**Project Title:** Use of Chlorophyll Meters to Assess Nitrogen Fertilization Requirements for Optimum Wheat Grain and Silage Yield and Quality

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## Abstract

Nitrogen fertilizer is the most used and often the most mismanaged nutrient input. Nitrogen management has tremendous implications on crop productivity, quality and environmental stewardship. Sufficient nitrogen is needed to optimum yield and quality. Soil and in-season plant tissue testing for nitrogen status are a time consuming and expensive process. Real time sensing of plant nitrogen status can be a useful tool in managing nitrogen inputs. The objectives of this project were to assess the reliability of remotely sensed non-destructive plant nitrogen measurements compared to wet chemistry data from sampled plant tissue, develop in-season nitrogen recommendations based on remotely sensed data for improved nitrogen use efficiency and assess the potential for determining yield and quality from remotely sensed data. Very good correlations were observed between early-season remotely sensed crop nitrogen status and nitrogen concentrations and subsequent fertilizer recommendations. The SPAD meter gave the most accurate readings. Early season fertilizer recommendation would be to apply 35 lbs N/A plus 12 lbs N/A for each unit difference measured between the crop and reference area. Once the crop was sufficiently fertilized meter readings became inconclusive and were of no benefit for determining nitrogen status, silage yield and protein and grain yield and protein.

## Introduction and Objectives

The southern San Joaquin Valley in 2009 produced 3.4 million tons of wheat silage valued at 76.5 million dollars on 212,000 acres and 389,800 tons of grain valued at 95.1 million dollars on 153,500 acres. Nitrogen requirements for wheat production are well established. The nitrogen requirement can be accurately determining by knowing the available soil nitrogen and the amount of added nitrogen. Much of the wheat silage acreage is fertilized with manure and irrigated with lagoon water. However, an accurate and thorough measurement of nitrogen levels in manure and lagoon water is rarely conducted. The over application of nitrogen has the potential to dramatically impact ground water through leaching and surface water from runoff. The quality of wheat silage, as determined by nutritional value either as energy or protein percent decreases as the plant develops. For optimum nutrition, it is recommended that wheat silage be harvested between the boot and early heading. This timing however, does not produce the most tonnage nor the most energy or protein per acre. For optimum grain production, it is recommended that split nitrogen applications be made with a majority of the nitrogen applied prior to heading. Nitrogen applications after heading may improve grain protein to meet acceptable protein levels. The use of remote sensing to determine nitrogen status in the plant is a quick method for determining if any additional nitrogen is required to produce optimum yield and quality.

Petegrove, et al. found that fifty percent of the variability in grain protein could be accounted for by flag leaf nitrogen content using transmittance/absorbance measurements made at Feekes 10.5. Murdock, et al. had correlation values between 0.88 and 0.95 for Feekes 6 meter reading and yield for both reflectance and transmittance/absorbance measurement methods. Wright, et al. overall had lower correlation ($R^2$) values with hand held meters than Murdock, et al. but they were higher than those from satellite imagery. Li, et al. observed nitrogen use efficiencies of 61.3, 51.0 and 13.1 % using sensor-based, soil minimum nitrogen management and traditional farmer practices, respectively. In an economic analysis, Biemacher, et al. determined that plant-sensing systems have the potential to increase profitability.

## Materials and Methods

Plots with various preplant nitrogen application rates were established at multiple locations across the southern San Joaquin Valley.

The primary sites were at the UCCE Kern Research Farm and UC Westside REC. A randomized complete block factorial design with three replications was used. These locations provided low initial nitrogen plot areas. Plots were 5 feet by 25 feet. Irrigation was sufficient to not be a limiting factor. Treatments at these locations included nitrogen applications of 0, 100, 200, and 300 lbs. nitrogen per acre applied at planting and at growth stage Feekes 5 nitrogen was applied so that each plot had received a total of 300 lbs N/acre. Additional plots at WSREC had 100 lbs N/A applied at planting and 0, 50, 100 and 150 lbs N/A applied in the spring prior to prior to Feekes 3 and nitrogen fertilizer applied at Feekes 8 so each plot received a total of 300 lbs N/A. Soil nitrogen level was tested before planting and after harvest. Plant nitrogen status was tested at Feekes 3, 6 and 8 and 10 (tillering through flag leaf extension). Plant nitrogen measurements were made by reflectance, transmittance/absorbance, and wet chemistry. One half of each plot was harvested for silage and the other half for grain. Each was sampled for nitrogen concentration at Dellavalle Laboratory, Inc. or Dairyland Lab, Inc.

Four sites were located on farmer fields harvested for grain. Nitrogen application rates at 2 fields were 125 lbs N/A below and above the farmer’s nitrogen application rate of 321 lbs N/A and the other was 100 lbs N/A above and below the farmer’s application rate of 225 lbs N/A. Two locations included an area that did not receive any preplant nitrogen, thus N rates were 220 or 125 lbs./A below and 125 lbs N/A above the farmer’s rate. These sites did not have equally applied nitrogen across the treatments as did the plots at West Side REC and Kern Research Farm. Plot areas were sampled at the same growth stages as previously described.

The two products used to remotely sense plant nitrogen content use either reflectance or light transmittance/absorbance. The reflectance method uses ambient and reflected light in the 660 and 840 nm wavelengths to calculate a relative chlorophyll index. This instrument is the Spectrum® FieldScout® CM 1000 NDVI Meter. The hand held device can measure areas from 1.5 inch to 4.5 inch diameter. This is the same methodology that is incorporated in aerial or satellite imagery. “Normalized difference vegetation index” or NDVI measurements were made with the instrument about 2 feet above the crop canopy with a 45 or 90 degree angle to the canopy. Measurements from reflected light are abbreviated CM 1000 45 or CM 1000 90 for the different angles.

The transmittance/absorbance instrument is a Konica Minolta SPAD 502 Plus, abbreviated SPAD for “Special Products Analysis Division.” The SPAD meter is clamped on a leaf and utilizes the 650 and 940 nm wavelengths to determine a relative chlorophyll index. Measurements were made at different locations on the plant leaf to determine the most representative spot. The CM 1000 NDVI meter displays the NDVI calculation (-1.0 to 1.0) whereas the SPAD meter readings are a relative index (-9.99 to 199.9) calculated from NDVI times a constant.

## Results

Good correlations ($R^2$>0.75) were observed between meter readings from both instruments and V5 nitrogen concentration (Figures 1 & 2). There were some differences between varieties at the different locations. The difference between the meter reading of the well fertilized treatment and the other treatments was calculated. Those differences (Table 3) had a good correlation for the CM 1000 45 ($R^2$=0.76). The CM 1000 90 correlation was not as good ($R^2$=0.59). Very good correlation was observed ($R^2$=0.85) for the SPAD meter readings (Figure 4).

No differences were observed for readings made at Feekes 8-9 (Tables 1, 2 & 4). Flag leaf chlorophyll meter readings were not well correlated to flag leaf nitrogen concentration for either measurement method (Figures 5 & 6). To achieve high yields and acceptable grain protein content, plant uptake of nitrogen exceeds the nitrogen concentration needed for maximum chlorophyll production.

There was no difference in yield or grain protein for fall fertilized or spring split applications at West Side REC (Tables 1 & 3). No differences in yield were observed for any of the treatments that received equal nitrogen fertilizer amounts (Tables 3 & 5). In the two farmer fields where no preplant nitrogen was added, yields were lower for those treatments (Table 6). Where a significant amount of nitrogen fertilizer was not applied preplant and additional nitrogen was not applied during the season flag leaf nitrogen was lower and was reflected in chlorophyll measurements. A lower preplant N fertilizer omission in farmer location #4 (Table 7) did not have as dramatic an effect on chlorophyll measurements although significantly different. Grain yield was lower for the no preplant fertilizer treatment.

There were no differences in SPAD measurements sampling the upper or lower leaf surface and no interaction with different nitrogen concentrations (Table 8). There was a significant difference when measurements were made along the leaf. Relative chlorophyll amounts increased as measurements were made from the leaf base to the leaf tip.

## Discussion, Conclusions and Recommendations

Early spring sampling of wheat plants can provide useful information on plant nitrogen status and the need for additional nitrogen fertilizer. The use of chlorophyll meters provides quick and accurate information needed for nitrogen fertilizer recommendations.

Grain yields were equivalent for all locations where total nitrogen applied was the same. The total nitrogen applied was greater than the typical amount (50 to 100 lbs N/A depending on yield potential). There was not a decline in yield for over fertilization that can occasionally occur. Where irrigation is correctly managed or winter rains do not leach fall applied nitrogen fertilizer there is no difference in grain yield based on timing of fertilizer application. The results may be different for lighter textured soils or where nitrogen fertilizer rate is not an over application. The spring fertilizer plots at the Kern Research Farm, which is a lighter textured soil, had a misapplication in the spring nitrogen fertilizer plots thus rendering that part of the experiment useless.

SPAD meter measurements should be made mid leaf on the upper most fully exposed leaf for greatest consistency and accuracy. Plants and leaves that are not representative of the field, under stress or insect damaged should not be used. Following recommendations from other research, CM 1000 measurements were made between 10:00 am and 2:00 pm and without shadows on the crop or meter for maximum ambient light. CM 1000 measurements made early in the season should be made with the instrument at a 45 degree angle from the crop. Too much bare soil can be included in the measurements made at a 90 degree angle early in the season thereby making those measurements less reliable. The 90 degree angle CM 1000 late-season measurements were more precise than the 45 degree angle measurements.

Early season nitrogen fertilizer recommendation is as follows:

Apply the expected full nitrogen fertilizer rate on a reference area at least three weeks prior to sampling with actively growing plants. The reference area should be representative of the field and can be several small areas throughout the field or a strip through the field. At Feekes 5 to 6, compare the readings from the reference areas to readings from the remainder of the field. Because individual plants vary, at least 30 readings should be made throughout the field and reference area. The difference between the averages of the readings will give an indication of the need for additional nitrogen fertilizer.

The nitrogen rate calculation is:

$$N = 35 + 12D$$

N = Recommended Nitrogen Rate in lbs N/A

D = Difference in SPAD meter reading between measured crop and reference area

or

$$N = 70 + 1660D$$

N = Recommended Nitrogen Rate in lbs N/A

D = Difference in CM 1000 NDVI meter reading between measured crop and reference area

Future experimentation should have smaller increments in nitrogen fertilizer applications to further refine the nitrogen fertilizer recommendation equations. Additional plots that are not fully fertilized should also be included to help determine optimum nitrogen fertilizer requirements and yield potential for the different soils and sites. The sites selected were a good representation of a range of soil types and yield potential. Another method to be investigated would be smaller more frequent nitrogen fertilizer applications. This would potentially be more adaptable to lighter textured soils than heavier textured soils because irrigations are more frequent and the potential for ground water contamination is greater.

**References**

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Figure 1. V5 Tissue Nitrogen Concentration versus NDVI Reading.

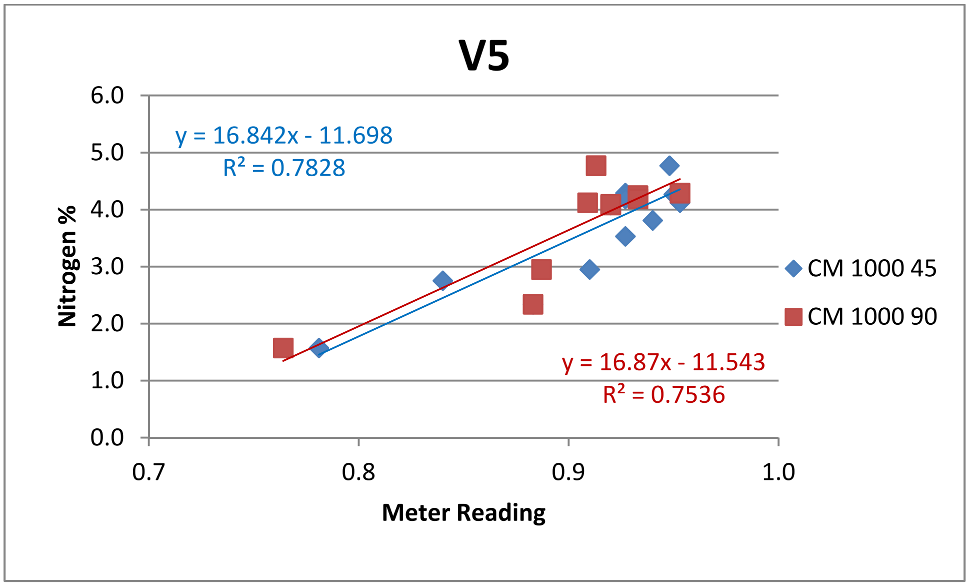


Figure 2. V5 Tissue Nitrogen Concentration versus SPAD Reading.

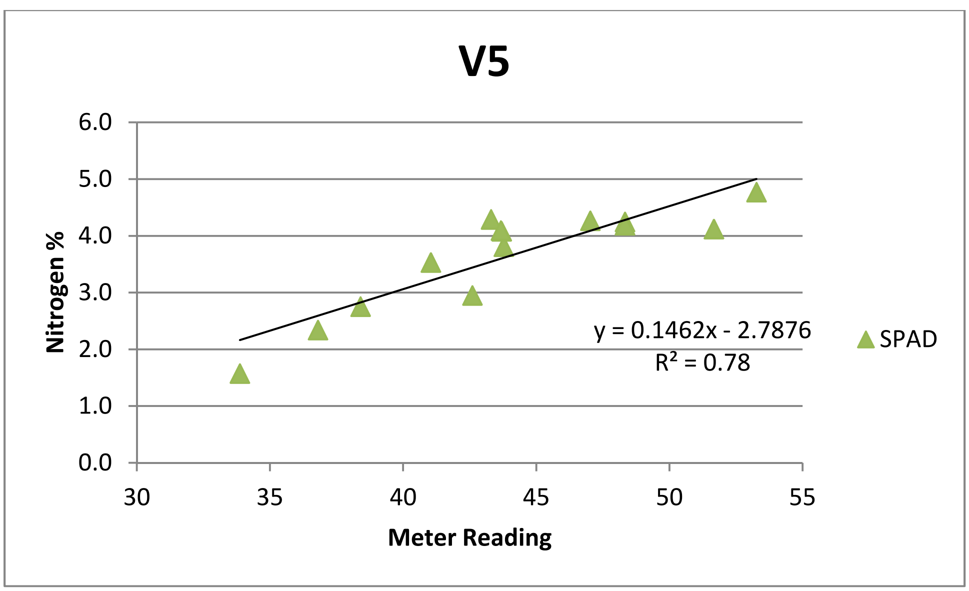

Figure 3. Recommended Nitrogen Rate versus NDVI Differential.

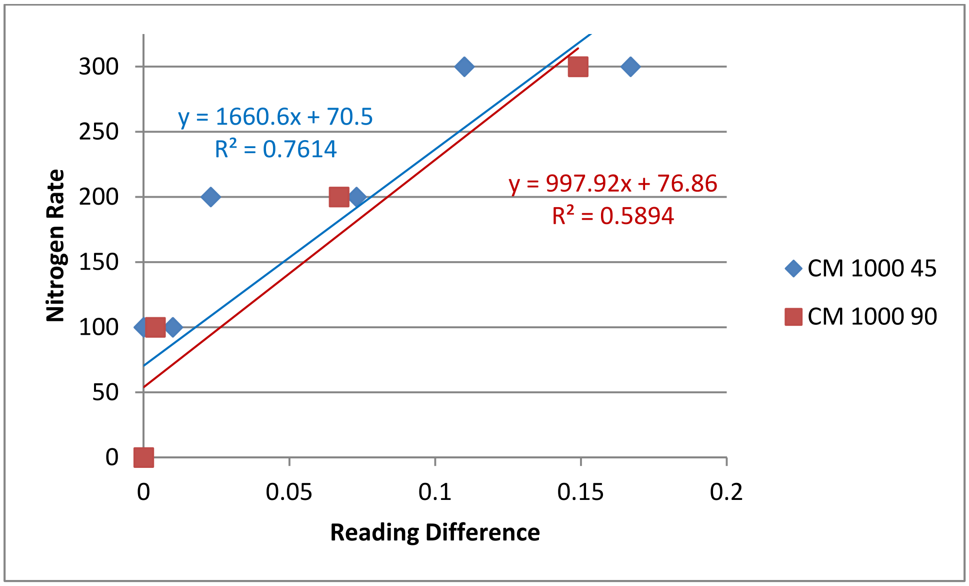


Figure 4. Recommended Nitrogen Rate versus SPAD Differential.

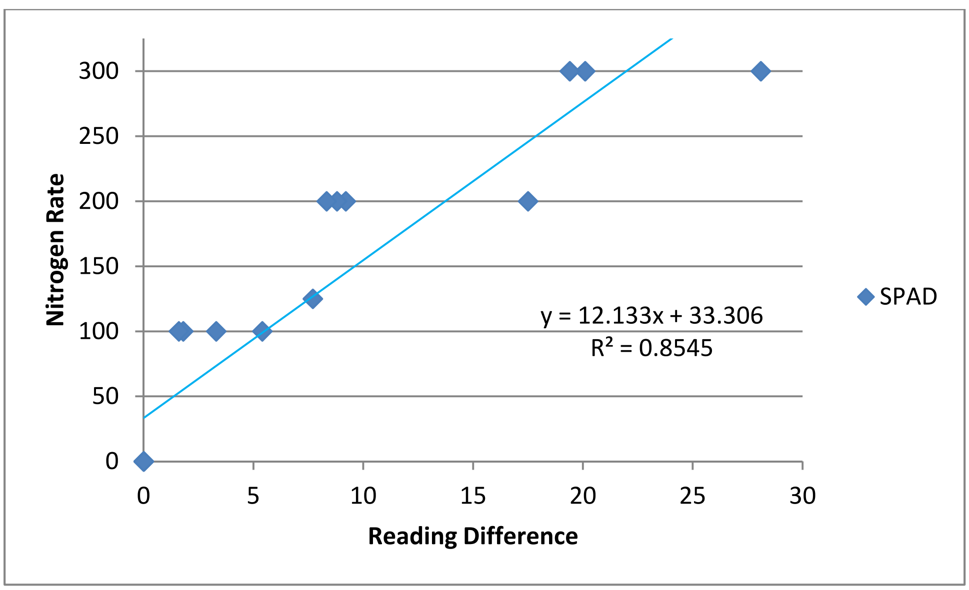

Figure 5. Flag Leaf Nitrogen Concentration versus NDVI reading.

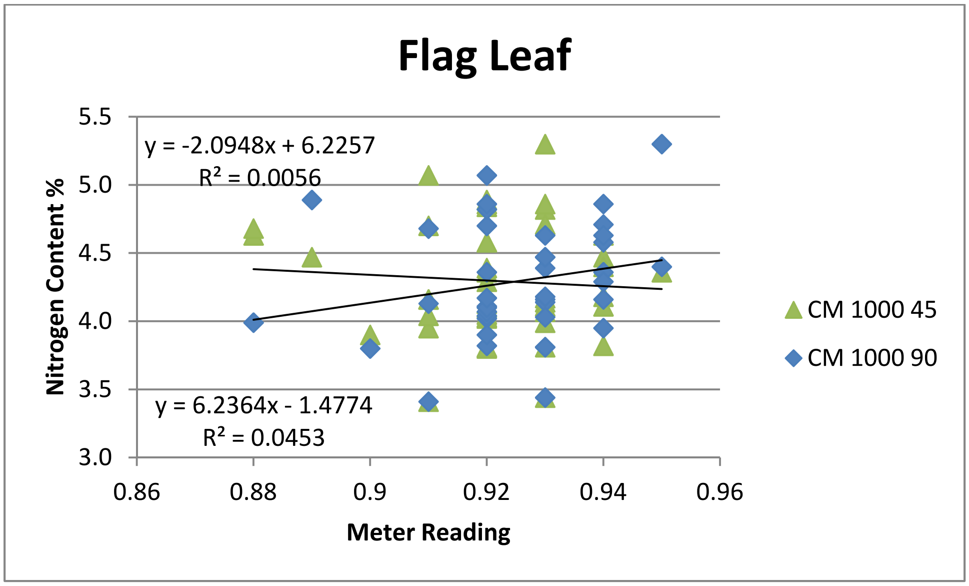


Figure 6. Flag Leaf Nitrogen Concentration versus SPAD reading.

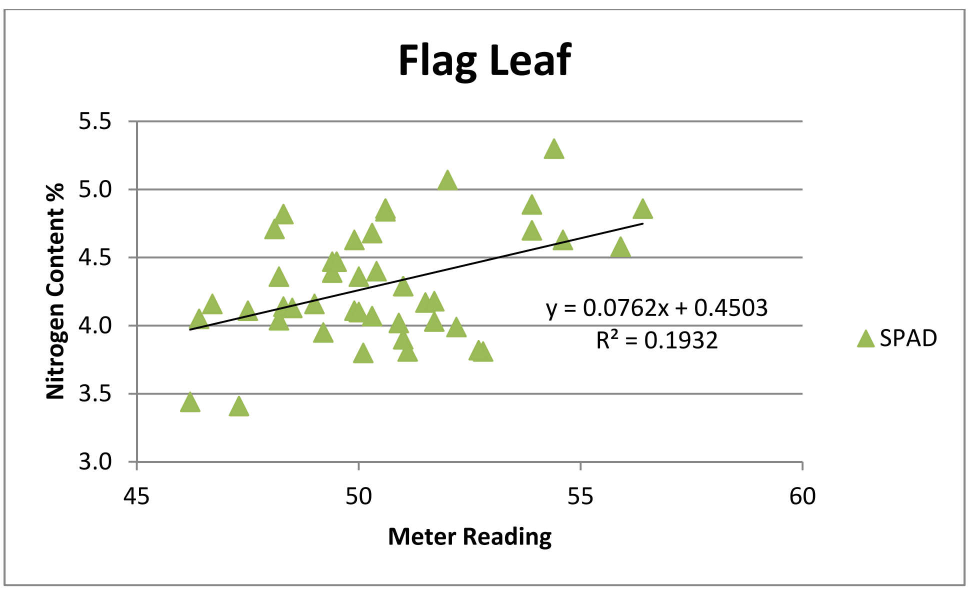

Table 1. West Side REC Spring Nitrogen Applications (100 lbs N/A applied at planting)

| | | 4/13 | | | | | |
|---|---|---|---|---|---|---|---|
| lbs N at Fekes 5 | lbs N at Fekes 8 | CM 1000 45† | CM 1000 90 | SPAD | N content | Grain Yield | Grain Protein |
| | | | | | % | lbs/A | % |
| 0 | 200 | 0.937 a | 0.937 | 48.7 | 4.10 | 6660 | 12.0 |
| 50 | 150 | 0.917 b | 0.917 | 49.1 | 3.96 | 6480 | 12.4 |
| 100 | 100 | 0.923 ab | 0.923 | 49.3 | 4.03 | 7010 | 12.3 |
| 150 | 50 | 0.930 ab | 0.913 | 52.3 | 4.24 | 7010 | 12.5 |
| $LSD_{0.05}$‡ | | 0.0199 | ns†† | ns | ns | ns | ns |
| CV%‡‡ | | 0.51 | 2.04 | 3.37 | 9.14 | 11.9 | 6.2 |

†Numbers followed by the same letter are not significantly different.
‡Least Significant Difference.
††Not Significantly Different.
‡‡Coefficient of Variation.

Table 2. West Side REC Fall Nitrogen Applications

| | | 4/13 | | | |
|---|---|---|---|---|---|
| lbs N at planting | lbs N at Fekes 5 | CM 1000 45 | CM 1000 90 | SPAD | N content |
| | | | | | % |
| 0 | 300 | 0.923 | 0.930 | 50.8 | 3.94 |
| 100 | 200 | 0.920 | 0.920 | 51.5 | 3.13 |
| 200 | 100 | 0.937 | 0.930 | 49.2 | 4.29 |
| 300 | 0 | 0.913 | 0.917 | 48.3 | 4.00 |
| $LSD_{0.05}$ | | ns | ns | ns | ns |
| CV% | | 1.76 | 0.83 | 2.84 | 3.76 |

Table 3. West Side REC Fall Nitrogen Applications

| lbs N at planting | lbs N at Fekes 5 | Silage Yield | Silage Protein | Grain Yield | Grain Protein |
|---|---|---|---|---|---|
| | | Tons/A | % | lbs/A | % |
| 0 | 300 | 10.5 | 8.73 | 6810 | 11.2 b |
| 100 | 200 | 9.3 | 9.80 | 6850 | 12.8 a |
| 200 | 100 | 10.2 | 10.56 | 7050 | 12.6 a |
| 300 | 0 | 10.2 | 9.07 | 6810 | 12.3 ab |
| $LSD_{0.05}$ | | ns | ns | ns | 1.1 |
| CV% | | 11.5 | 22.3 | 10.9 | 4.6 |

Table 4. Kern Research Farm Fall Nitrogen Applications

| | | 3/30 | | | |
|---|---|---|---|---|---|
| lbs N at planting | lbs N at Fekes 5 | CM 1000 45 | CM 1000 90 | SPAD | N content |
| | | | | | % |
| 0 | 300 | 0.92 | 0.93 | 54.3 | 3.81 |
| 100 | 200 | 0.91 | 0.93 | 52.2 | 3.73 |
| 200 | 100 | 0.91 | 0.94 | 51.4 | 4.05 |
| 300 | 0 | 0.92 | 0.93 | 50.3 | 3.63 |
| $LSD_{0.05}$ | | ns | ns | ns | ns |
| CV% | | 1.78 | 0.69 | 4.39 | 8.52 |

Table 5. Kern Research Farm Fall Nitrogen Applications

| lbs N at planting | lbs N at Fekes 5 | Silage Yield | Silage Protein | Grain Yield | Grain Protein |
|---|---|---|---|---|---|
| | | Tons/A | % | lbs/A | % |
| 0 | 300 | 9.88 | 10.4 | 5630 | 15.5 |
| 100 | 200 | 10.98 | 8.3 | 5970 | 15.6 |
| 200 | 100 | 11.19 | 8.8 | 5820 | 14.6 |
| 300 | 0 | 11.00 | 8.8 | 6270 | 12.9 |
| $LSD_{0.05}$ | | ns | ns | ns | 1.4 |
| CV% | | 8.9 | 8.3 | 7.2 | 4.8 |

Table 6. Farmer location #1.

| | | Flag Leaf | | | | |
|---|---|---|---|---|---|---|
| lbs N at planting | N Total | CM 1000 45 | CM 1000 90 | SPAD | Nitrogen Content | Grain Yield |
| | | | | | % | lbs/A |
| 0 | 80 | 0.88 | 0.87 b | 41.0 b | 3.27 b | 6910 b |
| 220 | 300 | 0.90 | 0.92 a | 47.2 a | 3.85 ab | 8455 a |
| 345 | 425 | 0.90 | 0.92 a | 47.9 a | 4.30 a | 8080 a |
| $LSD_{0.05}$ | | ns | 0.02 | 4.33 | 0.63 | 9.0 |
| CV% | | 8.9 | 1.6 | 4.2 | 7.4 | 3.7 |

Table 7. Farmer location #4.

| lbs N at planting | N Total | Flag Leaf CM 1000 45 | Flag Leaf CM 1000 90 | Flag Leaf SPAD | Flag Leaf Nitrogen Content | Grain Yield |
|---|---|---|---|---|---|---|
| | | | | | % | lbs/A |
| 0 | 100 | 0.92 | 0.90 b | 47.9 ab | 4.29 | 5090 |
| 125 | 225 | 0.88 | 0.90 b | 45.7 b | 4.31 | 6420 |
| 250 | 350 | 0.91 | 0.93 a | 49.4 a | 4.32 | 6480 |
| $LSD_{0.05}$ | | ns | 0.02 | 2.4 | ns | 16.2† |
| CV% | | 2.3 | 1.0 | 2.2 | 2.7 | 10.2 |

†$LSD_{0.10}$

Table 8. SPAD meter position measurements.

| Leaf Position | Nitrogen Concentration | Meter Reading | | Meter Reading |
|---|---|---|---|---|
| Upper Surface | | 41.0 | | |
| Lower Surface | | 41.6 | | |
| $LSD_{0.05}$ | | ns | | |
| | Low N | 26.7 c | | |
| | Medium N | 46.9 b | | |
| | High N | 50.4 a | | |
| $LSD_{0.05}$ | | 2.5 | | |
| Upper Surface | Low N | 25.9 | | |
| | Medium N | 47.5 | | |
| | High N | 49.7 | | |
| Lower Surface | Low N | 27.4 | Near Base | 39.6 a |
| | Medium N | 46.3 | Mid Leaf | 41.7 ab |
| | High N | 51.1 | Near Tip | 45.3 a |
| $LSD_{0.05}$ | | ns | | 4.3† |
| CV% | | 6.5 | | 7.5 |

†$LSD_{0.10}$