

Relationship Between Coefficient of Variation Measured by Spectral Reflectance and Plant Density at Early Growth Stages in Winter Wheat

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ABSTRACT

The use of by-plot coefficient of variation (CV) has not been evaluated in precision agricultural work. This study evaluated the relationship between CVs determined from normalized-difference vegetative index (NDVI) sensor readings, plant population, and sensing direction on NDVI values. Randomly selected plots, measuring 1 m² (2003) and 3 m² (2004), were established for this study. Plots in 2004 were divided into three 1 m² subplots with, 0 and 120 kg ha⁻¹ fall-applied N, and 80 kg ha⁻¹ topdress nitrogen (N). Sensor reading of subplots were taken at Feekes 5 and 7 using the Green Seeker hand-held sensor. Results showed that the relationship between vegetative RI (RI_{NDVI}) and harvest RI (RI_{Harvest}) improved with increasing CV values. The prediction of RI_{Harvest} was improved when CV was integrated into the RI_{NDVI} calculation. RI_{Harvest} can be better predicted with RI_{NDVI} when the CV of spectral radiance measurements is used in the RI_{NDVI} equation.

Keywords: coefficient of variation, response index, plant stand, normalized vegetative index, winter wheat

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INTRODUCTION

Globally, approximately 82 million t of nitrogenous fertilizers were applied in 2001 (FAO, 2002). Cereal grains accounted for 60% of the total nitrogen (N) fertilizer applied in 1994 (FAO, 1995). Only 33% of the fertilizer N used for cereal-grain crops is removed in the grain (Raun and Johnson, 1999).

Plant N losses in winter wheat (*Triticum aestivum* L.) have accounted for between 21% (Harper et al., 1987) and 41% (Daigger et al., 1976) of the total N lost using N¹⁵. Loss of gaseous N due to denitrification is reported to range from 10% (conventional tillage) to 22% (no-till) in corn (*Zea mays*) (Hilton et al., 1994). In addition, fertilizer N losses in surface runoff range between 1% (Blevins et al., 1996) and 13% (Chichester and Richardson, 1992) of the total N applied. Lower levels of losses due to runoff are usually associated with no-till conditions. An additional pathway for N loss is through leaching of nitrate (NO₃⁻) when applied in excess of crop need. In cooler temperate climates, NO₃-N losses through tile drainage have approached 26 kg N ha⁻¹ yr⁻¹ under conventional tillage of corn when only 115 kg N ha⁻¹ was applied (Drury et al., 1996).

While plant loss accounts for a very large portion of N loss, the soil environment also accounts for a high percentage of this loss. If any one of the pathways can be restricted and loss thus reduced, the benefit will be significant. Johnson and Raun (2003) calculated that a 1% global increase in cereal nitrogen use efficiency (NUE) would have a value of \$235 million in N-fertilizer savings if yields were maintained. Raun et al. (2002) reported an improvement in NUE of >15% when N fertilization was based on optically sensed in-season estimated yield (INSEY).

The GreenSeeker hand-held optical sensor (NTech Industries, Ukiah, CA.), developed by Oklahoma State University, senses a 0.6×0.01 m area when held at a distance of approximately 0.6 to 1.0 m from the illuminated surface. The sensed dimensions remain approximately constant over the height range of the sensor. The sensor unit has self-contained illumination in both red (660 \pm 10 nm) and NIR (767 \pm 15 nm) bands. The device measures the fraction of emitted light in the sensed area that is returned to the sensor (reflectance). The algorithm currently used by N-Tech Industries, "WheatN1.0," includes several distinct components. Raun et al. (2005a) identified three specific ones: (1) mid-season prediction of grain yield, determined by dividing normalized difference vegetative index (NDVI) by the number of days from planting to sensing (estimate of biomass produced per day from planting to the specific date when sensor readings are collected); (2) estimation of temporally dependent responsiveness to applied N by placing non-N-limiting strips in production fields each year, and comparing these to the farmer practice (fertilizer response index); (3) determination of the spatial variability within each 0.4 m² area using the coefficient of variation (CV) from NDVI readings.

The results of previous work have shown that stand density and uniformity have an affect on grain yield. Weisz et al. (2001) reported that as plant stand or

tiller density increased, grain yield tended to increase, and the variation within the field to decrease. Nielsen (2001) showed that in corn, for every 2.56 cm standard deviation of plant-to-plant spacing, there was a decrease in yield of 1567 kg ha⁻¹ from the average yield of 9800 kg ha⁻¹. This finding indicated the need to make fertilization recommendations with stand density as a factor. Flowers et al. (2003) validated the use of aerial photography for determining winter wheat tiller density. Using the density estimates, they determined that basing N application on a critical density threshold had an 85.5% success rate. Lukina et al. (2000) observed that as the vegetation coverage increased, the CV of NDVI values decreased. Raun et al. (2001) showed that NDVI values from mid-season sensor readings could be used to predict yield. Combining NDVI and CV independently may result in improved prediction of yield potential.

Coefficient of variation is defined as the standard deviation divided by the mean (Tippett, 1952; Senders, 1958; Steel et al., 1997; Lewis, 1963). Steel et al. (1997) describe CV as a quantity of use to the experimenter in evaluating results from different experiments involving the same unit of measure, possibly conducted by different people. Little and Hills (1978) suggested that CV can be used to compare experiments involving different units of measurement and/or plot sizes. The CV is a relative measure of variation and varies with every comparison of what is considered large or small, and only experience with similar data can determine its meaning (Steel et al., 1997).

In an evaluation of 62 wheat field research projects, Taylor et al. (1997) observed that mean yield and CV were negatively correlated. The work of Taylor et al. (1997) also showed that CVs decreased with corresponding decreases in plot size. Washmon et al. (2002) suggested that if within-field CVs could be predicted, the potential response to added nutrients could also be established and in-season nutrient additions adjusted accordingly. They further stated that the mid-season CV of a field could be equated to the response index, which is currently used by various researchers to determine topdress fertilizer rates.

Raun et al. (2005a) predicted that when CV was low, a responsive field element should be capable of greater yield than when a similarly responsive field element CV was large. In testing this concept, it was observed that YP_{N-CV} (predicted yield with added N using INSEY and the CV at the time of sensing) values more closely followed observed yield than did YP_N (predicted yield using the INSEY equation) values. Morris et al. (2005) noted that when plot CVs of NDVI readings were >18, maximum yields could not be achieved when N fertilizer was delayed until mid-season. When plot CVs were <18, delaying all N fertilization until mid-season resulted in maximum yields and increased NUE.

The GreenSeeker sensor collects more than 10 readings within each 0.4 m^2 traveling at 10 mph (Raun et al., 2005a). Raun et al. (2005a) stated that the 10 readings collected from each 0.4 m^2 were considered sufficient to obtain a composite sample to estimate reliably the average from such a small area,

understanding that the 10 sensor readings were representative of the variability from the same 0.4 m^2 surface area.

The variable-rate method is a vast improvement on the use of 15 soil samples to represent a unit area that could range from a few acres to several hundred acres (Johnson et al., 2000). If the goal is to maximize crop NUE, the use of averaged NDVI values obtained from spectral-reflectance measurements presents a problem. Currently, two 0.4 m^2 areas with similar NDVIs would receive the same treatment, but might need two different N rates. A good stand of nutrient-deficient wheat may have the same average NDVI as a poor stand of nutrient-enriched wheat. The ability to index plant-stand density on the go may provide the solution needed. The effect of plant population and tiller density on the GreenSeeker sensor's ability to determine yield potential correctly has not yet been assessed.

The objectives of this work were to determine the relationship between the CV of measurements of NDVI and plant population at early growth stages. Sensing direction in relation to the crop-row direction on the CV from spectral radiance measurements was also evaluated, in addition to the change in CV over time.

MATERIALS AND METHODS

Experimental sites were established in Oklahoma, at the EFAW research farm in Stillwater, the Hajek farm in Hennessey, the Lake Carl Blackwell Research Station near Stillwater, and the Perkins Research Station near Perkins in the spring of 2003. The same sites were used in 2004 excluding the Hajek farm. The soil classification for the sites is listed in Table 1. All planting and management dates are reported in Table 2.

In 2003, 30 plots were randomly selected at the Hajek farm and the EFAW farm. Forty-five plots were randomly selected at the Perkins station and Lake Carl Blackwell farm. Plots were established after germination at Feekes 1

Table 1 Soil classification for all experimental sites (Lake Carl Blackwell, Perkins, EFAW, and Hajek Farm) in 2003–2004

| Location | Soil classification | | | |
|---------------------|--|--|--|--|
| Lake Carl Blackwell | Port fine-silty, mixed, superactive, thermic Cumulic Haplustoll | | | |
| Perkins Research | Konawa Teller association | | | |
| | Konawa fine-loamy, mixed, active, thermic Ultic Haplustalf | | | |
| | Teller fine-loamy, mixed, active, thermic Udic Argiustoll | | | |
| EFAW | Norge fine silty, mixed, active, thermic Udic Paleustoll | | | |
| Hajek Farm | Shellabarger fine-loamy, mixed, superactive, mesic Udic Argiustoll | | | |

| Location | Crop year | Planting date | Variety | Seeding rate (kg ha ⁻¹) | Topdress date |
|---------------------|--------------|------------------|----------|--|------------------|
| Lake Carl Blackwell | 2003 | 10/01/02 | Intrada | 101 | |
| | 2004 | 10/7/03 | Jagalene | 95.3 | 3/30/04 |
| Perkins | 2003 | 10/14/02 | Jagger | 101 | |
| | 2004 | 9/26/03 | Jagger | 89.7 | 2/20/04 |
| EFAW | 2003 | 10/8/02 | 2174 | 89.7 | _ |
| | 2004 | 11/10/03 | OK 101 | 72.9 | 3/30/04 |
| Hajek Farm | 2003 | 10/20/02 | Custer | 89.7 | — |

 Table 2

 Planting date, variety, seeding rate, and topdress application dates for all experimental sites (Lake Carl Blackwell, Perkins, EFAW, and Hajek Farm) in 2003–2004

(emergence). The plots were established at this stage so that the plots would be oriented with seed rows. Plot size measured 1.48×1.48 m, with each plot containing eight rows spaced 15 cm apart. In total, 150 plots were used.

In 2004, the experiment was modified to include three N treatments (0, 120 kg ha⁻¹ fall applied, and 80 kg ha⁻¹ topdress), each applied to a plot of 1.48×4.44 m. The treatment structure was the same for all plots. Each plot was randomly selected within each location. Twenty-five plots were established at EFAW, Lake Carl Blackwell, and Perkins at Feekes 1.

Plant-stand density was estimated for each plot at Feekes 1 by counting all plants within four rows randomly selected in each plot. This count was preformed prior to tillering; therefore, each shoot was recorded as a plant. The 120 kg ha⁻¹ N was applied at plot establishment in fall and the 80 kg ha⁻¹ N was topdressed at Feekes 6 (first node visible) using urea (46-0-0, N-P-K).

Spectral-radiance measurements were taken using the GreenSeeker handheld optical sensor unit. The device used a technique to measure crop reflectance and to calculate NDVI. The equation for NDVI is shown below:

$$NDVI = \frac{\rho_{NIR} + \rho_{Red}}{\rho_{NIR} + \rho_{Red}}$$
(1)

where ρ_{NIR} was the fraction of emitted NIR radiation returned from the sensed area (reflectance) and ρ_{Red} was the fraction of emitted red radiation returned from the sensed area (reflectance).

During the 2003 season at the EFAW research station and Lake Carl Blackwell farm locations, sensing was performed once a week until maturity. Readings were taken from Feekes 5 (leaf sheaths strongly erect) through Feekes 8 (flag leaf visible) at the Hajek farm and Perkins Research Station. A total of four sensor passes were made on each plot, holding the sensor approximately 75 to 100 cm above the crop canopy. The sensor path was parallel to the seed rows; two passes were made midway between the second and third rows, then the sixth and seventh rows. Two sensor passes were taken perpendicular to the seed rows with the passes made parallel to the plot borders and offset approximately 30 cm from the plot borders. Time for each pass was three seconds. Approximately 30 NDVI readings were collected with each pass. In 2004, sensing began at all locations in January at or near Feekes 3 (tillers formed) and continued until physiological maturity. Also in 2004, a fifth pass was added to the sensing plan, directed at a 45-degree angle to the planting direction on each subplot, for the purpose of complete evaluation of sensing direction.

For the 2003 season, wheat head counts were taken at maturity by counting the number of heads in the rows, which had been used to estimate plant population. In both seasons, a 1 m² section from the center of each plot or subplot was harvested at maturity using a hand sickle, cutting slightly above the crown. The harvested samples were dried, weighed, and threshed using a mechanized thresher. Sample grain was then weighed to determine yield of each plot. Total grain N content was determined using the Carlo Erba NA 1500 Series 2 nitrogen analyzer (Milan, Italy) on the 2004 samples.

Two response indices (RI), harvest RI ($RI_{Harvest}$) and vegetative RI (RI_{NDVI}), were calculated using the following equations (Raun et al., 2002):

$$RI_{Harvest} = \frac{Yield_{N-Rich}}{Yield_{Check}}$$
(2)

where $\text{Yield}_{N-\text{Rich}} = \text{yield}$ of the N-rich plot and $\text{Yield}_{\text{Check}} = \text{Yield}$ NDVI of the control plot, and

$$RI_{NDVI} = \frac{NDVI_{N-Rich}}{NDVI_{Check}}$$
(3)

where $NDVI_{N-Rich}$ = average NDVI of the N-rich plot and $NDVI_{Check}$ = average NDVI of the control plot.

Several equations were evaluated to improve the ability of the RI_{NDVI} to predict final yield with inclusion of CVs. The data were subjected to statistical analysis using SAS (SAS Institute, 2002). Simple regression was the primary form of trend analysis for both measured and calculated response variables, however, Cate-Nelson, linear-linear, and linear-plateau models were also investigated.

RESULTS

Using the Cate-Nelson model across all site-year combinations, a critical CV range of 17–20 was determined (Figure 1). Figure 2 illustrates the change in CV of treatments over time, from January until physiological maturity; this trend was similar at all three locations in the 2004 crop year. The maximum CV



Figure 1. Relationship between the CV of NDVI readings and winter wheat plant population (seven locations, 2003–2004, multiple seeding rates, and six varieties). The critical CV range of 17–20, identified as the region between the two vertical lines, was determined using the Cate-Nelson model.

occurred near Feekes 6 (stem elongation) growth stage at all locations. The CV was also affected by N treatments, but the trend of CV over time was generally the same across treatments (Figure 2).

The linear relationship ($R^2 = 0.17$) between $RI_{Harvest}$ and RI_{NDVI} over all three locations is shown in Figure 3. When these data were separated by CV



Figure 2. Change in mean CV, from NDVI readings collected from three N treatments within the row in winter wheat, over time at the EFAW Research Farm, Stillwater, OK (2004).

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Figure 3. Comparison of $RI_{Harvest}$ (yield of N-rich plot/yield of control) versus RI_{NDVI} (NDVI of the N-rich plot/NDVI of the control) for all CV data.

values, it can be seen that with increasing CV there was an improvement in the relationship between $RI_{Harvest}$ and RI_{NDVI} . When the CV of the fertilized plot was 10 or less, there was no linear relationship between $RI_{Harvest}$ and RI_{NDVI} ($R^2 = 0.002$) (Figure 4). Figure 5 shows the linear relationship of the two RIs



Figure 4. Comparison of $RI_{Harvest}$ (yield of N-rich plot/yield of control) versus RI_{NDVI} (NDVI of the N-rich plot/NDVI of the control) when the CV of the fertilized plot was ≤ 10 .

1990



Figure 5. Comparison of RI_{Harvest} (yield of N-rich plot/yield of control) versus RI_{NDVI} (NDVI of the N-rich plot/NDVI of the control) when the CV of the fertilized plot was $10 < and \le 25$.

when the CV was between 10 and 25 ($R^2 = .19$). Figure 6 shows the relationship of the two when the CV was greater than 25 ($R^2 = .30$).

Incorporating the CV into the calculation of RI as $RI_{NDVI-CV}$ significantly improved the relationship with $RI_{Harvest}$ ($R^2 = 0.38$) (Figure 7) when compared



Figure 6. Comparison of $RI_{Harvest}$ (yield of N-rich plot/yield of control) versus RI_{NDVI} (NDVI of the N-rich plot/NDVI of the control) when the CV of the fertilized plot was >25.



Figure 7. Comparison of $RI_{Harvest}$ (yield of N-rich plot/yield of control) versus $RI_{NDVI-CV}$ {(NDVI of the N-rich plot/NDVI of the control) × (max CV – control CV/max CV – critical CV)}.

with that of RI_{NDVI} and RI_{Harvest}. The best equation was:

$$RI_{NDVI-CV} = \frac{NDVI_{N-Rich}}{NDVI_{Check}} \times \frac{CV_{Max} - CV_{Check}}{CV_{Max} - CV_{Critical}}$$
(4)

where NDVI_{N-Rich} and NDVI_{Check} are as defined as in Equation 3: CV_{Max} = maximum coefficient of variation, CV_{Check} = coefficient of variation of NDVI readings taken from the control plot, and $CV_{Critical}$ = critical coefficient of variation.

The CV_{Max} , which is the highest observed CV, used in the calculation was 45. A $CV_{Critical}$ of 17, derived from Figure 1, was employed. These values, CV_{Max} and $CV_{Critical}$, were expected to change in different production environments and for different crops.

The regression analysis of the relationship between average NDVI readings collected in the direction of the seed row versus those collected from the same area moving perpendicular to the seed row is reported in Figure 8 for EFAW, Lake Carl Blackwell, Perkins, and Hennessey in 2003 and 2004. The trend line fits a linear relationship, with a slope not significantly different from 1.0 and an intercept not significantly different from 0, with an R² of 0.97. Three thousand six hundred sixty observations, compiled from two years of readings over seven locations, were used in the regression. A highly significant linear relationship (R² = 0.97) was also found between NDVI readings collected in the direction of the seed row and those taken from the same area moving at a 45-degree angle across the seed rows.

The relationship between the CV of NDVI readings taken with the seed row and perpendicular to the seed row was determined to be quadratic (R^2 of 0.78) (Figure 9). At high CV, where plant stand was poorest, the values of the CVs collected with the row and perpendicular to the row tended to converge.

1992



Figure 8. Comparison of with-seed row NDVI readings versus across-seed row NDVI readings (seven locations, 2003–2004, multiple seeding rates, and six varieties, n = 3660).

DISCUSSION

In this study, seven site years were used to evaluate the plant population's effect on CVs of NDVI readings. No one site year could be used to identify a critical CV because there was little difference in population at any one site. The critical CV value determined from this study corresponds with the results presented by Morris et al. (2005), who observed that plots of winter wheat with a CV



Figure 9. Relationship between with-seed row CV of NDVI readings and across-seed row CV of NDVI readings.

greater than 18 were unable to recover completely from early-season N stress. This result clearly indicated that in areas of a field where CV exceeded the critical level of approximately 20, it was recognized that the crop would not be able to utilize the additional N. This finding has potential use in variable-rate application, where the amount of N fertilizer applied could be reduced.

Raun et al. (2005b) observed a peak in CV in corn at the V6 stage and inferred that the peak could represent the best time to apply in-season foliar N fertilizer, as this was the time when spatial variability of NDVI values was greatest. Similar results were found in this study. The first peak in CV occurred near the Feekes 6 growth stage. This result coincides with the time when spatial variability was greatest. This finding in turn suggests the time when variable-rate technology could have the greatest benefit—at least for detection of yield potential. It is necessary to apply topdress N prior to Feekes 6, because it is much easier to damage the crop with applicator traffic soon after stem elongation. In Oklahoma, topdress-N application timing is commonly determined by weather and field conditions. Often, the topdress-N application is made from December through March, typically well before the crop reaches Feekes 6.

In this small plot experiment, the linear relationship between the $RI_{Harvest}$ and the RI_{NDVI} was found to be poor. (Figure 3). This result suggests that at this scale, we were not able reliably to predict yield response to added N midseason. Hodgen et al. (2005) found that the relationship between $RI_{Harvest}$ and RI_{NDVI} was strong, with a R^2 of 0.75. In their study, location averages were used to determine the relationship, while this study used each individual plot.

The linear relationship between vegetative RI and harvest RI was improved with increasing CV (Figures 4–6). The $RI_{Harvest}$ and RI_{NDVI} showed the best linear relationship when the CV of the fertilized plot was greater than 25. A linear relationship was still seen with CVs of between 10 and 25, but at a lower significance level. When the CV was less than 10, there was no relationship between $RI_{Harvest}$ and RI_{NDVI} .

The data indicate that when the CV is at high levels, the calculation for RI_{NDVI} is appropriate. However, at a CV of less than 10, RI_{NDVI} was unable to predict $RI_{Harvest}$. In light of this limitation, $RI_{NDVI-CV}$ was introduced to incorporate CV into the RI equation. The $RI_{NDVI-CV}$, which is a derivative of RI_{NDVI} that includes the CV of the control plot, has a much better relationship with $RI_{Harvest}$ ($R^2 = 0.37$) (Figure 7). This improvement of more than 50% indicated the ability of CV to identify the reduced yield potential in those plots with poor stands. An improvement in RI estimation should improve variable-rate N application and NUE.

Figure 8 demonstrates the linearity in the relationship of NDVI readings as a direction of travel over the seed row. This issue becomes important when considering that fertilizer applicators will be outfitted with the sensors. Unlike in research where sensor direction can be carefully adjusted, the direction of the movement of sensors on an applicator is much more difficult to control when studying winter wheat. Figure 8 shows a strong significant linear relationship with a slope of 1 and an intercept of 0. These results indicate that NDVI readings were independent of direction of travel. Determining of the relationship between with-seed row CV and perpendicular-to-seed row CV will also make it possible incorporate to the use of CV into variable-rate N application.

CONCLUSIONS

It was observed that when CVs were greater than approximately 20, the plant population was poor, with <100 plants/m². The ability of the crop to respond to added N was evaluated using several response indices ($RI_{Harvest}$, RI_{NDVI} , and $RI_{NDVI-CV}$). It was found that $RI_{NDVI-CV}$ equation 4 provided improved prediction of $RI_{Harvest}$ compared with to the more conventional RI_{NDVI} equation 3. It was suggested that when this equation is implemented into the algorithm, variable-rate applicators will apply less N over areas that have CVs greater than 20. The reduction in N applied reduces the expense to farmers and the risk of N being lost to the environment.

The observation that CV reached a peak at Feekes 5–6 suggests that current timing of application may have to be changed in order to maximize the efficiency of the technology. As to application direction, it was beneficial to see that it did not matter in which direction the sensors were traveling across the seed row, because the NDVI values would remain the same. This finding was extremely important in that it suggests the applicators need not follow any rigid guidelines for the equipment to perform properly.

Integrating CV into N-fertilization algorithms will be more challenging with the discovery that across the seed row, CV was consistently higher than in other directions. The CVs can be used as an estimate of variation in plantstand densities by identifying the areas where the plant stand is so poor that N application would be unnecessary. The use of CV from NDVI readings could improve upon the efficiency with which variable topdress N is applied.

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