3-N concentration; next, the leaf was reconstructed by combining the N or NO_3-N content of the petioles and leaflets. Finally, leaf N or NO_3-N content was calculated back to a concentration by dividing content by total dry weight of the leaf (leaflets plus petioles).

Ground-based reflectance was measured on the same days as the tissue samples. Scans were taken 2 m above the canopy to give an approximate field-of-view diameter of 1 m. On 1 July and 6 Aug. in 2010 (47 and 83 days after emergence [DAE], respectively), and on 6 July and 29 July in 2011 (43 and 66 DAE, respectively), thirteen subsamples were taken per treatment plot. Subsamples were taken over the third row from the alley at 1.5, 3.4, 5.2, 7.0, 8.8, 10.7, and 12.5 m alongside the plots, as well as over the fourth, fifth, and sixth rows at each end of the plot. On all other dates (31 and 59 DAE in 2010; and 30, 56, and 79 DAE in 2011), three subsamples were taken over the third row from the alley at 3.4, 7.0, and 10.7 m alongside the plots. Subsamples within each plot were averaged for further analysis. Percent reflectance was recorded at 16 narrowband wavelengths (10-14 nm bandwidth) through the visible and near-infrared regions; narrowbands were centered at 460, 510, 560, 610, 660, 710, 760, 810, 870, 900, 950, 1000, 1320, 1480, 1500, and 1720 nm.

Vines were mechanically killed on 10 Sept. 2010 (118 DAE) and 14 Sept. 2011 (113 DAE), and tubers were mechanically harvested from the third and fourth rows from the alley in each treatment plot approximately 1-2 weeks after the vines were killed. Tubers were sorted into weight classes for total and graded yield. Grade A yield was determined by subtracting undersized tuber yields (85 g) from the total yield.

Yield data from the study were analyzed using PROC MIXED (SAS Institute, 2008) with N treatments and variety considered as fixed variables and years and replications (nested within years) considered as random variables. For the main effects and interactions of the fixed variables, pairwise comparisons of the least square means were made using the *lsmeans / pdiff* option of the MODEL statement (α = 0.05). The PDMIX800 macro (Saxton, 1998) was used to place treatment means into letter groupings based on the pairwise comparisons. The interactions between the fixed effects and years were assessed by year-specific inference using best linear unbiased predictors (BLUPs) as described by Littell et al. (2006). Since random effects cannot be tested in the *lsmeans* statement, the *estimate* statement was used to make pairwise comparisons between treatment means for the interactions of the random variables (α = 0.05).

For tissue samples and LAI data, years were analyzed separately for each sampling date. Pairwise comparisons of the least square means were made using the *lsmeans / pdiff* option of the MODEL statement of the MIXED procedure of SAS (SAS Institute, 2008; α = 0.05). The PDMIX800 macro (Saxton, 1998) was used to place treatment means into letter groupings based on the pairwise comparisons.

2.4. Yield prediction

Linear regression analyses were performed between Grade A yield and the various tissue measurements/LAI using the REG procedure of SAS (SAS Institute, 2008). The coefficients of determination (r^2) were used to evaluate the ability of in-season field measurements to be used as an indicator of potato N status as it was reflected by Grade A tuber yield. Regression was performed separately for each date and variety.

Formulas for three narrowband spectral indices (NDI2, NDVI, and SR8; Table 3-3) were applied to the Cropscan data. Narrowband NDI2 and SR8 were chosen because they performed very well in predicting leaf N using canopy-level hyperspectral data from the same field plots (Nigon et al., 2012). In that analysis, there were also other narrowband indices that performed very well, but they could not be used in this study because all wavelengths needed for their calculation were not obtained by the Cropscan sensor.

Narrowband NDVI did not perform particularly well in that analysis, but it was included in this study for comparison with the other indices and previously published research.

Linear regression models were used to determine the relationships comparing leaf N concentration, LAI, and Grade A yield to that of chlorophyll meter readings and each of the spectral indices at different growth stages. Their dependence on leaf N concentration and LAI was determined by comparing their r^2 values for leaf N concentration with their r^2 values for LAI.

3. Results and discussion

3.1. Weather

Mean monthly rainfall and temperatures for the 2010 and 2011 growing seasons (April through September) are compared with 30 year averages from a weather station near Becker, MN (1971-2000; Midwest Regional Climate Center) in Table 3-4. In addition to the 80 cm of rainfall in 2010, 32 cm of irrigation was applied for a total of 112 cm of water. In 2011, 26 cm of irrigation was applied in addition to the 64 cm of rainfall for a total of 90 cm of water over the growing season. Overall, 2010 and 2011 were much wetter than the average growing season. In 2010, the average temperature over the growing season was 1.0 °C above average, but in 2011, it was 0.2 °C below average.

Because the timing of rainfall was different in each year, the timing of NO_3-N leaching was also likely different, thereby influencing available plant N differently in each year. In 2010, heavy rainfall occurred in June; in 2011, heavy rainfall occurred in May and July. In Central Minnesota for later maturing processing varieties, June can be primarily characterized by the vegetative and tuber initiation stages of potato growth, and July can be primarily characterized by the tuber bulking stage. Nitrogen uptake rates are highest at the end of tuber initiation and throughout tuber bulking, and N stress during these stages has different effects on potato development (Ojala et al., 1990). Nitrogen stress before the end of tuber initiation (e.g., in 2010) can reduce leaf area and stimulate early tuber bulking, and N stress before the end of tuber bulking (e.g., in 2011) can reduce tuber bulking rate and ultimately reduce tuber yield and size (Ojala et al., 1990).

3.2. Tuber yield

Grade A tuber yield was assessed for each year and variety (Table 3-5) since their main effects were statistically significant (α=0.05; analysis of variance not shown). Nitrogen treatments significantly affected yield; in every case, the 34 N early treatment always resulted in significantly less yield than the other N treatments, and the 270 N split and 270 N split +s treatments always had the highest statistically significant yields. However, the treatments with the 270 kg N ha^{-1} split rate were statistically similar to 180 N split in both years for AR, and were statistically similar to both 180 N split and 270 N early in 2011 for RB. The 270 N early treatment had significantly lower yields than the treatments with the 270 kg N ha^{-1} split rate in both years for AR and in 2010 for RB. Errebhi et al. (1998) and Westermann et al. (1988) reported similar yield results with split applications of N fertilizer, especially during leaching years. In 2010 for RB, a lower yield was observed for the 180 N split treatment than for any of the treatments with the 270 kg N ha^{-1} rate.

3.3. In-season plant tissue analysis

Petiole, leaflet, and whole leaf total N and NO_3-N concentrations were assessed by variety (Fig. 3-1 and Fig. 3-2, respectively) since the main effect of variety was statistically significant for most tissue samples on most sample dates (α=0.05; analysis of variance not shown). The only exceptions to this occurred with some of the tissue samples for the main effect of variety on the early sample dates in each year (i.e., 31 DAE in 2010 and 30 DAE in 2011) and the latest sample date in 2011 (i.e., 79 DAE). The significant effect of variety observed in this study was different from that observed by Williams and Maier (1990), who observed that the effect of variety on critical nutrient ranges for NO_3-N were not significant for Atlantic, Coliban, and Kennebec varieties. As the rate of applied N fertilizer increased (Tables 3-1 and 3-2), total N and NO_3-N concentrations of tissue samples also increased (Figs. 3-1 and 3-2). When the main effect of variety was significant for tissue samples, RB usually had higher total N and NO_3-N concentrations than AR. It is likely that the differences in petiole, leaflet, and whole leaf total N and NO_3-N concentration between varieties in the present study is because AR is slower to emerge and is a longer season variety than RB (Whitworth et al., 2011). Assuming each variety in this study had similar relative N sufficiency over the growing season (in terms of maximizing tuber yield), the difference in total N and NO_3-N concentration between varieties suggests that absolute sufficiency ranges for total N and NO_3-N concentrations vary by variety. Absolute sufficiency ranges should be slightly lower for AR than for RB, especially before ~50 DAE.

Throughout the season, the patterns in total N and NO_3-N concentrations between leaf and leaflet data were very similar since a large proportion of the leaf dry weight consisted of the leaflet dry weight and very little consisted of the petiole dry weight. Tissue N and NO_3-N concentrations for all N treatments were highest on the first sample date in each year (i.e., 31 DAE in 2010 and 30 DAE in 2011). After the first sample date, tissue N concentrations for the N treatments that had post-emergence N split applied were stable throughout the season (Fig. 3-1). Tissue NO_3-N concentrations for the N treatments that had post-emergence N split applied were also relatively stable after the first sample date, although not as stable as tissue N concentrations (Fig. 3-2). Alternatively, tissue N and NO_3-N concentrations for the 270 N early treatment generally continued to decrease as the season progressed; this decrease was likely due to a high rate of soil NO_3-N leaching. Because of the high rainfall totals in June and July (Table 3-4), the 2010 and 2011 growing seasons in central Minnesota had high leaching losses. Decreased N use efficiencies and increased NO_3-N leaching rates have been shown to result from early applications of N fertilizer when compared to split applications of post-emergence N fertilizer, especially during heavier leaching years (Errebhi et al., 1998; Westermann et al., 1988). Regardless of variety, year, or tissue sample, N and NO_3-N concentrations for 270 N early fell below the 270 N split treatments after ~56 DAE and before ~66 DAE (Figs. 3-1 and 3-2). By the last sample date in each year, N and NO_3-N concentrations for 270 N early also fell below the 180 N split treatment in most cases. This did not occur for

AR tissue N in 2011 or for RB leaflet NO_3-N in 2010 (the tissue concentrations for the 270 N early and 180 N split N treatments were statistically similar to each other in these cases).

Several reports have recommended N nutrient sufficiency levels for potatoes based on leaf N and/or petiole NO_3-N concentration (Table 3-6). Not all recommendations were consistent with each other, and they were not usually variety-specific. For comparison purposes in this study, however, we assume that leaf N sufficiency levels are 40 g kg^{-1} at the vegetative stage (Mills & Jones, 1996) and 35 g kg^{-1} at the tuber bulking stage (Rosen & Eliason, 2005; Westermann, 1993); these sufficiency levels are highlighted in gray in Fig. 3-1. Based on these values, we observe that many of the N treatments are at or above the N sufficiency levels, including the 34 N early treatment in many cases. Alternatively, for petiole NO_3-N, we assume sufficiency levels to be 17,000 mg kg^{-1} at the vegetative stage, 11,000 mg kg^{-1} at the tuber bulking stage, and 6000 mg kg^{-1}at the maturation stage (Rosen & Eliason, 2005); these sufficiency levels are highlighted in gray in Fig. 3-2. According to the petiole NO_3-N sufficiency levels, all of the N treatments in this study are below or barely meet the minimum N sufficiency levels on most measurement dates. The discrepancy in N sufficiency between these two laboratory analysis techniques for tissue samples collected in this study is very obvious, and could easily lead to divergent N fertilizer recommendations. This discrepancy could be due to inaccurate recommendations for either leaf N or petiole NO_3-N, but could also be due to NO_3-N leaching and the timing of sampling in relation to the last N fertilizer application. Petiole NO_3-N concentration can be so sensitive that significant variability has been shown from differences in sample times during the day (Zhang et al. 1996).

The days between N fertilizer applications relative to sampling dates are listed in Table 3-2. Tissue NO_3-N concentrations were more dynamic than tissue N concentrations with respect to the time that passed between the last split application of N fertilizer and the sampling date. In other words, tissue NO_3-N was more responsive to change than tissue N in the days immediately following post-emergence N applications (e.g., within seven days). For example, on 59 DAE in 2010 and 79 DAE in 2011, tissue samples were collected within about 7 days after the last N fertilizer application. On these dates, tissue NO_3-N concentrations increased from the previous sampling date whereas tissue N concentrations remained about the same or decreased from the previous sampling date. This observation occurred for all tissue samples, but it was especially apparent for the leaflet and leaf samples.

Once soil NO_3-N is absorbed by the roots of potato plants, it may be either reduced and synthesized into amino acids while being stored in the root tissue, or may be transported across the root and deposited in the xylem for movement toward the shoots (Haynes, 1986). Portions of both nitrate and amino acids can be transported to the stem and petiole cells; the bulk of this is transported further into the leaflets where it is mostly reduced to ammonium and stored as amino acids (Haynes, 1986). Therefore, the stems and petioles are the plant organs that transport recently absorbed N, and the leaflets are the plant organs that store N. This signifies that petiole NO_3-N concentration is a good indicator of the current plant uptake of N, and leaf or leaflet total N is a good indicator of the total plant uptake of N over the season thus far. For N fertilizer recommendations based on in-season tissue samples, the data from this study suggest that both the growth stage and the number of days that passed since the last N application are important considerations if using tissue NO_3-N analysis, but only the growth stage is important to consider if using total N analysis.

Leaflet and leaf total N concentrations for the 34 N early treatment were significantly lower than all other N treatments for both varieties on all sample dates (Fig. 3-1). In contrast, tissue NO_3-N concentrations from 180 and 270 N treatments anytime on or after ~43 DAE in both years were similar to the 34 N early treatment (Fig. 3-2). The primary reason for this was that NO_3-N concentrations for the 34 N early treatment did not change significantly over the season and the NO_3-N concentrations for the other N treatments decreased as the season progressed. This resulted in tissue NO_3-N concentrations from the the other N treatments becoming statistically similar to those of the 34 N early treatment as the growing season progressed, especially for the 180 N split and 270 N early treatments on the later sample dates (270 N early concentrations were likely affected by NO_3-N leaching). Tissue NO_3-N concentrations for the 34 N early treatment were nearly 0 mg kg^{-1} at every growth stage, and as the season progressed, the other N treatments approached this 0 mg kg^{-1} concentration, especially if N fertilizer was not split applied. Alternatively, total N concentrations for the 34 N early treatment tended to continually decrease over the season, and there was usually always a significant difference between the 34 N early treatment and the other N treatments. Biemond and Vos (1992) also observed this relationship between leaf N and leaf NO_3-N for the Bintje variety. These differences can be compared visually in Figs. 3-1 and 3-2; the tissue samples analyzed for NO_3-N concentration (Fig. 3-2) tended to have "fan shaped" patterns among N treatments over sample dates whereas the tissue samples analyzed for total N concentration (Fig. 3-1) behaved similarly among N treatments over sample dates (i.e., the rate at which total N decreased over the season was relatively similar between 34 N early and the other treatments).

When using tissue samples for determining N fertilizer recommendations, growers are not usually concerned about extreme N stress conditions since they do not typically occur on commercial farms. Instead, it is more important to be able to detect early differences in areas of the field that have small or moderate N stress levels. To test the ability of tissue samples to detect low amounts of N stress in this experiment, tissue N and NO_3-N analyses for the 180 N split and 270 N early treatments were directly compared to the same analyses for the 270 N split treatments (data not shown). Among the tissue samples analyzed for both N and NO_3-N concentration, there were no clear advantages of one approach over another at detecting differences in the small to moderate N stress situations.

3.4. Leaf area index

The main effect of N was highly significant for leaf area index (LAI) on all sample dates; the main effect of variety was significant on all sample dates except 47 DAE in 2010 and 66 DAE in 2011. For simplicity, LAI values are presented for both varieties (Fig. 3-3). On most sample dates, LAI for the 34 N early treatment was statistically lower than all other N treatments. LAI values for the remaining N treatments were variable, however; on most sample dates, there was no statistical difference in LAI for these remaining N treatments. Some of the variability in LAI values between N treatments and over sample dates may be due to the limitations of the meter used to measure LAI.

3.5. In-season N stress prediction

Leaf area index, as well as total N and NO_3-N in petiole, leaflet, and whole leaf samples, were related to Grade A tuber yield using linear regression models on each sample date (Fig. 3-4). It is important to note that the relationships between in-season plant measurements and yield should be interpreted with caution because yield can be affected by stresses that occur anytime during the season, including stresses that occur after in-season measurements. In this study, we assume that any external stress that occurred during the season occurred in all experimental treatments; therefore, differences in the relationships between in-season plant measurements and yield can be directly compared, and any differences can be primarily attributed to the response of the treatment on each respective sample date. None of the sample dates showed consistent relationships between tissue samples/LAI and Grade A yield over both varieties and/or both years. Leaflet and whole leaf N generally had higher r^2 values over sample dates than petiole N or any of the tissue samples analyzed for NO_3-N. Petiole NO_3-N concentration, however, had relatively similar or slightly higher r^2 values on the earliest sample date in each year (i.e., 31 DAE in 2010 and 30 DAE in 2011). Compared to the other tissue samples, leaflet NO_3-N had poor r^2 values on almost every sample date. The r^2 values for LAI were statistically significant on most sample dates, but there was no sample date over both years or both varieties that clearly showed a good relationship with Grade A yield. Overall, the tissue samples and LAI did not do a good job at consistently predicting Grade A tuber yield; among sample dates, results were variable over year and variety.

3.5.1. Non-invasive methods for predicting leaf N

There are many non-invasive sensing methods available for predicting canopy structure parameters such as plant biomass, LAI, leaf chlorophyll, and plant N content/concentration (Duchenne et al., 1997). Among these non-invasive sensing methods, the chlorophyll meter is the most widely used, and research on potato has shown that it can be effective at identifying the need for in-season adjustments to N fertilizer rates and timing (Gianquinto et al., 2004; Ledent et al., 2006; Vos and Bom, 1993; Wu et al., 2007). The chlorophyll meter predicts relative chlorophyll based on the transmittance of light energy through leaflets of potato plants at 650 nm and 940 nm; these wavelengths correspond to the red and near-infrared regions of the electromagnetic spectrum, respectively (Goffart et al., 2008). Another non-invasive sensing method that is still under investigation for N stress predictions is the use of hand-held multispectral radiometers (e.g., Cropscan). Cropscan measures canopy-level spectral reflectance at as many as to 16 user-defined wavelengths. This allows for the calculation of spectral indices that can be used for N stress prediction (Goffart et al., 2008).

Chlorophyll meter readings and three narrowband spectral indices (i.e., NDVI, NDI2, and SR8; calculated from Cropscan measurements; Table 3-3) were measured and used to predict leaf N concentration, LAI, and Grade A tuber yield as determined by the r^2 values from linear regression models (Fig. 3-5). In general, the chlorophyll meter readings and all three of the indices had a good relationship with leaf N concentration at all sample dates for both varieties. The r^2 values representing the relationship between leaf N and chlorophyll meter readings tended to improve as the season progressed. By the last sample date in each year, the r^2 values were near or above 0.80. The r^2 values representing the relationships between leaf N concentration and each of the indices generally started high and decreased as the season progressed. The r^2 values for the chlorophyll meter readings were lower than the r^2 values of the best indices on the first sample date in each year and each variety, but they were higher than the r^2 values of the best indices by the last sample date in both years and both varieties. This can possibly be attributed to interference from background soil for the spectral measurements. The chlorophyll meter readings were not influenced by LAI/canopy closure because they depend only on measurements from individual leaflets. Alternatively, since canopy-level spectral measurements are influenced by many external factors other than leaf chlorophyll content, such as the effect of soil background (Daughtry et al., 2000), the relationships between leaf N concentration and spectral data were likely negatively affected, especially on the later sampling dates when the crop began to senesce. Overall, NDI2 had the best relationship with leaf N concentration on the early to mid sampling dates, and the chlorophyll meter readings had the best relationship with leaf N concentration on the late sampling dates.

3.5.2. Non-invasive methods for predicting LAI

The r^2 values for the relationship between LAI and the indices/chlorophyll meter readings were not as high as they were for leaf N concentration, and they were variable over sample dates (Fig. 3-5). For RB, at 59 DAE in 2010 and 56 DAE in 2011, neither the chlorophyll meter nor any of the indices were significantly different. For the most part, the r^2 values among the indices were similar at each growth stage, and the r^2 values for the chlorophyll meter were either similar or lower than the r^2 values for the indices.

3.5.3. Non-invasive methods for predicting Grade A tuber yield

Overall, the spectral indices (especially NDVI and NDI2) predicted Grade A tuber yield better than the chlorophyll meter (Fig. 3-5). The r^2 values for NDVI and NDI2 gradually increased as the season progressed; they ranged from 0.40-0.65 at the first sample date in each year, and ranged from 0.55-0.85 by the last sample date in each year. The r^2 values for the chlorophyll meter varied with each variety throughout the growing season. For RB, the r^2 values were highest on the early sample dates, and then they decreased and were lowest by the last sample date. For AR, the r^2 values were lowest on the first sample date, they peaked mid-season (between ~47 DAE and ~56 DAE), and then decreased on the later sample dates. This difference between varieties can possibly be attributed to AR being slower to emerge and having a longer season than RB (Whitworth et al., 2011).

Overall, spectral indices such as NDVI and NDI2 predicted Grade A tuber yield better than tissue N or NO_3-N concentrations. This suggests that plant nutrient measurements alone (e.g., leaf N or petiole NO_3-N) may not be best suited to detect N stress in a potato crop as it relates to Grade A tuber yield. Perhaps measurements that combine plant nutrient concentration and biomass/leaf area index have a better potential to predict crop

N stress for in-season N fertilizer recommendations than nutrient measurements alone. Two invasive plant measurements that do this are plant N content and the calculation of the Nitrogen Nutrition Index (NNI). The NNI is calculated as the ratio between current N concentration in the dry biomass and the critical N concentration. The critical N concentration is defined as the minimum N concentration necessary to achieve maximum growth rate (Ulrich, 1952), and is calculated based on the dry biomass of the crop and certain estimated parameters (Goffart et al., 2008). Averaged over three sampling dates, Bélanger et al., (2001) reported an R^2 of 0.71 for the relationship between NNI and yield for an irrigated Russet Burbank potato crop. The authors did not report an r^2 value for the relationship between leaf N concentration and yield and therefore, the two invasive sampling techniques cannot be directly compared.

Canopy-level spectral data likely has a very good relationship with the NNI since they are each influenced by the effects of both plant N concentration and biomass/LAI (Daughtry et al., 2000). If this is indeed the case, canopy-level spectral data would be better suited to be a predictor of crop N stress for in-season fertilizer recommendations since it is a non-invasive measurement and could be easily measured at a precise scale so that within-field spatial variability is accounted for.

4. Conclusions

This two year study showed that narrowband spectral indices such as NDVI or NDI2 can predict Grade A tuber yield better than plant tissue measurements which are currently considered as the best management practice for determining the rate and timing of in-season N fertilizer applications (Rosen and Bierman, 2008). Petiole NO_3-N concentration is very responsive to N fertilizer applications that occur within about 7 days of the sampling date, and is therefore a good indicator of current plant N uptake. However, since petiole NO_3-N concentrations depend on growth stage as well as the amount of days that have passed since the last N application, petiole NO_3-N concentration may not be the best indicator of overall crop N status. Nitrate-nitrogen concentrations were always higher in petioles than whole leaves or leaflets, and total N was always higher in whole leaves and leaflets than petioles. In general, this indicates that the diagnostic criteria should change based on the tissue part sampled and the form of N tested. Across growth stages, whole leaf N was able to predict Grade A tuber yield much better than petiole NO_3-N.

Plant nutrient concentration alone was not able to adequately detect in-season N stress in a potato crop as it relates to Grade A tuber yield. Instead, data that is influenced by biomass/LAI in addition to plant nutrient concentration (e.g., canopy-level spectral reflectance data) was best suited to determine overall crop N status. Future research should be conducted: (i) using more N fertilizer rates and timings than those used in this study, (ii) to determine the relationship between canopy-level spectral data and the Nitrogen Nutrition Index (NNI), and (iii) to evaluate the variability of spectral data over growth stages, years, and varieties so feasibility of N recommendations can be determined.

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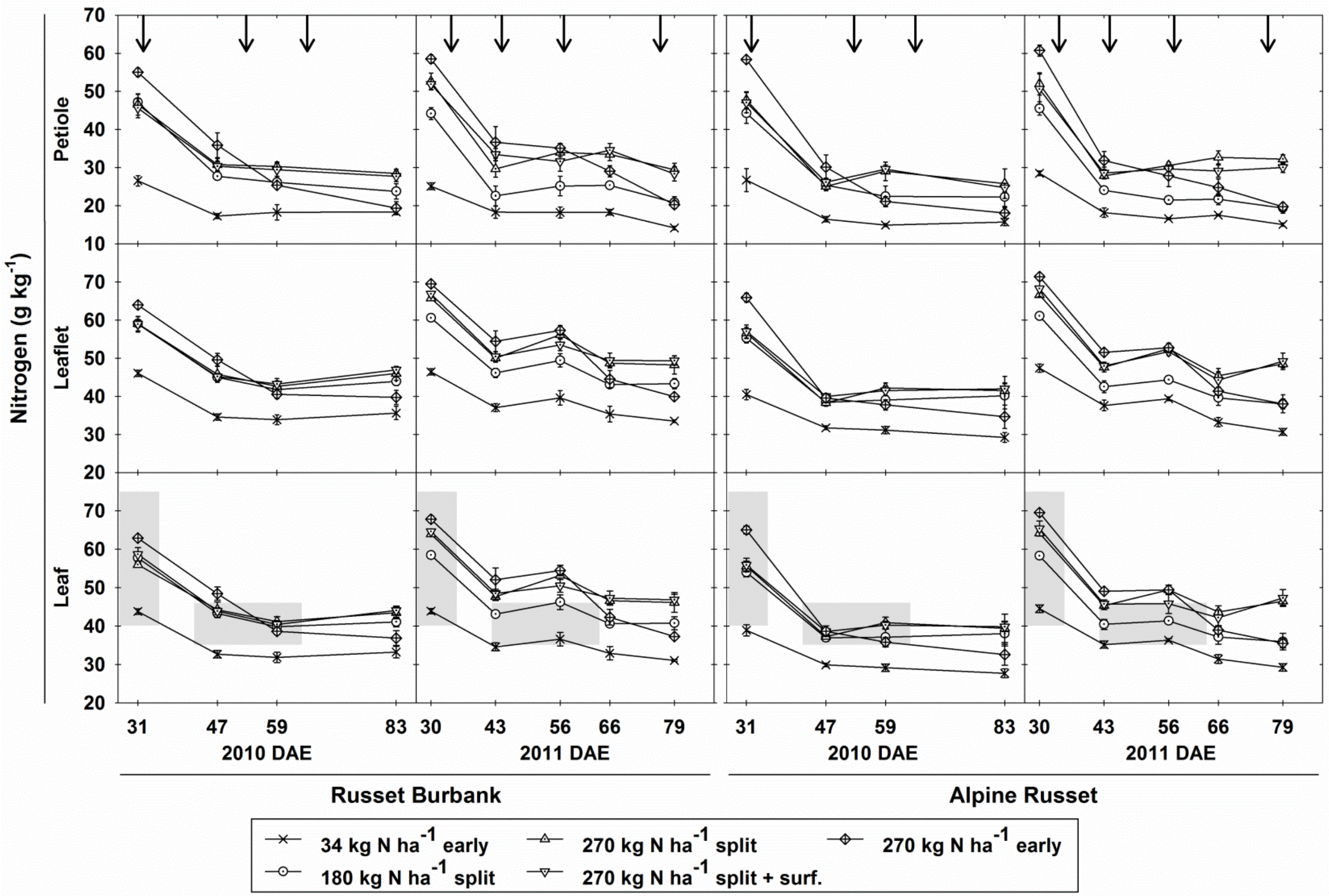

Fig. 3-1. Petiole, leaflet, and whole leaf nitrogen (N) concentrations under various N treatments for Russet Burbank and Alpine Russet potato varieties on different days after emergence (DAE) in 2010 and 2011. Means are presented with standard error bars, and gray shaded areas represent leaf N sufficiency levels according to Rosen & Eliason (2005) and Westermann (1993). Arrows signify when split applications of post-emergence N fertilizer were applied.

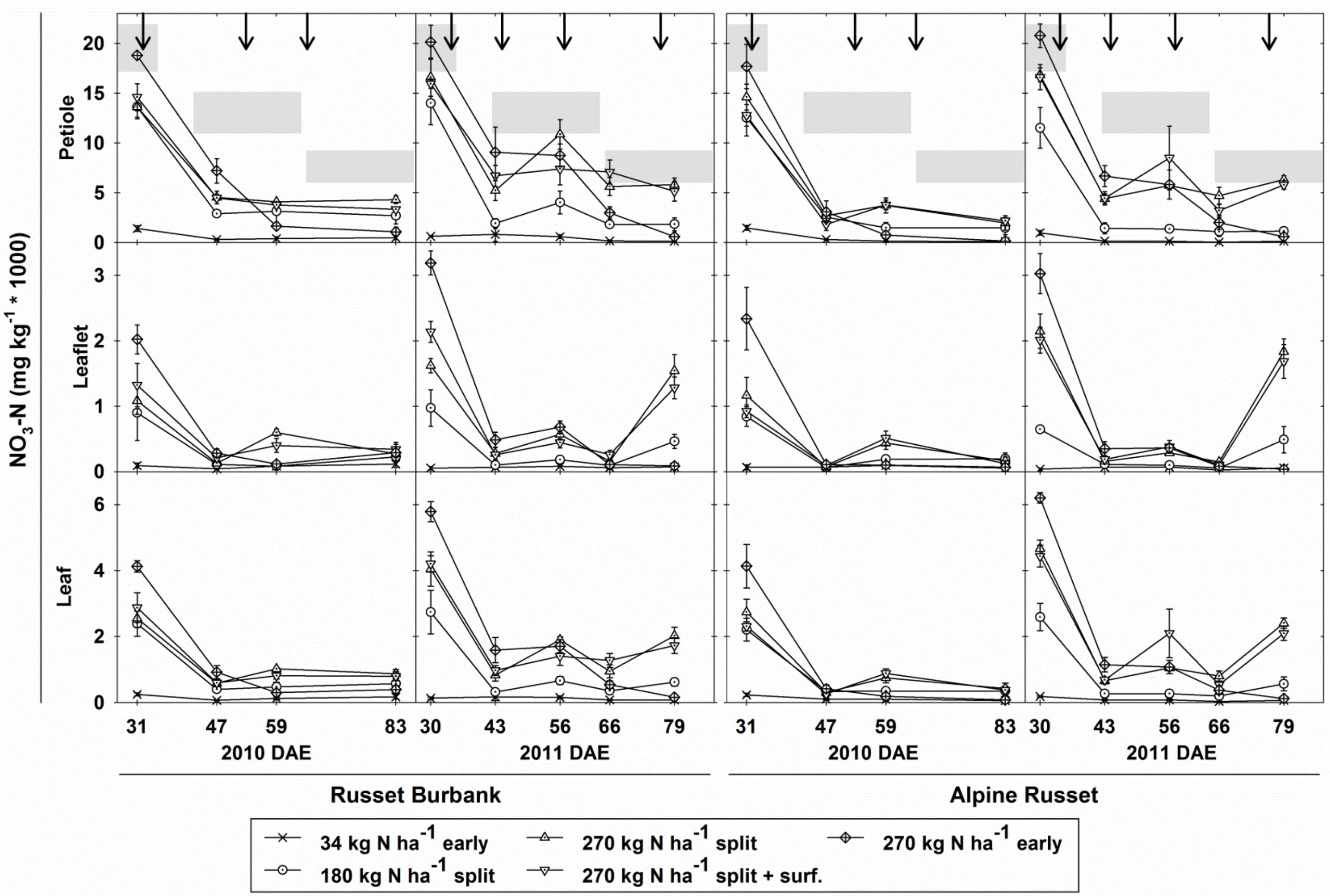

Fig. 3-2. Petiole, leaflet, and whole leaf nitrate-nitrogen (NO_3-N) concentrations under various N treatments for Russet Burbank and Alpine Russet potato varieties at different days after emergence (DAE) in 2010 and 2011. Means are presented with standard error bars, and gray shaded areas represent petiole NO_3-N sufficiency levels according to Rosen & Eliason (2005). Arrows signify when split applications of post-emergence N fertilizer were applied.

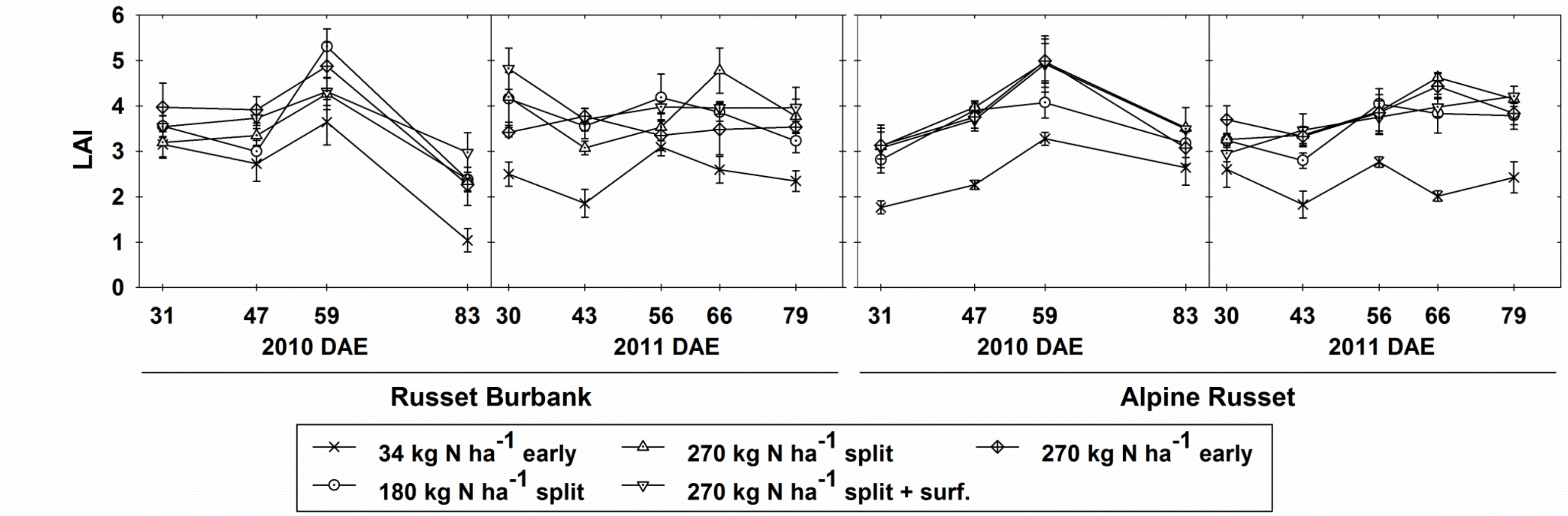

Fig. 3-3. Leaf area index (LAI) under various N treatments for Russet Burbank and Alpine Russet potato varieties at different days after emergence (DAE) in 2010 and 2011. Means are presented with standard error bars.

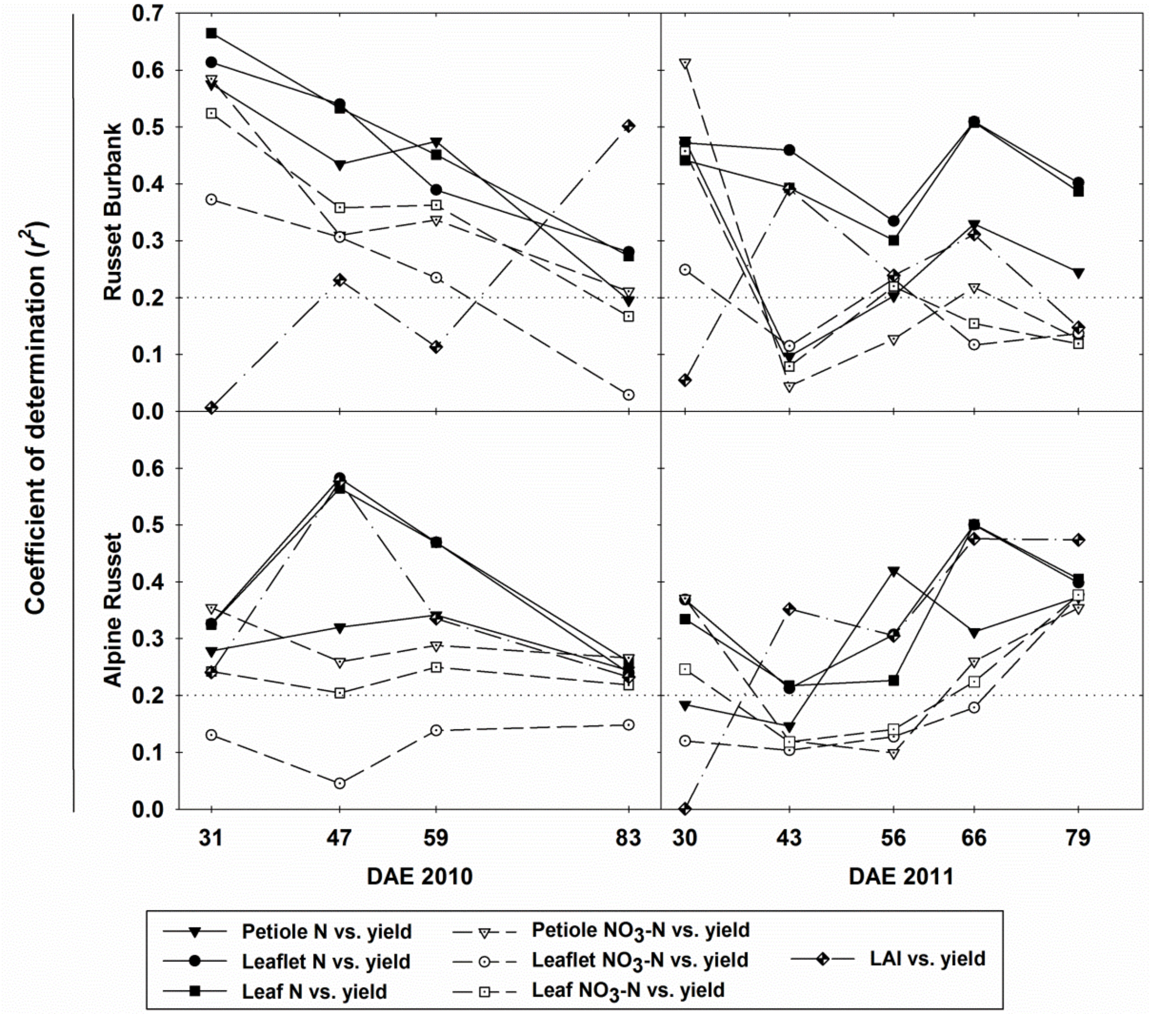

Fig. 3-4. Coefficient of determination (r^2) as a function of days after emergence (DAE) for the relationship with Grade A tuber yield. Values below the dotted line are not statistically significant (α =0.05).

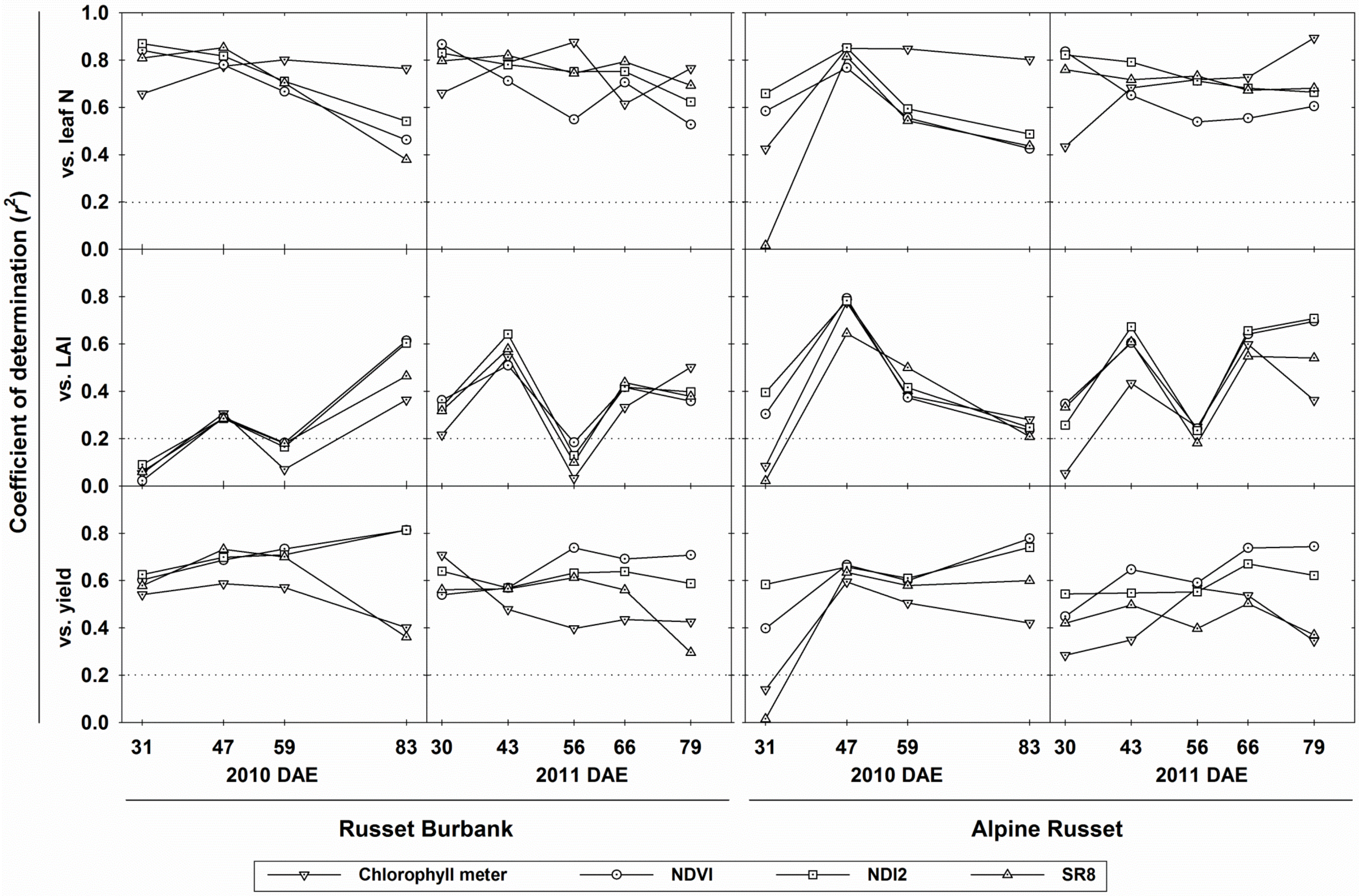

Fig. 3-5. Coefficient of determination (r^2) for the relationships between each ground truth measurement (leaf N concentration [top], leaf area index [LAI; middle], and Grade A tuber yield [bottom]) and predictor (chlorophyll meter readings, NDVI, NDI2, and SR8) as it varies with days after emergence (DAE). Values below the dotted line are not statistically significant (α =0.05).

Table 3-1. Rate and timing of nitrogen (N) fertilizer treatments for 2010 and 2011.

Nitrogen fertilizer treatment	Timing of application					Total N
	planting	emergence		post-emergence		
		2010	2011	2010	2011	
	----------------------------	kg N ha^{-1}	----------------------------			
34 N early	34	0	0	0	0	34
180 N split	34	78	90	17 × 4[‡]	11.2 × 5[‡]	180
270 N split	34	124	124	28 × 4[‡]	22.4 × 5[‡]	270
270 N split + s[†]	34	124	124	28 × 4[‡]	22.4 × 5[‡]	270
270 N early	34	124	124	112	112	270

[†] A soil surfactant (IrrigAid Gold) was used in this treatment, and was applied at a rate of 10 L ha^{-1}.

[‡] On average, about two weeks passed between split applications of post-emergence N.

Table 3-2. Actual nitrogen (N) fertilizer applied at the time of data acquisition for N treatments with split applications of post-emergence fertilizer.

Year	DAE	Growth stage	Days after last N application	Days before next N application	N Treatment	
					180 N split	270 N split
					-- kg N ha^{-1} --	
2010	31	Vegetative	12	1	129	186
	47	Tuber bulking	15	6	146	214
	59	Tuber bulking	6	6	163	242
	83	Maturation	18	-	180	270
2011	30	Vegetative	14	4	135	180
	43	Tuber bulking	9	1	146	203
	56	Tuber bulking	12	1	158	225
	66	Maturation	9	10	169	248
	79	Maturation	3	-	180	270

Table 3-3. Spectral indices evaluated in this study.

Index	Name		Formula	Developed for	Developed by
NDI2	Normalized difference index 2	=	$(R_{870} - R_{710})/(R_{870} - R_{660})$	Chlorophyll	Datt (1999)
NDVI	Normalized difference vegetation index	=	$(R_{810} - R_{660})/(R_{810} + R_{660})$	Structure	Lichtenthaler et al. (1996)
SR8	Simple ratio 8	=	$R_{870}/(R_{560} \times R_{710})$	Chlorophyll	Datt (1998)

†R_i = reflectance at wavelength i (nanometers)

Table 3-4. Mean monthly rainfall and air temperature data for 2010 and 2011 growing seasons and the 30-year mean.

Month	Rainfall			Temperature		
	2010	2011	30-year mean	2010	2011	30-year mean
	--------------	cm	--------------	--------------	°C	--------------
April	4.6	6.9	6.0	11.6	6.2	7.6
May	6.6	16.8	8.2	14.9	13.1	14.8
June	26.2	7.8	11.3	19.0	18.8	19.3
July	7.1	22.5	10.5	22.6	23.9	21.7
August	15.0	8.3	11.7	22.7	20.7	20.4
September	20.1	1.5	7.3	14.0	15.2	15.3

Table 3-5. Grade A tuber yield under different N treatments in 2010 and 2011 for Russet Burbank and Alpine Russet potato varieties. Different letters signify differences between N treatments within year and variety.

N treatment	Grade A tuber yield (Mg ha^{-1})			
	Russet Burbank		Alpine Russet	
	2010	2011	2010	2011
34 early	31.6 d[†]	27.5 b	42.5 c	34.3 c
180 split	45.4 c	42.1 a	53.4 ab	48.6 a
270 split	51.5 a	43.1 a	55.0 a	49.2 a
270 split + s	50.2 ab	41.0 a	54.6 a	49.1 a
270 early	48.7 b	41.8 a	50.9 b	42.6 b

[†] Means followed by the same letter within a measurement date and variety are not significantly different (α=0.05).

Table 3-6. Optimum N sufficiency levels for leaf total nitrogen (N) and petiole nitrate-nitrogen (NO_3-N) concentrations by growth stage and variety. Information is based on recommendations from previous publications.

Growth stage	Variety	Optimum N sufficiency	Reference
Total N		------- $g\ kg^{-1}$ -------	
Tuber bulking[†]	"Many" different varieties	>35	Westermann (1988)
Vegetative[‡]	-[§]	40-60	Mills and Jones (1996)
Tuber bulking[‡]	-[§]	30-40	Mills and Jones (1996)
-[§]	-[§]	35-45	Rosen and Eliason (2005)
NO_3-N		------ $mg\ kg^{-1}$ ------	
Vegetative[†]	-[§]	17000-22000	Rosen and Eliason (2005)
Vegetative[†]	Russet Burbank	>22000	Gardner and Jones (1975)
Tuber initiation[†]	Atlantic, Coliban, and Kennebec	27000-30000	Williams and Maier (1990)
Tuber bulking[†]	-[§]	11000-15000	Rosen and Eliason (2005)
Tuber bulking[†]	Russet Burbank	>15000	Westermann (1988)
Tuber bulking[†]	Russet Burbank	>19000	Gardner and Jones (1975)
Tuber bulking[†]	Atlantic, Coliban, and Kennebec	18000-23000	Williams and Maier (1990)
Maturation[†]	-[§]	6000-9000	Rosen and Eliason (2005)
Maturation[†]	Atlantic, Coliban, and Kennebec	10000-16000	Williams and Maier (1990)
Maturation[†]	Russet Burbank	>14000	Gardner and Jones (1975)

[†] Tissue samples were collected from the most recent fully-developed leaf.

[‡] Tissue samples were collected from the 25 most recent fully-developed leaves.

[§] Not indicated

Chapter 4.

HYPERSPECTRAL IMAGERY FOR DETECTING NITROGEN STRESS IN TWO POTATO VARIETIES

Chapter Summary

In order to use remotely sensed spectral data for determining rates and timing of variable rate nitrogen (N) applications at a commercial scale, the most reliable indicators of crop N status must be determined. This study evaluated the ability of chlorophyll meter readings and spectral indices/models to predict N stress in potatoes based on measurements of leaf N concentration to be used for variable rate application of N fertilizer. The measurements and spectral indices/models that were evaluated include: (i) a list of 17 broadband and 82 narrowband previously published spectral indices; (ii) partial least squares regression (PLS) models using reflectance spectra, first derivative reflectance spectra, and both as independent variables; and (iii) chlorophyll meter readings. Two hyperspectral images were acquired with an AISA-Eagle hyperspectral camera in both 2010 and 2011. Each year, experimental treatments included five N treatments with varying rates and timing of N fertilizer and two potato varieties, Russet Burbank (RB) and Alpine Russet (AR). Based on the coefficient of determination (r^2) and the root mean squared error of cross-validation (RMSECV), the best PLS models could predict leaf N concentration the best, and the chlorophyll meter readings and the best narrowband indices could predict leaf N concentration better than the best broadband indices. Because canopy-level hyperspectral images can predict leaf N concentration at a similar or better accuracy than chlorophyll meter readings, they can be much more advantageous for variable rate application since canopy-level images can account for spatial variation whereas chlorophyll meter readings cannot. Applying the NSI formula to plant measurements and spectral indices/models made them mostly insensitive to the effects of variety; normalizing data to a reference value is useful for properly interpreting spectral measurements among varieties. Based on analyses in this study, the best predictor of N stress was determined to be the PLS model using derivative reflectance as input for its independent variables (r^2 = 0.79, RMSECV = 14% for RB; r^2 = 0.77, RMSECV = 13% for AR). However, the best technique for determining N stress level for variable rate application of N fertilizer using the NSI was determined to be MTCI because the combination of its good r^2 values, RMSECVs, and accuracy assessment. Results from the means separation of the NSIs suggest that spectral indices/models may be a better predictor of N stress than plant tissue samples since spectral data is affected by both tissue N concentration and leaf area index.

Keywords: Hyperspectral imagery, chlorophyll meter, broadband indices, narrowband indices, partial least squares regression, nitrogen sufficiency index (NSI), accuracy assessment, coefficient of variation, nitrogen, potato

1. Introduction

Potato (*Solanum tuberosum* L.) is an important crop worldwide, ranking first in 2010 among all other non-cereal food crops with over 324 million Mg produced (FAOSTAT, 2010). The coarse textured soils typically used for irrigated potato production are relatively low in organic matter and cation exchange capacity, and therefore, are generally low in soil nutrient reserves. The low inherent soil fertility of these soils, coupled with the large nitrogen (N) requirement for a potato crop leads to high fertilizer inputs (Dean, 1994). Potato plants are relatively shallow rooted compared to other field and vegetable crops and are very sensitive to water stress (Bailey, 2000; Lesczynski & Tanner, 1976). This leads to below average nutrient uptake and poor nitrogen use efficiency (NUE), which potentially results in high rates of nitrate leaching (Lynch et al., 2012; Rosen & Bierman, 2008; Zvomuya et al., 2003). Groundwater nitrate concentrations are often reported to be significantly higher in irrigated than non-irrigated cropland (Burkart & Stoner, 2008). Previous studies have shown that only about a third to a half of applied N is recovered by an irrigated potato crop in years of moderate to heavy leaching (Errebhi, Rosen, Gupta, & Birong, 1998; Waddell et al., 2000). Therefore, to optimize production and to minimize environmental N losses for irrigated potato production, precise N management is important.

One strategy used to maximize NUE by minimizing the quantity of N lost from the root zone is to apply N fertilizer in split applications. Matching the timing and rate of N fertilizer with the N needs of the crop during different growth stages will help to optimize crop uptake while minimizing N losses (Canter, 1997; Errebhi et al., 1998). Increased NUE and yields have been shown to result from split applications of N fertilizer during tuber initiation and bulking, especially during leaching years (Errebhi et al., 1998; Westermann et al., 1988). The use of fertigation provides a convenient method to split apply post-emergence N applications. The challenge lies in the ability of the producer to estimate the appropriate rates and timing of split N applications so that fertilizer N best matches crop demands. This is particularly a challenge because crop uptake rates and soil N transformations/losses depend on the interaction of many complicated (and sometimes unpredictable) factors throughout the growing season, including: fertilizer source, soil fertility, soil type/CEC, and weather conditions.

A logical strategy for determining the exact rate and timing of post-emergence N applications is to make adjustments based on in-season plant monitoring. Since this approach uses plant measurements during the growing season, it is able to account for the effect of seasonal weather conditions on crop N availability (Meisinger et al., 2008). The current best management practice for potato production in Minnesota is to base the rate and timing of post-emergence N fertilizer applications on petiole nitrate-nitrogen (NO_3-N) concentrations (Rosen & Bierman, 2008).Threshold sufficiency levels (for RB) are 17,000, 11000, and 6000 mg kg^{-1} NO_3-N for the early vegetative, mid tuber growth, and tuber bulking/maturation stages, respectively (Rosen & Bierman, 2008).

The opportunity exists to use remote sensing to predict crop biophysical parameters that depend on crop N uptake such as tissue N or NO_3-N concentration and/or leaf chlorophyll concentration. Hyperspectral imagery is a powerful research tool that can provide information necessary to determine the best techniques to monitor crop N status over a variety of conditions and growth stages. Hyperspectral data acquired during the growing season can be used to monitor crop N status because spectral characteristics of green-leaved vegetation change as leaf chlorophyll content changes, and N is closely related to chlorophyll in plant cell metabolism (Stroppiana et al., 2012). Hundreds of spectral indices have been developed with the aim of predicting particular biophysical parameters while minimizing the effects of solar irradiance and soil background (Jackson & Huete, 1991; Rees, 2001). Chemometric models that use all hyperspectral wavebands (e.g., partial least squares regression or principal components analysis) have been found to predictive crop biophysical parameters very well, especially for leaf N concentration (Cohen et al., 2010; Hansen & Schjoerring, 2003; Nguyen et al., 2006).

Research on aerial based hyperspectral imagery for N sufficiency in potatoes, specifically using chemometric models to make variable rate management decisions under various cultural conditions, has not been published. The objectives of this study were: (i) to evaluate the ability of chlorophyll meter readings, narrowband and broadband indices, and partial least squares regression (PLS) models to predict leaf N concentration in two potato varieties; (ii) to evaluate the ability of chlorophyll meter readings and spectral indices/models to classify nitrogen sufficiency index (NSI) stress levels into pre-determined stress classes via an accuracy assessment, and (iii) to evaluate the differences in variability within and across experimental treatments for petiole NO_3-N and leaf N concentration, chlorophyll meter readings, and spectral indices/models after being normalized by an NSI.

2. Materials and methods

2.1. Study site

Field experiments were conducted over two years (2010-2011) at the University of Minnesota Sand Plain Research Farm (45°23’N, 95°53’W) near Becker, MN. The soil at this location is classified as an excessively drained Hubbard loamy sand (sandy, mixed, frigid Typic Hapludoll) comprised of 82% sand, 10% silt, and 8% clay. The available water holding capacity in the upper 150 cm of soil is 10 cm. During the growing season (April – September), the 30-year average (1971-2000) temperature and rainfall are 16.5° C and 550 mm, respectively (Midwest Regional Climate Center). The previous crop in both years was non-irrigated cereal rye (*Secale cereal* L.).

To determine pre-plant soil inorganic N, soil samples from the upper 60 cm were collected from each whole plot on 23 March and 6 April in 2010 and 2011, respectively. The samples were air-dried, ground to pass a 2-mm sieve, and analyzed for KCl extractable NO_3-N and NH_4-N using a Wescan N analyzer (Carlson et al., 1990). Pre-plant soil inorganic N (NH_4-N + NO_3-N) was 15 kg ha^{-1} in 2010 and 20 kg ha^{-1} in 2011.

2.2. Experimental design

This field study was set up as a randomized complete block design with a split-split plot restriction on randomization replicated four times. The whole plot treatment was irrigation rate (i.e., unstressed and stressed); to evaluate the hypotheses in this particular study, only data from the unstressed plots were used. Therefore, the data in this experiment were analyzed using a randomized complete block design with a split plot restriction on randomization.

The subplot treatment included a low, medium, and high N rate (i.e., 34, 180, and 270 kg N ha^{-1}) with variable timing of post-emergence N applications for the high rate, for a total of five N treatments (i.e., 34 early, 180 split, 270 split, 270 split + surfactant (s), and 270 early; Table 4-1). On average, about two weeks passed between split applications of post-emergence N for treatments 180 split, 270 split, and 270 split + s. Treatments 270 split and 270 split + s had the same rate and timing of N application, but a soil surfactant (IrrigAid Gold) was applied to 270 split + s at a rate of 10 L ha^{-1} to investigate the effects of the surfactant on soil moisture and N uptake under different irrigation regimes; although determining the effects of the soil surfactant was not an objective of this part of study, data from the 270 split + s N treatment were included in the dataset since the soil surfactant had little or no statistical significance on the parameters measured. Because treatments 180 split, 270 split, and 270 split + s had split applications of post-emergence N fertilizer, actual N applied at the time of data acquisition varied (Table 4-2). All N applications were completed by the later image date in 2010, but one post-emergence N application remained at the later image date in 2011. In both years, the planting and emergence N source was mono-ammonium phosphate and urea, respectively. Post-emergence N was split applied four times by hand as a 1:1 mixture of urea and ammonium nitrate in 2010 and five times by spray boom and tractor as 28% urea-ammonium nitrate solution in 2011. All post-emergence N was irrigated in immediately following application.

The sub-subplot treatment consisted of two potato varieties (i.e., Russet Burbank and Alpine Russet). Russet Burbank (RB), the most widely grown processing potato variety in the U.S. and in Minnesota, has a mid-season maturity class of 111-120 days after planting (Whitworth et al., 2011). Alpine Russet (AR) has a late-season maturity class of 121-130 days after planting, and is a relatively new variety grown in Minnesota. Compared to RB, it has similar yields, superior processing quality after long-term storage, and greater resistance to water-stress induced tuber defects (Whitworth et al., 2011).

In both years, whole “B” seed and cut “A” seed were used for the RB and AR varieties, respectively. Seed was hand planted in furrows with 90 cm row spacing and approximately 30 cm spacing between seed pieces within rows. Each plot consisted of seven 13.7 m rows. Planting dates were 16 April 2010 and 29 April 2011 and plant emergence occurred on 15 May 2010 and 24 May 2011. Two days after plant emergence in each year, emergence fertilizer was applied and rows were mechanically hilled. Chemicals were applied as needed during the season for the control of pests, disease, and weeds according to standard practices in the region (Engel et al., 2012).

To estimate N supplied by precipitation and irrigation, water samples were collected. The concentrations of NO_3-N and NH_4-N from the samples were determined using a Wescan N analyzer. Following reduction of NO_3-N to NH_4-N using granular zinc, NO_3-N was determined as the difference between NH_4-N + NO_3-N and NH_4-N (Carlson, 1986; Carlson et al., 1990). Total N supplied by rainfall + irrigation was 53 and 41 kg N ha^{-1} in 2010 and 2011, respectively.

2.3. Aerial image acquisition

Aerial hyperspectral imagery was acquired with an Airborne Imaging Spectrometer for Applications (AISA Eagle) visible/near-infrared hyperspectral imaging sensor (SPECIM, Spectral Imaging, Ltd., Oulu, Finland) by the Center for Advanced Land Management Information Technologies (CALMIT) from the University of Nebraska-Lincoln, USA. The AISA Eagle is a complete pushbroom system, consisting of a hyperspectral sensor head, a miniature GPS/INS sensor, and a data acquisition unit in a rugged PC with a display unit and power supply. It has a 1,000 pixel swath width and was configured to capture imagery in 63 narrowbands (2.3 nm spectral resolution) covering the visible and near-infrared portions of the solar spectrum from 401 to 982 nm; bandwidths ranged from 8.8 - 9.6 nm.

Images were captured from approximately 1900 m above the ground between 10 a.m. and 3 p.m. local time on 1 July 2010, 6 August 2010, 6 July 2011, and 29 July 2011; in the experimental year, these dates corresponded to 47, 83, 43, and 66 days after plant emergence (DAE), respectively. The second image date in 2010 was at a later growth stage than the second image date in 2011; this was due to equipment problems and weather-related scheduling conflicts, which resulted in apparent differences in the data that were likely due to a difference in actual crop maturity rather than a difference between years.

2.4. Image processing

A post-processing software package, CaliGeo, was used for radiometric correction (using NIST traceable calibrations), rectification (using a C-Migits III GPS/INS unit manufactured by Systron Donner Inertial Division, Walnut Creek, CA, USA), and geo-referencing. Geographic coordinates of plot corners were acquired with a GPS unit (0.3 m accuracy), which allowed in-house geo-referencing to minimize image distortion. The spatial resolution of the rectified images was 1.0 m. ENVI software (Version 4.8, Exelis, Inc., USA) was used for all subsequent image analysis.

2.5. Plant sampling and field measurements

Tissue samples (petioles and leaflets) were collected on 1 July and 5 August in 2010 and 7 July and 28 July in 2011; these dates were on the same day or within one day of image acquisition. The fourth leaf from the apex of the shoot was sampled on twenty plants in the fifth row from the alley in each treatment plot. Relative chlorophyll (SPAD-502, Spectrum Technologies, Plainfield, IL) was immediately measured at a central point on the terminal leaflet between the midrib and leaf margin. The twenty measurements from each plot were averaged to represent a single value for each treatment plot and were recorded. Following chlorophyll readings, leaflets were stripped from the petiole, and then both the leaflets and the petioles were saved separately for analysis.

Leaflet and petiole samples were oven-dried at 60°C, each tissue part was weighed for dry matter yield, and then each was ground separately with a Wiley mill to pass a 2-mm sieve. Total N concentration in ground tissue samples was determined with a combustion analyzer (Elementar Vario EL III, Elementar Americas Inc., Mt. Laurel, NJ) following the methods of Horneck and Miller (1997). Following extraction with water, NO_3-N concentration was determined using a Wescan analyzer after reduction to NH_4 using granular zinc following the methods of Carlson (1986) and Carlson et al. (1990). To obtain total N concentrations for the entire leaf, petiole and leaflet N concentrations were first used to calculate N content on a dry weight basis by multiplying dry matter by N concentration; next, the leaf was reconstructed by combining the N content of the petioles and leaflets, and finally, leaf N content was calculated back to a concentration.

Tubers were mechanically harvested from the third and fourth rows from the alley on 28 September 2010 and 22 September 2011 (i.e., 165 and 146 days after planting in 2010 and 2011, respectively). Tubers were weighed on a fresh weight basis to determine total tuber yield.

Leaf N concentration data were analyzed using PROC MIXED (SAS Institute, 2008) with N treatments and variety considered as fixed variables and replications considered as random variables. For the main effects and interactions of the fixed variables, pairwise comparisons of the least square means were made using the *lsmeans / pdiff* option of the MODEL statement ($\alpha = 0.05$). The PDMIX800 macro (Saxton, 1998) was used to place treatment means into letter groupings based on the pairwise comparisons.

Yield data were analyzed using PROC MIXED (SAS Institute, 2008) with N treatments and variety considered as fixed variables and years and replications (nested within years) considered as random variables. For the main effects and interactions of the fixed variables, pairwise comparisons of the least square means were made using the *lsmeans / pdiff* option of the MODEL statement of the MIXED procedure of SAS (SAS Institute, 2008; $\alpha = 0.05$). The PDMIX800 macro (Saxton, 1998) was used to place treatment means into letter groupings based on the pairwise comparisons. The interactions between the fixed effects and years were assessed by year-specific inference using best linear unbiased predictors (BLUPs) as described by Littell et al. (2006).

2.6. Spectral data

In this study, four types of spectral data were used to evaluate the ability of canopy-level reflectance to be used as an indicator of N stress: (i) multispectral broadband and hyperspectral narrowband reflectance; (ii) first derivative reflectance of hyperspectral narrowbands; (iii) selected published broadband and narrowband indices; and (iv) predicted values from partial least squares regression based on narrowband reflectance and/or first derivative reflectance.

Broadband data were obtained by aggregating hyperspectral data into four broad spectral regions: blue (427-518 nm), green (508-572 nm), red (657-695 nm), and NIR (770-828 nm). The first derivative reflectance was calculated for a given wavelength as the slope of the reflectance between neighboring bands.

2.6.1. Spectral indices

Reflectances for all broadband and narrowband wavelengths, first derivative reflectances for all narrowband wavelengths, and a comprehensive list of 17 broadband and 82 narrowband previously published spectral indices were used in a preliminary analysis to determine the wavelengths and/or indices that had the best overall ability to detect N stress. The correlation coefficient (*r*) was calculated for each of these wavelengths/indices using the CORR procedure of SAS (SAS Institute, 2008) in order to determine the wavelengths/indices that had the best relationships with leaf N concentration on each image date. To determine which wavelengths and indices performed best overall, r^2 was averaged for each wavelength/index among the four image dates. The indices that had the best average r^2 from this preliminary analysis are listed in Table 4-3, and were used for comparison in all subsequent analysis techniques for detecting N stress. There were no broadband/narrowband wavelengths or first derivative wavelengths that performed particularly well across all image dates. Narrowband NDVI did not perform particularly well, but it is included for comparison with other indices and previously published research.

2.6.2. Partial least squares regression

Partial least squares regression (PLS) is a method for constructing predictive models that is especially useful when there are many independent variables which are highly collinear (Phatak & Jong, 1997). PLS uses data compression by finding a few new factors (often called *latent variables* or *components*) that will play a similar role as the many independent variables. Each latent variable (LV) is computed as certain linear combinations of the amplitudes of the original independent variables. The LVs then use the relevant structural information from the many independent variables to linearly predict the response variables. Tobias (1995) provides a detailed discussion of the basic PLS concepts and a description of statistical options, while Yeniay and Goktas (2002) describe the underlying PLS algorithm and compare it to other prediction methods.

Three PLS models were constructed across image dates, and three PLS models were constructed for each variety by image date combination, each using different sets of independent spectra. Independent spectra for the three models included: (i) hyperspectral reflectance only; (ii) first derivative reflectance only; and (iii) both hyperspectral and first derivative reflectance. The independent spectra were used to predict leaf N concentration in the models. There were 20 samples used as input for each of the models constructed within image dates, and 80 samples used as input for each of the models constructed across image dates.

The PLS models were calibrated and cross-validated by a full one-at-a-time cross-validation using the PLS procedure of SAS (SAS Institute, 2008). This option calibrated the PLS models iteratively using leaf N and spectral data from all treatment plots except for one; in each iteration, a different sample was left out from the dataset until every sample had been left out once. Using the *CVTEST (STAT = PRESS)* option of the PLS procedure of SAS (SAS Institute, 2008), model predictions were made for the least number of LVs in which the predicted residual sum of squares (PRESS) were not significantly greater than those of the model with the minimum PRESS. The PRESS is used as an indication of the predictive power of a model, and is calculated according to Eq. (4-1). It simulates prediction by repeatedly fitting the model, while leaving out the observation that it is attempting to predict each time the model is fit (Méndez Mediavilla et al., 2008), similar to the procedures used for the 'one-at-a-time' cross-validation of the PLS model discussed above. Since the observed samples y_i are not used in fitting the model, the predicted values $\hat{y}_{(i)}$ and their residuals are completely independent of y_i. This independence of the predicted residuals enables the PRESS to be a true assessment of the prediction capabilities of the model (Méndez Mediavilla et al., 2008).

$$PRESS = \sum_{i=1}^{n}(y_i - \hat{y}_{(i)})^2 \qquad \text{Eq. (4-1)}$$

where PRESS is the predicted residual sum of squares; y_i is the observed value of sample i; $\hat{y}_{(i)}$ is the predicted value of sample i not included in the model formulation; and n is the total sample number.

2.7. Pixel extraction

To extract pixel data from individual treatment plots for subsequent analysis, regions of interest (ROIs) were created for each treatment plot. Pixels that were most influenced by the effects of bare soil were semi-automatically excluded from being used in data analysis; this was done by including pixels in the ROIs only if they were above a minimum threshold when tested against a narrowband Greenness index (Smith et al., 1995) image. This technique effectively filtered out pixels that were most influenced by bare soil during growth stages with full canopy cover (e.g., pixels representing border plants). The number of pixels included in each ROI ranged from 15 to 78 among each of the four image dates; each pixel represented an area of 1.0 m^2, and each treatment plot had an area of ~88 m^2.

2.8. Data analysis

Spectral data were always kept in image format until it was necessary to perform statistical procedures outside of ENVI. Several analysis techniques were performed and were used as metrics to evaluate the spectral indices/wavelengths that were best able to determine N stress. These analysis techniques included: (i) linear regression analysis; (ii) normalized nitrogen sufficiency; (iii) calculation of the coefficient of variation across N treatments, and (iv) accuracy assessment.

2.8.1. Prediction analyses

A linear regression analysis with leaf N concentration was performed for chlorophyll meter readings and spectral indices/models using the REG procedure of SAS (SAS Institute, 2008) in order to obtain the coefficient of determination (r^2). The root mean squared error of cross-validation (RMSECV) was calculated from the PRESS according to Eq. (4-1) and Eq. (4-2), and is therefore a measure of a model's ability to predict new samples. The RMSECV was used together with r^2 to compare the predictive capability of the spectral data (RMSECV values were normalized by calculating the ratio between RMSECV values and the range of predicted values). The measurements and spectral indices/models with the highest r^2 and lowest RMSECV were considered to be the best. Prediction analyses for chlorophyll meter readings and spectral indices/models were performed within and across image dates so they could be more precisely evaluated.

$$RMSECV = \sqrt{\frac{PRESS}{n}} \qquad \text{Eq. (4-2)}$$

where RMSECV is the root mean squared error of cross-validation; *PRESS* is the predicted residual sum of squares (Eq. (4-1)); and *n* is the total sample number.

2.8.2. Normalized nitrogen sufficiency

A nitrogen sufficiency index (NSI) was applied to the plant measurements and spectral indices/models in order to normalize them for comparative purposes. By applying an NSI to the spectral data, values were normalized to a non-limiting N reference area (Peterson et al., 1996). The NSI was calculated using the higher average replicate value of either 270 split or 270 split + s as the reference according to Eq. (4-3). For NG and TCARI/OSAVI$_{(705,\ 750)}$, the index values representing the treatments with the most N stress were higher than those representing the treatments with the least N stress; therefore, the reciprocal of the NSI was used for these indices. Before applying the NSI formula for DD/MSAVI and TCARI/OSAVI$_{(705,\ 750)}$, a value of 2 was added to all pixels since these indices yielded negative values for some pixels.

$$NSI = \frac{N_i}{N_{ref(270\ split)}} \times 100 \qquad \text{Eq. (4-3)}$$

where NSI is nitrogen sufficiency index; N_i is the measured value of plant measurements, spectral indices, or PLS predictions from the non-reference treatment plot; and $N_{ref(270\ split)}$ is the reference value – the higher average replicate value between 270 split and 270 split + s was used.

On each image date, an analysis of variance was conducted for the plant measurements and several selected indices/models that were normalized by an NSI (Eq. (4-3)). Data were analyzed as a randomized complete block design with four replications and a split plot restriction on randomization using the MIXED procedure of SAS with Type III ANOVA method at α=0.05 (SAS Institute, 2008). N treatment and variety were considered as fixed effects, while replication was considered as random. The PDMIX800 macro (Saxton, 1998) was used to place treatment means into letter groupings based on the pairwise comparisons. Each image date was analyzed separately.

2.8.3. Coefficient of variation

The coefficient of variation (CV) was calculated to estimate the relative amount of variability present in plant measurements and selected spectral indices/models; the CV is useful because it helps to compare measurements that have different mean values and ranges. In this study, the CV describes the amount of relative variability present among N treatments for a particular plant measurement or spectral index/model; those with larger CVs have a greater range of values among N treatments. The relative fluctuation (RF) in CVs among image dates was calculated to determine the plant measurements and spectral indices/models that had the most consistent variability among image dates (Eq. (4-4)). Petiole NO_3-N concentration was included in the CV analysis because the best management practice for potato production in Minnesota is to base post-emergence N fertilizer applications on petiole NO_3-N (Rosen & Bierman, 2008); this gave us the opportunity to directly compare petiole NO_3-N to other potential plant or spectral measurements. Because TCARI/OSAVI$_{(705,\ 750)}$ and DD/MSAVI yielded negative values for some pixels, their CVs were not valid; transformations applied in order to correct this inherently resulted in an underestimation of the actual variability present for these indices.

$$RF = \frac{CV_{max} - CV_{min}}{CV_{avg}} \times 100 \qquad \text{Eq. (4-4)}$$

Where RF is relative fluctuation; CV_{max} and CV_{min} are the maximum and minimum coefficient of variation values among image dates, respectively; and CV_{avg} is the average coefficient of variation among image dates.

2.8.4. Accuracy assessment

An accuracy assessment was performed in order to evaluate the quality of chlorophyll meter readings and spectral indices/models to be able to classify N stress levels into pre-defined ranges. NSI values for leaf N concentration and spectral data were divided into three classes (Table 4-4), which represented data from the N treatments in Table 4-1. A confusion matrix was constructed to calculate the overall accuracy and Cohen's kappa coefficient (k) for the chlorophyll meter readings and spectral indices/models; the principal diagonal entries represented the correct classification. The proportion of the total number of correctly classified instances among the total number of instances in the confusion matrix represented the 'overall accuracy' of the classification. To accommodate for the effects of chance agreement, the coefficient k was calculated based on the overall accuracy and the proportion of units expected by chance agreement (Eq. (4-5); Cohen, 1960). The percentage of cases in which the predicted N stress class was higher and lower than the N stress class as determined by leaf N concentration (i.e., percentage of classifications in which N stress was overestimated and underestimated, respectively) was reported. The percentage of incorrect classifications by two class levels (i.e., high cost errors) was also reported.

$$k = \frac{N\sum_{i=1}^{r} x_{ii} - \sum_{i=1}^{r}(x_{i+} \times x_{+i})}{N^2 - \sum_{i=1}^{r}(x_{i+} \times x_{+i})} \qquad \text{Eq. (4-5)}$$

where k is Cohen's kappa coefficient; r is the number or rows and columns in the confusion matrix; x_{ii} is the number of instances along the principal diagonal; x_{i+} is the marginal total in row i; x_{+i} is the marginal total in column i; and N is the total number of observations (Tso & Mather, 2009).

3. Results and discussion

3.1. Leaf N concentration and tuber yield

Nitrogen treatment and variety significantly affected leaf N concentration on all image dates (Table 4-5); therefore, differences in leaf N are reported among N treatments for each of the image dates and each variety (Table 4-6). As the rate of applied N fertilizer increased (refer to Tables 4-1 and 4-2), leaf N also increased. Leaf N was more sensitive to N treatments in 2011 than in 2010, and was higher in RB than in AR across all N treatments and image dates. In both varieties, leaf N was above the threshold sufficiency level (i.e., 35 g kg^{-1}; Westermann, 1993) on all image dates for 270 split and 270 split + s. Leaf N for 180 split was statistically similar to 270 split and 270 split + s for both varieties and both image dates in 2010, but was lower than 270 split and 270 split + s for both varieties and both image dates in 2011. Leaf N was consistent from the first to the second image date in each year for all N treatments that had post-emergence N split applied. Alternatively, for 270 early, leaf N dramatically dropped off on the second image date for both varieties and both years.

Total tuber yield was assessed for each of the N treatments and in both years of the study (Table 4-7); the main effect of variety was not statistically significant ($\alpha=0.05$). Overall, tuber yield was higher in 2010 despite lower leaf N levels. The N treatments that had post-emergence N split applied had the highest tuber yields numerically among N treatments. In 2011, 270 early had a statistically lower yield than 270 split, despite similar overall N rates. Errebhi et al. (1998) and Westermann et al. (1988) reported similar yield results with split applications of N fertilizer, especially during leaching years.

3.2. Spectral data

A major limitation to the widespread use of remote sensing for making N fertilizer recommendations is the knowledge of spectral algorithms that are reliable over a variety of conditions (e.g., soil types, growth stages, cultivars, weather, etc.; Samborski et al., 2009). By understanding the behavior of spectral data over a range of conditions, coefficients can be determined and used in an attempt to standardize the relationship between conditions. For this reason, spectral data in this study were primarily evaluated based on their ability to detect N stress on each image date and for each variety.

Hyperspectral data provide many opportunities for analysis that do not exist for multispectral data, especially for identifying and modeling plant biophysical parameters such as N stress (Aspinall et al., 2002). In this study, narrowband indices showed a slight improvement in r^2 and RMSECV over broadband indices (Tables 4-8 and 4-9); further improvement was observed when the entire spectra was used via PLS models.

3.2.1. Reflectance and first derivative spectra

The reflectance spectra and its first derivative for a potato crop under conditions of low (34 kg N ha^{-1} early) and high (270 kg N ha^{-1} early) N fertilizer application rates are shown in Fig. 4-1. Reflectance is greatest in the green (i.e., ~500-600 nm) and near-infrared regions (i.e., ~750-1000 nm) and lowest in the red region (i.e., ~600-680 nm).

These are the regions of the spectrum that the spectral reflectance values differed the most between the low and high N rates. This is because these regions are most influenced by leaf chlorophyll content (Jongschaap & Booij, 2004), and were therefore indirectly related to leaf N concentration in this study. A decrease in reflectance in the visible regions of the spectrum for the high N rate is a consequence of greater absorption by chlorophyll; an increase in reflectance in the near-infrared region of the spectrum for the high N rate is a consequence of more scattering in the mesophyll cells (Goffart et al., 2008; Rees, 2001).

Peaks in the derivative spectra occur at the green- (i.e., ~527 nm) and red-edge regions (i.e., ~732 nm; Fig. 4-1). These peaks show distinct differences between the low and high N rates. Also, the position of the peak shifts to a shorter wavelength for the low N rate. Two local peaks have been observed in derivative spectra of vegetation at the red-edge (Chen et al., 2010; Le Maire et al., 2004; Zarco-Tejada et al., 2002). The shift in the position of the peak to a shorter wavelength occurs as a consequence of lower leaf area index and/or chlorophyll content (Filella & Peñuelas, 1994). Data from this study show two distinct peaks for the low N rate (at ~704 and ~722 nm), but only one obvious peak (at ~732 nm) for the high N rate. The relatively large bandwidth (~9 nm) of the spectral data from this study is the likely reason for only being able to observe one peak for the high N rate.

3.2.2. Partial least squares regression

Partial least squares regression models were developed using the reflectance and/or the first derivative reflectance spectra as inputs for its independent variables. The number of extracted latent variables (LVs) for each of the PLS models that used data across image dates were as follows: RB, reflectance spectra only = 3; RB, derivative spectra only = 2; RB, both reflectance and derivative spectra = 2; AR, reflectance spectra only = 4; AR, derivative spectra only = 2; AR, both reflectance and derivative spectra = 3. For the PLS models constructed using data within image dates, all except four used only one extracted LV; the models that used two extracted LVs were: (i) RB, 66 DAE, both reflectance and derivative spectra; (ii) AR, 43 DAE, reflectance spectra only, (iii) derivative spectra only, and (iv) both reflectance and derivative spectra (refer to the superscripts in Tables 4-8 and 4-9).

Since the final predictive function calibrated to estimate the response is a linear combination of the spectral data, the different LVs can be defined by their respective loading weights. The loading weights quantify the contribution of different wavelengths to the final model based on centered and scaled predictors, which allows the model to achieve an optimal fit. The loading weights for the LVs of each of the PLS models constructed across image dates are shown in Fig. 4-2. The models/parts of the models that used reflectance spectra as the input for the predictors (Figs. 4-2a, 4-2c, 4-2d, and 4-2f) had similar loading curves for each of the first two LVs. Two important zones were identified in each of these PLS models by showing either a shift or peak in the loading weight at ~545 and ~695-742 nm. Similar zones were identified by Hansen and Schjoerring (2003) for PLS models that calibrated reflectance spectra to estimate crop biophysical parameters such as green biomass, chlorophyll concentration and density, N concentration and density, and leaf area index in wheat. The models/parts of the models that used derivative reflectance spectra as the input for the predictors (Figs. 4-2b, 4-2c, 4-2e, and 4-2f) also had similar loading curves for each of the first two LVs. In each of these models, two important zones were identified by showing shifts or peaks in the loading weights at ~481-545 and at ~647-741nm. These zones coincide with the peaks observed for the first derivative reflectance (Fig. 4-1).

3.3. Nitrogen stress prediction

Leaf N concentration was significantly different between varieties on each image date (Table 4-5), so linear regression analysis was performed separately for each variety. The r^2 values based on the linear relationship between leaf N and each of the measurements or spectral indices/models and their corresponding root mean squared error of cross-validation (RMSECV) values were reported within and across image dates for RB (Table 4-8) and AR (Table 4-9).

NDVI did not perform well because it is a structural index that is only sensitive to crop cover up to percent covers of 90; after 90% cover, NDVI values plateau (Barnes et al., 2000). Therefore, NDVI was a poor indicator of leaf N concentration if canopy cover was >90%, even when leaf N concentrations varied among N treatments. Since a preliminary analysis was performed to select and test only the best broadband and narrowband indices, the r^2 values and corresponding RMSECV for the reported indices were very similar overall (excluding NDVI, the range in r^2 and RMSECV between the best and worst of the reported indices was always less than 0.15 and 12% for r^2 and RMSECV, respectively).

The indices that resulted in the best r^2 values and corresponding RMSECV varied by image date and variety (Tables 4-8 and 4-9). The best r^2 values and corresponding RMSECV were always better for the narrowband indices than for the broadband indices. The best broadband indices were NG and GRVI for RB and AR, respectively, and the best narrowband index was DCNI for both varieties. DCNI performed better than chlorophyll meter readings on two of the four image dates for RB and on one of the four image dates in 2011. Improvement in r^2 over broadband indices fluctuated from 0.5 – 23.0% and 0.2 – 25.0% for RB and AR, respectively; improvement in the RMSECV over broadband indices fluctuated from 0.9 – 35.3% and 0.1 – 38.1% for RB and AR, respectively.

The PLS models that were constructed using first derivative reflectance spectra only and both reflectance and first derivative reflectance spectra always had the same or better r^2 values and corresponding RMSECV than the models constructed using reflectance spectra only (Tables 4-8 and 4-9). The PLS models that used both reflectance spectra and first derivative reflectance spectra showed an improvement over the best narrowband indices on three of the four image dates for both RB and AR. Improvement in r^2 over the best narrowband index for each image date fluctuated from 1.2 – 8.2% for RB and 0.0 – 4.9% for AR; improvement in the RMSECV over the best narrowband index for each image date fluctuated from 4 – 30% for RB and 2 - 23% for AR. PLS models showed an improvement over chlorophyll meter readings on three of the four image dates for RB and on two of the four image dates for AR.

Reflectance/derivative spectra, chlorophyll meter readings, spectral indices, and PLS models were also evaluated for their performance over all image dates. Evaluation over all image dates is helpful in determining the measurements, indices, and models that can best predict N stress regardless of growth stage. A correlogram comparing r^2 as a function of wavelength for the relationship between leaf N and reflectance/first derivative reflectance for both varieties was constructed to determine the spectral areas most influential in detecting N stress (Fig. 4-3). Across most wavelengths, r^2 values for reflectance was noticeably higher in RB than for AR; in both varieties, the highest r^2 values for reflectance were observed in the green to red regions (i.e., ~527 – 657 nm) the red-edge region (i.e., ~695 – 704 nm) and the near-infrared region (i.e., ~750 – 900 nm). Across wavelengths, r^2 values for first derivative reflectance were much more variable than for raw reflectance; the highest r^2 values for first derivative reflectance were observed in the green-edge region (i.e., ~518 nm) and the beginning and end of the red-edge region (i.e., ~695 nm and ~732 nm, respectively).

None of the reflectance or first derivative reflectance wavelengths passed the preliminary analysis for consideration as one of the best overall measures of N stress (preliminary analysis was described in section *2.6.1.*). This was due to inconsistency among image dates. Compared to the best spectral indices, the r^2 values representing the relationship between leaf N and reflectance/first derivative spectra performed poorly overall across image dates (Fig. 4-3; Tables 4-8 and 4-9). Some wavelengths had high r^2 values for a specific image date, but results were not consistent over image dates (data not shown). As the field of precision agriculture progresses, the measurements or spectral indices/models used to determine variable rate management decisions based on N stress may vary depending on the growth stage. If this scenario someday exists, spectral measurements to be used to determine crop N stress should be evaluated based on accuracy for individual growth stages.

Over image dates, the broadband index with the best r^2 values and RMSECV was NG for both varieties; the narrowband indices with the best r^2 values and RMSECV were NDI2 and R-M/MSAVI for RB (Table 4-8), and DCNI and NDI2 for AR (Table 4-9). These indices performed better than the best broadband indices for both varieties; they performed at a similar level to that of the chlorophyll meter readings for RB, and performed slightly below that of the chlorophyll meter readings for AR. Each of the PLS models across image dates resulted in improvement over the best narrowband indices. The best performing PLS models for each variety were those constructed using first derivative spectra only. Improvements in PLS r^2 values over chlorophyll meter readings were 7% and 30% for RB and AR, respectively; improvements in RMSECV over chlorophyll meter readings were 1% and 25% for RB and AR, respectively.

Overall, spectral data showed some improvements over the chlorophyll meter readings, although most improvements were minimal (Tables 4-8 and 4-9). Canopy-level spectra are not only influenced by leaf chlorophyll content, but also by external factors such as leaf area index and the background soil effects (Daughtry et al., 2000). Chlorophyll meter readings are largely unaffected by these external factors, but this comes at the expense of spatial information. Canopy-level imagery contains spatial information, and can therefore provide an advantage over chlorophyll meter readings for variable rate application of N fertilizer, especially if it can predict N stress with comparable accuracy. In addition, tissue samples used for the chlorophyll meter readings in this study were the same tissue samples used to determine leaf N concentration. Alternatively, canopy-level spectral data in this study included information from plants other than those in which tissue samples were taken for leaf N concentration determination. Therefore, this provided a bias in which the r^2 and RMSECV values of the chlorophyll meter readings were favored over those of the canopy-level spectral data. It is reasonable to expect the spectral indices and PLS models to improve in performance if the tissue samples came from the same plants that are represented in the canopy-level imagery.

3.4. Practical use of the nitrogen sufficiency index

A nitrogen sufficiency index (NSI; Eq. (4-3)) can be applied to plant measurements or spectral data over a large scale and at a relatively low cost, making it a very practical approach for determining relative N stress (Tremblay et al., 2011). NSI values were calculated for the plant measurements and spectral indices/models (Tables 4-10 and 4-11). On the first image dates in each year, the NSI values for the 270 kg N ha^{-1} early treatment were typically above 100%. This occurred because on these image dates, the 270 early treatment received substantially more N fertilizer than the NSI reference treatments (i.e., 270 split and 270 split + s; Table 4-2).

3.4.1. Accuracy assessment

An accuracy assessment was performed using confusion matrices in which leaf N concentration was compared to chlorophyll meter readings and the spectral indices/models. Prior to conducting the accuracy assessment, treatment data for each measurement and spectral index/model were normalized using a nitrogen sufficiency index (NSI; Eq. (4-3)) and were placed into predetermined N stress classes (Table 4-4). The measurements and spectral indices/models with the highest overall accuracies/kappa coefficients, as well as those with the most balanced misclassified treatment plots (between overestimated and underestimated stress levels) were considered to be the best.

The measurements and spectral indices/models with the best accuracies and kappa coefficients were not necessarily those that had the best r^2 or RMSECV values. Among the spectral indices, the highest overall accuracies and kappa coefficients were observed for DCNI in RB, and for CCI and MTCI in AR (Table 4-12). DD/MSAVI and MTCI were the most consistent across varieties in providing good r^2 values, RMSECVs, and accuracies/kappa coefficients. DD/MSAVI and MTCI had relatively similar accuracies/kappa coefficients as the chlorophyll meter readings and PLS models that used first derivative reflectance as the independent input variables. Averaged across varieties, the overestimated and underestimated stress for: chlorophyll meter readings were 7% and 16%, respectively; DD/MSAVI were 9% and 17%, respectively; MTCI were 14% and 11%, respectively; and PLS RD were 6% and 19%, respectively. Of these four N stress predictors, MTCI was the only one that did not have a substantially higher percentage of underestimated stress than overestimated stress; instead, MTCI slightly overestimated the N stress classes. For N fertilizer recommendations, it is perhaps better to have a slightly higher percentage of overestimated stress than underestimated stress since a grower may run the risk of losing yield if fertilizer is applied below the optimum rate due to misclassified areas of underestimated stress.

Classification of data into predefined stress classes is a good way to evaluate the usefulness of a measurement for variable-rate application decisions at a commercial scale (Cohen et al., 2010). Performing an accuracy assessment was a way to evaluate the quality of the measurements and spectral indices/models based on their ability to classify N stress levels into pre-defined ranges. When similar accuracies, kappa coefficients, and high cost errors were observed, the balance between the percentage of overestimated and underestimated stress was a useful way to determine the measurements and spectral indices/models best suited for determining crop N stress for variable rate application.

3.4.2. Analysis of variance

The main effect of nitrogen rate/timing was highly significant for all NSI plant measurements and spectral indices/models (Table 4-13). In many cases, the main effect of variety, as well as the N by variety interaction, was not significant for these NSI plant measurements and spectral indices/models. Before the NSI transformation, the main effect of variety for leaf N concentration was significant on every image date (Table 4-5); however, after the NSI transformation, it was no longer significant, mostly because the references used for the NSI transformations were variety dependent (i.e., they were selected within variety). This general transition of statistical significance following the NSI transformation occurred for many of the plant measurements and spectral indices/models, especially on the second image date in each year (data before the NSI transformation are not presented). This transition also occurred for the N by variety interaction for many of the plant measurements and spectral indices/models. Plant measurements and spectral indices/models that are insensitive to external factors such as variety can be useful over a broader range of environmental conditions. Different varieties may have different N stress thresholds and different N fertilizer requirements, so the user should be aware of these differences while making fertilizer recommendations.

3.4.3. Variability in nitrogen sufficiency index values

NSI values of plant measurements and spectral indices/models differed over N treatments and image dates (Tables 4-10 and 4-11). The variation among N treatments corresponded to different levels of relative N stress as determined by that plant measurement or spectral index/model. The variation over N treatments was different for each plant measurement and spectral index/model. The coefficient of variation (CV) was calculated on each image date in order to objectively measure the spread of NSI values among N treatments for each plant measurement and spectral index/model. In order to make observations about the variability among image dates, the relative fluctuation (RF) of the CVs among image dates was calculated. High RF values indicate inconsistent variability among image dates. This inconsistency is the result of a high sensitivity to absolute N stress among image dates according to the respective plant measurement or spectral index/model used; in other words, the inconsistency is a result of differing NSI values for similar N treatments across image dates.

For most plant measurements and spectral indices/models, the RF values were smaller for RB than for AR (Table 4-14). DCNI and NG had the smallest RF values for RB and AR, respectively. If an NSI is used, plant measurements and spectral indices/models with smaller RF values are better for N stress determination over a broader range of growth stages and years (i.e., they will have more consistent NSI values among growth stages and years).

The CVs for petiole NO_3-N concentration were over five times as high as those for leaf N concentration (Table 4-14). There were large discrepancies in the CVs between petiole NO_3-N and spectral data, and overall similarities in the CVs between leaf N and spectral data. This confirms the suggestion by Cohen et al. (2010) that the variability in aerial hyperspectral imagery might be very similar to the variability in leaf N, but not to that of petiole NO_3-N. NDVI1, SR8, and MTCI had the highest CVs among spectral indices and PLS models (each had a CV greater than 20% for RB and greater than 19% for AR).

Although high CVs are ideal for observing differences between N treatments for a specific plant measurement or spectral index/model, the CV does not have much dependence on the measurement or spectral index/model’s ability to predict leaf N concentration. Instead, the CV can be used as a general indicator of the theoretical range of N stress levels for plant measurements and spectral indices/models, and should be used only after determining the ability of the measurement or spectral index/model to predict N stress. Described more precisely, a measurement or spectral index/model with a high

CV simply means that it has a larger range in NSI values (i.e., there is less saturation over the range of NSI values).

The CVs of plant measurements and spectral indices/models are very important for being able to make fertilizer recommendations. Assuming a fixed NSI threshold and similar prediction accuracies between two spectral indices/models, an index/model with a higher CV will tend to overestimate N stress and one with a lower CV will tend to underestimate N stress. Therefore, in order for NSIs to be universal across plant measurements and spectral indices/models for fertilizer recommendations, they should be standardized based on their CVs.

3.5. Means separation

A means separation was performed within variety and image date to determine the ability of each plant measurement and spectral index/model to distinguish between N treatments. The treatment groupings for the spectral indices/models can be directly compared to those for the plant measurements; this is a way to determine if a spectral index/model has similar separation capabilities among N treatments as the ground truth plant measurements. Differences among treatment groupings between petiole NO_3-N and leaf N were similar in most cases (Tables 4-10 and 4-11). When treatment groupings were different between petiole NO_3-N and leaf N, petiole NO_3-N was usually more conservative at detecting differences among N treatments (i.e., petiole NO_3-N usually detected less groups than leaf N). However, because petiole NO_3-N concentration depends on the growth stage as well as the number of days since the last N fertilizer application (Nigon et al., 2012), different results might be obtained if measurements were taken shortly after post-emergence fertilizer applications instead of shortly before applications. Mean groupings for the chlorophyll meter readings were usually very similar to that of leaf N. However, in many cases, spectral indices/models were actually less conservative than leaf N at detecting differences. In other words, many of the spectral indices/models were able to distinguish between the N treatments even better than the plant measurements. This is likely because canopy-level spectral data is affected by leaf area index in addition to plant biophysical parameters such as leaf N concentration (Daughtry et al., 2000).

Petiole NO_3-N and leaf N measurements were not the most precise indicators of N stress based on means separation of the N treatments. This suggests that simple plant nutrient measurements do not detect crop N stress to the full degree; measurements that combine plant nutrient concentration and biomass/leaf area index have the best potential to predict crop N stress so accurate N fertilizer recommendations can be made. The Nitrogen Nutrition Index (NNI) is one measurement that does this; Duchenne et al., (1997) and Bélanger et al., (2001) report the use of an NNI for a potato crop.

The NNI is calculated as the ratio between current N concentration in the dry biomass and the critical N concentration. The critical N concentration is defined as the minimum N concentration necessary to achieve maximum growth rate (Ulrich, 1952), and is calculated based on the dry biomass of the crop and certain estimated parameters (Goffart et al., 2008). Because the NNI considers both plant nutrient concentration and biomass, it is likely a very good indicator of crop N status; however, it is a very cumbersome measurement and is not feasible to be able to account for within-field spatial variability. It is reasonable to suggest, however, that canopy-level spectral data would have a very good relationship with the NNI since they are both influenced by the effects of plant N concentration and biomass/leaf area index (Daughtry et al., 2000). If this is the case, canopy-level spectra would be better suited to be an indicator of crop N stress for fertilizer recommendations since it is able to precisely account for spatial variability.

4. Conclusions

The measurements and spectral indices/models best suited for predicting N stress to be used for variable rate N fertilizer management decisions varied depending on whether data were analyzed within or across image dates. Overall, the best spectral indices had similar r^2 values, RMSECVs, overall accuracies, and kappa coefficients as the chlorophyll meter readings; however, because the spectral indices used canopy-level imagery, these techniques provide an advantage over chlorophyll meter readings since imagery can better account for spatial variability. Across image dates (two seasons) for both varieties (Russet Burbank and Alpine Russet), the PLS models, especially those that used first derivative reflectance spectra as independent variables, were determined to be able to predict leaf N concentration the best in potatoes (i.e., they had the highest r^2 and lowest RMSECV values). However, the best predictors of leaf N concentration based on r^2 and RMSECV values did not necessarily have the best accuracies when classifying the treatment plots into predetermined NSI stress classes. Because MTCI performed well across image dates and varieties for all evaluative measures in this study (r^2, RMSECV, and the accuracy assessment), it was considered to be the best technique for determining N stress level for variable rate application of N fertilizer for a broad range of conditions.

By applying the NSI formula to plant measurements and spectral indices/models, in many cases, the main effect of variety and the N × variety interaction became statistically insignificant. By transforming a plant measurement or spectral index/model so it is insensitive to external factors such as variety, it can be useful over a broader range of environmental conditions. For NSIs to be useful for variable rate fertilizer management, the user must be aware of the variability imposed by the spectral index/model relative to the variability of leaf N concentration; spectral indices/models with more variability than leaf N concentration may tend to overestimate N stress level, and those with less variability may tend to underestimate N stress level.

Because canopy-level spectral data are affected by biomass/leaf area index in addition to plant biophysical parameters (Daughtry et al., 2000), many of the spectral indices/models were able to distinguish between the N treatments better than petiole NO_3-N and leaf N concentration according to a means separation analysis. Regarding the use of hyperspectral data for variable rate N fertilizer management in potatoes, future research should be focused on determining the relationship between canopy-level spectral data and the Nitrogen Nutrition Index (NNI). In addition, the findings of this study should be verified using more N fertilizer rates and timings as well as other sensor platforms with a similar spatial resolution. Work could also be done to tweak the formulas of spectral indices so their CVs are more similar to the CVs of leaf N concentration. This could result in spectral indices that have both a high predictive capability (high r^2 values, low RMSECV values) and a high accuracy according to an accuracy assessment of different N stress levels.

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6. References

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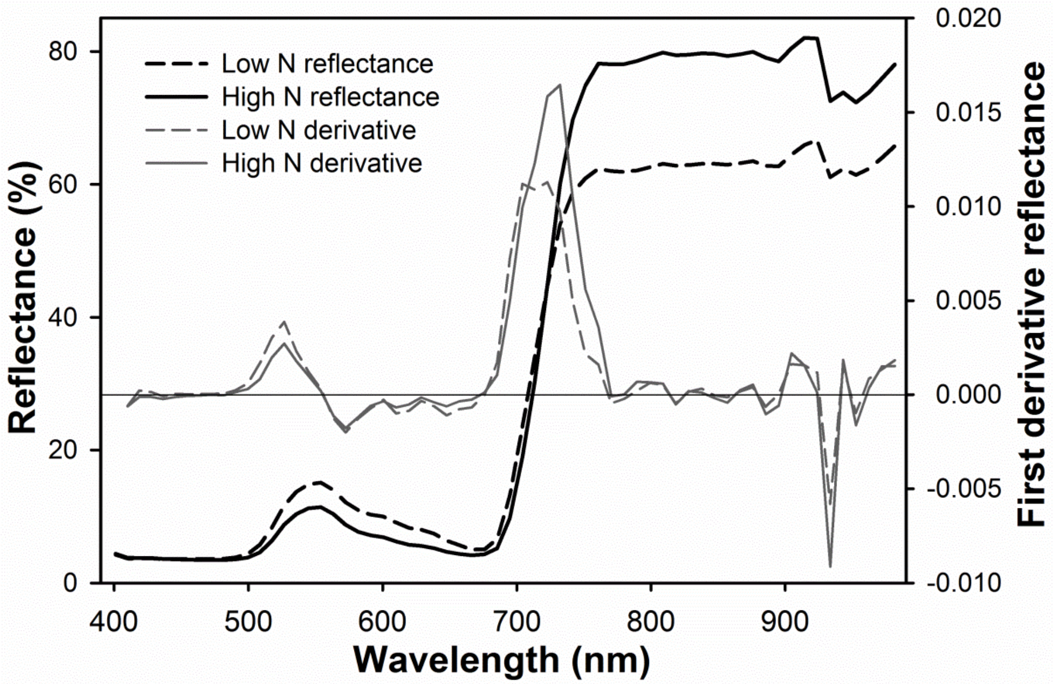

Fig. 4-1. Aerial-based canopy-level hyperspectral reflectance and first derivative reflectance spectra at all AISA Eagle narrowbands (2.3 nm bandwidth) for a Russet Burbank potato canopy at 47 days after emergence (1 July 2010) in Becker, MN. Low N corresponds to the 34 kg N ha^{-1} early treatment, and high N corresponds to the 270 kg N ha^{-1} early treatment. Similar trends among spectral frequencies were observed for the Alpine Russet variety.

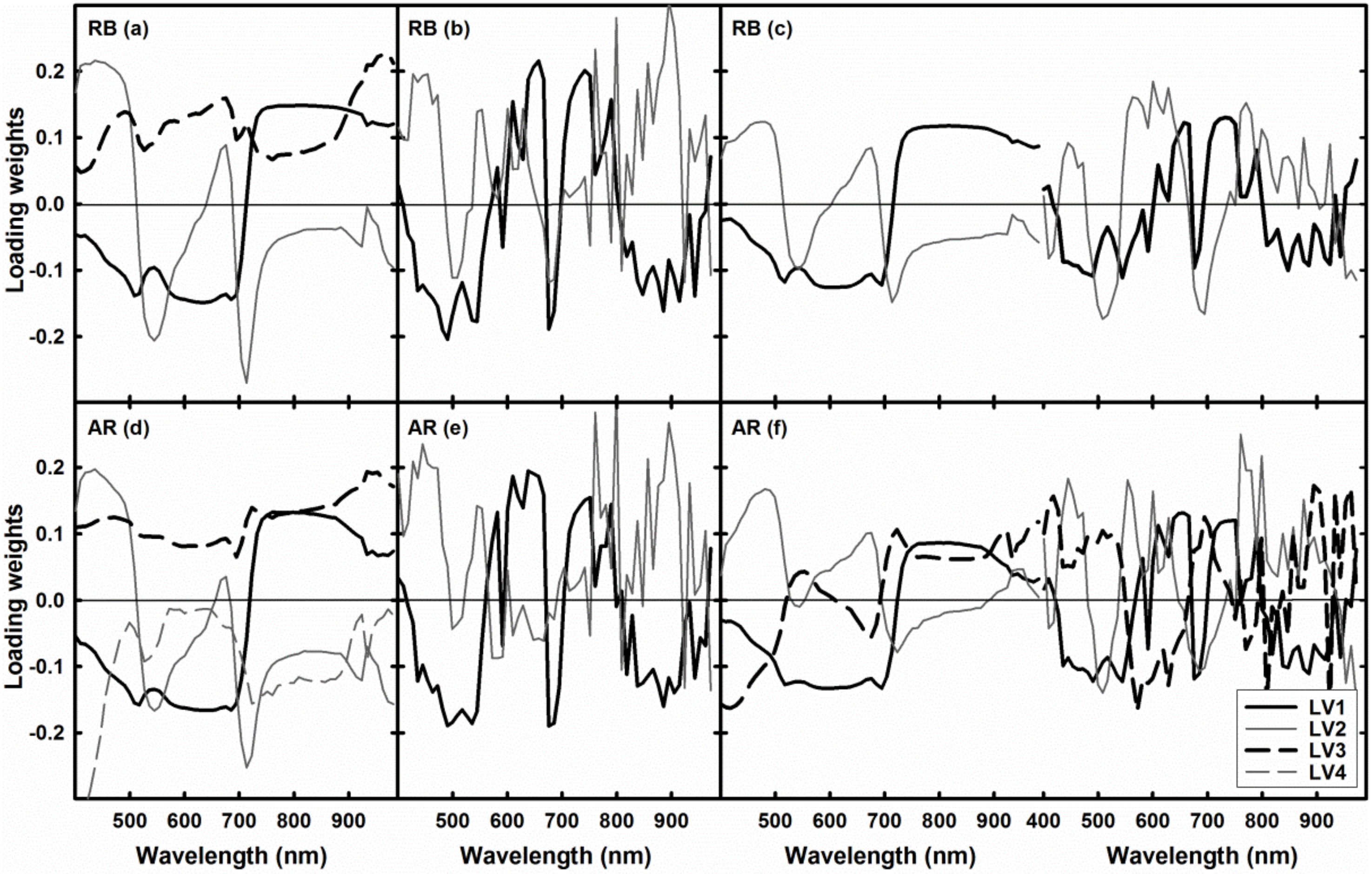

Fig. 4-2. Loading weights for the combination of predictors (spectra) that comprise each of the latent variables (LVs) for the PLS models calibrated to estimate leaf nitrogen. The top and bottom subplots represent relevant loading weights for the models that used Russet Burbank (RB) and Alpine Russet (AR) input data, respectively; from left to right, the subplots represent relevant loading weights for the models that used reflectance spectra (a,d), first derivative reflectance spectra (b,e), and both (c,f) as the input for the predictors. High absolute values of loading weight coefficients indicate high importance of that wavelength in the PLS model.

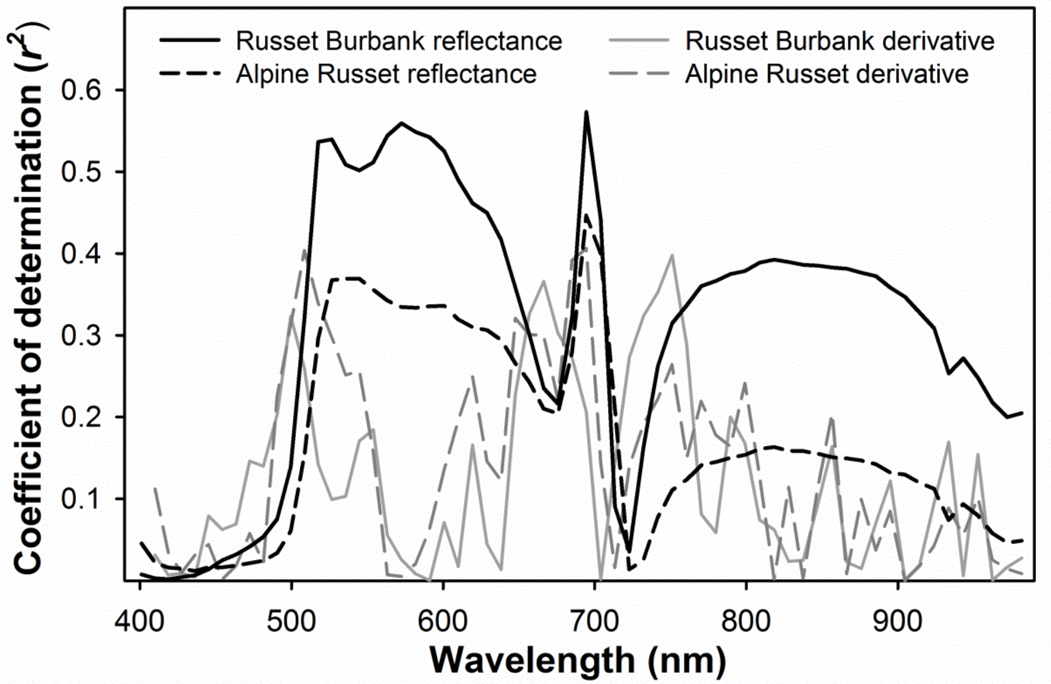

Fig. 4-3. Correlogram showing the coefficient of determination (r^2) as a function of wavelength derived from linear regression analysis of leaf nitrogen concentration against canopy reflectance and reflectance derivative for Russet Burbank and Alpine Russet varieties across all image dates.

Table 4-1. Rate and timing of nitrogen (N) fertilizer treatments for 2010 and 2011.

Nitrogen fertilizer treatment	Timing of application					Total N
	planting	emergence		post-emergence		
		2010	2011	2010	2011	
	kg N ha^{-1}					
34 N early	34	0	0	0	0	34
180 N split	34	78	90	17 × 4[‡]	11.2 × 5[‡]	180
270 N split	34	124	124	28 × 4[‡]	22.4 × 5[‡]	270
270 N split + s[†]	34	124	124	28 × 4[‡]	22.4 × 5[‡]	270
270 N early	34	124	124	112	112	270

[†] A soil surfactant (IrrigAid Gold) was used in this treatment, and was applied at a rate of 10 L ha^{-1}.

[‡] On average, about two weeks passed between split applications of post-emergence N.

Table 4-2. Actual nitrogen (N) fertilizer applied at the time of data acquisition for N treatments with split applications of post-emergence fertilizer.

N fertilizer treatment	2010		2011	
	47 DAE	83 DAE	43 DAE	66 DAE
	kg N ha^{-1}			
180 kg N ha^{-1} split	146	180	139	166
270 kg N ha^{-1} split	214	270	203	248

Table 4-3. Summary of the best performing indices evaluated in this study. A total of 17 broadband and 82 narrowband previously published indices were used for the initial analysis.

Index	Name	Formula[†]	Developed for	Developed by
Broadband indices				
GNDVI	Green normalized difference vegetation ind	$= (R_{NIR} - R_G)/(R_{NIR} + R_G)$	Chlorophyll	Gitelson et al. (1996)
GRVI	Green ratio vegetation index	$= R_{NIR}/R_G$	Nitrogen	Sripada et al. (2006)
NG	Normalized green	$= R_G/(R_{NIR} + R_R + R_G)$	Nitrogen	Sripada et al. (2006)
Narrowband indices				
CCI	Canopy chlorophyll index	$= D_{723}/D_{704}$	Chlorophyll	Sims et al. (2006)
DCNI	Double − peak canopy nitrogen index	$= \frac{(R_{723} - R_{704})/(R_{704} - R_{666})}{(R_{723} - R_{666} + 0.03)}$	Nitrogen	Chen et al. (2010)
DD/MSAVI	$\frac{\text{Double difference index}}{\text{Modified soil adjusted vegetatino index}}$	$= \frac{(R_{751} - R_{723}) - (R_{704} - R_{666})}{0.5[2 \times R_{799} + 1 - \sqrt{(2 \times R_{799} + 1)^2 - 8 \times (R_{799} - R_{666})}]}$	Chlorophyll	Haboudane et al. (2008)
MTCI	MERIS terrestrial chlorophyll index	$= (R_{751} - R_{713})/(R_{713} - R_{676})$	Chlorophyll	Dash and Curran (2004)
NDI2[‡]	Normalized difference index 2	$= (R_{847} - R_{713})/(R_{847} - R_{676})$	Chlorophyll	Datt (1999)
NDVI	Normalized difference vegetation index	$= (R_{799} - R_{676})/(R_{799} + R_{676})$	Structure	Lichtenthaler et al. (1996)
NDVI1[‡]	Normalized difference vegetation index	$= (R_{751} - R_{732})/(R_{713} + R_{723})$[§]	Chlorophyll	Vogelmann et al. (1993)
R-M/MSAVI	$\frac{\text{Red − edge model index}}{\text{Modified soil adjusted vegetation index}}$	$= \frac{(R_{751}/R_{723}) - 1}{0.5[2 \times R_{799} + 1 - \sqrt{(2 \times R_{799} + 1)^2 - 8 \times (R_{799} - R_{666})}]}$	Chlorophyll	Haboudane et al. (2008)
SR2[‡]	Simple ratio 2	$= R_{799}/R_{554}$	Chlorophyll	Buschmann and Nagel (1993)
SR8[‡]	Simple ratio 8	$= R_{857}/(R_{554} \times R_{704})$	Chlorophyll	Datt (1998)
TCARI/OSAVI$_{(705,750)}$	$\frac{\text{Transformed chlorophyll absorption ratio index}_{(705,750)}}{\text{Optimized soil − adjusted vegetation index}_{(705,750)}}$	$= \frac{3[(R_{751} - R_{704}) - 0.2 \times (R_{751} - R_{554}) \times (R_{751}/R_{704})]}{(1 + 0.16) \times (R_{751} - R_{704})/(R_{750} + R_{704} + 0.16)}$	Chlorophyll	Wu et al. (2008)
Viopt1[‡]	Optimal vegetation index 1	$= 100 \times (\ln R_{761} - \ln R_{732})$	Nitrogen	Jasper et al. (2009)
Viopt2[‡]	Optimal vegetation index 2	$= R_{761}/R_{732}$	Nitrogen	Jasper et al. (2009)

[†] R_i = reflectance at wavelength i (nanometers); D_i = first derivative reflectance at wavelength i (nanometers)

[‡] Named in this study

[§] The bands in the numerator of NDVI1 were switched to the formula shown so the index yielded positive values.

Table 4-4. Nitrogen (N) stress classes for the accuracy assessment; based on N sufficiency index (NSI) ranges.

Nitrogen stress class	NSI range (%)
High	<80
Moderate	80-95
Low	>95

Table 4-5. Analysis of variance for leaf nitrogen concentration at different days after emergence in 2010 and 2011.

Source of variation	2010		2011	
	47 DAE	83 DAE	43 DAE	66 DAE
	-------------------	P > F	-------------------	
Nitrogen treatment (N)	<0.0001	<0.0001	<0.0001	<0.0001
Variety (V)	<0.0001	<0.0001	0.011	0.0004
N × V	0.002	0.903	0.484	0.646

Table 4-6. Leaf nitrogen concentration for Russet Burbank and Alpine Russet under various nitrogen treatments at different days after emergence (DAE) in 2010 and 2011.

Variety	N Treatment	2010		2011	
		47 DAE	83 DAE	43 DAE	66 DAE
Russet Burbank	34 early	3.27 c[†]	3.32 b	3.46 d	3.29 c
	180 split	4.33 b	4.11 a	4.31 c	4.06 b
	270 split	4.43 b	4.35 a	4.77 b	4.66 a
	270 split + s	4.39 b	4.41 a	4.85 b	4.73 a
	270 early	4.84 a	3.69 b	5.21 a	4.23 b
Alpine Russet	34 early	2.99 b	2.77 c	3.52 d	3.14 c
	180 split	3.70 a	3.80 a	4.05 c	3.72 b
	270 split	3.72 a	3.94 a	4.53 b	4.36 a
	270 split + s	3.86 a	3.98 a	4.57 ab	4.22 a
	270 early	3.86 a	3.26 b	4.90 a	3.89 b

[†] Means followed by the same letter within an image date and variety are not significantly different (α=0.05).

Table 4-7. Potato tuber yield under different nitrogen treatments in 2010 and 2011.

N treatment	Tuber yield (Mg ha^{-1})	
	2010	2011
34 early	45.16 b[†]	40.09 c
180 split	55.15 a	52.70 ab
270 split	58.27 a	53.18 a
270 split + s	57.49 a	52.28 ab
270 early	54.72 a	49.00 b

[†] Means followed by the same letter within an image date and variety are not significantly different (α=0.05).

Table 4-8. Coefficient of determination (r^2) and root mean squared error of cross-validation (RMSECV) representing the relationship between leaf N concentration and the measurement or spectral index/model for predicting N stress in a Russet Burbank potato crop. Results are presented for different days after emergence (DAE) in 2010 and 2011, as well as for all dates combined. All r^2 values were statistically significant at $\alpha = 0.05$. Reported RMSECV was normalized by calculating the ratio between RMSECV values and the range of predicted values.

Measurement or spectral index / model	2010				2011				All dates *(n=80)*	
	47 DAE (*n=20*)		83 DAE *(n=20)*		43 DAE *(n=20)*		66 DAE *(n=20)*			
	r^2	RMSECV (%)	r^2	RMSECV (%)	r^2	RMSECV (%)	r^2	RMSECV (%)	r^2	RMSECV (%)
Ground measurements										
Chlorophyll meter	0.77	19	0.76	17	0.79	18	0.78	27	0.74	14
Broadband indices										
GNDVI	0.77	20	0.57	28	0.77	21	0.81	17	0.65	18
GRVI	0.76	20	0.58	29	0.80	19	0.83	16	0.69	18
NG	0.77	19	0.59	28	0.77	21	0.81	17	0.69	17
Narrowband indices										
CCI	0.79	19	0.63	24	0.79	18	0.78	19	0.69	17
DCNI	0.79	18	0.73	18	0.81	16	0.75	21	0.70	16
DD/MSAVI	0.79	19	0.62	25	0.80	18	0.80	18	0.71	16
MTCI	0.78	19	0.63	26	0.81	17	0.80	18	0.72	16
NDI2	0.78	19	0.66	22	0.79	19	0.81	18	0.74	14
NDVI	0.58	28	0.47	34	0.73	23	0.75	21	0.31	28
NDVI1	0.78	19	0.59	29	0.82	17	0.80	19	0.72	17
R-M/MSAVI	0.79	19	0.67	21	0.83	16	0.78	20	0.73	16
SR2	0.76	20	0.60	28	0.81	18	0.84	16	0.69	18
SR8	0.74	20	0.71	19	0.82	16	0.77	19	0.70	17
TCARI/OSAVI$_{(705, 750)}$	0.76	19	0.61	25	0.82	16	0.80	18	0.69	20
Viopt1	0.79	19	0.62	25	0.82	17	0.82	18	0.72	16
Viopt2	0.79	18	0.62	25	0.82	17	0.82	18	0.72	17
Partial least squares regression										
PLS RR	0.76	20	0.62	23	0.76	20	0.80	18	0.76	16
PLS RD	0.81	16	0.68	20	0.84	15	0.83	17	0.79	14
PLS RR & RD	0.81	16	0.67	21	0.84	15	0.90[†]	11[†]	0.76	15

[†] Indicates a partial least squares regression model that used two extracted latent variables.

Table 4-9. Coefficient of determination (r^2) and root mean squared error of cross-validation (RMSECV) representing the relationship between leaf N concentration and measurement or spectral index/model for predicting N stress in an Alpine Russet potato crop. Results are presented for different days after emergence (DAE) in 2010 and 2011, as well as for all dates combined. All r^2 values were statistically significant at $\alpha = 0.05$. Reported RMSECV was normalized by calculating the ratio between RMSECV values and the range of predicted values.

Measurement or spectral index / model	2010				2011				All dates *(n=80)*	
	47 DAE *(n=20)*		83 DAE *(n=20)*		43 DAE *(n=20)*		66 DAE *(n=20)*			
	r^2	RMSECV (%)	r^2	RMSECV (%)	r^2	RMSECV (%)	r^2	RMSECV (%)	r^2	RMSECV (%)
Ground measurements										
Chlorophyll meter	0.85	15	0.80	16	0.68	20	0.85	20	0.59	18
Broadband indices										
GNDVI	0.82	15	0.53	30	0.73	23	0.66	25	0.46	26
GRVI	0.80	16	0.56	28	0.78	21	0.70	23	0.42	30
NG	0.82	16	0.54	30	0.74	23	0.68	25	0.49	25
Narrowband indices										
CCI	0.83	16	0.62	24	0.81	20	0.65	25	0.49	26
DCNI	0.82	16	0.70	19	0.86	15	0.61	25	0.57	20
DD/MSAVI	0.85	15	0.57	27	0.81	19	0.64	25	0.50	24
MTCI	0.84	16	0.59	26	0.83	17	0.65	25	0.51	26
NDI2	0.84	16	0.60	25	0.81	18	0.66	25	0.57	21
NDVI	0.76	17	0.47	33	0.65	29	0.56	29	0.27	37
NDVI1	0.84	15	0.56	27	0.83	18	0.65	25	0.51	28
R-M/MSAVI	0.83	16	0.63	22	0.89	13	0.62	25	0.55	22
SR2	0.81	16	0.57	28	0.79	19	0.70	23	0.40	31
SR8	0.77	18	0.68	20	0.83	16	0.65	22	0.49	27
TCARI/OSAVI(705, 750)	0.82	16	0.67	20	0.83	17	0.65	24	0.42	29
Viopt1	0.84	15	0.56	26	0.83	17	0.68	24	0.51	26
Viopt2	0.84	15	0.56	26	0.83	16	0.68	24	0.50	27
Partial least squares regression										
PLS RR	0.83	15	0.61	22	$0.89^{\dagger}$	$14^{\dagger}$	0.66	26	0.76	14
PLS RD	0.83	15	0.71	19	$0.93^{\dagger}$	$10^{\dagger}$	0.70	22	0.77	13
PLS RR & RD	0.84	15	0.71	19	$0.93^{\dagger}$	$10^{\dagger}$	0.70	22	0.77	14

†Indicates a partial least squares regression model that used two extracted latent variables.

Table 4-10. Mean nitrogen sufficiency index (NSI) values for selected plant measurements and spectral indices/models for the Russet Burbank variety at different years and days after emergence (DAE).

Year	DAE	N treatment	Plant measurements			Broadband Indices	Narrowband indices						Partial least squares regression
			Petiole NO_3-N	Leaf N	Chlorophyll meter	NG	DCNI	MTCI	NDI2	NDVI	NDVI1	SR8	PLS RD
2010	47	34 early	7 c[†]	74 c	71 c	68 d	69 d	58 d	79 d	95 b	48 d	51 d	68 c
		180split	64 b	98 b	97 b	94 c	95 c	93 c	97 c	99 a	91 c	90 c	96 b
		270 split	100b	100b	99 b	100ab	100b	100b	100b	100a	100b	100ab	99 b
		270 split + s	99 b	99 b	100b	97 bc	100b	100b	100b	99 a	99 b	95 bc	100b
		270 early	159a	109a	104a	104a	105a	106a	102a	100a	108a	107a	105a
	83	34 early	11 c	75 c	71 c	67 c	66 c	48 c	77 c	76 c	39 c	55 c	78 c
		180split	63 ab	93 ab	93 a	87 b	88 b	79 b	92 b	95 b	75 b	80 b	90 b
		270 split	100a	99 a	98 a	101 a	100 a	100 a	100 a	100 a	100 a	100 a	100 a
		270 split + s	78 a	100 a	100 a	100 a	99 a	99 a	100 a	100 a	100 a	97 a	99 a
		270 early	25 bc	84 bc	83 b	87 b	81 b	76 b	90 b	97 ab	74 b	74 b	86 b
2011	43	34 early	12 c	71 d	71 c	69 d	73 d	56 d	80 d	93 c	47 d	57 d	78 d
		180split	29 c	89 c	90 b	92 c	91 c	88 c	96 c	99 b	85 c	86 c	94 c
		270 split	78 b	98 b	94 b	99 b	99 b	99 b	100b	100a	98 b	98 b	99 b
		270 split + s	100ab	100b	100a	100b	100b	100b	100b	100a	100a	100b	100b
		270 early	135a	107a	101a	103a	102a	105a	102a	100a	106a	104a	103a
	66	34 early	2 d	70 c	73 c	70 d	66 d	55 d	77 d	94 c	47 d	55 c	70 d
		180split	25 cd	86 b	92 b	89 c	87 c	82 c	92 c	99 b	79 c	82 b	89 c
		270 split	79 ab	99 a	100a	100a	100a	100a	100a	100a	100a	100a	100a
		270 split + s	100a	100a	99 a	99 ab	99 a	97 ab	99 ab	100ab	97 ab	99 a	98 a
		270 early	42 bc	90 b	92 b	95 b	93 b	91 b	96 b	99 ab	90 b	91 ab	94 b

[†] Means followed by the same letter within an image date and variety are not significantly different ($\alpha=0.05$).

Table 4-11. Mean nitrogen sufficiency index (NSI) values for selected plant measurements and spectral indices/models for the Alpine Russet variety at different years and days after emergence (DAE).

Year	DAE	N treatment	Plant measurements			Broadband Indices	Narrowband indices						Partial least squares regression
			Petiole NO_3-N	Leaf N	Chlorophyll meter	NG	DCNI	MTCI	NDI2	NDVI	NDVI1	SR8	PLS RD
2010	47	34 early	11 b[†]	77 b	75 b	71 c	75 c	61 c	82 c	96 b	50 c	59 c	71 d
		180split	95 a	96 a	97 a	97 b	97 b	96 b	98 b	100a	95 b	95 b	96 c
		270 split	100a	97 a	98 a	100ab	100ab	100b	100ab	100a	100b	100ab	100ab
		270 split + s	68 a	100a	100a	99 b	98 b	99 b	99 b	100a	98 b	97 b	98 bc
		270 early	114a	100a	100a	105a	103a	105a	102a	100a	107a	107a	104a
	83	34 early	5 b	70 c	55 d	68 c	62 d	48 c	74 c	86 b	40 c	51 c	69 c
		180split	67 a	96 a	94 b	88 b	87 b	82 b	93 b	97 a	77 b	80 b	89 b
		270 split	90 a	99 a	100ab	98 a	98 a	96 a	99 a	99 a	95 a	98 a	97 a
		270 split + s	100a	100a	104a	100a	100a	100a	100a	100a	100a	100a	100a
		270 early	7 b	82 b	81 c	87 b	80 c	76 b	90 b	97 a	73 b	74 b	87 b
2011	43	34 early	3 c	77 d	83 c	67 d	83 d	59 d	84 d	87 c	46 d	64 d	82 c
		180split	32 c	88 c	92 b	92 c	93 c	88 c	96 c	99 b	84 c	88 c	93 b
		270 split	99 b	99 b	99 a	99 b	99 b	98 b	99 b	100a	96 b	98 b	100a
		270 split + s	100b	100b	100a	100ab	100ab	100ab	100a	100a	100a	100a	100a
		270 early	150a	107a	100a	102a	101a	102a	101a	100a	103a	102a	102a
	66	34 early	1 d	72 d	68 d	73 c	75 c	60 c	80 c	94 c	51 c	63 c	78 d
		180split	23 cd	85 c	88 c	91 b	89 b	86 b	94 b	99 ab	85 b	84 b	90 c
		270 split	100a	100a	100a	98 a	99 a	99 a	100a	100a	99 a	97 a	99 a
		270 split + s	68 ab	97 ab	99 a	100a	100a	100a	100a	100a	100a	100a	100a
		270 early	43 bc	89 bc	93 b	93 b	94 ab	89 b	96 b	99 b	87 b	92 ab	95 b

[†] Means followed by the same letter within an image date and variety are not significantly different (α=0.05).

Table 4-12. Comparison of classification results using an accuracy assessment between leaf N concentration and measurements or spectral indices/models for Russet Burbank and Alpine Russet potato varieties. All measures of accuracy are reported across all image dates.

Measurement or index / model	Russet Burbank *(n=80)*					Alpine Russet *(n=80)*				
	Overall accuracy	Kappa	Overestimated stress	Underestimated stress	High cost errors	Overall accuracy	Kappa	Overestimated stress	Underestimated stress	High cost errors
						%				
Ground measurements										
Chlorophyll meter	79	66	8	14	0	75	60	6	19	0
Broadband indices										
GNDVI	56	23	3	41	0	51	15	1	48	3
GRVI	75	60	14	11	3	74	58	10	16	0
NG	75	60	11	14	0	73	56	9	19	0
Narrowband indices										
CCI	78	64	13	10	3	79	66	8	14	0
DCNI	78	64	10	13	0	70	52	9	21	0
DD/MSAVI	76	62	10	14	0	74	58	8	19	0
MTCI	73	57	18	10	3	79	66	10	11	1
NDI2	74	57	6	20	0	59	31	5	36	0
NDVI	51	13	3	46	3	49	8	0	51	9
NDVI1	71	55	19	10	4	74	59	16	10	4
R-M/MSAVI	75	60	11	14	0	73	56	9	19	0
SR2	75	60	14	11	3	75	60	9	16	0
SR8	66	48	24	10	3	76	63	14	10	0
$\text{TCARI/OSAVI}_{(705,750)}$	60	32	8	33	0	56	27	5	39	0
Viopt1	75	61	14	11	3	73	57	13	15	0
Viopt2	50	10	0	50	0	50	11	0	50	5
Partial least squares regression										
PLS RR	75	60	11	14	0	70	52	8	23	0
PLS RD	78	63	6	16	0	71	53	6	23	0
PLS RR & RD	73	55	10	18	0	76	62	6	18	0

Table 4-13. Analysis of variance for selected plant measurements and spectral indices/models at different days after emergence (DAE) in 2010 and 2011.

			Plant measurements			Broadband indies	Narrowband indices						Partial least squares regression
Year	DAE	Source of variation	Petiole NO_3-N	Leaf N	Chlorophyll meter	NG	DCNI	MTCI	NDVI	SR8	NDI2	NDVI1	PLS RD
2010	47	Nitrogen [N]	**[†]	**	**	**	**	**	**	**	**	**	**
		Variety [V]	-	-	-	*	**	-	*	**	**	-	-
		N × V	-	**	*	-	**	**	-	-	**	-	-
	83	Nitrogen	**	**	**	**	**	**	**	**	**	**	**
		Variety	-	-	-	-	-	-	**	-	-	-	*
		N × V	-	-	-	-	-	-	**	-	-	-	-
2011	43	Nitrogen	**	**	**	**	**	**	**	**	**	**	**
		Variety	-	-	**	*	**	-	**	*	**	-	-
		N × V	-	-	**	-	**	-	**	**	**	-	**
	66	Nitrogen	**	**	**	**	**	**	**	**	**	**	**
		Variety	-	-	-	-	**	-	-	-	-	-	*
		N × V	-	-	-	-	*	-	-	-	-	-	*

[†] ** and * are significant at 0.01 and 0.05; - is nonsignificant.

Table 4-14. Coefficient of variation (CV) for plant measurements and selected spectral indices/models for Russet Burbank and Alpine Russet potato varieties at different days after emergence (DAE) in 2010 and 2011.

Measurement or spectral index /model	Russet Burbank						Alpine Russet					
	2010		2011		AVG	Relative fluctuation	2010		2011		AVG	Relative fluctuation
	47 DAE	83 DAE	43 DAE	66 DAE			47 DAE	83 DAE	43 DAE	66 DAE		
	%						%					
Plant measurements												
Petiole NO3-N	66	71	83	81	75	0.22	71	105	78	85	85	0.40
Leaf N	14	13	15	14	14	0.18	10	19	12	13	14	0.64
Chlorophyll meter	13	14	13	14	13	0.10	11	23	8	11	13	1.16
Broadband indices												
NG	15	14	14	14	14	0.10	14	14	15	12	14	0.23
Narrowband indices												
DCNI	15	15	13	15	14	0.19	11	19	8	12	12	0.88
MTCI	20	23	21	21	21	0.17	18	25	18	18	20	0.39
NDI2	9	9	9	10	9	0.08	8	11	7	8	8	0.51
NDVI	4	8	4	4	5	1.00	3	6	5	3	4	0.73
NDVI1	25	28	27	26	26	0.13	24	30	25	23	25	0.30
SR8	24	21	21	21	22	0.13	20	26	16	17	20	0.47
Partial least squares regression												
PLS RD	12	10	14	13	12	0.28	9	16	12	11	12	0.56

Chapter 5.

HYPERSPECTRAL IMAGERY FOR THE DETECTION OF NITROGEN STRESS IN POTATO FOR IN-SEASON MANAGEMENT

This chapter has been published in the Proceedings of the 11th International Conference on Precision Agriculture. Table and figure numbering has been modified to correspond to thesis conventions.

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Chapter Summary

Potato (*Solanum tuberosum*, L.) yield and quality are highly dependent on the availability of nitrogen (N) during the crop's critical growth stages. Canopy-level hyperspectral imagery has been shown to be an effective research tool for determining the best wavelengths and/or indices for the detection of N stress in a number of crops. Research findings from hyperspectral imagery can be applied to active sensors as a way to increase the effectiveness of real-time, variable rate N applications, but limited data exist for potato production. A field study was conducted in 2010 and 2011 at the Sand Plain Research Farm in Becker, MN on a Hubbard loamy sand soil to evaluate the effects of water stress, N fertilizer rate/timing, the varieties Russet Burbank (RB) and Alpine Russet (AR), and growth stage on the ability of canopy-level hyperspectral imagery (401-982 nm) to detect N stress in a potato crop. The ability of canopy-level narrowband reflectance to detect differences in potato N status was evaluated by performing linear regression between hyperspectral narrowband reflectance and leaf N concentration as a means to distinguish the optimum wavelengths to detect N stress. As the potato crop matured, the coefficient of determination (r^2) decreased at most hyperspectral wavelengths, especially in the NIR region. The far green and near red regions from ~582-610 nm, and those at the beginning of the red-edge from ~685-695 nm were the best performing wavelengths for detecting N stress overall. Canopy reflectance was able to predict leaf N among image dates more consistently for RB than for AR.

Keywords: Hyperspectral imagery, nitrogen stress, potato, in-season fertilizer applications

1. Introduction

Well-drained, coarse-textured soils with soil moisture conditions that are maintained to be nearly constant via irrigation are ideal for potato production (Phene and Sanders, 1976). The sandy loam and loamy sand soils used for potato production in Minnesota are typically low in organic matter and cation exchange capacity (CEC), and therefore, have a relatively low reserve of soil nutrients. The low inherent soil fertility, coupled with a large nitrogen (N) requirement for potato production results in high N fertilizer inputs (Dean, 1994). Overhead irrigation is used to meet water requirements, but unpredicted rainfall following irrigation can enhance nitrate leaching. In addition, potato plants have a relatively shallow root system and poor nutrient use efficiency (NUE), which can potentially result in further N losses (Rosen and Bierman, 2008; Zvomuya et al., 2003).

The goal of N management for potato production is to provide the crop with a sufficient quantity of available N throughout the growing season to optimize yields, while minimizing the quantity of N lost from the root zone. However, managing N is a challenge because there are usually many unknown factors involved. One strategy used by producers in an attempt to optimize crop N uptake is to match the timing and rate of fertilizer N with the needs of the crop during different growth stages (Errebhi et al., 1998). A minimum of three split N applications (planting, emergence, and post-emergence) is a recommended best management practice for irrigated potato on coarse-textured soils in Minnesota (Rosen and Bierman, 2008). If fertigation is available, additional split N applications can be made post-emergence. Increased NUE and yields have been shown to result from split applications of N fertilizer during tuber initiation and early bulking (Errebhi et al., 1998; Westermann et al., 1988).

Crop N uptake and soil N losses depend on the interaction of many complicated and sometimes unpredictable factors, including: fertilizer source, soil organic matter, soil texture/CEC, and the weather conditions. Each of these can influence crop N uptake rates or soil N transformations throughout the growing season. While fertigation is a convenient method to apply frequent split applications of N fertilizer throughout the season, diagnostic tools are needed to help guide appropriate rates and timing of N applications that best match crop N demands. A logical strategy for determining the appropriate rate and timing for post-emergence N applications is to make adjustments based on in-season plant monitoring. This approach relies on plant measurements during the growing season, and therefore, is able to account for the effect of seasonal weather conditions on crop N availability (Meisinger et al., 2008).

Hyperspectral reflectance acquired during the growing season can be used to monitor crop N status since: (1) spectral characteristics of green-leaved vegetation change as leaf chlorophyll content changes, and (2) N is closely related to chlorophylls in plant cell metabolism (Miao et al., 2009; Stroppiana et al., 2012). Reflectance of vegetation, specifically in the visible (i.e., 400-670 nm) and near-infrared (NIR; i.e., ~670-1000 nm) regions of the spectrum, are most influenced by leaf chlorophyll content (Jongschaap and Booij, 2004). The absorption of light energy by chlorophyll results in low leaf reflectance in the visible region, and the scattering of light energy by the mesophyll results in high leaf reflectance in the NIR region (Rees, 2001). As leaf chlorophyll content increases, leaf reflectance in the visible and NIR regions generally decrease and increase, respectively (Goffart et al., 2008; Rees, 2001). In addition to the optical properties of leaves, N deficiencies affect canopy architecture and leaf area (Moran et al., 2004), which can also affect the spectral characteristics of a crop at the canopy level.

Research on the use of hyperspectral imagery and development of appropriate indices for the potato crop under various cultural conditions is limited. Therefore, the objective of this study was to evaluate the potential for using canopy-level hyperspectral reflectance as an indicator of potato N status in two varieties subjected to various N treatments and irrigation regimes.

2. Materials and methods

2.1. Study site

Field experiments were conducted over two years (2010-2011) at the University of Minnesota Sand Plain Research Farm (45°23’N, 95°53’W) near Becker, MN. The soil at this location is classified as an excessively drained Hubbard loamy sand (sandy, mixed, frigid Typic Hapludoll) comprised of 82% sand, 10% silt, and 8% clay. The available water holding capacity in the top 120 cm of soil is 10 cm. During the growing season (April – September), the 30-year average temperature and rainfall are 16.5° C and 550 mm, respectively (MRCC, 1971-2000). The previous crop in both years was non-irrigated cereal rye (*Secale cereal* L.).

2.2. Experimental design

Each year, the experiment was set up as a randomized complete block design with a split-split plot restriction on randomization, replicated four times. The whole plot treatment was irrigation rate (unstressed and stressed). Irrigation was applied with an overhead sprinkler system. A water balance method was used to schedule irrigation rate for the unstressed treatment (Wright, 2002). The stressed plots were irrigated at a rate in which total water equaled approximately 89% and 81% of the unstressed plots in 2010 and 2011, respectively and was calculated from total water applied between emergence and vine kill. Timing was variable between years and depended on weather conditions. On average, approximately four days elapsed between irrigation applications.

The subplot treatment included a low, medium, and high N rate (i.e., 34, 180, and 270 kg N ha^{-1}) with variable timing of post-emergence N applications for the high rate, for a total of five N treatments (i.e., 34 N early, 180 N split, 270 N split, 270 N split + surfactant (s), and 270 N early; Table 5-1). The 270 N split and 270 N split + s had the same rate and timing of N application, but a soil surfactant (IrrigAid Gold) was applied to 270 N split + s at a rate of 10 L ha^{-1} on top of the hill just prior to emergence to investigate the effects of the surfactant on N uptake under different irrigation regimes. On average, about two weeks passed between split applications of post-emergence N for the 180 N split, 270 N split, and 270 N split + s treatments. Because the 180 N split, 270 N split, and 270 N split + s treatments had split applications of post-emergence N fertilizer, actual N applied at the time of data acquisition varied. All N applications were completed by the later image date in 2010, but one post-emergence N application remained at the later image date in 2011. The 270 N early treatment had the same N rate as the 270 N split treatments, but all post-emergence N was applied early. In both years the planting and emergence N source was mono-ammonium phosphate and urea, respectively. Post-emergence N was split applied four times by hand as a 1:1 mixture of urea and ammonium nitrate in 2010 and five times by spray boom and tractor as 28% urea-ammonium nitrate solution in 2011. All post-emergence N was irrigated in immediately following application.

The sub-sub plot treatment consisted of two potato varieties, Russet Burbank (RB) and Alpine Russet (AR). Russet Burbank, the most widely grown processing potato variety in the U.S. and in Minnesota, has a mid-season maturity class of 111-120 days after planting (Whitworth et al., 2011). Alpine Russet, which has a late-season maturity class of 121-130 days after planting, is a relatively new variety that is being grown in Minnesota. Compared to RB, it has similar yields, superior processing quality after long-term storage, and greater resistance to water-stress induced tuber defects (Whitworth et al., 2011).

In both years, whole “B” seed and cut “A” see were used for the RB and AR varieties, respectively. Seed was hand planted in furrows with 90 cm row spacing and approximately 30 cm spacing between seed pieces within rows. Each plot consisted of seven 13.7 m rows. Planting dates were 16 April 2010 and 29 April 2011 and plant emergence occurred on 15 May 2010 and 24 May 2011. Two days after plant emergence in each year, emergence fertilizer was applied and rows were mechanically hilled. Chemicals were applied as needed during the season for the control of pests, disease, and weeds according to standard practices in the region (Engel et al., 2012).

2.3. Data collection

Leaf samples were collected on 1 July and 5 August in 2010 and 7 July and 28 July in 2011. The fourth leaf from the apex of the shoot was sampled from twenty plants in the fifth row from the alley in each sub-sub plot. Tissue samples were oven-dried at 60°C and ground with a Wiley mill to pass a 2 mm screen. Total N concentration was determined with an Elementar Vario EL III combustion analyzer (Horneck and Miller, 1997).

Aerial hyperspectral imagery was acquired with an Airborne Imaging Spectrometer for Applications (AISA Eagle) visible/near-infrared hyperspectral imaging sensor

(SPECIM, Spectral Imaging, Ltd., Oulu, Finland) by the Center for Advanced Land Management Information Technologies (CALMIT) from the University of Nebraska-Lincoln, USA. The AISA Eagle is a complete pushbroom system, consisting of a hyperspectral sensor head, a miniature GPS/INS sensor, and a data acquisition unit in a rugged PC with a display unit and power supply. It has a 1,000 pixel swath width and was configured to capture imagery in 63 narrowbands (2.3 nm spectral resolution) covering a spectral range from 401 to 982 nm; band widths ranged from 8.8 - 9.6 nm. Images were captured with 1.0 m spatial resolution on 1 July [47 days after emergence (DAE)] and 6 August (83 DAE) in 2010 and on 6 July (43 DAE) and 29 July (66 DAE) in 2011. The flyover for the late growth stage was much later in 2010 than in 2011 due to equipment problems and weather-related scheduling conflicts; this resulted in apparent differences in the data that were likely due to a difference in actual crop maturity rather than a difference between years.

2.4. Image processing

A post processing software package, CaliGeo, was used for radiometric correction (using NIST traceable calibrations), rectification (using a C-Migits III GPS/INS unit manufactured by Systron Donner Inertial Division, Walnut Creek, CA, USA), and geo-referencing. Geographic coordinates of plot corners were acquired with a GPS unit (0.3 m accuracy), which allowed in-house geo-referencing to minimize image distortion. ENVI software (Version 4.8, Exelis, Inc.) was used for all further image processing.

To extract plot-based pixel data for all analyses, regions of interest (ROIs) were created for every treatment plot on each image date. Pixels that were most affected by bare soil were automatically excluded from ROIs by including only pixels that were above a minimum threshold value using a narrowband Greenness index (Smith et al., 1995) image. This technique was an effective way to exclude pixels that were affected by bare soil during growth stages with full canopy cover. Since plant senescence had already occurred for the low and medium N rates on the later image date in 2010 (i.e., 83 DAE), each pixel was affected by bare soil in these treatment plots to some extent; this resulted in ROIs that included pixels that were partially affected by bare soil on this date.

2.5. Statistical analysis

Data were analyzed as a randomized complete block design with four replications and a split-split restriction on randomization using the MIXED procedure of SAS with Type III ANOVA method at α=0.05 (SAS Institute, 2008). Pairwise comparisons of the least square means were made using the *lsmeans / pdiff* statement (α = 0.05) to determine significant differences among treatment means of the main effects and interactions in Table 5-2 (individual treatment means are not presented, but trends are discussed if effects were significant). Irrigation, N treatment, and variety were considered as fixed effects, while replication was considered as random.

To evaluate the effectiveness of canopy-level narrowband reflectance as an indicator of potato N status, the coefficient of determination was calculated between potato leaf N and canopy reflectance at all AISA Eagle narrowbands. The CORR procedure of SAS (SAS Institute, 2008) was performed to obtain a correlation coefficient (r) for each image date and variety, and then r was squared to obtain the coefficient of determination (r^2), which represents the quality of the relationship between the predictor (e.g., canopy reflectance) and the modeled values (e.g., leaf N concentration).

3. RESULTS AND DISCUSSION

Results are divided into three sections based on the type of data presented. The first two sections (reflectance spectra and linear regression analysis) begin with results that were similar between varieties, followed by results that were unique to each variety. The third section (analysis of variance) is divided into sub-sections for the main effects and interactions, and is presented last because the wavelengths selected for analysis were obtained from regions in which consistent trends were observed across image dates and varieties from linear regression analysis.

Canopy-level hyperspectral reflectance in the 400-980 nm range was obtained from the potato crop growing under five N treatments (Table 5-1, Fig. 5-1). Reflectance data were averaged across irrigation treatments to simplify the results in this study. This approach can be justified because the main effect of irrigation and its interactions were not statistically significant in most cases for leaf N and hyperspectral reflectance across image dates (Table 5-2). Furthermore, differences in r^2 were very small across and between irrigation, even in the limited cases where there was a significant effect of irrigation on leaf N or hyperspectral reflectance.

Due to above average rainfall in early- to mid-summer in both 2010 and 2011, there was little difference in total water (rainfall + irrigation) until ~55 DAE in both years.

3.1. Reflectance spectra

Reflectance spectra are presented for only 2011 image dates since similar trends were observed between years at most wavelengths (Figs. 5-1 and 5-2). Reflectance in the red and NIR regions (i.e., 600-980 nm) showed similar trends between varieties, but varied among N application rates and between image dates (Figs. 5-1 and 5-2).

On 43 DAE, reflectance was lowest for the higher N rates in the red region, and reflectance was highest for the higher N rates in the NIR region (Figs. 5-1 and 5-2). On 66 DAE, a different trend was observed, but only between the treatments with the high rates in which the timing of post-emergence N varied. The 270 N split treatments had lower reflectance in the red region and higher reflectance in the NIR region than the 270 early treatment, even though the 270 N split treatments had not yet received all post-emergence N applications by 66 DAE (i.e., total N applied at this time was higher for 270 N early than for the 270 N split treatments; Figs. 5-1 and 5-2). This suggests that there is less N uptake on 66 DAE if all post-emergence N is applied early than if it is split applied; the lower N uptake for 270 N early is likely due to losses of N fertilizer from the system, probably via leaching.

Reflectance in the visible spectrum (i.e., 400-680 nm) peaked in the middle of the green region [i.e., 554 nm (Fig. 5-1)] in both varieties on both image dates. The trend for reflectance among N application rates in the green region (i.e., 500-600 nm) varied by variety, primarily on 43 DAE. c (Fig. 5-1). This finding is consistent with previous reports of canopy reflectance in RB potato (Wang and Rosen, 2004), as well as with what is typical spectral behavior of vegetation in general (Goffart et al., 2008; Rees, 2001). Conversely, in AR, there was less difference in reflectance between N rates, and reflectance was lowest for the low N rate and highest for the medium N rate (Fig. 5-2). A similar trend was observed in potatoes grown in India for the K. Chandramukhi variety at 45 days after planting (Jain et al., 2007), although data from that study at a later growth stage was similar to the trends observed in RB in this study. Discussion was not provided by the authors regarding this difference, but in both cases, it is likely due to an external factor. Visual observations on this date suggest that it could be due to either the effects of bare soil or the effects of crop flowering (bare soil was present in the 34 N early treatment plots, and flowering was most prevalent in the 34 N early treatment plots, but only for AR). This trend was not observed in the 2010 data on 47 DAE, but there was less difference in reflectance between N rates for AR than for RB – AR was flowering for the 34 N early treatment on this date, but there was little bare soil.

3.2. Prediction of leaf nitrogen concentration from hyperspectral imagery

The r^2 representing the relationship between potato leaf N concentration and canopy reflectance varied as a function of hyperspectral wavelength (Figs. 5-3 and 5-4). For both varieties, r^2 decreased at most wavelengths as the crop matured and was especially true in the NIR region. Also, a dip in r^2 was observed in the green region for both varieties. For RB, the dips occurred only in the later image dates, and for AR, the dips occurred in both image dates in 2011 only. A connection between the two varieties regarding the occurrence of these dips cannot be explained from the data in this study since the dips only occurred in one year for AR. With the exception of the NIR region, the trends of r^2 as a function of hyperspectral wavelength (e.g., the spectral location of peaks and dips) differed between varieties.

3.3. Prediction trends in Russet Burbank

In general, canopy reflectance was better correlated to RB potato leaf N on the earlier image dates (i.e., 47 and 43 DAE for 2010 and 2011, respectively) than on the later image dates in each year (i.e., 83 and 66 DAE for 2010 and 2011, respectively; Fig. 5-3). On the earlier image dates, r^2 was consistent as a function of wavelength between years, and the highest r^2 occurred in the green and red regions from ~518-638 nm, at the beginning of the red-edge from ~695-704 nm, and throughout most of the NIR region from ~751-924 nm (Fig. 5-3). In these regions, r^2 ranged from 0.74-0.78 in 2010 and from 0.76-0.82 in 2011. On the later image dates, r^2 was noticeably different as a function of wavelength between years. This is likely due to the 17 day difference in image acquisition relative to crop emergence on the later image date between years (i.e., imagery was captured 83 DAE in 2010 and 66 DAE in 2011). Because of this, data on the later image date cannot be directly compared between years.

There are, however, notable trends that can be observed by examining r^2 as a function of wavelength based only on DAE (i.e., examining the shape of the r^2 curves). As the potato crop matured, r^2 generally decreased for nearly every wavelength (Fig. 5-3). This trend is especially apparent throughout the entire NIR region, as well as in the red region from ~610-638 nm. The same trend also occurred with the peak at the beginning of the red-edge region from ~685-695 nm; this peak diminished as DAE increased (Fig. 5-3). On the later image dates (i.e., 66 and 83 DAE), maximum r^2 values were higher in the visible region than in the NIR region; the highest r^2 values for 66 and 83 DAE occurred at 610 and 582 nm, respectively.

A dip in r^2 in the green region occurred from ~518-572 nm on the later image date in each year. This dip is more pronounced at 83 DAE than 66 DAE, suggesting that the ability to detect N stress using this wavelength region decreases as growth stage progresses. On all image dates, the lowest r^2 occurred in the red edge around 713 nm, although this varied slightly depending on the growth stage (Fig. 5-3). The 563 nm band at 43 DAE had the highest r^2 (0.82) of all wavelengths for RB.

These results suggest that the differences in reflectance for RB potato with differing degrees of N stress become less apparent during the later growth stages. The optimal date to obtain hyperspectral measurements over a RB potato canopy to detect N stress cannot be precisely determined based solely on this data; however, earlier dates appear to be better correlated with leaf N concentration.

3.4. Prediction trends in Alpine Russet

The peaks in r^2 were relatively consistent as a function of wavelength for AR on the earlier image dates between years. Peaks occurred in the green and red regions from ~572-610 nm, in the beginning of the red-edge from ~695-704 nm, and throughout most of the NIR region from ~751-924 nm (Fig. 5-4). Throughout most of the NIR region from ~751-924 nm, r^2 ranged from 0.70-0.72 in both years (Fig. 5-4).

In the green region, a dip occurred in r^2 from ~536-554 nm for both image dates in 2011; however, the dip was much less pronounced on 66 DAE (Fig. 5-4). The formation of these dips was not consistent with either of the image dates in 2010, and is likely a result of the unexpected similarity in reflectance among N treatments in the green region (Fig. 5-2). These dips may have formed because of an external factor that was only present in 2011, but currently there is no definitive explanation for their occurrence. If these dips are present in other AR data, it could potentially cause poor performance for narrowband indices that use those wavelengths to detect N stress.

The relationship between leaf N concentration and canopy reflectance decreased as the crop matured (Fig. 5-4). This trend is especially apparent throughout the entire NIR region, but is also evident in the visible and beginning of the red-edge regions from ~400-704 nm (with the exception of the 83 DAE image date). The r^2 values on 83 DAE in the blue and red regions were remarkably higher than for any of the other image dates. The 527 nm band on this date had the highest r^2 (0.74) of all wavelengths for AR (Fig. 5-4). This could have occurred because of a combination of the cooler night-time temperatures in 2010 and the late season maturity class of AR, which likely resulted in better overall growing conditions during the late growth stages in 2010 for AR; however, this does not explain the low r^2 in the NIR region.

3.5. Analysis of variance for selected spectra and leaf nitrogen concentration

Analysis of variance for main effects (irrigation, N treatment, and variety) and their interactions was performed for canopy reflectance at four narrowband wavelengths (536, 601, 695, and 799 nm) and for leaf N (Table 5-2). These wavelengths were selected because these were the regions where differences in peaks and dips were evident for the relationship between leaf N concentration and reflectance between varieties and/or among image dates. The 536 nm band was near the reflectance peak in the visible region and was also near the central location of the dips that were observed on the later image dates for RB and in 2010 for AR. The 601 and 695nm bands were the locations of two of the r^2 peaks on all image dates and both varieties. The 799 nm band was selected to represent the NIR wavelengths since r^2 was consistent throughout most of the NIR region. Because the three-way interaction for all wavelengths and leaf N concentration on all image dates was not significant, only main effects and two-way interactions are discussed.

3.5. Main effects

The main effect of N was highly significant for all selected variables, and the main effect of variety was significant for most of the selected variables. The 695 nm band on the later image date in 2010 was the only selected variable that did not have a significant variety effect (Table 5-2). The main effect of irrigation was only significant for 536, 601, and 799 nm on the earlier image date in 2010. The differences in statistical significance for these wavelengths on the earlier image dates between years may be due to differences in water stress because of rainfall the day prior to image acquisition in 2011.

Among the interactions for the selected variables, N by variety was the most significant, followed by irrigation by variety (Table 5-2). The irrigation by N interaction was statistically significant for only leaf N on the later image date in 2010.

3.6. Nitrogen by variety

The two-way interaction between N and variety for leaf N was only significant on the earlier image date in 2010. On this date, leaf N was greatest for the treatments with the

270 kg N ha^{-1} rate in each variety; leaf N in RB was greater than AR for all N treatments except 34 N early, in which case it was statistically similar in both varieties. Similar to leaf N on the earlier image date in 2010, reflectance at 536, 601, and 695 nm on the earlier image dates in each year was greater for RB than for AR; however, reflectance was greater for 180 N split than for the treatments with the 270 kg N ha^{-1} rate in RB, but not in AR. More simply, reflectance at these wavelengths was much more static across N rates in AR than in RB. When there was a significant interaction between N and variety at these wavelengths on the later image date in 2011, reflectance for the 270 N split treatments tended to be similar in both varieties, but reflectance for 34 N early, 180 N split, and 270 N early was greater in RB than in AR. Since high reflectance at these wavelengths suggests an N stress, there is strong evidence of N stress for 34 N early and 180 N split in both varieties. Based on reflectance for the 270 N early treatment on the later image dates, the 601 and 695 nm bands show evidence for being able to detect the early stages of an N stress in RB only – reflectance at these wavelengths for 270 N early is greater in RB than in AR, even though reflectance for 270 N early is statistically similar to the 270 N split treatments in each variety.

The N by variety interaction was significant for reflectance at 799 nm on each image date in 2011, as well as on the later image date in 2010. On the earlier image date in 2011, reflectance for the 34 N early treatment was lower than the other N treatments to a greater extent in AR that in RB. This is the opposite of what was observed at 536, 601, and 695 nm on the earlier image date in 2011 – at the 536, 601, and 695 nm bands, reflectance decreased with increasing N rate and tended to be more static in AR than in RB; however, at the 799 nm band, reflectance increased with increasing N rate and tended to be more static in RB. On the later image date in 2011 at the 799 nm band, reflectance was similar between varieties for all N treatments except 270 N early, in which reflectance in AR was lower than in RB. It is also notable that reflectance for 270 N early in RB was similar to the 270 N split treatments, but reflectance for 270 N early in AR was lower than for the 270 N split treatments, suggesting an N stress for 270 N early in AR; this provides contradicting evidence towards the trends observed with reflectance at 601 nm and 695 nm for this image date.

On the later image date in 2010 at 799 nm, reflectance was similar among the treatments with the 270 kg N ha^{-1} rate in RB, but reflectance was greater for 270 N split + s than either 270 N split or 270 N early in AR. This suggests that the added soil surfactant either increased N uptake or prolonged growth in RB compared to the other N treatments. It is also notable that reflectance on the later image date in 2010 at 799 nm is greater in AR than in RB; this is different on all other image dates at 799 nm, and provides evidence that AR continues to grow at the dates when RB is beginning to senesce. Although reflectance is greater in AR on 83 DAE, leaf N is greater in RB, which suggests that there is (1) a difference in reflectance at 799 nm due to physiological differences in leaf composition between the varieties on this date, or (2) a background soil effect on reflectance in RB that decreases reflectance at 799 nm, even though leaf N is still relatively high.

3.7. Irrigation by variety

The two-way interaction between irrigation and variety was only significant at a few wavelengths for any particular image date, and they were not consistent over image dates (Table 5-2). Significant differences were likely due to either (1) rainfall that had occurred prior to the earlier image date in 2011, which resulted in a lack of any apparent water stress, or (2) differences in the amount of bare soil present with each variety on the later image date in 2010.

3.8. Irrigation by nitrogen

The two-way interaction between irrigation and N was significant for only leaf N on the later image date in 2010 (Table 5-2). Leaf N on this date was similar for 180 N split, 270 N split, and 270 N split + s in the unstressed irrigation, but was lower for 180 N split than for either of the 270 N split treatments. In addition, the added soil surfactant was also the reason for this significant response – leaf N was statistically similar for the 270 N split treatments in the unstressed irrigation, but was higher for 270 N split than for 270 N split + s in the stressed irrigation. The general lack of interaction between irrigation and N is confirmed both by spectral reflectance and leaf N data.

4. Conclusions

Using leaf N samples to determine in-season crop N status is a slow, destructive process that is not spatially representative. The use of canopy-level hyperspectral imagery to estimate crop N status is much faster and non-destructive, and can also account for spatial variability. However, the ability of hyperspectral narrowbands to accurately detect N stress depended on potato variety, growth stage, and sometimes, on the wavelengths used for prediction.

Both varieties clearly demonstrated that as the potato crop matured, the relationship between leaf N and wavelength decreased for most wavelengths, especially throughout the entire NIR region. Regardless of variety, the best performing wavelengths for detecting N stress across all image dates were those at the end of the green and beginning of the red regions from ~582-610 nm and those at the beginning of the red-edge from ~685-695 nm. However, the relationship between canopy reflectance and leaf N showed a more consistent trend among image dates for RB than for AR (on a relative scale). Consistent trends among image dates are more likely to lead to more accurate prediction of N stress, regardless of growth stage.

Reflectance in the NIR region could be used to predict N stress for both varieties, but only in the early growth stages since the ability of NIR wavelengths to predict crop N stress decreases as growth stage progresses. However, reflectance at 799 nm on 83 DAE was able to detect subtle differences in leaf N that reflectance at 536, 601, and 695 nm were not able to, but only for AR. hyperspectral data may not be absolutely necessary in the NIR region since r^2 was consistent across most of the NIR region, suggesting that NIR broadband reflectance obtained from a multispectral sensor would likely be as effective in predicting N stress as an NIR narrowband obtained from a hyperspectral sensor.

The presence or absence of an N stress regarding the timing of N application for a particular variety on a particular date, sometimes depended on which wavelengths were used. For example, on the later image date in 2010, reflectance at 601 and 695 nm suggested an N stress in RB when post-emergence N was applied early; however, reflectance at 799 nm on the same date suggested an N stress in AR when post-emergence N was applied early (based on the N by variety interactions).

Measurements of leaf N concentration and canopy-level hyperspectral reflectance should be obtained at more frequent intervals throughout the growing season to further understand (1) the behavior of the dips in r^2 in the green region and (2) the effects of timing of split applications of post-emergence N on reflectance.

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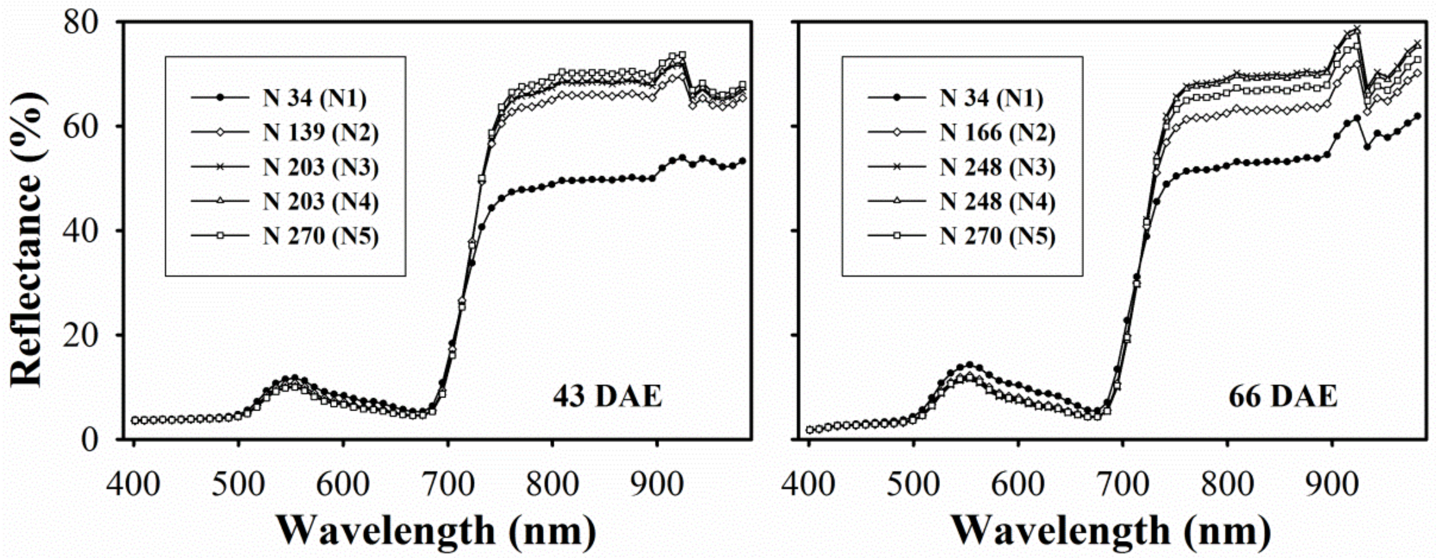

Fig. 5-1. Reflectance spectra of Russet Burbank potato canopy under five nitrogen treatments at 43 (left) and 66 (right) days after emergence (DAE) in Becker, MN (2011). Treatments are labeled according to actual nitrogen applied at the time of image acquisition in kg N ha^{-1} (refer to Table 5-1).

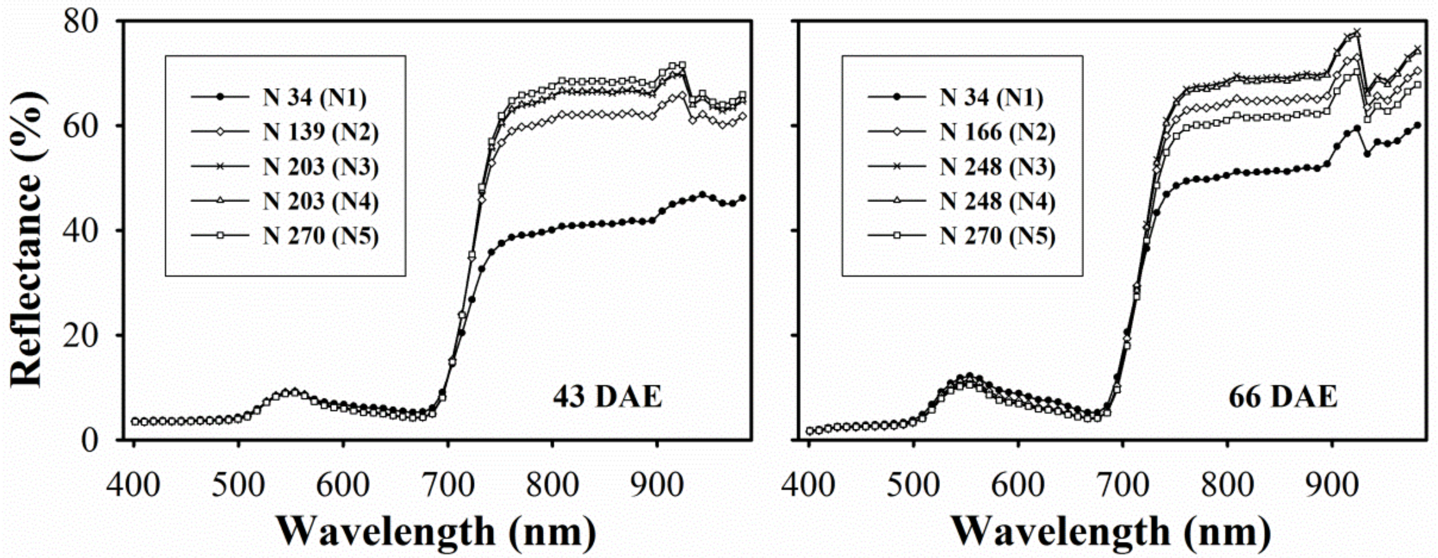

Fig. 5-2. Reflectance spectra of Alpine Russet potato canopy under five nitrogen treatments at 43 (left) and 66 (right) days after emergence (DAE) in Becker, MN (2011). Treatments are labeled according to actual nitrogen applied at the time of image acquisition in kg N ha^{-1} (refer to Table 5-1).

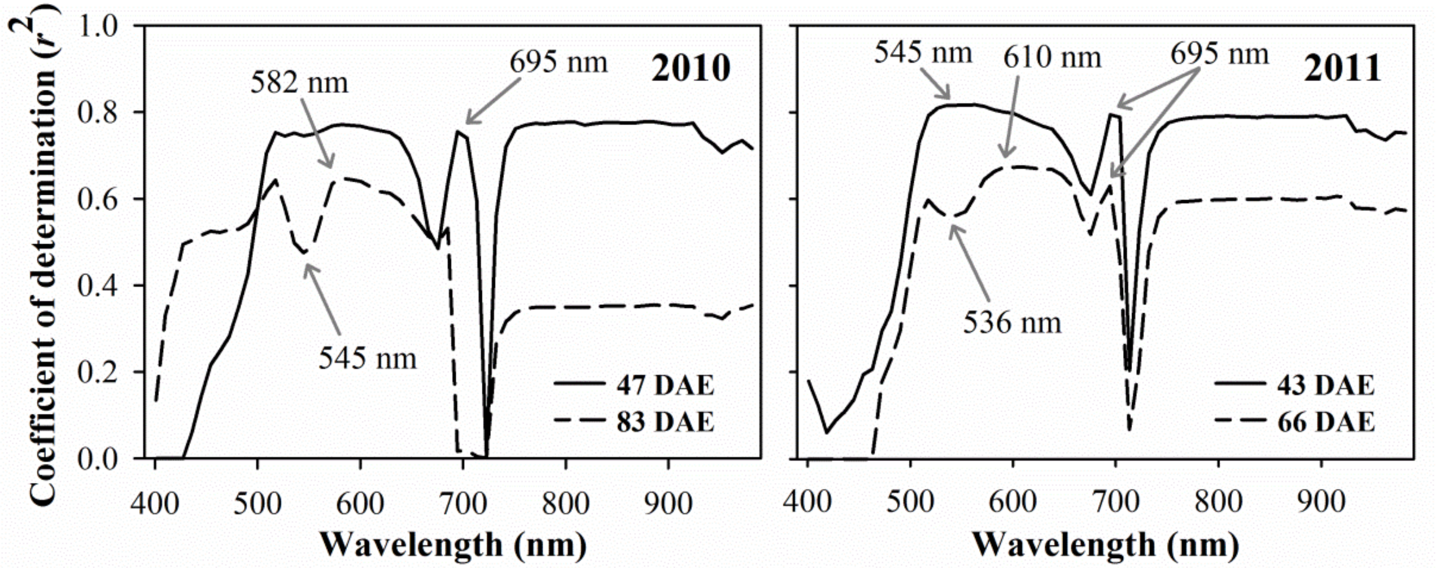

Fig. 5-3. Coefficient of determination (r^2) for the relationship between leaf nitrogen concentration and canopy reflectance at all AISA Eagle wavelengths (401-982 nm) for Russet Burbank potato. Data are grouped by days after emergence (DAE) for 2010 (left) and 2011 (right).

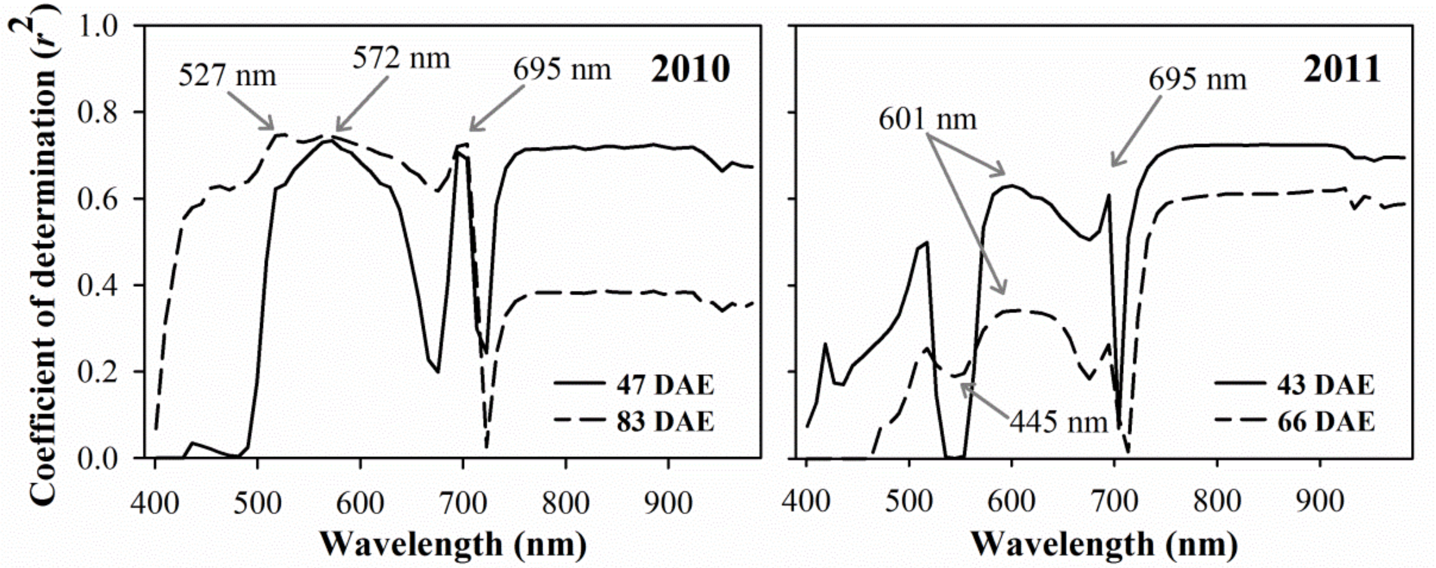

Fig. 5-4. Coefficient of determination (r^2) for the relationship between leaf nitrogen concentration and canopy reflectance at all AISA Eagle wavelengths (401-982 nm) for Alpine Russet potato. Data are grouped by days after emergence (DAE) for 2010 (left) and 2011 (right).

Table 5-1. Rate and timing of nitrogen (N) fertilizer treatments for 2010 and 2011.

Nitrogen fertilizer treatment	Timing of application					Total N
	planting	emergence		post-emergence		
		2010	2011	2010	2011	
	---------------------------- kg N ha^{-1} ----------------------------					
34 N early	34	0	0	0	0	34
180 N split	34	78	90	17 × 4[‡]	11.2 × 5[‡]	180
270 N split	34	124	124	28 × 4[‡]	22.4 × 5[‡]	270
270 N split + s[†]	34	124	124	28 × 4[‡]	22.4 × 5[‡]	270
270 N early	34	124	124	112	112	270

[†] A soil surfactant (IrrigAid Gold) was used in this treatment, and was applied at a rate of 10 L ha^{-1}.

[‡] On average, about two weeks passed between split applications of post-emergence N.

Table 5-2. P-values of canopy reflectance for the analysis of variance of four narrowband wavelengths and leaf N on the four image dates.

Selected variables			Main effects			Interactions		
			nitrogen	variety	irrigation	N × var	irr × var	irr × N
2010	47 DAE	536 nm	**[†]	**	*	**	*	-
		601 nm	**	**	*	**	*	-
		695 nm	**	**	*	**	*	-
		799 nm	**	**	-	-	-	-
		leaf N	**	**	-	**	-	-
	83 DAE	536 nm	**	-	-	*	-	-
		601 nm	**	**	-	-	-	-
		695 nm	**	-	-	-	-	-
		799 nm	**	**	-	*	*	-
		leaf N	**	**	-	-	-	*
2011	43 DAE	536 nm	**	**	-	**	-	-
		601 nm	**	**	-	**	*	-
		695 nm	**	**	-	**	*	-
		799 nm	**	**	-	**	-	-
		leaf N	**	*	-	-	-	-
	66 DAE	536 nm	**	**	-	**	-	-
		601 nm	**	**	-	**	-	-
		695 nm	**	**	-	**	-	-
		799 nm	**	*	-	*	-	-
		leaf N	**	**	-	-	-	-

[†] ** and * are significant at $\alpha = 0.01$ and $\alpha = 0.05$; - is nonsignificant.

Chapter 6.

EVALUATION OF THE NITROGEN SUFFICIENCY INDEX FOR USE WITH HIGH RESOLUTION, BROADBAND AERIAL IMAGERY IN A COMMERCIAL POTATO FIELD

Chapter Summary

Canopy-level spectral data has the potential to be a reliable tool for making in-season nitrogen (N) management decisions for a potato (*Solanum tuberosum*, L.) crop. The nitrogen sufficiency index (NSI) can be used to normalize spectral measurements so spectral data is calibrated to local conditions. This study verified the ability of spectral imagery to be used for predicting N stress, and then evaluated the implications of using high spatial resolution broadband spectral imagery for determining N prescriptions at different growth stages after being normalized by an NSI. Canopy-level aerial images were obtained on 30, 56, and 79 days after emergence (DAE) with a Redlake MS4100 multispectral camera. Imagery was acquired for research plots (each plot represented an area of 98 m^2), as well as for a commercial potato field (59 ha). In the research plots, experimental treatments included five N treatments with varying rates and timing of N fertilizer and two potato varieties, Russet Burbank (RB) and Alpine Russet (AR). The research plots showed that the Green Ratio Vegetation Index (GRVI) predicted N stress based on leaf N concentration the best among all broadband spectral indices tested on individual dates (r^2 values ranged from 0.61 – 0.83 and root mean square error of cross-validation ranged from 15 – 27% among image dates and varieties). NSI values that used two standard deviations above the mean value of the over-fertilized commercial field as the reference were substantially higher than NSI values that used the mean value of the over-fertilized commercial field as the reference; this is evidence that luxury consumption of N fertilizer does occur in potatoes to some degree. GRVI normalized by an NSI that used the recommended rate and timing from the research plots as a reference could detect areas of the commercial field that were most unsuitable for supplemental N fertilizer applications on different measurement dates. On 56 and 79 DAE, most of the commercial field was above the GRVI NSI over-sufficiency threshold level of 120% (the mean pixel values were greater than 127% for both varieties). Therefore, most of the areas of the commercial field were determined to be unsuitable for supplemental N fertilizer applications on these dates. Because of differences in potato variety, growth stage, or other local conditions, reference areas are needed in order to make accurate recommendations. The images with sub-meter spatial resolution obtained by the Redlake

MS4100 provided spectral and spatial information for the commercial field that could not be detected by the coarser spatial resolutions at the early and late growth stages, but provided similar information at the growth stage in which peak vegetative growth occurred. The coefficient of variation of GRVI images with 0.25 m spatial resolution was 120% lower on 30 DAE and 77% lower on 79 DAE than GRVI images with 1.0 m spatial resolution, but was similar on 56 DAE. The authors propose a practical approach and the implications associated with using spectral data for in-season N management. To determine the degree to which luxury consumption affects spectral data in potatoes, future research should address differences between spectral measurements made both before and after split applications of post-emergence N fertilizer; over-fertilized plots should be compared to plots that are fertilized at the recommended rate and timing.

Keywords: High spatial resolution imagery, broadband indices, nitrogen sufficiency index (NSI), chlorophyll meter, nitrogen, potato

1. Introduction

The coarse textured soils typically used for irrigated potato (*Solanum tuberosum*, L.) production are relatively low in organic matter and cation exchange capacity, and therefore, are generally low in soil nutrient reserves. The low inherent soil fertility of these soils, coupled with the large nitrogen (N) requirement for a potato crop, leads to high fertilizer inputs (Dean, 1994). Potato plants are relatively shallow rooted compared to other field and vegetable crops and are very sensitive to water stress (Bailey, 2000; Lesczynski & Tanner, 1976). This leads to below average nutrient uptake and poor nitrogen use efficiency (NUE), which potentially results in high rates of nitrate leaching (Lynch et al., 2012; Rosen & Bierman, 2008; Zvomuya et al., 2003). Previous studies have shown that only about a third to a half of applied N is recovered by an irrigated potato crop in years of moderate to heavy leaching (Errebhi et al., 1998; Waddell et al., 2000). Therefore, precise nitrogen management for irrigated potato is important in order to optimize production and to minimize environmental N losses.

Matching the timing and rate of N fertilizer with the N needs of the crop during different growth stages will help to optimize crop uptake while minimizing N losses (Canter, 1997; Errebhi et al., 1998). Increased NUE, N uptake, and yields have been shown to result from split applications of N fertilizer during tuber initiation and bulking, especially during leaching years (Errebhi et al., 1998; Westermann et al., 1988; Nigon et al., 2012a). The use of fertigation provides a convenient method to split apply post-emergence N applications. The challenge lies in the ability of the producer to estimate the appropriate rates and timing of split N applications so that fertilizer N best matches crop demands. A logical strategy for determining the exact rate and timing of post-emergence N applications is to make adjustments based on in-season plant monitoring. Since this approach uses plant measurements during the growing season, it is able to account for the effect of seasonal weather conditions on crop N availability (Meisinger et al., 2008).

The opportunity exists to use aerial or satellite imagery to predict crop biophysical parameters that depend on crop N uptake such as tissue N concentration and/or leaf chlorophyll concentration. Since plant N status is closely related to chlorophyll in plant cell metabolism, spectral imagery can be used to monitor crop N status because the spectral characteristics of green-leaved vegetation change as leaf chlorophyll content changes (Stroppiana et al., 2012). There are a variety of multispectral and hyperspectral cameras on the market that can be used to obtain canopy-level spectral imagery. The spatial resolution capabilities of these cameras can vary, and this affects the amount of variability that is detected by the spectral imagery (Woodcock & Strahler, 1987).

The nitrogen sufficiency index (NSI) can be applied to spectral imagery in order to normalize the measurements to a non-limiting N reference area so that the ability of a producer to make N management decisions can be enhanced (Peterson et al., 1996). NSIs based on spectral images can be implemented across a very large spatial scale, and therefore, can be a very practical diagnostic tool for prescribing split applications of post-emergence N fertilizer (Tremblay et al., 2011). An NSI depends on the use of a reference value, which has traditionally been a measurement from an over-fertilized, N-rich area of the field (Blackmer & Schepers, 1995; Denuit et al., 2002; Sripada et al., 2008). Normalizing data to these over-fertilized, N-rich areas of the field eliminates the need to develop a specific calibration function for each set of variables or local conditions (e.g., soil type, variety, etc.; Shapiro et al., 2006). Over-fertilized reference plots may not be suitable for determining supplemental N in a potato crop because previous research has shown that spectral measurements for plots with medium N rates have not been significantly different from spectral measurements for over-fertilized plots (Denuit et al., 2002; Olivier et al., 2006). Furthermore, over-fertilization of irrigated potato results in reduced tuber yields (Rosen & Bierman, 2008). Over-fertilized reference plots may not be sufficient for determining supplemental N in a potato crop because plots or fields without N fertilizer are also needed to show that chlorophyll meter readings are significantly different between the over-fertilized and under-fertilized plots.

To our knowledge, research investigating the use of aerial-based broadband spectral indices after being normalized by an NSI has not been published for potato. Broadband spectral data from aerial imagery is able to account for within-field spatial variation, and therefore, can be a more practical approach than chlorophyll meter readings for determining in-season N prescriptions. The objectives of this study were: (i) to verify that spectral measurements can indeed be a good indicator of N status in a potato crop, (ii) to determine how NSI values can vary at different growth stages because of differences in the NSI reference used, (iii) to identify whether estimates of N stress can be made in a commercial field without using a reference N strip, and (iv) to investigate how the variability within a commercial potato field changes with spatial resolution at different growth stages.

2. Materials and methods

2.1. Study sites

Field experiments were conducted in 2011 at the University of Minnesota Sand Plain Research Farm (45°23’N, 95°53’W) near Becker, MN. In addition, image data and plant samples were acquired for a 59 ha center-pivot irrigated commercial field which was located 3.2 km southeast of the experimental plots. The northern ~60% was planted into the Alpine Russet variety, and the southern ~40% was planted into the Russet Burbank variety. There is approximately a 2.5 m change in elevation from the highest point of the field (northeast corner) to the lowest point of the field (southeast corner); this equates to an average slope less than 0.5%. The soils at both locations are predominantly classified as an excessively drained Hubbard-Mosford complex (sandy, mixed, frigid Typic Hapludolls). In the upper 30 cm of the soil profile, the textures of the Hubbard and the Mosford series are loamy sand and sandy loam, respectively. In the upper 150 cm of soil, the available water holding capacities of the Hubbard and the Mosford series are 10 and 12 cm, respectively. During the growing season (April – September), the 30-year average temperature and rainfall (1971-2000) are 16.5° C and 550 mm, respectively (Midwest Regional Climate Center).

2.2. Experimental design of research plots

The field study at the research farm was set up as a randomized complete block design with a split-split plot restriction on randomization replicated four times. The whole plot treatment was irrigation rate (i.e., unstressed and stressed); to evaluate the hypotheses in this particular study, only data from the unstressed plots were used. Therefore, the data from the field study were analyzed using a randomized complete block design with a split plot restriction on randomization. The treatments from the research plots are described in detail by Nigon et al. (2012c); only data from the 2011 field experiment were used in the current study. The subplot treatment included a low, medium, and high N rate with variable timing of post-emergence N applications for the high rate, for a total of five N treatments. The N treatments were: 34 early, 180 split, 270 split, 270 split + surfactant (s), and 270 early (Table 6-1). For the treatments that had split applications of post-emergence N, actual N applied at the time of data acquisition varied (Table 6-2). The sub-subplot treatment consisted of two potato varieties (i.e., Russet Burbank and Alpine Russet).

To estimate N supplied by precipitation and irrigation, water samples were collected using a plastic bottle with a screw-on cover; samples were frozen until analysis. The concentrations of NO_3-N and NH_4-N from the samples were determined using a Wescan N analyzer. Following reduction of NO_3-N to NH_4-N using granular zinc, NO_3-N was determined as the difference between NH_4-N + NO_3-N and NH_4-N (Carlson, 1986; Carlson et al., 1990). Total N supplied by rainfall + irrigation was 41 kg N ha^{-1}.

2.3. Aerial image acquisition

Aerial multispectral imagery was acquired with a Redlake MS4100 3-Charge Coupled Device (CCD) digital multispectral camera by the Upper Midwest Aerospace Consortium (UMAC) from the University of North Dakota, Grand Forks, ND. The Redlake MS4100 uses three CCD imaging sensors and has a 1920 × 1080 pixel resolution. The CCD sensors captured energy at wavelengths ranging from 775-825 nm, 650-695 nm, and 505-575 nm to correspond to broadbands centered in the near-infrared (NIR), red, and green regions of the electromagnetic spectrum, respectively. The Redlake MS4100 captured images of the treatment plots and commercial field at a 0.25 m spatial resolution from approximately 235 m above the ground between 1000 h and 1500 h local time on 23 June, 19 July, and 11 August 2011; in the experimental year, these dates corresponded to

30, 56, and 79 DAE, respectively (Table 6-2). ENVI software (Version 4.8, Exelis, Inc., USA) was used for image analyses.

2.4. Plant sampling and field measurements

Tissue samples (petioles and leaflets) were collected on or within one day of each image date, as well as on 43 and 66 DAE (Table 6-2), at both the treatment plots and the commercial field. Sampling methods, chlorophyll meter measurements, tissue analysis, and harvesting methods were conducted as described in detail by Nigon et al., (2012b).

Leaf N concentration and yield data were analyzed using PROC MIXED (SAS Institute, 2008) with N treatments and variety considered as fixed variables and replications considered as random variables. For the main effects and interactions of the fixed variables, pairwise comparisons of the least square means were made using the *lsmeans / pdiff* option of the MODEL statement (α = 0.05). The PDMIX800 macro (Saxton, 1998) was used to place treatment means into letter groupings based on the pairwise comparisons.

2.5. Image analysis

Spectral data were always kept in image format until it was necessary to perform statistical procedures outside of ENVI.

2.5.1. Spectral indices

A preliminary analysis was conducted in the research plots using a list of 17 previously published broadband spectral indices on four dates over two years in order to determine the indices that had the best overall ability to detect N stress. The correlation coefficient (r) was calculated for each of these indices using the CORR procedure of SAS (SAS Institute, 2008) in order to determine the indices that had the best relationships with leaf N concentration on each date. To determine which indices performed best overall, r^2

was averaged for each index among the four dates. Some of the indices that had the best average r^2 from this preliminary analysis are listed in Table 6-3, and were used for comparison in all subsequent analyses in this study. Narrowband NDVI did not perform particularly well in the preliminary analysis, but it was included for comparison with other indices and previously published research.

2.5.2. Pixel extraction

To extract pixel data for subsequent analysis, regions of interest (ROIs) were created for each treatment plot at the research farm and for each variety at the commercial field. Depending on the analysis, different pixels were used within each treatment plot or field area. The treatment plot images were used for the prediction analysis; ROIs were created to only include pixels representing the middle of the plots. The commercial field images were used for the normalized nitrogen sufficiency and variability analyses. For the normalized nitrogen sufficiency analysis, pixels that were most influenced by the effects of bare soil were excluded from being used in data analysis; this was done by including pixels in the ROIs only if they were above a minimum threshold when tested against structural indices. A broadband NDVI image (Rouse et al., 1974) was used for the multispectral imagery (i.e., imagery obtained on 30, 56, and 79 DAE). This technique effectively filtered out pixels that were most influenced by bare soil so that only the pixels that represented a high proportion of vegetation were used in the NSI analysis. For the variability analysis, all the pixels within the commercial field were used.

2.5.3. Prediction analyses

A linear regression analysis with leaf N concentration was performed for chlorophyll meter readings and spectral indices from the treatment plots at the research farm using the REG procedure of SAS (SAS Institute, 2008) in order to obtain the coefficient of determination (r^2). The root mean squared error of cross-validation (RMSECV) was calculated from the predicted residual sum of squares (PRESS) according to Eq. (6-1) and

Eq. (6-2). The PRESS (Eq. (6-1)) is used as an indication of the predictive power of a model. It simulates prediction by repeatedly fitting the model, while leaving out the observation that it is attempting to predict each time the model is fit (Méndez Mediavilla et al., 2008). Since the observed samples y_i are not used in fitting the model, the predicted values $\hat{y}_{(i)}$ and their residuals are completely independent of y_i. This independence of the predicted residuals enables the PRESS to be a true assessment of the prediction capabilities of the model (Méndez Mediavilla et al., 2008).

The RMSECV was used together with r^2 to compare the predictive capability of the spectral data (RMSECV values were normalized by calculating the ratio between RMSECV values and the range of predicted values). The chlorophyll meter readings and spectral indices with the highest r^2 and lowest RMSECV were considered to be the best. Prediction analyses for chlorophyll meter readings and spectral indices were performed within and across dates so they could be more precisely evaluated.

$$PRESS = \sum_{i=1}^{n} (y_i - \hat{y}_{(i)})^2 \qquad \text{Eq. (6-1)}$$

where PRESS is the predicted residual sum of squares; y_i is the observed value of sample i; $\hat{y}_{(i)}$ is the predicted value of sample i not included in the model formulation; and n is the total sample number.

$$RMSECV = \sqrt{\frac{PRESS}{n}} \qquad \text{Eq. (6-2)}$$

where RMSECV is the root mean squared error of cross-validation; *PRESS* is the predicted residual sum of squares (Eq. (6-1)); and n is the total sample number.

2.5.4. Normalized nitrogen sufficiency

A nitrogen sufficiency index (NSI) was applied to spectral indices of the commercial field in order to normalize them to a “non-limiting” N reference area (Peterson et al., 1996), so the within-field variability could be compared among indices. The NSI was calculated using one of three different reference values according to Eq. (6-3), and they were critiqued for their practicality at a large scale.

$$NSI = \frac{N_i}{N_{ref}} \times 100 \qquad \text{Eq. (6-3)}$$

where NSI is nitrogen sufficiency index; N_i is the measured pixel value of the spectral indices; and N_{ref} is the reference value.

The three reference values used in this study were: (i) the higher average replicate value of either 270 split or 270 split + s from the research plots, (ii) the mean value of the commercial field, or (iii) two standard deviations above the mean value of the commercial field. These references were chosen to represent three scenarios that are likely to be implemented by potato growers. The first used a reference that was obtained from plots in which the University of Minnesota recommended rate and timing of N fertilizer had been applied. The second scenario is to use the mean value from an over-fertilized reference plot. Because 105 kg N ha^{-1} more than the recommended N fertilizer rate was applied to the commercial field in the current study, the mean value of the entire commercial field served as the reference for this scenario. The third scenario is to use the maximum value from an over-fertilized reference plot; two standard deviations above the mean value of the entire commercial field served as the reference value for this scenario in order to eliminate the possibility that an outlier was used as the reference.

The reference values were calculated separately for each sample date and each variety. For NG, the index values representing the treatments with the most N stress were higher than those representing the treatments with the least N stress; therefore, the reciprocal of the NSI was used for these indices (this resulted in the NSI values for these indices to be slightly over-estimated compared to the NSI values for the other indices).

2.5.5. Variability as it changes with spatial resolution

The coefficient of variation (CV) was calculated to estimate the relative amount of variability present in the broadband spectral indices at various spatial resolutions; this was done in order to determine the spatial resolution in which optimum spatial variability is achieved. To obtain images with different spatial resolutions, pixel aggregates were calculated from the original images (i.e., the images with the finest spatial resolution) so each new pixel represented an average spectral value of the aggregated pixels. In this study, the CV describes the amount of variability present among all pixels within either variety of the commercial field for a particular spectral index. Therefore, the spatial resolutions and indices with larger CVs have a greater range of values for the pixels within the commercial field. The relative fluctuation (RF) in CVs among spatial resolutions was calculated to determine the spectral indices that had the most consistent variability across spatial resolutions (Eq. (6-4)).

$$RF = \frac{CV_{max} - CV_{min}}{CV_{avg}} \times 100 \qquad \text{Eq. (6-4)}$$

where RF is relative fluctuation; CV_{max} and CV_{min} are the maximum and minimum coefficient of variation values among spatial resolutions for a specific spectral index, respectively; and CV_{avg} is the average coefficient of variation among spatial resolutions.

3. Results and discussion

3.1. Tuber yield and leaf N concentration

The main effect of variety for total tuber yield in research plots was not statistically significant (α=0.05), so tuber yield was presented across varieties (Table 6-4). The N treatments that had post-emergence N split applied had the highest tuber yields numerically among N treatments. The 270 N early treatment had a statistically lower yield than 270 N split, despite similar overall N rates. Errebhi et al. (1998) and Westermann et al. (1988) reported similar yield results with split applications of N fertilizer, especially during leaching years. The 34 N early treatment had significantly lower yields than all other N treatments. Tuber yield data for the commercial field was similar to the 270 N split treatments from the research plots (personal communication; data not presented).

Nitrogen treatment and variety significantly affected leaf N concentration in research plots on all dates (analysis of variance not shown); therefore, differences in leaf N are reported among N treatments for each of the measurement dates and each variety (Table 6-5). Within sampling dates, leaf N increased as the rate of applied N fertilizer increased (refer to Tables 6-1 and 6-2). Leaf N was above the threshold sufficiency level of 35 g kg^{-1} (Westermann, 1993) for both varieties, for all N treatments except the 34 N early treatment, and on all sample dates. Refer to Nigon et al. (2012b) for a detailed discussion of leaf N concentration among N treatments, between varieties, and over the course of the growing season from the research plots. Leaf N concentration in the commercial field was substantially higher than the 270 N split treatments on 56 and 66 DAE for both varieties. By 79 DAE, leaf N in the commercial field was comparable to that of the 270 N split treatments for RB, and was substantially lower for AR. Leaf samples in the commercial field were not collected on 30 and 43 DAE.

3.2. Nitrogen stress prediction

Because, leaf N concentration in the research plots was significantly different between varieties on each measurement date (data not shown), the linear regression analysis was performed separately for each variety. The r^2 values based on the linear relationship between leaf N and the chlorophyll meter readings or spectral indices and their corresponding root mean squared error of cross-validation (RMSECV) values were reported within and across dates for both RB and AR (Table 6-6). The chlorophyll meter readings and spectral indices with the highest r^2 and lowest RMSECV were considered to be the best.

The indices that resulted in the best r^2 values and corresponding RMSECV varied by date and variety (Table 6-6). Based on the frequency of "best" r^2 and RMSECV values, the overall best broadband indices in the research plots within dates was NG for RB and GRVI for AR. GNDVI did have very similar r^2 and RMSECV values for both varieties, however. All broadband indices performed better than the chlorophyll meter readings on 30 DAE for both varieties; on the later dates, the chlorophyll meter readings performed better than the broadband indices on every date.

Chlorophyll meter readings and spectral indices from the research plots were evaluated over all dates in order to determine which measurement or index could best predict N stress over all dates (i.e., regardless of growth stage). Over all dates, the r^2 and RMSECV values for the chlorophyll meter readings and the broadband indices were significant; however, the chlorophyll meter readings and spectral indices did not have as strong of a relationship with leaf N concentration over all dates as they did within dates (r^2 values ranged from 0.39 to 0.68 and RMSECV values ranged from 18 to 34% over all dates). GRVI was able to predict N stress better than all other broadband indices, and was able to predict N stress better than the chlorophyll meter for the RB variety (Table 6-6).

Compared to the other indices, NDVI could predict leaf N concentration the best on 30 DAE. It is probable that NDVI did not perform well on the other dates because it is a structural index that is most sensitive to crop cover up to 90% crop closures; after 90% closure, NDVI values plateau (Barnes et al., 2000), even when leaf N concentrations vary among N treatments. Since a preliminary analysis was performed to select the best broadband indices, the r^2 values and corresponding RMSECV for the reported indices were roughly similar overall (the range in r^2 and RMSECV between the best and worst indices on each date was always less than 0.14 and 9% for r^2 and RMSECV, respectively). These N stress prediction analyses verify that broadband spectral indices have a good relationship with leaf N concentration, particularly within dates.

3.3. Feasibility of using a nitrogen sufficiency index

Contrary to previous research on potatoes (Denuit et al., 2002; Olivier et al., 2006), chlorophyll meter readings for the high N rate in the current study were different from chlorophyll meter readings for the lower N rates that were above 0 kg N ha^{-1}; this was the case on all dates for the AR variety, and all but the first date for the RB variety (data not shown). This shows that a lack of significance between N treatments is not the primary reason that using an over-fertilized reference plot for calculating a nitrogen sufficiency index (NSI) is unsuitable for a potato crop, as suggested by Denuit et al. (2002) and Olivier et al. (2006). However, because over-fertilization of N negatively affects tuber yield, it is plausible that spectral data representing a potato crop canopy would be affected by N over-fertilization. An NSI sufficiency threshold of 95% is used in many studies (Varvel et al., 1997; Zebarth et al., 2002). However, it is important to understand how the choice of a reference plot can affect NSI values (and therefore, the optimum NSI sufficiency threshold) used to determine the rate and timing of supplemental N fertilizer. Any variation in how an NSI reference plot is managed can ultimately influence the conditions in which supplemental N fertilizer would be applied. To determine differences among NSI references, three NSI reference scenarios were compared by applying them to several spectral analysis techniques (chlorophyll meter readings and four broadband indices) from a commercial potato field (Tables 6-7 and 6-8).

3.3.1. Variation within reference scenarios

There were differences in the variation of NSI values in the commercial potato field among the analysis techniques (i.e., the chlorophyll meter readings or the spectral indices) for a specific reference scenario. This is apparent by observing the different standard deviations among the analysis techniques within a reference scenario. There are two basic approaches to deal with this variation so that NSIs can be used for accurate fertilizer management decisions (assuming the different analysis techniques have an equal ability to predict crop N stress). The first approach is to determine a sufficiency threshold level based on the variation present for a specific analysis technique. This can be effective because although NSI values change among analysis techniques within a reference scenario, the actual N stress for a given field (or location within a field) does not change. The second approach is to select the analysis technique to use based on its coefficient of variation (CV; Nigon et al., 2012c). This approach should be used if it is desirable to use a fixed NSI sufficiency threshold (e.g., 95%); the user would select the analysis technique whose variation results in a desired NSI sufficiency threshold.

Differences in NSI values due to growth stage are mostly normalized since the NSI reference value changes with growth stage in order to accommodate differences in local conditions. However, before the spectral measurements are normalized, differences among growth stages are apparent (Fig. 6-2). Among the dates in which images of the commercial field were collected, the absolute values of GRVI peaked on 56 DAE (when peak vegetative growth occurred; Fig. 6-2). The same was true for the chlorophyll meter readings and the other spectral indices (NG reached a minimum; data not presented).

Regardless of the date, variety, or reference scenario used, the rank in CV (from most to least) was (1) GRVI, (2) NG, (3) GNDVI, and (4) NDVI; this was similar to the results of Nigon et al. (2012c). GRVI always had the most variation, and consequently, has the greatest likelihood to include pixel values below a fixed NSI sufficiency threshold. In contrast, NDVI has the least likelihood to include pixels below a fixed NSI sufficiency threshold. It is difficult to objectively compare chlorophyll meter readings to the spectral indices because there were only a limited number chlorophyll meter readings collected; the spectral indices included thousands of pixels, whereas the chlorophyll meter readings included mean values for only a couple locations in the commercial field.

3.3.2. Differences among NSI reference scenarios

A lower NSI value is meant to indicate a higher degree of N stress, and therefore suggests that N fertilizer should be applied to the crop. The scenario in which the mean value of the commercial field was used as the reference also used data from the commercial field for N_i in the NSI formula (Eq. (6-3)); this resulted in mean values of 100% (or close to 101% for NG because the reciprocal was used). The standard deviations vary slightly for the NSIs among the different reference scenarios, but the CV does not. This indicates that no matter which reference is used to calculate an NSI, the relative spread among the data will always be similar for the same chlorophyll meter reading or spectral index. The standard deviations and ranges included in Tables 6-7 and 6-8 can be used to observe NSI differences from an absolute perspective.

The different NSI values among the reference scenarios for a specific variety × date × spectral index combination in Tables 6-7 and 6-8 indicate that the absolute NSI value depends on the reference being used. For example, the mean NSI for GNDVI on 30 DAE for the RB variety is 105% when the recommended N treatment from the research plots was used as the reference, but the mean NSI value changed to 100% and 93% when the mean and two standard deviations above the mean of the commercial field were used as references, respectively (Table 6-7). Although the absolute NSI values differ among the reference scenarios, the N status of the actual crop does not change; therefore, differences among reference scenarios would lead to inaccurate supplemental N recommendations if a fixed NSI sufficiency threshold is used across all scenarios. The NSIs that used the recommended N treatment from research plots as the reference had the highest mean values of the three reference scenarios. This reference scenario is the most conservative, and has the least likelihood of over-application of supplemental N fertilizer. In contrast, the NSIs that used two standard deviations above the mean from the commercial field as the reference had the lowest mean values (this scenario has the highest likelihood of over-application of supplemental N fertilizer).

Tuber yields for the commercial field were slightly lower than that of the research plots for the 270 N split treatments (personal communication). The lower tuber yields for the commercial field that was over-fertilized with N fertilizer is similar to previous research on irrigated potato in central Minnesota. Rosen & Bierman (2008) found that tuber yield plateaus and actually declines for N fertilizer rates greater than ~270 kg ha^{-1}. The commercial field had more applied N than the treatment plots with the recommended rate on each date except 30 DAE (Table 6-2), and had higher leaf N concentrations than the treatment plots with the recommended rate on 56 and 66 DAE (Table 6-5). Although leaf samples were not collected on 30 or 43 DAE, it is likely that leaf N concentration was also higher in the commercial field on 43 DAE. This implies that over-fertilization in the commercial field resulted in increased leaf N concentrations. For both varieties, all NSI values were above 100% when the recommended rate from research plots was used as a reference (Tables 6-7 and 6-8). This demonstrates that over-fertilization in the commercial field also resulted in increased NSI values for all spectral analysis techniques. Measurements from the research plots were typically collected immediately prior to N fertilization, however, and there was potentially a slight N stress present during the times of the measurements (Tables 6-1 and 6-2).

Suppose we use 95% as a fixed NSI sufficiency threshold to determine supplemental N fertilizer applications. It is important to note that using a fixed NSI sufficiency threshold among multiple spectral analysis techniques that have different CVs (chlorophyll meter readings or spectral indices) will likely result in different N sufficiency results (Nigon et al., 2012c). This could ultimately lead to conflicting N fertilizer recommendations for an equivalent level of crop N stress among spectral analysis techniques. Because the mean NSI values were all above 100% when the recommended rate from research plots was used as the reference (Tables 6-7 and 6-8), most (or all) of the commercial field would not receive supplemental N fertilizer. This depends on which spectral analysis technique is used; techniques with low standard deviations and low CVs (e.g., GNDVI and NDVI) are the least likely to show pixels to be below the 95% NSI sufficiency threshold, and techniques with higher standard deviations (e.g., chlorophyll meter readings, GRVI, and NG) are most likely to show pixels to be below the 95% NSI sufficiency threshold. The spectral indices were normally distributed (Fig. 6-1), so a very small proportion of NSI values occurred outside of the 95% confidence limits (two standard deviations above and below the mean). The histograms of GRVI NSI values show that there are very few pixels below the 95% NSI sufficiency threshold on all measurement dates for RB and for all dates but 30 DAE for AR (Fig. 6-1). This analysis (using the recommended rate as a reference) suggests that supplemental N fertilizer should not be applied; any applications would essentially result in an over-application of N fertilizer.

When the reference was two standard deviations above the mean value from the commercial field, the mean NSI values were all below 100%. The mean NSI values for GRVI and NG were less than 95% on most dates for each variety, and the NSI values for chlorophyll meter readings were less than 95% on three of the four dates for RB (Table 6-7) and one of the four dates for AR (Table 6-8). Using the 95% confidence limits to gauge the distribution of NSI values, most locations of the commercial field were at or below the 95% NSI sufficiency threshold on several dates. This analysis (using two standard deviations above the mean as a reference) suggests that locations of the commercial field which had average NSI values would actually need additional N fertilizer applications in several circumstances, even though we know N fertilizer was being over-applied on most dates. Thus, we can rule out this approach for determining the rate and timing of supplemental N fertilizer.

When the mean value of the commercial field was used as a reference, the mean NSI values were 100% (or slightly above 100% for NG), and supplemental N fertilizer would not be recommended for most pixels (using an NSI sufficiency threshold of 95%). Because N fertilizer was over-applied to the commercial field on most dates at a uniform rate, it is difficult to draw conclusions using data from the commercial field alone.

The over-fertilized N reference area used for the NSI calculation in potatoes is based on the idea that chlorophyll/greenness (as measured by spectral analysis techniques) reaches a maximum value in which the potential yield is also at a maximum. In other words, the use of an NSI that uses an over-fertilized reference disregards the possibility that the maximum yield potential could occur at a chlorophyll/greenness level lower than the maximum NSI level (i.e., if luxury consumption affects chlorophyll/greenness level). Therefore, any differences between an adequately fertilized reference area and an over-fertilized reference area can cause misleading recommendations if an over-fertilized reference area is used to calculate NSI values; this would ultimately result in the over-application of N fertilizer to the crop.

3.3.3. Using the nitrogen sufficiency index for a practical purpose

In order to determine the rate and timing of post-emergence N fertilizer using plant-based remote sensing approaches, an N sufficiency threshold should be established. A method to determine this sufficiency threshold is to evaluate the linear relationship between leaf N concentration and the spectral index as N rate increases using data from the research plots. In this study, GRVI was selected to be used because it has a high CV and has a good relationship with leaf N concentration, however, other indices would also work well. Fig. 6-3 shows the relationship between leaf N concentration and GRVI after being normalized by an NSI for both varieties and all three image dates. The lower threshold sufficiency level for leaf N concentration is 40 g kg^{-1} at 30 DAE and 35 g kg^{-1} at 56 and 79 DAE (Rosen & Eliason, 2005; Westermann, 1993). These threshold levels are represented in Fig. 6-3 by the horizontal dotted lines in each plot. The lower threshold sufficiency level for GRVI NSI is assumed to be the point in which the linear model crosses below the leaf N concentration threshold. The GRVI NSI level at the point in which this occurred was between ~66% (on 30 DAE for RB) and ~80% (on 79 DAE for AR) for the different plots in Fig. 6-3. In order to be confident that yields will not suffer because of underestimation of actual N stress, the less conservative 80% GRVI NSI value was chosen to be the lower threshold sufficiency level. Therefore, a plant with a GRVI NSI value below 80% should receive supplemental post-emergence N fertilizer in order to optimize yields.

The commercial field was over-fertilized throughout the season, and there was very little N stress throughout the field. Using the recommended rate from the research plots as an NSI reference, GRVI resulted in mean NSI values well over 100% on most measurement dates; there were very few pixels below 100% (Tables 6-7 and 6-8; Fig. 6-1). However, these data can be used to show which areas in the commercial field are most *unsuitable* for supplemental N fertilizer applications by using an over-sufficiency threshold.

Because it was determined that GRVI has a lower threshold sufficiency level of 80%, a symmetrical difference above the 100% reference level (i.e., 120%) was determined to be the threshold at which crop N was over-sufficient (for comparison purposes, a vertical dotted line is shown at this 120% threshold level in Fig. 6-1). Fig. 6-4 shows several GRVI NSI ranges for the RB portion of the commercial field on 30, 56, and 79 DAE. Blue and purple represent areas that have over-sufficient N fertilizer, red represents areas that are N stressed, and yellow and green represent areas that are sufficient or may be on the borderline of either stressed or over-sufficient. The white areas were masked out because they had a high probability of being pure soil or mixed pixels.

On 30 DAE, there were few pixels that were above the 120% threshold (Fig. 6-1); this is illustrated in Fig. 6-4 by the small proportion of blue and purple pixels on 30 DAE. On 56 DAE, the mean pixel value increased dramatically (129% for RB and 142% for AR), and a large proportion of the pixels were above 120%. This transition can be easily seen in Fig. 6-4. On 79 DAE, the mean pixel value was well above 120% again; between 56 and 79 DAE, there was a similar proportion of pixels above and below 120% (Figs. 6-1 and 6-4).

Overall, this analysis demonstrates that there was a sufficient quantity of N fertilizer applied to the commercial field on 30 DAE; however, it provides evidence that most of the commercial field was unsuitable for supplemental N fertilizer applications on 56 and 79 DAE. For this analysis, a single threshold value was used to determine areas unsuitable for supplemental N fertilizer applications, but growers should keep in mind that the threshold is just an estimation and may change based on growth stage. Fundamental agronomic practices regarding N fertilizer applications late in the growing season should be considered.

3.4. Effects of spatial resolution on within-field variability

The highest coefficients of variation (CVs) were observed for GRVI and NG, and the lowest CVs were observed for GNDVI and NDVI. On all dates, AR had higher CVs than RB; this was due to: (i) poorer overall emergence in AR and (ii) the center pivot being located in the AR section of the field.

Coarser spatial resolutions generally decreased the coefficient of variation (CV) for spectral indices in the commercial field (Table 6-9). This was primarily because there is a much greater likelihood of detecting pure pixels when images have a finer spatial resolution. When the spatial resolution becomes coarser, pixels generally become more mixed, and spectral variability in the dataset is lost. On all dates except 56 DAE, the finest spatial resolution resulted in the highest CV (Table 6-9). On 56 DAE, the CVs between the images with 0.25 m and 1 m spatial resolutions were exactly the same (after rounding). There was ~1.0 m between rows of the planted potato crop, so for the images with a 0.25 m spatial resolution, four pixels covered the width of an entire crop row (including the inter-row). Because mixed pixels were not masked out before this analysis, pixels could be a representation of the crop rows, inter-rows, or both. Therefore, it is reasonable to suggest that the variability on the dates before and after 56 DAE is due to bare soil showing through the crop canopy. The images with the 0.25 m spatial resolution could detect this variability, and the images with the 1 m spatial resolution could not.

On 56 DAE, however, the imagery with the 0.25 m spatial resolution was not able to detect any more variability than was detected by the imagery with the 1 m spatial resolution. This suggests that vegetative growth likely peaked on ~56 DAE and canopy cover was at or near 100%, which resulted in not being able to detect any additional spectral information with sub-meter spatial resolution imagery. Before this date, vegetative growth was still progressing, and after this date, the plant canopy began to fall over and, by 79 DAE, senesce. Fig. 6-2 illustrates how additional information was detected by the 0.25 m spatial resolution images on 30 and 79 DAE, but not on 56 DAE (the crop rows were planted West-East). Because there was only ~1 m between crop rows, factors detectable at a spatial scale greater than or equal to 1 m were the cause of the lost information for the images that had spatial resolutions greater than 1 m. It is reasonable to assume that an image that has a lower CV has a greater proportion of mixed pixels. On dates before and after peak vegetative growth (which was estimated to be ~56 DAE in the current study), images with finer spatial resolutions have a higher CV, and therefore, also have a greater proportion of pure pixels. Thus, there is value in obtaining imagery at sub-meter spatial resolutions in order to determine supplemental N fertilizer applications, especially at growth stages before and after ~56 DAE in the current study (Fig. 6-2).

The relative fluctuation (RF) of the CVs over spatial resolutions was lowest for GNDVI and NDVI. Low RF values are most desirable since they indicate consistent values over spatial resolutions. Low variability over spatial resolutions is important for a spectral index because it demonstrates its robustness over a variety of circumstances. If a grower uses imagery with a coarser spatial resolution than 0.25 m, less variability will be sacrifices if indices with low RF values are used; however, the ability of the indices used to predict crop N stress should also be a strong consideration when selecting a spectral index to be used to determine the rate and timings of split-applications of N fertilizer.

4. Conclusions

This study showed that high resolution broadband imagery can be a useful tool for detecting N stress in a potato crop while accounting for within-field spatial variability. Of the broadband spectral indices tested within measurement dates from the research plots, GRVI predicted leaf N concentration with the most certainty over all image dates, and actually performed better than the chlorophyll meter in some circumstances; GRVI also had the highest coefficient of variation from the commercial field. For the circumstances of this study, when the recommended rate and timing from the research plot was used as a reference, a GRVI NSI threshold of 120% was able to determine the areas of the commercial field that were most unsuitable for supplemental N fertilizer applications. Because of differences in potato variety, growth stage, or other local conditions, reference areas are needed in order to make accurate recommendations. Different NSI threshold levels should be established depending on: (1) the inherent variability/CV of the spectral index, (2) the NSI reference used, and (3) the growth stage.

The references used to determine the NSIs can drastically change supplemental N fertilizer recommendations. The data in this study make it difficult to provide concrete evidence as to which NSI reference is best in every circumstance. However, NSI values that used two standard deviations above the mean value of the over-fertilized commercial field as the reference were substantially higher than NSI values that used the mean value of the over-fertilized commercial field as the reference; this is evidence that luxury consumption of N fertilizer does occur in potatoes to some degree. Two standard deviations above the mean value of an over-fertilized reference should not be used at any growth stage, and the mean value of an over-fertilized reference should not be used at the later growth stages.

A major problem in the current study was that spectral measurements were collected immediately prior to post-emergence N fertilizer applications (when there was likely some N stress present). A similar experiment should be conducted to investigate the difference between using an over-fertilized reference plot and an adequately fertilized reference plot for NSI determination; however, instead of taking spectral measurements only immediately prior to post-emergence N fertilizer applications, measurements should be taken both before and ~2-4 days after fertilization (when the crop has been given time to recover from the stress). This would provide an NSI reference that is truly a representation of the recommended N rate. Specifically, the NSI references that should be evaluated include an over-fertilized and an adequately fertilized reference plot, both with spectral measurements collected shortly after fertilization. These spectral data could be used to determine the degree to which luxury consumption of N fertilizer occurs in a potato crop. Perhaps spectral measurements collected shortly after fertilization could serve as the NSI reference when determining the supplemental fertilizer rate, timing, and location for the next split application of post-emergence N in that same year.

5. Acknowledgements

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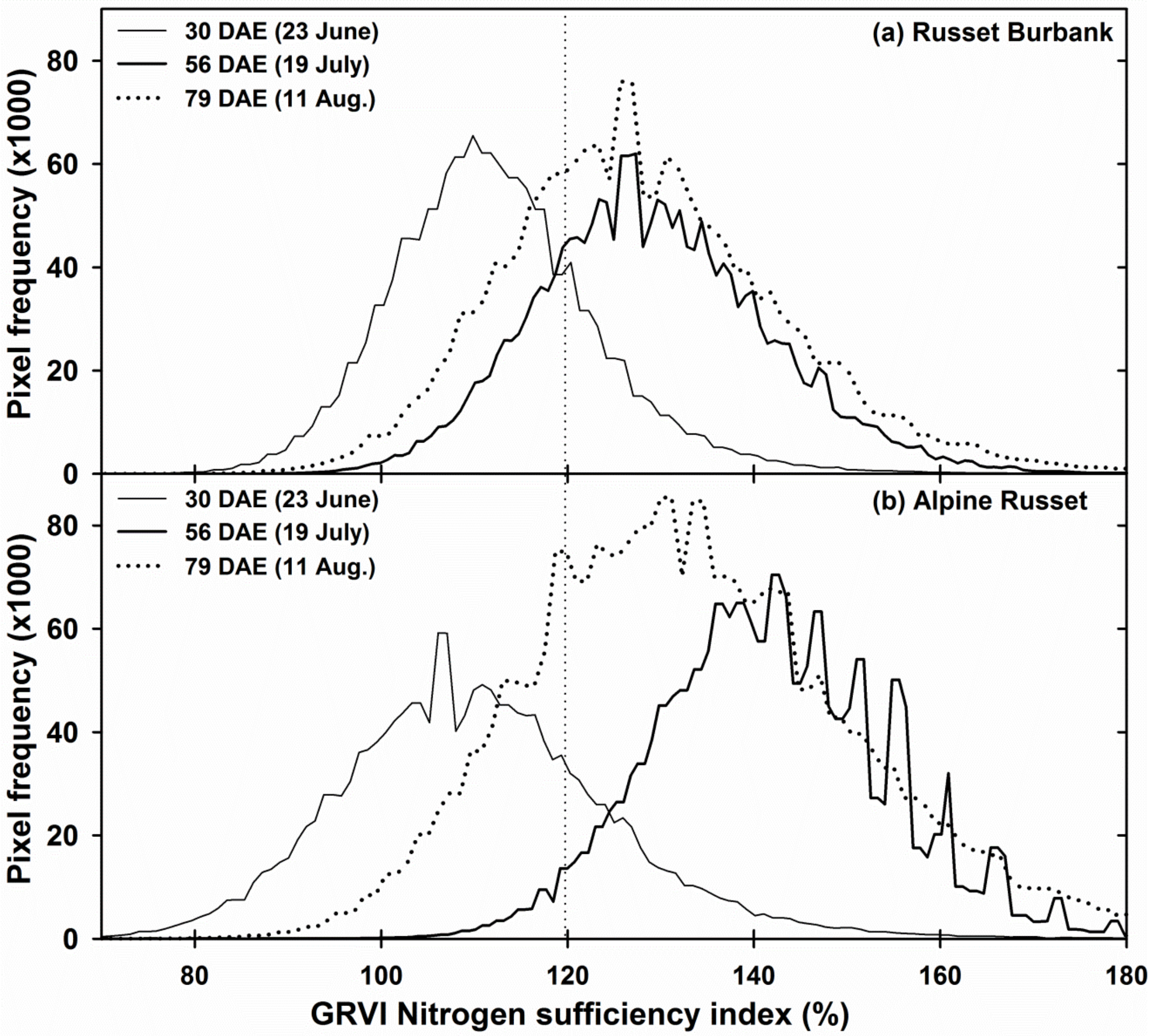

Fig. 6-1. Histograms for the broadband GRVI nitrogen sufficiency index for Russet Burbank (a) and Alpine Russet (b) throughout the 2011 growing season.

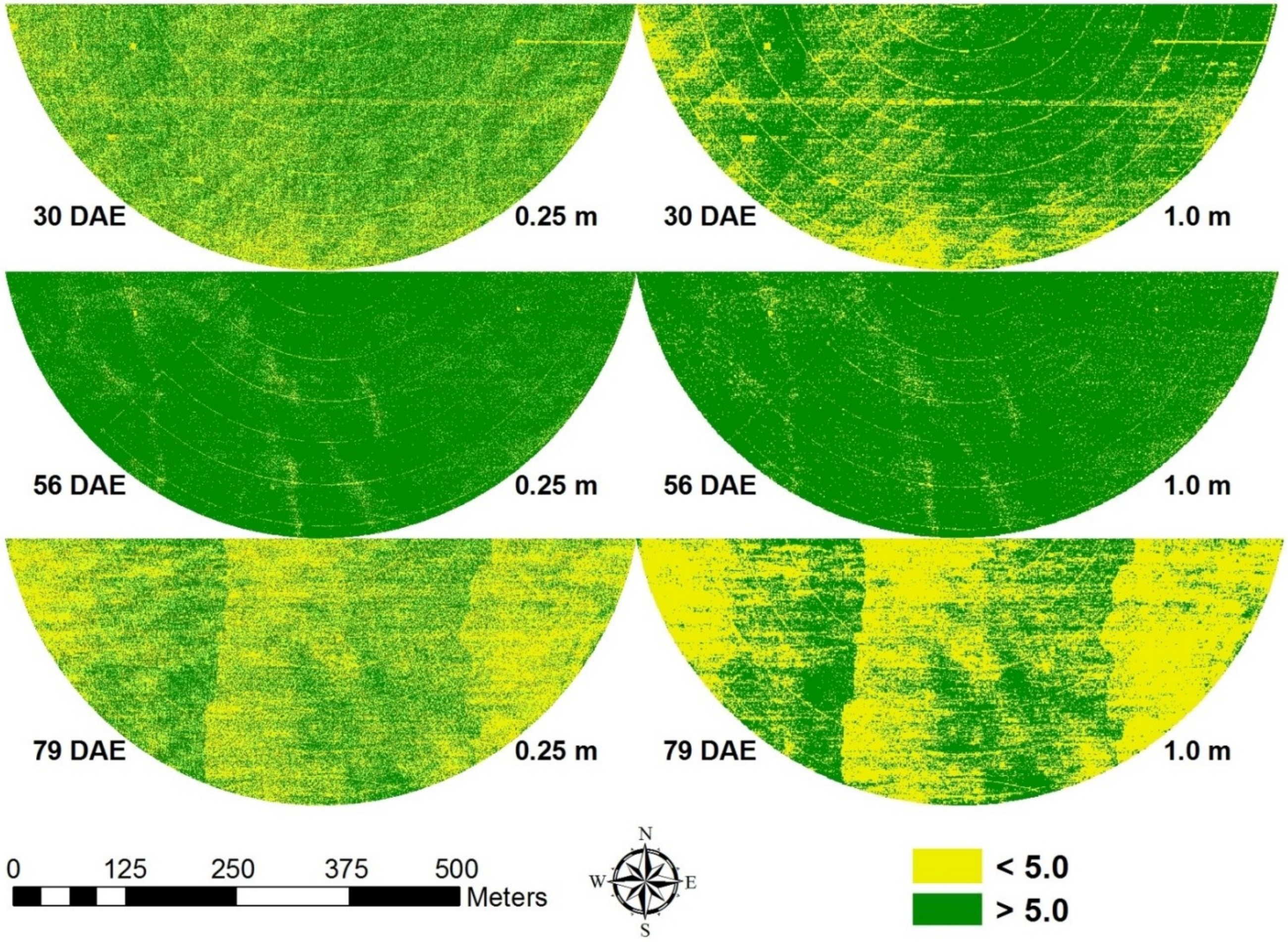

Fig. 6-2. The effect of spatial resolution on detectable variability in a commercial potato field. The images represent a broadband GRVI index on three dates throughout the growing season for 0.25 and 1.0 m spatial resolution. The area of the field in the images is the portion of the commercial field planted into the Russet Burbank variety.

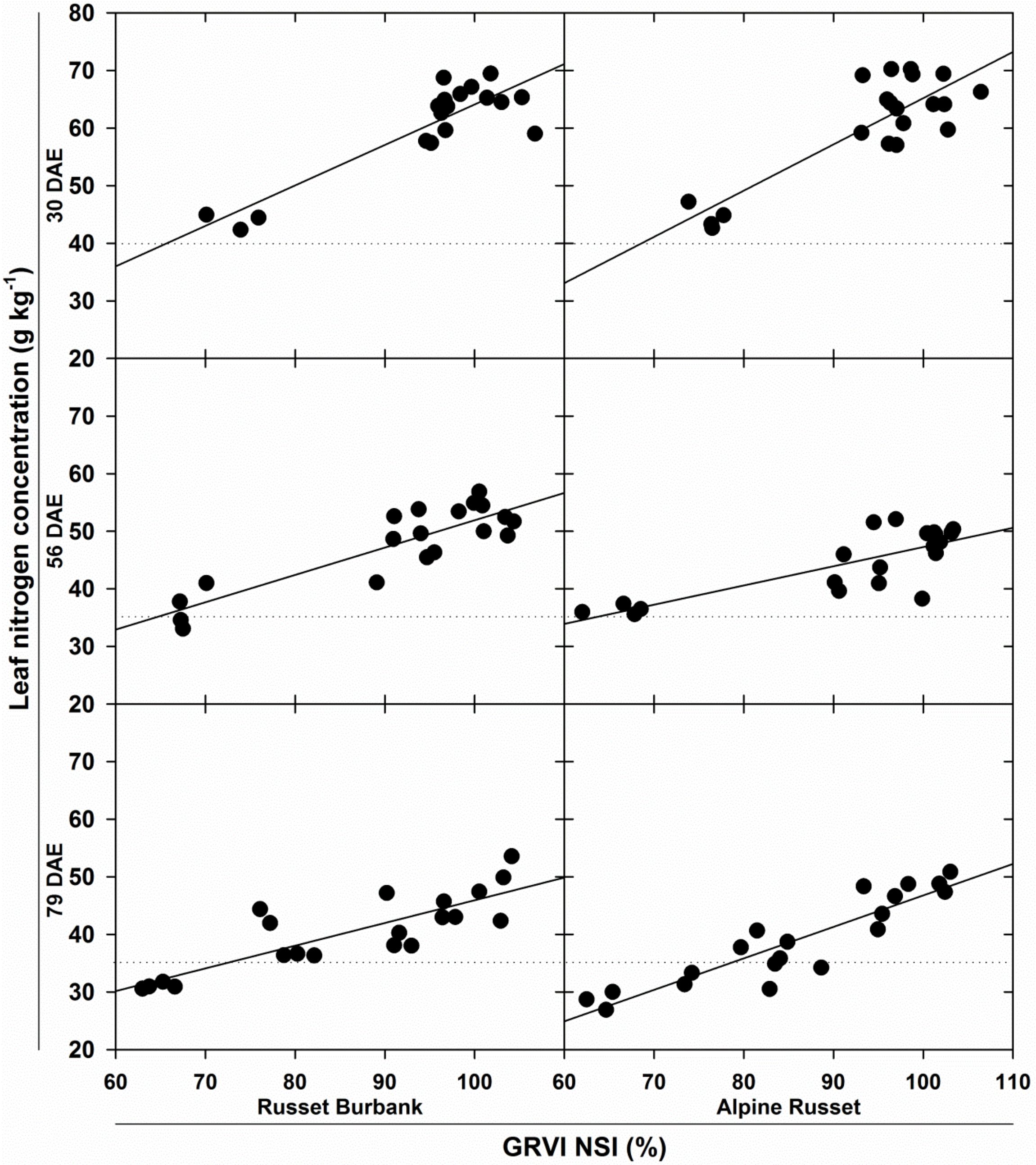

Fig. 6-3. The linear relationship between leaf nitrogen concentration and broadband GRVI for both varieties on 30, 56, and 79 days after emergence (DAE). Points represent data from the research plots after they were normalized by a nitrogen sufficiency index in which the recommended rate from the research plots were used as a reference. The dotted reference line represents the lower threshold sufficiency level for leaf nitrogen concentration (Rosen & Eliason, 2005; Westermann, 1993).

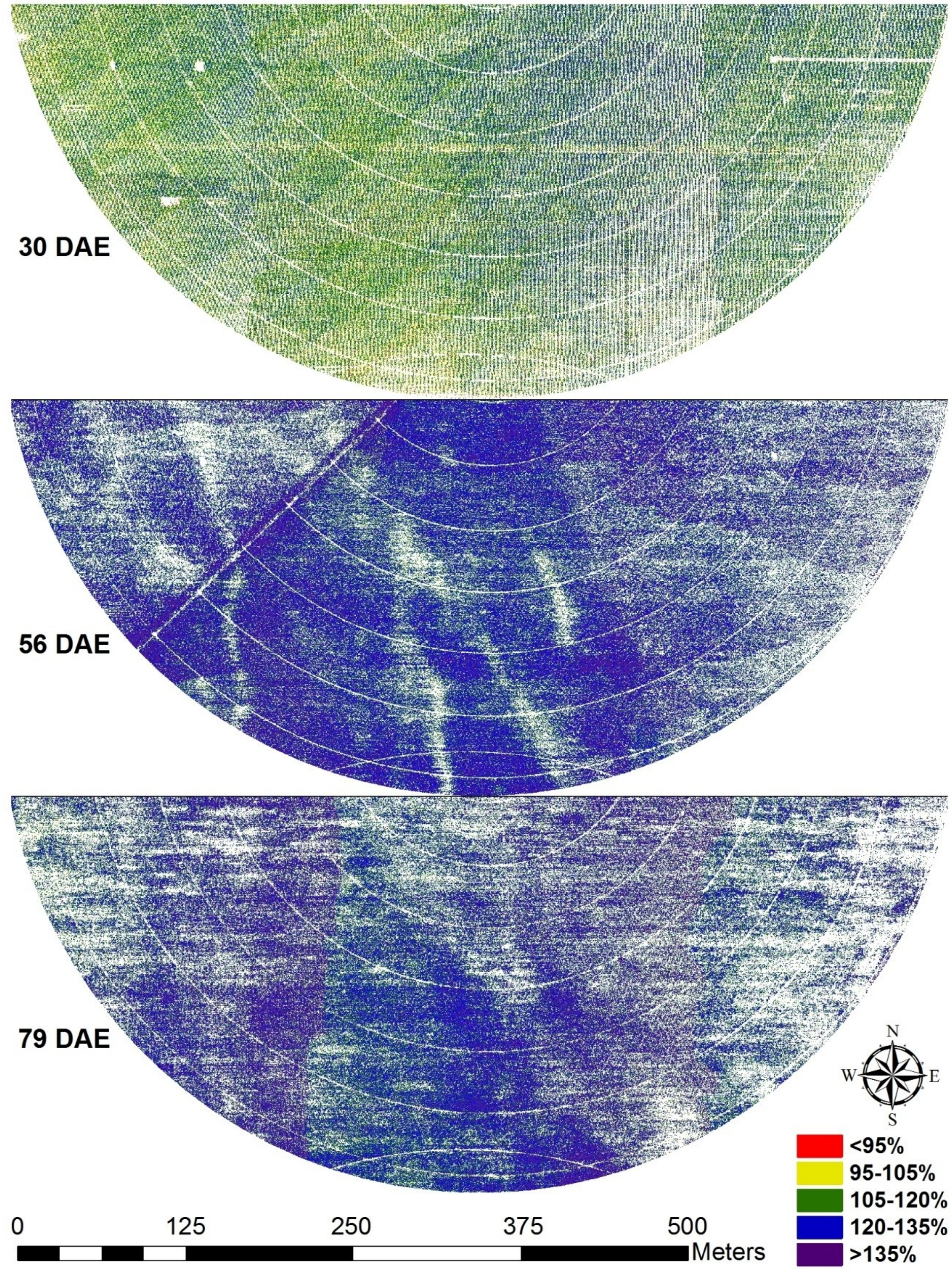

Fig. 6-4. Broadband GRVI index for the Russet Burbank variety on 30, 56, and 79 days after emergence (DAE). Images are shown after they were normalized by a nitrogen sufficiency index using the recommended rate from the research plots as a reference.

Table 6-1. Nitrogen (N) fertilizer treatments for the research plots. On average, about two weeks passed between split applications of post-emergence N for treatments 180 split, 270 split, and 270 split + s.

Nitrogen fertilizer treatment	Timing of application			Total N
	planting	emergence	post-emergence	
	------------------------	kg N ha^{-1}	------------------------	
34 N (control)	34	0	0	34
180 N split	34	90	11.2 × 5[†]	180
270 N split	34	124	22.4 × 5[†]	270
270 N split + s	34	124	22.4 × 5[†]	270
270 N early	34	124	112	270

[†] Split applications of post-emergence nitrogen fertilizer occurred at 16, 34, 44, 57, and 76 days after emergence.

Table 6-2. Growth stage, actual nitrogen (N) fertilizer applied throughout the season for the N treatments with split applications of post-emergence fertilizer, and the image source for each sample date.

Date	DAE	Growth stage	N Treatment[†]			Data acquired	
			180 N split	270 N split	Commercial	Redlake MS4100[‡]	Leaf samples
			----------------	kg N ha^{-1}	----------------		
23 June	30	Vegetative	135	180	178	✓	✓
6 July	43	Tuber bulking	146	203	228		✓
19 July	56	Tuber bulking	158	225	279	✓	✓
29 July	66	Maturation	169	248	325		✓
11 Aug.	79	Maturation	180	270	375	✓	✓

[†] *180 N split* and *270 N split* refer to N treatments from the research plots, and *Commercial* refers to the commercial field.

[‡] The Redlake MS4100 is a multispectral camera that acquired images in 3 broadbands (i.e., near-infrared, red, and green regions) at 0.25 m spatial resolution (operated by the Upper Midwest Aerospace Consortium [UMAC], University of North Dakota, Grand Forks, ND).

Table 6-3. Summary of the indices evaluated in this study. This list was developed primarily based on the results of a preliminary analysis from research plots using 17 broadband and 82 narrowband previously published indices on four dates over two years.

Index	Name		Formula†	Developed for	Developed by
Broadband indices					
GNDVI	Green normalized vegetation index	=	$(R_{NIR} – R_G)/(R_{NIR} + R_G)$	Chlorophyll	Gitelson et al. (1996)
GRVI	Green ratio vegetation index	=	R_{NIR} / R_G	Nitrogen	Sripada et al. (2006)
NDVI	Normalized difference vegetation index	=	$(R_{NIR} – R_R)/(R_{NIR} + R_R)$	Structure	Rouse et al. (1974)
NG	Normalized green	=	$R_G / (R_{NIR} + R_R + R_G)$	Nitrogen	Sripada et al. (2006)

† R_i = reflectance at spectral region i; $_G$ = green region (508-572 nm), $_R$ = red region (657-695 nm), and $_{NIR}$ = near-infrared region (770-828 nm).

Table 6-4. Potato tuber yield under different nitrogen treatments from research plots.

N treatment	Tuber yield
	--- Mg ha^{-1} ---
34 early	40.09 c[†]
180 split	52.70 ab
270 split	53.18 a
270 split + s	52.28 ab
270 early	49.00 b

[†] Means followed by the same letter are not significantly different (α=0.05).

Table 6-5. Leaf nitrogen concentrations for Russet Burbank and Alpine Russet potato varieties at different days after emergence under various nitrogen (N) treatments from the research plots, as well as from the over-fertilized commercial field.

N Treatment	Days after emergence				
	30	43	56	66	79
Russet Burbank	-------------------------		g kg^{-1}	-------------------------	
34 early	44 d[†]	35 d	32 b	33 c	31 c
180split	58 c	43 c	40 a	41 b	41 b
270 split	64 b	48 b	41 a	47 a	46 a
270 split + s	65 b	49 b	40 a	47 a	47 a
270 early	68 a	52 a	39 a	42 b	37 b
Commercial field	-	-	64	54	45
Alpine Russet					
34 early	45 d	35 d	29 c	31 d	29 c
180split	58 c	40 c	37 ab	37 c	36 b
270 split	64 b	45 b	41 a	44 a	46 a
270 split + s	65 b	46 ab	40 a	42 ab	47 a
270 early	70 a	49 a	36 b	39 bc	36 b
Commercial field	-	-	63	47	39

[†] Means followed by the same letter within a measurement date and variety are not significantly different (α=0.05).

Table 6-6. Coefficient of determination (r^2) and root mean squared error of cross-validation (RMSECV) representing the relationship between leaf nitrogen (N) concentration and chlorophyll meter reading/spectral indices for predicting N stress in Russet Burbank and Alpine Russet potato varieties from the research plots. Results are presented for different days after emergence (DAE), as well as for all dates combined. All values were significant at α=0.05. Reported RMSECV was normalized by calculating the ratio between RMSECV values and the range of predicted values.

Spectral analysis technique	r^2				RMSECV (%)			
	Days after emergence			All dates	Days after emergence			All dates
	30	56	79		30	56	79	
Russet Burbank								
Chlorophyll meter	0.66	0.88	0.76	0.66	24.3	13.8	17.1	18.2
GNDVI	0.80	0.79	0.69	0.67	16.0	18.1	24.5	18.9
GRVI	0.78	0.79	0.69	0.68	16.7	17.4	24.1	18.7
NDVI	0.82	0.71	0.59	0.63	15.8	21.9	30.5	21.2
NG	0.80	0.80	0.71	0.63	16.0	17.5	23.4	20.6
Alpine Russet								
Chlorophyll meter	0.43	0.72	0.89	0.58	37.7	19.4	11.7	21.2
GNDVI	0.75	0.59	0.79	0.43	18.7	28.1	17.3	30.0
GRVI	0.73	0.61	0.83	0.41	19.2	26.6	15.0	33.9
NDVI	0.81	0.49	0.74	0.40	17.2	34.9	19.2	31.2
NG	0.72	0.63	0.80	0.39	19.5	26.2	16.7	32.1

Table 6-7. Means, standard deviations (SDs), and ranges of chlorophyll meter readings (CM) and four broadband spectral indices from the commercial field for different days after emergence (DAE) for the Russet Burbank variety normalized by a nitrogen sufficiency index using three different nitrogen references: (i) the higher average replicate value of either 270 split or 270 split + s from the research plots, (ii) the mean value of the commercial field, and (iii) two standard deviations above the mean value of the commercial field.

Spectral index	Nitrogen sufficiency index reference								
	Treatment plots			Mean[†]			2 SDs above mean[†]		
	Mean	SD	Range	Mean	SD	Range	Mean	SD	Range
	------------------------------------			%			--------------------------------------		
30 DAE									
CM	-[‡]	-	-	-	-	-	-	-	-
GNDVI	105	4.1	73	100	3.9	69	93	3.6	64
GRVI	113	11.4	244	100	10.1	217	83	8.4	180
NDVI	106	2.5	23	100	2.3	21	96	2.2	20
NG	109	9.2	205	101	8.5	188	84	7.1	157
56 DAE									
CM	109	5.3	8	100	4.9	7	91	4.4	6
GNDVI	111	3.6	45	100	3.3	41	94	3.1	39
GRVI	129	12.6	200	100	9.8	155	84	8.2	130
NDVI	112	2.2	20	100	2.0	17	96	1.9	17
NG	120	10.0	162	101	8.4	136	84	7.0	113
79 DAE									
CM	118	9.9	14	100	8.4	12	86	7.2	10
GNDVI	113	5.5	75	100	4.8	66	91	4.4	60
GRVI	128	15.3	314	100	11.9	244	81	9.6	197
NDVI	109	2.0	21	100	1.9	20	96	1.8	19
NG	121	12.0	251	101	10.0	210	81	8.1	170

[†] References labeled as *Mean* and *2SDs above mean* correspond to pixel values from the commercial field.

[‡] Chlorophyll meter readings were not taken on 30 DAE.

Table 6-8. Means, standard deviations (SDs), and ranges of chlorophyll meter readings (CM) and four broadband spectral indices from the commercial field for different days after emergence (DAE) for the Alpine Russet variety normalized by a nitrogen sufficiency index using three different nitrogen references: (i) the higher average replicate value of either 270 split or 270 split + s from the research plots, (ii) the mean value of the commercial field, and (iii) two standard deviations above the mean value of the commercial field.

Spectral index	Nitrogen sufficiency index reference								
	Treatment plots			Mean[†]			Two SDs above mean[†]		
	Mean	SD	Range	Mean	SD	Range	Mean	SD	Range
	--			%			--		
30 DAE									
CM	-[‡]	-	-	-	-	-	-	-	-
GNDVI	105	5.6	82	100	5.4	78	90	4.8	71
GRVI	110	14.7	241	100	13.3	218	79	10.5	172
NDVI	103	3.8	29	100	3.7	28	93	3.4	26
NG	111	12.3	211	101	11.3	192	79	8.8	150
56 DAE									
CM	111	6.1	9	100	5.5	8	90	4.9	7
GNDVI	113	2.9	45	100	2.6	40	95	2.4	38
GRVI	142	12.9	193	100	9.0	136	85	7.7	115
NDVI	115	1.7	20	100	1.4	18	97	1.4	17
NG	130	10.4	158	101	8.0	122	85	6.7	103
79 DAE									
CM	115	2.6	4	100	2.3	3	96	2.2	3
GNDVI	114	5.7	75	100	5.0	66	91	4.5	60
GRVI	134	18.3	290	100	13.7	216	79	10.7	170
NDVI	109	2.4	23	100	2.2	21	96	2.1	20
NG	125	14.4	235	101	11.6	190	79	9.0	147

[†] References labeled as *Mean* and *2SDs above mean* correspond to pixel values from the commercial field.

[‡] Chlorophyll meter readings were not obtained on 30 DAE.

Table 6-9. Coefficient of variation (CV) of selected broadband spectral indices in Russet Burbank and Alpine Russet potato varieties for different days after emergence (DAE) and at different spatial resolutions in the commercial field.

Spectral index	Russet Burbank								Alpine Russet						
	0.25 m	1 m	5 m	10 m	30 m	AVG	Relative fluctuation		0.25 m	1 m	5 m	10 m	30 m	AVG	Relative fluctuation
								%							
30 DAE															
GNDVI	4.6	2.2	1.5	1.2	2.8	2.5	1.38		12.1	9.1	7.4	6.7	6.0	8.3	0.74
GRVI	11.0	5.0	3.3	2.9	4.1	5.3	1.55		21.3	14.0	11.3	10.4	9.3	13.3	0.90
NDVI	5.1	2.7	1.9	1.5	4.3	3.1	1.15		18.5	14.9	12.2	11.0	9.8	13.3	0.66
NG	8.8	3.8	2.5	2.2	3.3	4.1	1.58		14.8	9.2	7.3	6.7	6.0	8.8	1.01
56 DAE															
GNDVI	3.9	3.9	1.7	1.5	2.7	2.7	0.86		4.9	4.9	2.9	2.4	2.4	3.5	0.72
GRVI	10.9	10.9	4.7	4.3	4.8	7.1	0.93		12.3	12.3	6.5	5.8	5.2	8.4	0.84
NDVI	3.5	3.5	1.7	1.6	3.7	2.8	0.76		6.2	6.2	3.8	3.1	3.3	4.5	0.68
NG	8.9	8.9	3.5	3.2	4.2	5.8	0.99		11.4	11.4	6.1	5.2	4.9	7.8	0.83
79 DAE															
GNDVI	5.6	3.2	2.4	2.2	2.6	3.2	1.07		6.3	4.5	3.7	3.4	3.1	4.2	0.75
GRVI	13.1	7.4	5.6	5.1	4.9	7.2	1.13		15.5	10.6	8.9	8.4	7.7	10.2	0.77
NDVI	3.5	2.3	1.7	1.6	2.8	2.4	0.81		4.7	3.7	2.9	2.5	2.3	3.2	0.75
NG	10.3	5.6	4.1	3.7	3.7	5.5	1.20		12.4	8.2	6.8	6.4	5.8	7.9	0.83

OVERALL CONCLUSIONS

1. Nitrogen use, tuber yield, and tuber quality

Both years in this study (2010 and 2011) were high leaching years. Rainfall from May through July was 399 mm in 2010 and 472 mm in 2011. During these months, there were 24 leaching days due to rainfall in 2010 and 30 leaching days due to rainfall in 2011. This high leaching, coupled with the poor inherent ability of potato to recover nitrogen (N) from the soil, resulted in nitrogen uptake efficiencies to be only 50% in 2010 and only 43% in 2011. Plant dry matter increased with increasing N fertilizer rate, but nitrogen use efficiency declined with increasing N fertilizer rate. This illustrates that plant dry matter produced per unit of N decreased as N supply increased. The timing of N fertilizer did not significantly affect plant dry matter or nitrogen use efficiency within the 270 kg ha^{-1} N fertilizer rate. However, the highest N uptake or N use efficiencies were not best for optimizing tuber yield or quality. The treatments with the lowest N uptake or N use efficiencies (i.e., the treatments with the 270 kg N ha^{-1} rate) always had significantly similar or higher total yield, Grade A yield, and proportion of tubers >170 g than the treatments with the highest N uptake or N use efficiencies (i.e., the treatments with the 34 or 180 kg N ha^{-1} rates). Therefore, tuber yields and quality characteristics should be used in conjunction with N efficiencies when determining optimum N fertilizer rates. Higher yields and N uptake efficiencies were measured when post-emergence N was split applied within the 270 kg ha^{-1} N rate.

Insufficient supplemental water during critical growth stages negatively affected tuber yield/quality and plant N uptake, but surprisingly had a positive overall effect on chip color. Irrigation did not significantly affect nitrogen efficiencies. However, because the unstressed irrigation treatment provided more available N to the crop than the stressed irrigation treatment, there was a confounding effect that made it difficult to identify the effects of irrigation regime on N uptake and N use efficiencies. Overall, specific gravity was not influenced by water stress or N stress in this study. The incidence of hollow heart decreased both with water and N stress, and the proportion of misshapen tubers was not affected by water stress, but decreased as N rate increased. It was also difficult to identify the effect of N treatment on tuber sugar concentration and its relationship to light reflectance because of differences in responses between sugar concentration and chip color. However, nitrogen stress that occurred with the 34 N treatment clearly resulted in the highest tuber sucrose and glucose levels and the lowest light reflectance.

2. Russet Burbank vs. Alpine Russet

Both varieties had similar quantities of plant dry matter, but Alpine Russet had higher plant N uptake. This resulted in higher N uptake efficiency for Alpine Russet, but higher N utilization efficiency for Russet Burbank. The overall N use efficiency was similar between varieties. In general, Alpine Russet contained more sucrose in both ends of the tuber and more glucose in the bud end, and Russet Burbank contained more glucose in the stem end, resulting in lower light reflectance in the stem end at harvest in Russet Burbank, and lower light reflectance in the bud end after storage in Alpine Russet. When the main effect of variety was significant for tissue samples, Russet Burbank usually had higher total N and nitrate-nitrogen (NO_3-N) concentrations than Alpine Russet. It is likely that the differences in petiole, leaflet, and whole leaf total N and NO_3-N concentration between varieties was because Alpine Russet is slower to emerge and is a longer season variety than Russet Burbank (Whitworth et al., 2011), suggesting that sufficiency ranges should be slightly lower for Alpine Russet than for Russet Burbank, especially before ~50 DAE. Absolute reflectance values were typically greater for Russet Burbank than for Alpine Russet. Within measurement dates, the relationship between leaf N concentration and chlorophyll meter readings, spectral indices, or partial least squares regression models were relatively similar between varieties. However, when analyzed over measurement dates, Russet Burbank had a much better relationship with leaf N concentration than Alpine Russet. This illustrates that it is more important to normalize spectral measurements to that of local conditions (e.g., by using a nitrogen sufficiency index) for Alpine Russet than for Russet Burbank.

3. Tissue part and form of nitrogen for detecting in-season crop stress

Nitrate-nitrogen (NO_3-N) concentrations were always higher in petioles than whole leaves or leaflets, and total N was always higher in whole leaves and leaflets than petioles. In general, this indicates that the diagnostic criteria should change based on the tissue part sampled and the form of N tested. Tissue total N and NO_3-N concentrations were highest early in the growing season, but they leveled off as the season progressed. Tissue NO_3-N was more responsive to recent N fertilizer applications than tissue total N, whereas tissue total N was a better indication of cumulative N stress at the time of sampling. Narrowband spectral indices predicted Grade A tuber yield better than plant tissue measurements which are currently considered as the best management practice. Canopy-level spectral data such as aerial imagery or Cropscan readings had a much better relationship with leaf N concentration than petiole NO_3-N concentration. This is primarily because canopy-level reflectance measurements depend very little on petioles since the leaflets account for almost all of the total leaf area at a canopy-scale. Therefore, the use of canopy-level spectral data for in-season N fertilizer recommendations is largely based on the assumption that leaf N concentration can be effectively used to determine the rate and timing of in-season fertilizer applications. Future research should investigate effectiveness of using leaf N concentration for in-season fertilizer recommendations.

4. Canopy-level spectral data for determining in-season nitrogen fertilizer prescriptions

Overall, the best spectral indices had similar r^2 values, RMSECVs, overall accuracies, and kappa coefficients as the chlorophyll meter readings; however, because the spectral indices used canopy-level imagery, these techniques provide an advantage over chlorophyll meter readings since imagery can better account for spatial variability. Across image dates, the partial least squares regression models, especially those that used first derivative reflectance spectra as independent variables, were determined to be able to predict leaf N concentration the best in potatoes. For spectral data to be useful for variable rate fertilizer management, the user must be aware of the variability imposed by the spectral index/model relative to the variability of leaf N concentration. Spectral indices/models with more variability than leaf N concentration may tend to overestimate N stress level, and those with less variability may tend to underestimate N stress level.

Canopy-level imagery with 0.25 m spatial resolution could detect variability due to bare soil in a potato crop canopy much better than imagery with 1 m spatial resolution. This is important in regards to using spectral data for making N fertilizer recommendations. Data with a coarser spatial resolution will largely consist of mixed pixels if there is not full vegetative canopy cover (i.e., there will be less pure vegetation pixels). Because there are a greater proportion of pure vegetation pixels in imagery with a finer spatial resolution, it is easier to mask out the pure soil and mixed pixels in order to get a good representation of the true spectral properties of the vegetation. In other words, the spectral data are different between images with different spatial resolutions. This ultimately has an influence in the diagnostic criteria that should be used for determining N fertilizer recommendations between data with different spatial resolutions.

There was some uncertainty because of inconsistencies between the devices used to obtain spectral measurements. This illustrates one of the ever-challenging problems with using generalized remote sensing data for crop management decisions. Because of differences in sensors, potato variety, growth stage, or other local conditions, normalizing measurements to reference areas must be done in order to make accurate recommendations.

5. Use of a nitrogen sufficiency index

The nitrogen sufficiency index can be used to normalize spectral measurements so spectral data is calibrated to local conditions. By applying the nitrogen sufficiency index to plant measurements and spectral indices/models, in many cases, the main effect of variety and the N × variety interaction became statistically insignificant. By transforming a plant measurement or spectral index/model so it is insensitive to external factors such as variety, it can be useful over a broader range of environmental conditions. The reference used to determine nitrogen sufficiency indices can drastically change supplemental N fertilizer recommendations if the diagnostic criteria used are not adjusted accordingly. Deciding on the best reference N rate for a potato crop can be a challenge because there is evidence that luxury consumption occurs at least to some degree and excessive N may result in lower yield.

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APPENDIX

Thermal Data for Evaluating In-Season Water Status of an Irrigated Potato Crop

Summary

Potato (*Solanum tuberosum* L.) tuber yield and quality depend on an adequate supply of water; however, too much water can be detrimental to tuber quality, and can increase the likelihood of nitrate-nitrogen to be lost to the groundwater. An experiment was conducted to evaluate the effects of irrigation, N treatment, and variety on the ability of ground- and aerial-based remotely sensed thermal data. Overall, daily soil matric tension was much more responsive to differences in water stress than daily canopy temperature measured by infrared radiometers. Three of the four image dates in this study showed a significant response in irrigation regimes for canopy temperature measurements measured from aerial imagery. Canopy temperature differences on 47 and 83 days after emergence (DAE) in 2010 were well over 1 °C between the irrigation regimes, but on 43 and 66 DAE, they were less than 0.2 °C. However, on both 83 DAE in 2010 and 66 DAE in 2011, within-field and within-plot differences were visible in the imagery. Applying these data to water stress indices such as the Crop Water Stress Index should be completed to investigate its full functionality.

1. Introduction/Literature review

The physiological functioning of a potato (*Solanum tuberosum* L.) plant, as well as tuber yield and quality, depend on an adequate supply of water. Both timing and quantity of irrigation must be considered to optimize crop water use (Dean, 1994), and in-season monitoring is necessary to be able to do this. The crop water status is a dynamic balance between the available supply of water (depends on weather and soil properties) and the evaporative demand of the crop (depends on weather and the crop growth stage; Bailey, 2000; De Neve et al., 2000). The primary methods for monitoring in-season soil or crop water status include: soil-based monitoring, crop-based monitoring, and through the calculation of a net water balance.

Soil water content, soil water potential, and soil hydraulic conductivity are all important measurements in the planning and management of irrigation using soil-based monitoring (Hsiao, 1990). To optimize crop water use, soil moisture should be somewhere between the refill point and field capacity of the soil (Bailey, 2000). Several techniques for measuring water content and water potential are described by Campbell and Mulla (1990) and De Neve et al. (2000). Hydraulic conductivity measurements require more involved techniques, but since they depend primarily on the soil type, experimental data for a particular soil can be applied to field conditions when hydraulic conductivity measurements are needed. In addition to soil type, unsaturated hydraulic conductivity is also dependent upon water content or matric potential, which makes it nonlinear function and much more complicated to measure (Jury & Horton, 2004). Soil water measurements can be used for irrigation scheduling in order to maintain soil water content throughout the growing season (Bailey, 2000). Advantages of this approach are that measurements are usually less variable, and are generally easier to obtain and interpret than crop measurements (De Neve et al., 2000). Soil water measurements also give advance warning of a limitation of available water, usually before plants undergo any stress (De Neve et al., 2000); however, they are only point measurements, so within-field spatial variability is not accounted for.

Possible plant-based measurements for irrigation scheduling purposes are those that either measure a direct aspect of plant water status, or those that measure plant processes that are known to respond sensitively to water deficits (Jones, 2004). Refer to Jones (2004) for basic discussion, and to Hsiao (1990) for specific procedures and a more in depth discussion of some of the most common plant-based measurements for irrigation scheduling. The remainder of this sub-section will include discussion of the use of remote sensing (primarily using thermal measurements) for the indirect measurement of plant physiological responses that are closely related to water deficits (i.e., stomatal conductance).

Just as remote sensing can be used to detect crop N stress, it can also be used to detect crop water stress. Some researchers have found that spectral reflectance data in the visible and NIR regions of the spectrum can be useful for detecting plant water stress. For example, Zarco-Tejada et al. (2003) showed that the reflectance derivative can detect water stress in a tree canopy because the reflectance derivative curve forms a double peak under stressed conditions. Also, Neale et al. (2005) discussed how the use of high resolution multispectral aerial imagery can be used to obtain reflectance-based crop coefficients which can be used to effectively estimate crop evapotranspiration (ET).

However, it is likely that the responses of reflectance in the visible and NIR regions of the spectrum can be confounded with other stresses (e.g., N stress) if they are present. Therefore, it is perhaps more practical to use passive sensors based on thermal emission in the thermal infrared (TIR) part of the spectrum (i.e., ~8,000-12,000 nm) to detect crop water stress. Use of TIR is based on the concept that leaf stomata close partially or fully under water stress, which results in lower transpiration rates, and ultimately alters leaf temperature (Gates, 1968). Cooler leaf temperatures will result for open stomata because of higher transpiration rates, and warmer leaf temperatures will result for closed stomata because of lower transpiration rates. More specifically, the difference in canopy and air temperature (T_c-T_a) is less in a stressed crop than it is in an unstressed crop under conditions of high irradiance (Vos & Haverkort, 2007).

Czajkowski et al. (2004) and Asner (2004) discussed the basic theory behind the interpretation of TIR signals obtained from sensors and how they can be used to estimate the surface temperature of a target. Several crop water stress algorithms have been developed in recent years because of the development of accurate, low cost infrared thermometers that make it possible to remotely measure crop canopy temperature (Hatfield et al., 2004). Jackson et al. (1977) summed daily T_c-T_a values (termed 'Stress Degree Day'; SDD); this can be used as a tool to detect water stress based on the concept that elevation of canopy temperature above air temperature is indicative of stomatal closure, and ultimately crop water stress. Idso et al. (1981) later determined that the linear relationship in T_c-T_a values and the corresponding vapor pressure deficit of the crop can be used to effectively identify potential evaporation of a crop canopy – this is an empirical formulation of the crop water stress index (CWSI). This provided a way to normalize the SDD parameter in order to reduce the effect of external environmental factors (except the effect of cloud cover; (Idso et al., 1981)). Jackson et al. (1981) developed a theoretical model for the CWSI to be used for the detection of crop stress. This model is not only dependent on T_c-T_a, but also on the unstressed baseline temperature and the upper limit of T_c-T_a (i.e., when the canopy is no longer transpiring). This method has been particularly useful in arid climates, but has been much less useful in more humid or cloudy climates (Jones, 2004). To reduce the model's sensitivity to environmental variations, Jones (1999) revised the original CWSI model to use wet and dry reference surfaces instead of an unstressed baseline temperature and an upper limit of T_c-T_a, respectively.

The CWSI is one of the more common methods in which crop canopy temperature is used to quantify crop water stress (Hatfield et al., 2004). Field measurements of crops growing under different ranges of water stress can be used to derive critical values of the CWSI (De Neve et al., 2000); these critical values can be used to monitor plant water status and for planning irrigation scheduling in wheat (Gontia & Tiwari, 2008; Yuan et al., 2004), sugar beets (Köksal & Yıldırım, 2011), and corn (Edalat et al., 2010).

Other calculations that can be made from canopy temperature measurements for plant water status monitoring and irrigation scheduling include stomatal conductance (Berni, et al., 2009; Blonquist et al., 2009), leaf water potential (Cohen et al., 2005), canopy stress index (CSI) (Rodriguez et al., 2005), or the water deficit index (WDI; (Moran et al., 1994)). The CSI is defined by Rodriguez et al. (2005) as the difference between canopy (T_c) and air temperature (T_a), normalized by vapor pressure deficit; they found that the CSI accounted for 80% of the variation in growth rate and yield due to variability in water supply. The WDI combines spectral vegetation indices with composite canopy temperature measurements to allow application of the CWSI theory while accounting for the effect of soil in partially vegetated fields (Moran et al., 1994).

Maintaining a soil water balance is the most widely used approach for scheduling irrigation for a potato crop (De Neve et al., 2000). It uses estimations of the change in daily soil moisture as the difference between inputs (irrigation and rainfall) and losses (crop ET, drainage, and runoff) to compute a soil water balance assessing the allowable water deficit of the soil (Pardossi & Incrocci, 2011). This approach avoids the need to make either direct or indirect measurements of either plant or soil water status (De Neve et al., 2000). ET can be calculated using a crop coefficient, a reference ET, and specific weather data (Pardossi & Incrocci, 2011). Pardossi and Incrocci (2011) and Wright (2002) discuss the basic methods required for use of the water balance approach. The water balance approach is not very accurate because of the difficulty in estimating ET, and errors are cumulative overtime; therefore, the calculated water balance should be recalibrated periodically by using actual soil measurements (Jones, 2004). This approach does not account for within-field spatial variability, but it can be implemented at a low cost, and overall, it is very feasible.

Little has been done in developing methodologies to combine spectral and temperature data obtained via remote sensing to differentiate between the N and water status of a crop. Tilling et al. (2007) conducted a study on wheat using multispectral and thermal imagery to spatially detect differences between N and water stress, but a similar study has not been conducted for a potato crop. A better understanding of the combined effects of N and water will help to evaluate the possibility of fusing spectral and thermal data for the purpose of adjusting both N fertilizer and water applications in real time. In a practical sense, this would be useful if the same equipment was used for fertilizer and irrigation applications (e.g., fertigation). This experiment is being conducted in collaboration with a group in Israel. The overall objective of the experiment is to evaluate the ability of spectral and thermal imagery/data to distinguish between water and N status in a potato crop. To better understand the methodology required to pursue this main objective, several sub-objectives relating to both spectral and thermal imagery were evaluated. This report was written to acknowledge that the overall objective of the experiment is still in progress, and to present some of the thermal data from the University of Minnesota experimental site.

2. Materials and methods

Refer to Nigon et al. (2012a) for information describing the study site and experimental design. Soil matric tension was measured with granular matrix soil moisture sensors (Watermark Model 200, Irrometer Co., Riverside, CA) in and below the root zone. Sensors were placed in several plots, which included both irrigation treatments, both varieties, and N management strategies 3 and 4 for both years (one or two replications). The sensors were packed with moist soil during installation to ensure good sensor to soil contact.

Aerial thermal imagery was acquired with a FLIR Systems ThermaCam SC640 by the Center for Advanced Land Management Information Technologies (CALMIT) from the University of Nebraska-Lincoln, USA. Images were captured from approximately 1900 m above the ground between 10 a.m. and 3 p.m. local time on 1 July 2010, 6 August 2010, 6 July 2011, and 29 July 2011; in the experimental year, these dates corresponded to 47, 83, 43, and 66 days after plant emergence (DAE), respectively. ENVI software (Version 4.8, Exelis, Inc., USA) was used for all subsequent image analysis.

Infrared radiometers were used to measure ground-based canopy temperature (Apogee Inst., Model SI-111). Each radiometer was mounted to a pole at the end of the fifth row from the alley about 2 m off the ground, and was aimed at a 45° angle to the crop canopy. Measurements were taken every second and were averaged and recorded every half hour.

3. Results and discussion

3.1. Weather and soil temperature

Refer to Chapter 2 for an overview of daily rainfall, daily irrigation, average daily air temperature, and 2011 average daily soil temperature.

3.2. Soil matric potential

In 2010, average daily soil matric tension at the seed depth stayed below ~20 kPa the entire season for the unstressed irrigation treatment (Fig A-1). Average daily soil matric tension for the stressed irrigation treatment was similar to the unstressed treatment until 15 days after emergence (DAE), and then gradually increased to over 100kPa by 50 DAE. After 50 DAE, it fluctuated between ~40 and ~110 kPa until harvest. In 2011, average daily soil matric tension at the seed depth stayed below ~25 kPa until 110 DAE (Fig A-2). Average daily soil matric tension for the stressed irrigation treatment was similar to the unstressed treatment until about 20 DAE. After 20 DAE, soil matric tension increased slightly and fluctuated between ~20 and ~60 kPa until 70 DAE. After 70 DAE, soil matric tension steadily increased to ~140 kPa and then remained constant at ~140 kPa until harvest. Soil matric tension below the seed depth was less dynamic than soil matric tension at the seed depth in both years and in both irrigation treatments.

3.3. Thermal remote sensing

Two non-invasive remote sensing techniques that measure canopy temperature were broadly evaluated in this study. The first was to use ground-based infrared radiometers to obtain canopy temperature over the course of the growing season. The second technique was to use aerial-based thermal imagery to obtain instantaneous temperature measurements of the entire research field. The infrared radiometers continuously record the apparent canopy temperature at desired time intervals, but they do not account for spatial variability. Thermal imagery can account for spatial variability, but it is not practical to obtain measurements at frequent time intervals.

3.3.1. Infrared radiometers

The infrared radiometers showed that canopy temperatures were very similar throughout 2010 (Fig. A-3), and in 2011, they generally showed that canopy temperatures were actually higher for the unstressed irrigation regime than the stressed irrigation regime (Fig. A-4). It was expected that higher canopy temperatures would be measured in the stressed irrigation regime due to the plants closing their stomates after the onset of water stress. With their stomates closed, evapotranspiration would cease, ultimately resulting in an increased heat flux due to a decreased rate of vapor exchange. The lack of difference between irrigation regimes for these measurements may be partially due to the canopy temperature being averaged daily (i.e., using both daytime and nighttime data). If stomates close, the heat conductance increases and leaf temperature will become more responsive to air temperature. If the air temperature decreases below leaf temperature at night, leaf temperatures for plants with closed stomates will actually decrease faster than plants with stomates open.

From plant emergence until just before harvest in both years, canopy temperature fluctuated between about 15 and 30 ^{o}C. In both years, canopy temperature increased to almost 30 ^{o}C at ~10-15 DAE, and then decreased to almost 15 ^{o}C during the next week. However, because of the small proportion of canopy closure at this growth stage, the increase in temperature was largely due to dynamic surface soil temperatures, not canopy temperature. Based on imagery from Nigon et al. (2012b), it was estimated that canopy closure was ~75% at 30 DAE in 2011.

3.3.2. Thermal imagery

Thermal imagery was obtained on two dates in each growing season, and the main effect of irrigation was statistically significant on all dates except 43 DAE in 2011 (Table A-1). On dates when the irrigation regime had a significant effect on canopy temperate, the stressed irrigation was always higher (Table A-2). On 43 DAE in 2011, soil matric tension in the stressed irrigation regime was only ~40 kPa higher than the unstressed irrigation regime, and this difference was apparently not enough to affect the canopy temperatures. The canopy temperature varied by image date and in magnitude between the irrigation regimes. In both years, the later image date had a higher canopy temperature. In 2010, there was well over a one degree difference between the irrigation regimes, but in 2011, the difference was less than 0.2 °C on both image dates.

On 83 DAE in 2010 and 43 DAE in 2011, the main effect of N treatment was significant; however, both the irrigation × N treatment and N treatment × variety interactions were significant on 83 DAE in 2010, and the N treatment × variety interaction was significant on 43 DAE in 2011. Fig. A-5 shows the mean canopy temperatures and means separation for all treatments on 83 DAE in 2010. Fig. A-6 shows the mean canopy temperatures and means separation for the N treatment × variety interaction on 43 DAE in 2011.

Because thermal images can account for spatial variability, they make it possible to determine the amount of spatial variability within individual treatment plots. Fig. A-7 shows canopy temperature for the research plots on 83 DAE in 2010 without the alleyways masked out. Between irrigation regimes, differences in temperature are most obvious in the alleyways, but there are apparent differences within the treatment plots. On 83 DAE in 2010, plants had begun to senesce, so differences in temperature may be partially due to soil. Differences in temperature within treatment plots were still visually apparent in the image on 66 DAE in 2011, even though the absolute range in temperatures was much lower than for 83 DAE in 2010 (Fig. A-8). The alleys were masked out in Fig. A-8, so visual differences between pixels were due entirely to differences in crop canopy temperature. Differences in temperature between irrigation regimes were still visible, especially in the east half of the field. There was standing water in the plots in the southeast corner on 66 DAE in 2011, and it is obvious that this caused especially low canopy temperatures. Because the main effect of N treatment and the N treatment × variety interaction were significant on both image dates from Figs. A-7 and A-8, differences within irrigation regime are likely due to N treatment.

4. Conclusions

Throughout the season, daily soil matric tension was much more responsive to differences in water stress than daily canopy temperature that was measured by the infrared radiometers. The lack of responsiveness for the daily canopy temperature was partially due to both daytime and nighttime data being used. Only daytime leaf canopy temperature data should be used for the determination of crop water stress. Three of the four image dates in this study showed a significant response in irrigation regimes for canopy temperature measurements measured from aerial imagery. There were large differences in the absolute values of canopy temperature among image dates, but within-field and within-plot differences were still apparent. Applying this data to water stress indices such as the Crop Water Stress Index should be completed to investigate its full functionality.

5. References

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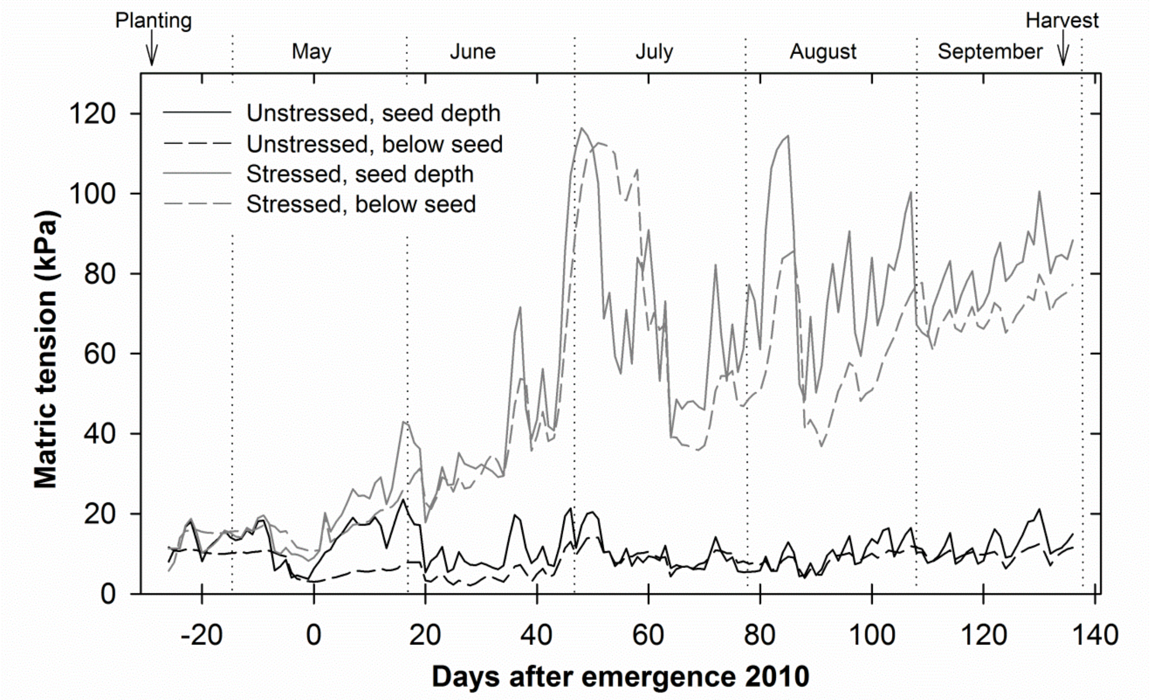

Fig. A-1. Average daily soil matric tension throughout the 2010 season for the unstressed and stressed irrigation regimes at and about 30 cm below the seed depth.

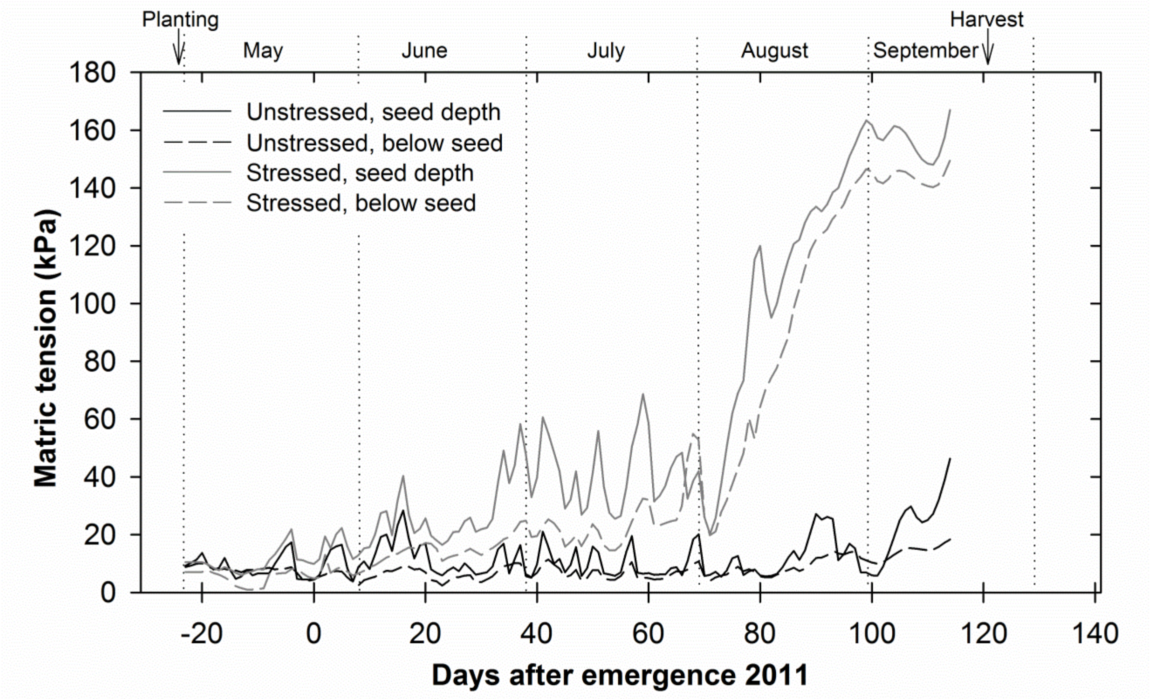

Fig. A-2. Average daily soil matric tension throughout the 2011 season for the unstressed and stressed irrigation regimes at and about 30 cm below the seed depth.

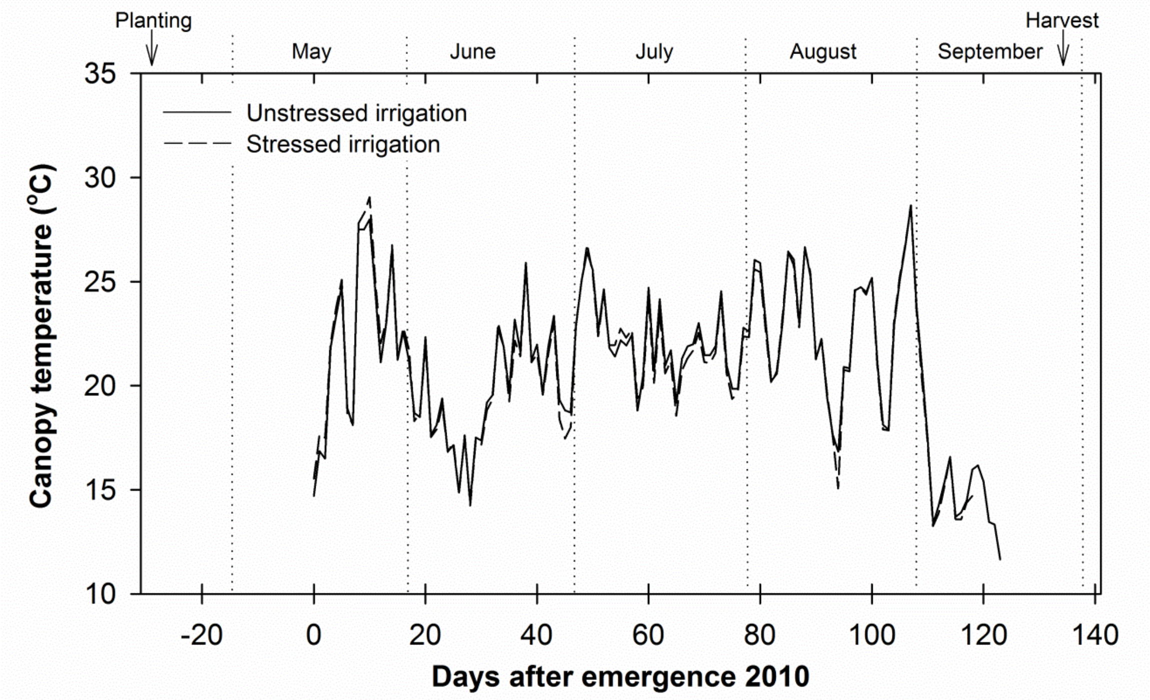

Fig. A-3. Average daily potato canopy temperature throughout the 2010 season for the unstressed and stressed irrigation regimes.

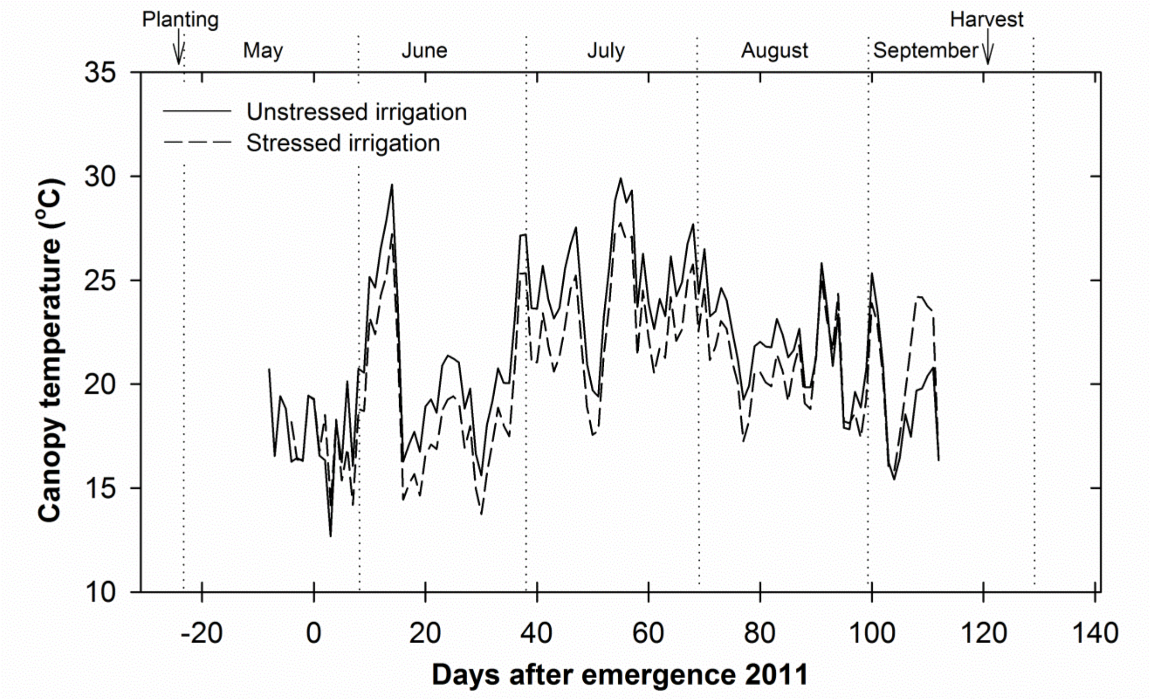

Fig. A-4. Average daily potato canopy temperature throughout the 2011 season for the unstressed and stressed irrigation regimes.

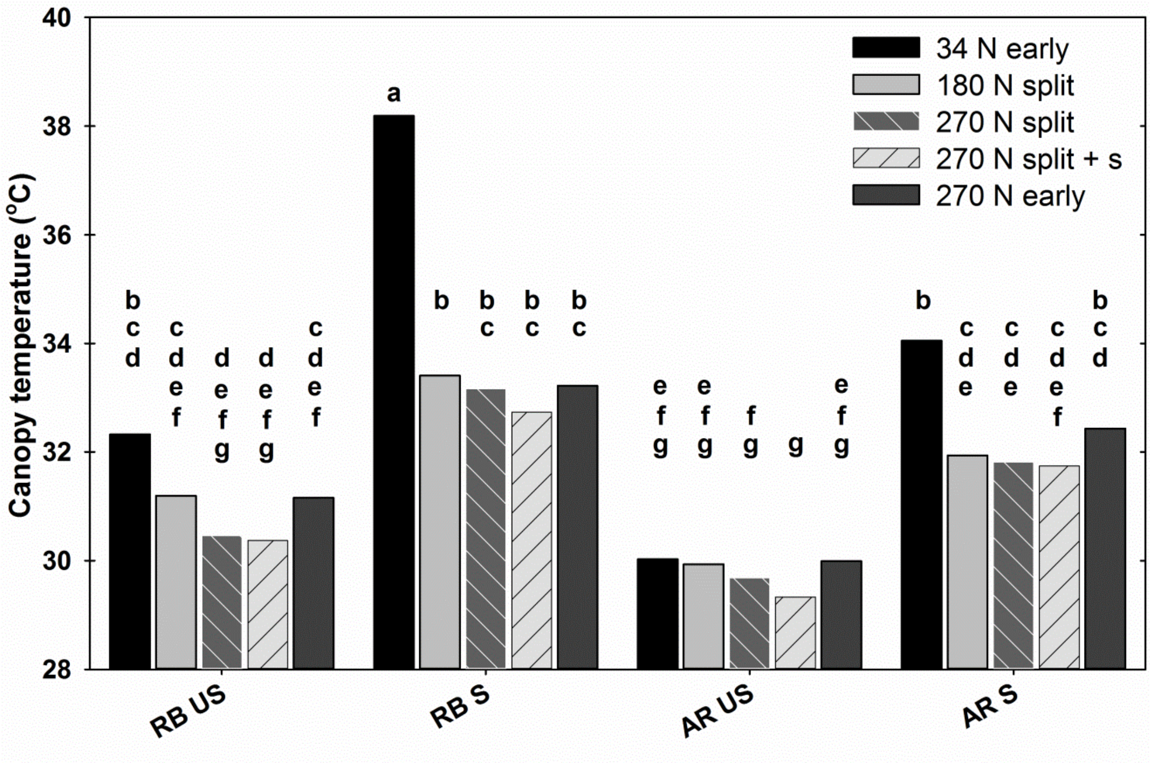

Fig. A-5. Mean values for canopy temperature as measured by the ThermaCam SC640 on 83 DAE in 2010 for the irrigation × N treatment interaction and the N treatment × variety interaction. Values with the same letter are not significantly different (α = 0.05). RB = Russet Burbank, AR = Alpine Russet; US = unstressed, S = stressed.

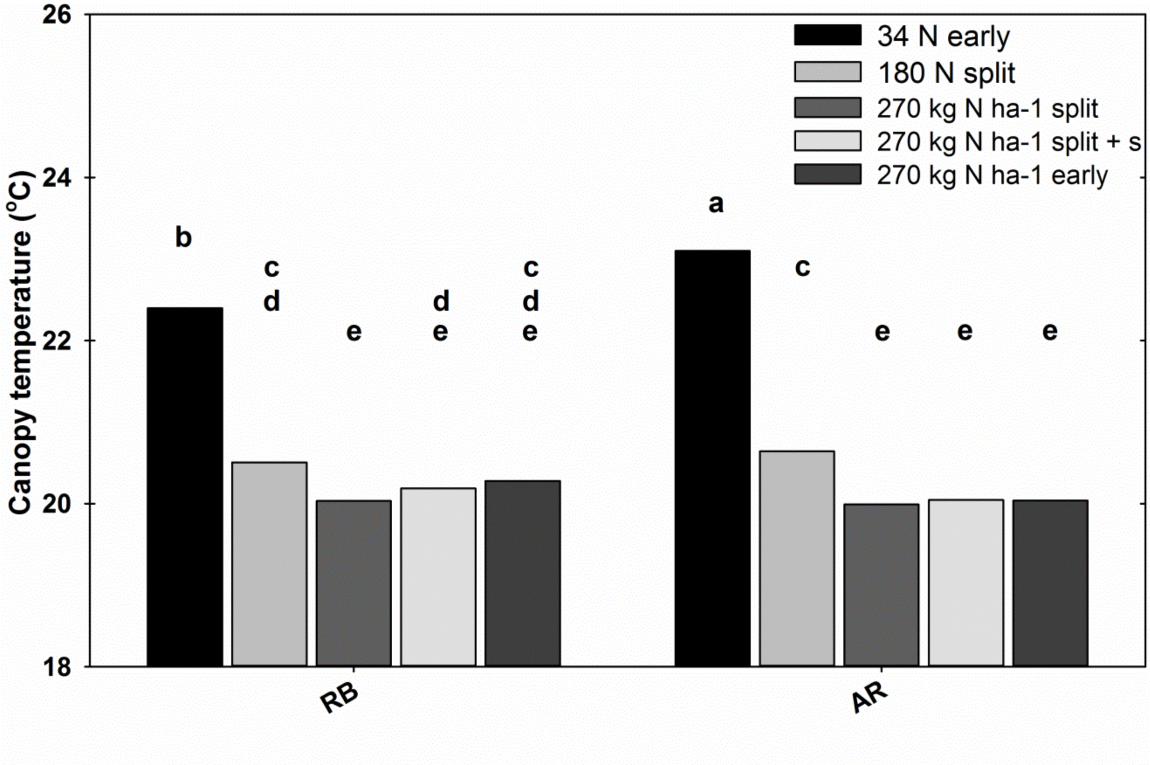

Fig. A-6. Mean values for canopy temperature as measured by the ThermaCam SC640 on 43 DAE in 2011 for the N treatment × variety interaction. Values with the same letter are not significantly different (α = 0.05). RB = Russet Burbank, AR = Alpine Russet.

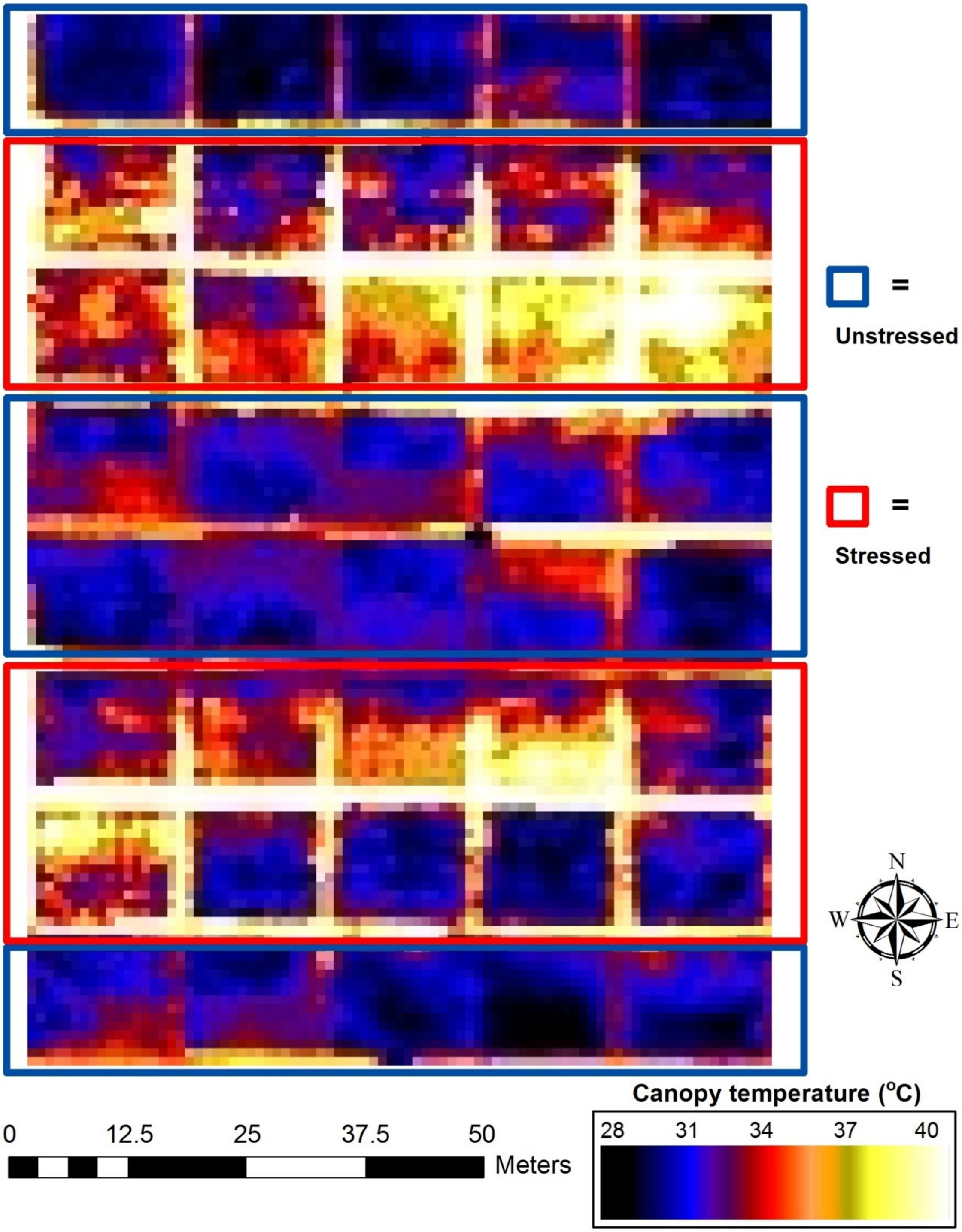

Fig. A-7. Thermal image of the research plots obtained from the ThermaCam SC640 on 83 days after emergence in 2010. Plots outlined in blue are unstressed irrigation treatments and plots outlined in red are stressed irrigation treatments.

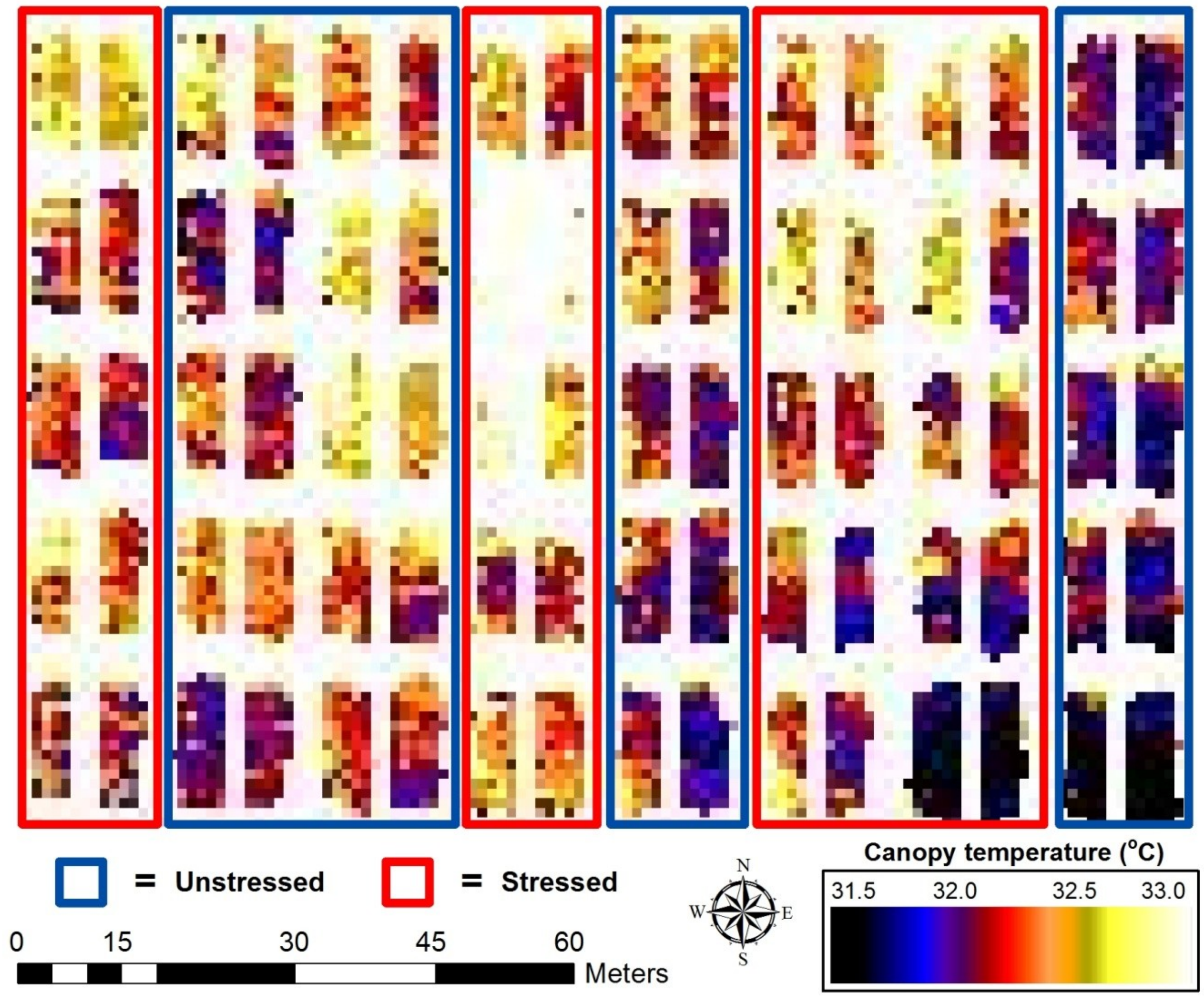

Fig. A-8. Thermal image of the research plots obtained from the ThermaCam SC640 on 66 days after emergence in 2011. Plots outlined in blue are unstressed irrigation treatments and plots outlined in red are stressed irrigation treatments.

Table A-1. Analysis of variance for canopy temperature as measured by the ThermaCam SC640 on two dates in each 2010 and 2011.

Effect	2010		2011	
	47 DAE	83 DAE	43 DAE	66 DAE
Irrigation [I]	*	*	-	**
N treatment [N]	-	***	***	-
Variety [V]	***	*	-	**
I × N	-	***	-	-
I × V	-	-	-	-
N × V	-	***	**	-
I × N × V	-	-	-	-

Table A-2. Main effects of irrigation, nitrogen (N) treatment, and variety on canopy temperature as measured by the ThermaCam SC640 on two dates in each 2010 and 2011.

Sources of Variation	2010		2011	
	47 DAE	83 DAE[†]	43 DAE[†]	66 DAE
Irrigation	-----------------	Temperature (°C)	-----------------	
Unstressed	27.4 b[‡]	30.4 b	20.8 a	32.2 b
Stressed	29.1 a	33.3 a	20.7 a	32.4 a
N Treatment				
34 N early	29.1a	33.6 a	22.7 a	32.5 a
180 N split	27.9a	31.6 b	20.6 b	32.4 a
270 N split	28.4a	31.3 b	20.0 c	32.3 a
270 N split + s	27.9a	31.0 b	20.1 c	32.3 a
270 N early	28.0a	31.7 b	20.2 c	32.3 a
Variety				
Russet Burbank	28.4 b	32.6 a	20.7 a	32.4 a
Alpine Russet	28.1 a	31.1 b	20.8 a	32.3 b

[†] Response variables that had at least one significant interaction; refer to Table A-1 and Figs. A-5 and A-6.

[‡] Means followed by the same letter are not significantly different (α=0.05).

Other Tables

Table A-3. Analysis of variance for potato tuber yields and size categories.

Effect	0-85 g	85-170 g	170-283 g	283-397 g	>397 g	#1 > 85 g	#2 > 85 g	Grade A yield	Total yield	>170g	> 283 g
Year [Y]	***[†]	-	***	***	***	**	-	***	***	***	***
Irrigation [I]	-	-	*	*	**	-	*	*	**	**	**
N treatment [N]	***	-	-	***	***	***	***	***	***	***	***
Variety [V]	***	***	***	***	***	***	-	***	-	***	***
I x N	-	-	-	-	-	-	-	-	-	-	-
I x V	*	**	-	**	***	-	-	-	-	*	**
I x Y	*	***	*	-	-	-	-	-	-	-	-
N x V	-	**	**	-	**	-	***	*	-	-	-
N x Y	-	-	-	-	-	-	-	-	-	-	-
V x Y	-	-	-	-	-	-	-	-	-	-	-
I x N x V	-	-	-	-	-	-	-	-	-	-	-
I x N x Y	-	-	-	-	-	-	-	-	-	-	-
I x V x Y	*	*	-	-	*	*	**	-	-	-	*
N x V x Y	*	***	-	-	*	-	-	-	-	-	-

[†] ***, **, and * are significant 0.001, 0.01, and 0.05, respectively; - is nonsignificant.

Table A-4. Tuber yields by size categories for all treatments over both years of the study.

Source of variation			Tuber Size Class							Grade A yield	Total yield	>170g	> 283 g
Irrigation	Variety	N treatment	0-85 g	85-170 g	170-283 g	283-397 g	>397 g	#1 > 85 g	#2 > 85 g				
			ton ha^{-1}									%	
Unstressed	Russet Burbank	34 N early	13.4 ab[†]	24.7 a	7.5 i	0.4 k	0.1 kl	23.7 gh	8.9 bcdef	32.6 h	46.0 g	17.0 h	0.9 h
		180 N split	10.0 cd	25.1 ab	20.1 cde	2.5 ij	0.6 hij	39.8 cdef	8.4 cdef	48.2 cde	58.3 abcd	39.3 def	5.1 fg
		270 N split	9.2 cd	24.3 ab	21.1 abc	5.6 cdef	1.1 gh	39.8 cdef	12.4 ab	52.2 bc	61.3 a	44.8 d	10.7 cde
		270 N split + s	9.3 cd	23.8 ab	20.3 bcd	5.0 defg	1.1 efgh	38.8 def	11.5 abc	50.2 cd	59.5 ab	43.6 de	10.0 cde
		270 N early	8.6 cde	23.5 abc	19.5 cdef	5.9 cdef	0.9 efgh	43.2 bcde	6.7 efgh	49.8 cd	58.4 abc	44.8 d	11.7 cd
	Alpine Russet	34 N early	5.7 f	20.1 cde f	16.3 efg	5.1 efgh	0.8 hijk	27.9 g	14.4 a	42.3 fg	48.0 g	45.1 d	11.7 de
		180 N split	4.4 fgh	16.3 fgh	22.9 abc	12.1 a	5.0 b	46.7 ab	9.5 bcd	56.3 ab	60.6 a	65.5 ab	27.8 a
		270 N split	4.1 h	13.6 hi	23.8 ab	12.5 a	7.5 a	48.1 a	9.3 bcde	57.4 a	61.5 a	71.0 a	32.2 a
		270 N split + s	4.3 gh	15.1 ghi	24.6 a	12.0 a	5.5 ab	46.6 ab	10.6 abc	57.2 a	61.5 a	68.1 a	28.1 a
		270 N early	4.4 gh	12.9 i	23.1 abc	11.0 ab	4.5 bc	45.1 ab	6.5 defg	51.5 c	55.9 bcde	68.6 a	27.2 a
Stressed	Russet Burbank	34 N early	15.1 a	18.6 efg	6.2 i	0.3 k	0.0 l	21.8 h	3.3 j	25.1 i	40.2 h	15.0 h	0.6 h
		180 N split	10.9 bc	25.0 a	13.4 gh	2.3 ij	0.3 ijkl	36.4 f	4.7 hij	41.0 g	51.9 f	29.7 g	4.7 fg
		270 N split	9.3 cd	22.0 abc d	17.1 def	3.4 hi	1.2 ghi	35.3 f	8.3 defg	43.6 efg	52.9 ef	39.8 def	7.8 ef
		270 N split + s	9.8 cd	24.6 a	15.8 fg	4.1 fgh i	1.0 ghi	38.0 ef	7.5 fghi	45.5 def	55.3 cdef	36.1 efg	8.5 def
		270 N early	8.8 d	21.9 abc d	16.7 defg	3.6 ghi	1.2 fgh	39.8 def	3.6 j	43.4 efg	52.2 f	39.3 def	8.4 def
	Alpine Russet	34 N early	7.1 e	20.0 bcd e	12.3 h	1.9 j	0.1 jkl	25.4 gh	9.0 bcde	34.3 h	41.5 h	32.1 fg	4.2 g
		180 N split	5.0 fgh	18.2 def g	20.8 abc	7.1 cde	2.6 efg	42.8 bcd	5.9 fghi	48.7 cd	53.8 ef	55.3 c	17.0 bc
		270 N split	5.5 fg	18.2 def g	20.1 bcde	7.6 cd	2.8 de	44.5 abc	4.1 ghij	48.6 cd	54.1 ef	54.3 c	17.8 b
		270 N split + s	5.7 fg	15.8 ghi	20.4 abc	8.6 bc	4.2 cd	44.0 abcd	5.0 ghij	49.0 cd	54.6 def	58.6 bc	21.6 b
		270 N early	5.1 fgh	16.3 fgh	20.6 abc	8.0 cd	2.6 def	44.2 abc	3.3 ij	47.5 cdef	52.7 ef	56.8 c	18.3 b

[†] Means followed by the same letter within a tuber size class are not significantly different (α=0.05).

Table A-5. Analysis of variance for petiole nitrate nitrogen (NO_3-N) and whole leaf nitrogen (N) at various days after emergence (DAE) throughout the 2010 growing season.

Effect	31 DAE		47 DAE		59 DAE		83 DAE	
	Petiole NO_3-N	Leaf N	Petiole NO_3-N	Leaf N	Petiole NO_3-N	Leaf N	Petiole NO_3-N	Leaf N
Irrigation [I]	**[†]	-	-	-	-	-	-	-
N treatment [N]	***	***	***	***	***	***	***	***
Variety [V]	-	*	***	***	**	***	***	***
I x N	-	-	-	-	-	-	-	*
I x V	-	*	-	-	-	-	-	-
N x V	-	**	**	***	-	-	***	-
I x N x V	-	-	-	-	-	-	-	-

[†] ***, **, and * are significant 0.001, 0.01, and 0.05, respectively; - is nonsignificant.

Table A-6. Analysis of variance for petiole nitrate nitrogen (NO_3-N) and whole leaf nitrogen (N) at various days after emergence (DAE) throughout the 2011 growing season.

Effect	30 DAE		43 DAE		56 DAE		66 DAE		79 DAE	
	Petiole NO_3-N	Leaf N	Petiole NO_3-N	Leaf N	Petiole NO_3-N	Leaf N	Petiole NO_3-N	Leaf N	Petiole NO_3-N	Leaf N
Irrigation [I]	-[†]	-	-	-	-	-	-	-	**	**
N treatment [N]	***	***	***	***	***	***	***	***	***	***
Variety [V]	-	*	**	***	***	***	***	***	-	***
I x N	-	-	-	-	-	-	-	-	***	-
I x V	-	-	-	-	-	-	-	-	-	-
N x V	-	-	-	-	*	-	*	-	-	*
I x N x V	-	-	-	-	-	-	-	-	-	-

[†] ***, **, and * are significant 0.001, 0.01, and 0.05, respectively; - is nonsignificant.

Table A-7. Petiole nitrate nitrogen (NO_3-N) and whole leaf nitrogen (N) treatment means at various days after emergence (DAE) throughout the 2010 growing season.

Source of variation			Petiole NO3-N				Leaf N			
Irrigation	Variety	N treatment	31 DAE	47 DAE	59 DAE	83 DAE	31 DAE	47 DAE	59 DAE	83 DAE
			mg kg^{-1}				g kg^{-1}			
Unstressed	Russet Burbank	34 N early	1414 g[†]	310 h	407 h	490 hi	43.8 i	32.7 f	31.8 f	33.2 gh
		180 N split	13631 ef	2895 defg	3148 bcde	2707 cd	57.7 defg	43.3 c	39.8 abcd	41.1 bcd
		270 N split	13619 ef	4532 cd	4087 abc	4306 ab	56.0 efgh	44.3 c	41.2 ab	43.5 abc
		270 N split + s	14603 cdef	4490 cd	3775 abcd	3342 bc	58.7 defg	43.9 c	40.4 abcd	44.1 ab
		270 N early	18788 bcd	7204 a	1657 efgh	1075 fghi	62.9 bc	48.4 a	38.6 abcde	36.9 efg
	Alpine Russet	34 N early	1472 g	306 h	147 h	102 i	38.9 j	29.9 gh	29.1 f	27.7 ij
		180 N split	12518 f	2560 fg	1497 fgh	1473 efgh	54.0 h	37.0 de	37.1 cde	38.0 def
		270 N split	14605 cdef	2696 efg	3735 abcd	1972 defg	55.6 gh	37.2 de	40.9 ab	39.4 cde
		270 N split + s	12769 f	1822 fgh	3777 abcd	2186 cdef	55.9 fgh	38.6 d	40.2 abc	39.8 bcde
		270 N early	17680 bc	3083 def	751 gh	144 i	65.0 ab	38.6 d	35.8 e	32.6 h
Stressed	Russet Burbank	34 N early	1671 g	490 h	335 h	184 hi	44.0 i	32.4 fg	31.1 f	32.0 h
		180 N split	14366 def	3008 def	2489 cdef	2560 cde	57.7 defg	42.9 c	40.5 abc	38.9 de
		270 N split	16054 cdef	5128 bc	4441 ab	5586 a	59.1 de	45.4 bc	43.5 a	46.9 a
		270 N split + s	15614 cdef	4337 cde	5407 a	4650 ab	58.8 def	42.8 c	43.2 a	43.7 ab
		270 N early	20639 ab	6666 ab	2228 defg	618 ghi	64.9 ab	47.7 ab	40.4 abc	34.3 fgh
	Alpine Russet	34 N early	3870 g	443 h	336 h	57 i	39.1 j	30.0 h	28.2 f	24.3 j
		180 N split	17018 bcde	1346 gh	2469 cdef	410 hi	60.0 cd	35.4 e	37.5 bcde	32.1 h
		270 N split	15987 cde	1925 fgh	3614 bcd	1912 def	58.3 defg	37.6 de	40.9 abc	39.5 cde
		270 N split + s	16325 cdef	2244 fg	3259 bcde	1960 def	60.4 cd	37.3 de	40.8 abc	37.5 def
		270 N early	20309 a	4253 cde	783 h	264 hi	67.5 a	39.5 d	35.9 de	30.9 hi

[†] Means followed by the same letter within a measurement date are not significantly different (α=0.05).

Table A-8. Petiole nitrate nitrogen (NO_3-N) and whole leaf nitrogen (N) treatment means at various days after emergence (DAE) throughout the 2011 growing season.

Source of variation			Petiole NO3-N					Leaf N				
Irrigation	Variety	N treatment	30 DAE	43 DAE	56 DAE	66 DAE	79 DAE	30 DAE	43 DAE	56 DAE	66 DAE	79 DAE
			mg kg^{-1}					g kg^{-1}				
Unstressed	Russet Burbank	34 N early	636 h[†]	828 hi	603 hi	175 h	125 f	43.9 h	34.6 i	36.6 k	32.9 ij	31.0 hi
		180 N split	13993 fg	1927 hi	4023 efg	1803 efgh	1843 bcd	58.5 g	43.1 gh	46.2 efghi	40.6 defg	40.7 cde
		270 N split	16616 def	5225 bcdef	10859 a	5630 abc	5813 a	63.9 def	47.7 cde	53.2 abc	46.6 ab	46.1 ab
		270 N split + s	15956 ef	6712 bc	7383 bcd	7089 a	5142 a	64.6 def	48.5 bcd	50.5 bcd	47.2 a	46.9 ab
		270 N early	20116 abc	9063 a	8750 ab	2986 def	596 def	67.8 abc	52.1 a	54.5 a	42.3 cdef	37.3 ef
	Alpine Russet	34 N early	975 h	150 i	136 i	47 h	140 f	44.5h	35.2 i	36.3 k	31.4 ij	29.2 hij
		180 N split	11510 g	1435 hi	1363 ghi	1100 fgh	1154 def	58.3g	40.5 h	41.4 j	37.2 gh	36.0 fg
		270 N split	16847 def	4408 defg	5728 cdef	4700 bcd	6313 a	64.2def	45.3 defg	49.4 def	43.6 abcd	46.5 ab
		270 N split + s	16599 def	4446 cdefg	8500 abc	3215 de	5793 a	65.4bcde	45.7 cdefg	45.8 ghi	42.2 cdef	47.3 a
		270 N early	20772 abc	6669 abc	5820 cdef	2012 efgh	586 def	69.5a	49.0 abc	49.4 def	38.9 efg	35.5 fg
Stressed	Russet Burbank	34 N early	855 h	102 i	268 i	56 h	62d f	44.0h	33.2 i	34.5 k	29.6 j	27.5 ij
		180 N split	12081 g	2083 ghi	2582 fghi	2812 def	1045 def	58.8g	43.6 efgh	44.5 hij	40.9 defg	38.5 def
		270 N split	17599 cde	5526 bcde	7330 bc	6261 ab	2698 bc	61.6f	47.3 cdef	50.8 abcd	44.4 abcd	43.3 bc
		270 N split + s	17715 cde	5107 cdef	6304 bcde	6767 a	1730 cde	62.9ef	47.5 cdef	49.9 cde	45.9 abc	41.2 cd
		270 N early	21832 ab	7712 ab	8040 abc	4056 cd	441 ef	68.0ab	51.6 ab	53.6 ab	44.3 abcd	37.1 ef
	Alpine Russet	34 N early	1401 h	126 i	195 i	77 h	48 f	44.3h	35.2 i	34.3 k	29.2 j	25.4 j
		180 N split	12535 g	759 i	1387 ghi	701 gh	414 f	56.4g	40.6 h	42.5 ij	34.7 hi	32.1 gh
		270 N split	18021 cde	3049 efgh	3545 efgh	4164 cd	3064 b	65.6bcd	41.9 gh	46.3 fgh	42.5 cdef	40.9 cde
		270 N split + s	19458 bcd	3255 fgh	3943 defgh	4684 bcd	1774 bcd	65.1cde	43.8 fgh	48.3 defgh	42.9 bcde	40.5 cde
		270 N early	22815 a	6384 abcd	5731 bcde	2767 defg	656 def	70.0a	48.0 bcd	48.5 defg	38.9 fg	35.5 fg

[†] Means followed by the same letter within a measurement date are not significantly different (α=0.05).