Evaluation of Fall and Spring Nitrogen Rates Effect on Cereal Rye Forage Crude Protein and Tillering Using NDVI and Canopeo to Make Infield Nitrogen Rate Decision

Kendra Stahl[1], Jason Hartschuh[2], and Alan Gahler[3]
1 Ohio State University Extension Crawford County – 808 Whetstone St Bucyrus Ohio 44820
2 Ohio State University Extension North Central Research Station – 1165 Gonawein Rd Fremont Ohio 43420
3 Ohio State University Extension Sandusky County – 2000 Countryside Dr, Suite D Fremont Ohio 43420

A paper from the Proceedings of the
16th International Conference on Precision Agriculture
21-24 July 2024
Manhattan, Kansas, United States

Abstract.

Fall applied nitrogen has been used to increase plant tiller and protein in wheat but less research has been done of its effects on cereal rye forage and how NDVI and Canopeo readings can be used to make nitrogen application management decisions. This study took place at the Ohio State University North Central Agricultural Research Station in Fremont, Ohio. The experiment is a randomized complete block split-plot design with four nitrogen rates in the fall (0, 30, 60, and 90 lbs/ac) and in the spring an additional four nitrogen rates over the fall rates of 25, 50, 75, and 100 pounds per acre in rye. Rye was planted in October and harvested in May. NDVI readings were taken in late November before fall dormancy and in the spring after greenup to determine the greenness and density of green matter in an area. Canopeo readings were taken at the same time as NDVI readings to determine the percent of live green vegetation in an area. Fall nitrogen rates increased tillering and have significantly increased spring yield. The results of the NDVI and Canopeo readings were evaluated together to determine if fall applied N increases the tillering density and protein in rye. We are investigating if spring NDVI readings can be used to assist with making spring nitrogen application management decisions.

Keywords.
Nitrogen, NDVI, Canopeo, Cereal Rye, spring, fall

The authors are solely responsible for the content of this paper, which is not a refereed publication. Citation of this work should state that it is from the Proceedings of the 16th International Conference on Precision Agriculture. EXAMPLE: Last Name, A. B. & Coauthor, C. D. (2024). Title of paper. In Proceedings of the 16th International Conference on Precision Agriculture (unpaginated, online). Monticello, IL: International Society of Precision Agriculture.

Introduction

Quantifying cover crop forage nitrogen (N) content in the spring has traditionally been done by cutting a biomass sample from a known area, drying it, weighing it, and submitting the dried tissue to a lab to measure the N concentration, which is an accurate method, but is tedious, time consuming, and expensive (White, 2023). There has been development of mobile applications and handheld devices to measure the amount of ground cover or the amount of biomass in a field. The mobile apps are powerful tools which allow producers, crop consultants, researchers, and other end users to easily acquire, process, and annotate digital images in the field to obtain real-time, geo-referenced green canopy cover estimates (Patrigtnani & Ochsner, 2015). While there are multiple different software's out there that can measure canopy cover, we used Canopeo and Normalized Difference Vegetation Index (NDVI) to find a difference in between fall N treatments and spring N applications effect on yield. An NDVI sensor is a quick way to non-destructively estimate cover crop biomass N content (White, 2023). NDVI has been found to be equal or superior to other indices in predicting percent groundcover (Prabhakara et al., 2015). Canopeo is an automatic color threshold (ACT) image analysis tool. One known limitation of Canopeo is the need to keep the camera an adequate height above the canopy (Patrigtnani & Ochsner, 2015). Fall applied nitrogen has been used to increase plant tiller and protein in wheat but less research has been done of its effects on cereal rye forage and how NDVI and Canopeo readings can be used to make nitrogen application management decisions the follow spring. Altom et al. (1996) has found that spring forage yields contributed more towards total forage dry matter production than did fall-winter forage production. Nitrogen applications can also affect the nutrient and protein levels in a forage. Lyons et al. (2019) reported that spring N applications impacted crude protein (CP) concentration in 91% of their trials. Total forage yields have been found to be higher for fall applications compared to spring, especially at higher rates (150 and 200 lb/ac), but spring forage yields were found to contribute more total dry matter production compared to fall-winter production (Altom et al., 1996). The objectives of this experiment were (i) investigate if spring NDVI readings can be used to assist with making spring N application management decisions, and (ii) determine if fall applied N increases tillering density and protein in cereal rye. One hypothesis for this study is that fall N rates will increase tillering and significantly increase spring yield.

Methods and Materials

This study took place at the Ohio State University North Central Agricultural Research Station in Fremont, Ohio (41°18'43.4"N 83°10'15.4"W). The soil type at this location is a loamy fine sand (loamy, mixed, mesic Aquic Arenic Hapludalfs). The experiment is a randomized complete block split-plot design with four nitrogen rates in the fall before planting (0, 30, 60, and 90 pounds/acre (lbs/ac)) and in the spring before jointing an additional four nitrogen rates of 25, 50, 75, and 100 lbs/ac over the fall rates in cereal rye *(Secale cereale*). Nitrogen was applied as Urea. The cereal rye was planted in October 2023 and harvested in May 2024. Plots were harvested using a RCI 36A plot harvester harvesting a 91.44cm wide by 762cm area from the center of a 304cm wide plot. After harvest the samples were weighted, dried for two days, weighed again, and then sent off to Dairyland Laboratories Inc. (Battle Creek Michigan) to be analyzed.

NDVI

The NDVI scale runs from 0 to 1, with values closer to 1 indicating greater absorption of red light by the chlorophyll in plant leaves (White, 2023), meaning the closer to 1 the number is the more ground cover is in the field, and the higher the nitrogen content. A Greenseeker handheld sensor was walked across each plot held out from the person to make sure there was no possible contamination of data. NDVI readings were taken in late November before fall dormancy and in March after greenup to determine the greenness and density of green matter in an area. During analysis results of each plot within a treatment block was averaged before being analyzed statistically.

Canopeo

Canopeo is developed by Oklahoma State University (version 2.0, Stillwater, OK). This application uses color values in the red–green–blue (RGB) system to determine ground cover. The Canopeo app can be downloaded by going to https://canopeoapp.com/#/login. Canopeo uses a range of 0-100 as a percentage. The larger the number in this scale, the more ground cover there is. Canopeo readings were taken at the same time as NDVI readings to determine the percent of live green vegetation in an area.

Statistical Analysis

The data was analyzed in RStudio using packages agricolae (De Mendiburu, 2023) and ggplots2 (version 4.3.3, Rstudio Team, 2020). Forage plot yields and quality were analyzed using an ANOVA with Treatment, Snitrogen, Fnitrogen, the interaction between the treatments, and block. An LSD mean separation was used to determine if the significant difference between treatments. A correlation between NDVI, Canopeo, yield and the fall N rates was analyzed to observe if there was a positive or negative correlation between the measurements.

Results and Discussion

This study will specifically be focusing upon the Dry Matter per ton (DM_TONS), crude protein (CP), and total digestible nutrients (TDN). We looked at how these factors were affected by fall applied N (Fnitrogen), spring applied N (Snitrogen), Block, Fall:Spring N application interactions (F:S), and the total amount of N applied in the fall and spring (Fsnitrogen) (Table 1). The NDVI and Canopeo readings were analyzed in Rstudio looking at fall applied N influence on DM_TONS, CP, and TDN (Table 2).

	Fnitrogen	Snitrogen	Block	F:S	Fsnitrogen
DM_TONS	0.000000000711	0.000000308	0.0000069	0.28	0.000000000776
CP	0.00621	0.0000000000888	0.0042	0.62363	0.0000000794
TDN	0.3275	0.0334	0.1926	0.7871	0.285

Table 1. Table of p-values for interactions between dry matter per tons (DM_TONS), crude protein (CP) and total digestible nutrients (TDN) and fall applied nitrogen (Fnitrogen), spring applied N (Snitrogen), Block, Fall:Spring N application interactions (F:S), and the total amount of N applied in the fall and spring (Fsnitrogen). Means were separated at $p < 0.05$.

	Nfall	Block
Fall NDVI	0.0112	0.0113
Fall Canopeo	0.00661	0.00615

Spring NDVI	0.0000019	0.00545
Spring Canopeo	0.00146	0.53895

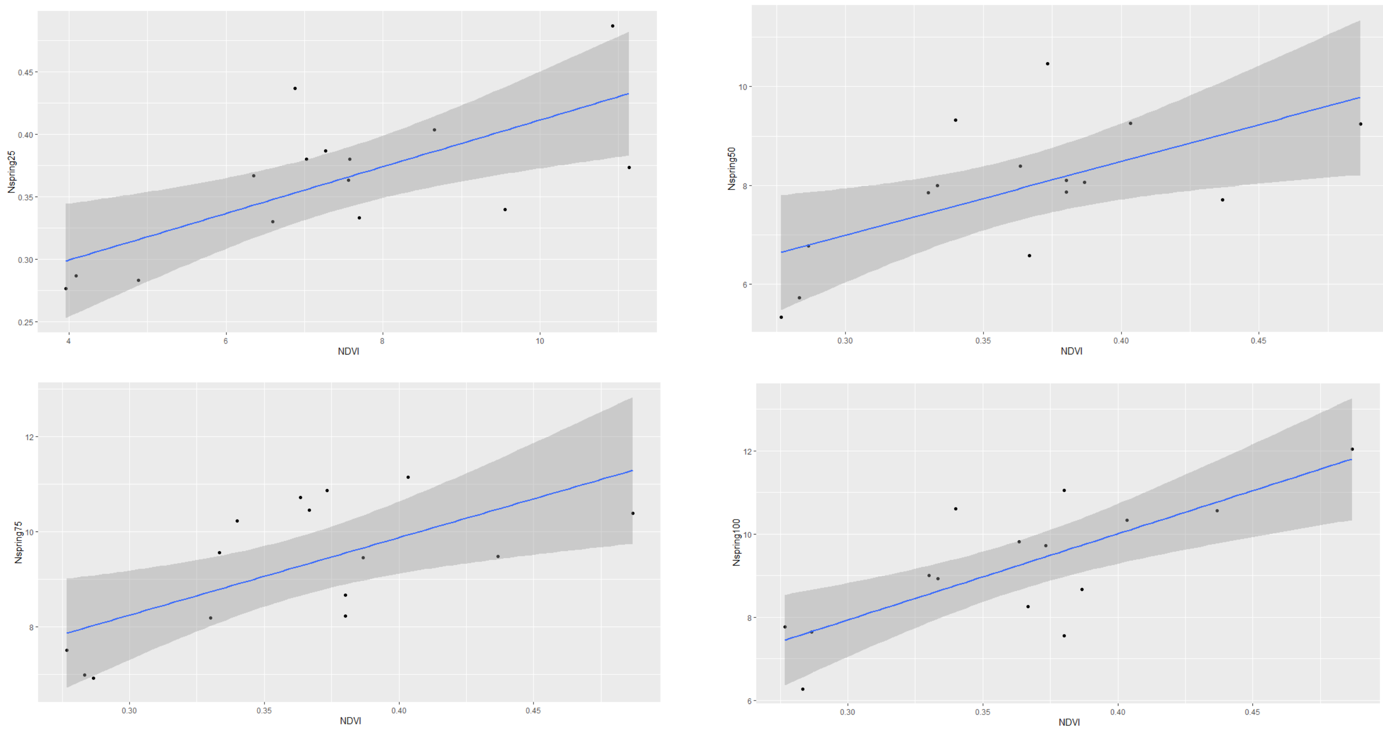
Table 2. Table of p-values for interactions between fall and spring NDVI and Canopeo readings and fall applied nitrogen (Nfall). Means were separated at $p < 0.05$.

We found that DM_TONS was found to be significantly affected by Fnitrogen, Snitrogen, and Fsnitrogen. The F:S interaction was not found to be significantly influenced with a p-value of 0.28. Crude protein was found to be significantly influenced by Fnitrogen, Snitrogen, and Fsnitrogen. The F:S interaction was not found to be significantly influenced with a p-value of 0.624. Only Snitrogen was found to significantly affect TDN with a p-value of 0.0334. Fnitrogen, F:S and Fsnitrogen did not statistically influence TDN. Nitrogen is a critical factor in forage yield and quality. The combinations of fall and spring nitrogen determine quality and yield. To maximize yield at least 30 pounds of fall nitrogen was needed. However, only 30 pounds of fall nitrogen was applied with 100 pounds of spring nitrogen yields can be maximized. Statically yields were the same when 90 pounds of fall nitrogen was applied in the fall and only 25 pounds in the spring. Spring nitrogen is the primary driver of crude protein, but 90 pounds of fall nitrogen statistically increases crude protein.

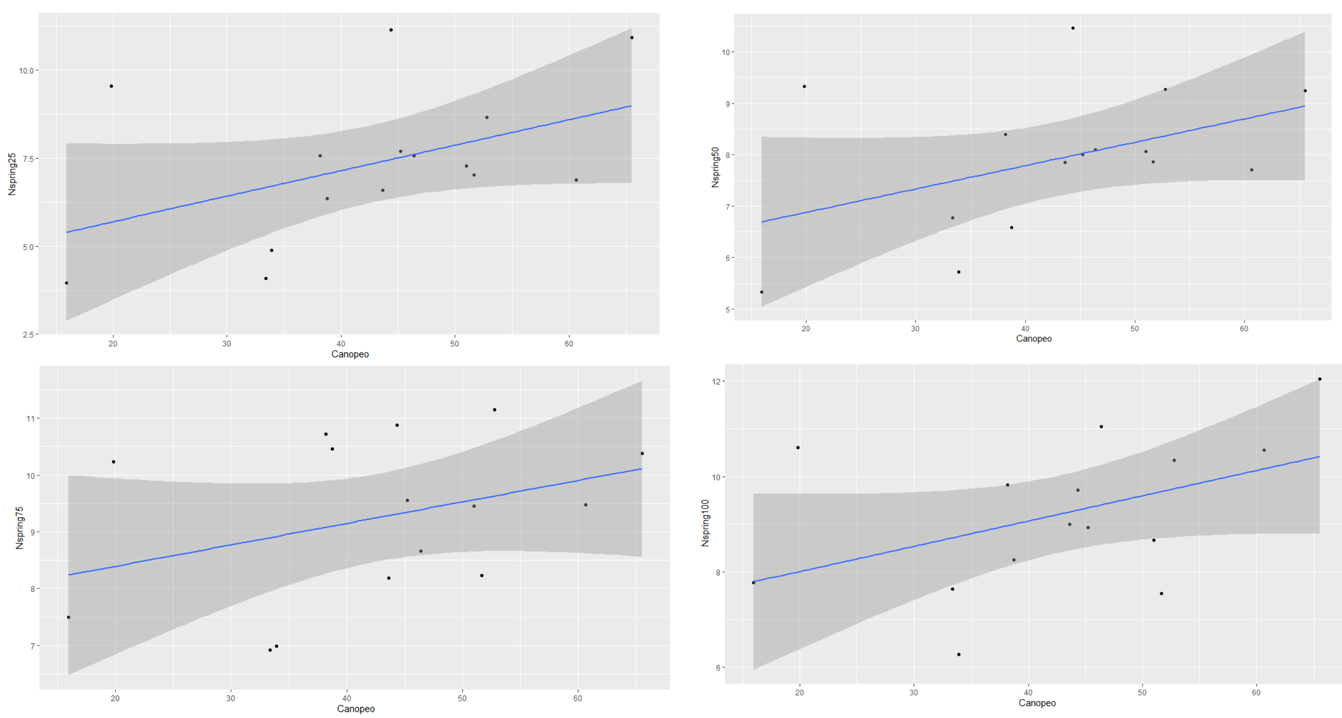
Treatment	DM_Tons	CP	TDN
N Fall			
0	6.56 c	11.39 b	63.69 a
30	8.26 b	11.3 b	63.60 a
60	9.17 a	11.23 b	63.08 a
90	9.45 a	12.61 a	61.84 a
N Spring			
25	7.37 b	9.86 d	64.17 a
50	7.77 b	10.68 c	63.89 a
75	9.18 a	12.55 b	63.06 a
100	9.17 a	13.45 a	61.08 b

Table 3. Dry matter yield and quality of Cereal rye for the fall and spring nitrogen treatments Results within the N spring or N fall section with the same letter are statistically the same. Spring and Fall nitrogen were analyzed separately since the interaction was not significant.

In Table 3 the fall nitrogen treatment had a significant yield increase through the 60 pounds of nitrogen treatment. However, the forage quality crude protein statistically increased through the 90-pound fall nitrogen treatment. As livestock producers use fall manure nitrogen on cereal rye forage crops at rates of over 90 pounds of available nitrogen only a small amount of spring nitrogen may be needed to maximize yield. Crude protein was only significantly impacted by the fall nitrogen treatment when 90 pound of fall nitrogen was applied and the TDN was not affected at all by fall nitrogen treatment. Crude protein was significantly affected by each 25-pound increase in spring nitrogen regardless of fall nitrogen rate. While the interaction was not significant greater crude protein increase were seen in the lowest fall nitrogen treatment. The ability to use NDVI to determine spring Nitrogen status will help growers make more informed decisions about spring nitrogen needs based on their forage needs. If maximizing yield is the goal applying most of their nitrogen in the fall can accomplish this. While a goal of max maximizing yield and quality requires at least 60 pounds of nitrogen and high spring nitrogen rates. TDN of cereal rye however was negatively affected by increased nitrogen rates. While our grow stage assessment showed that each treatment was at approximately the same growth stage on the primary tiller for harvest timing in the low nitrogen plots plants were shorter and secondary tillers were not as advanced.



Graph 1. Linear graph for Normalized Difference Vegetation Index (NDVI) and each spring applied nitrogen treatment. Spring nitrogen treatments included 25, 50, 75, and 100 lbs/ac named Nspring25, Nspring50, Nspring75, and Nspring100 respectively. Graphs made in Rstudio. Line of best fit.



Graph 2. Canopeo and each spring applied nitrogen treatment. Spring nitrogen treatments included 25, 50, 75, and 100 lbs/ac named Nspring25, Nspring50, Nspring75, and Nspring100 respectively. Graphs made in Rstudio. Line of best fit.

A Pearson correlation between fall and spring NDVI readings, fall and spring Canopeo readings and all of the spring applied N treatments (Nspring25, Nspring50, Nspring75, and Nspring100) was done to observe if there was a positive or negative correlation between ground cover measurement tools. We found the NDVI had a correlation with all of the spring applied N treatments, and Canopeo did not (Table 3). This shows that NDVI is a better tool to use to measure ground cover, and potential nitrogen available in the spring for the crop compared to Canopeo. This study showed that we could use NDVI to assess the crops fall nitrogen uptake which maybe a tool to determine spring nitrogen needs. Prabhakara et al. (2015) and Chianucci et al. (2018) found that NDVI and other digital indices can be effective to measure groundcover and canopy cover with accuracy compared to traditional assessment. These digital indices are a great resource to use, but no method can be 100% accurate and will still need quality control of the imagery classification (Chianucci et al., 2018). One strong advantage of using digital photography is that these methods remove the subjectivity of the operator, which increases the precision of canopy cover estimates, allowing reproducibility of measures, and user inspection of results of the classification, unlike other existing methods (Chianucci et al., 2018). Knowing how to correctly use the digital index is important. (Sunoj et al., 2021) recommends avoiding: (i) sunny imaging conditions that can cause streaks and shadows, (ii) areas with previous crop residues, (iii) areas impacted by tillage, (iv) wet areas, and (v) areas with weed infestations. Holding the camera parallel to the ground to avoid image distortion (Sunoj et al., 2021) and far enough away from the body is another point to remember when using these tools.

	NDVI	Canopeo
Nspring25	0.7	0.46
Nspring50	0.62	0.44
Nspring75	0.65	0.36
Nspring100	0.76	0.46

Table 3. Table of p-values for interaction between for Normalized Difference Vegetation Index (NDVI), Canopeo and the spring applied nitrogen treatment. Spring nitrogen treatments included 25, 50, 75, and 100 lbs/ac named Nspring25, Nspring50, Nspring75, and Nspring100 respectively. Means separated at $p < 0.05$.

The impact of N applications split between spring and fall on yield is important. Nitrogen applied in the fall or split between the fall and spring will result in more constant forage production throughout the season compared to one spring applications, though, spring applied N was found to increase yields up to 200 lbs/ac compared to other methods (Altom et al., 1996). The amount of N applied in the fall or spring can also impact the yield. High rates of N fertilizer are required for maximum rye-wheat-ryegrass forage yields (Altom et al., 1996). (Szuleta et al., 2023) found that the average yield was slightly higher in treatments that had N applications of 70 lbs/ac compared to 35 lbs/ac. Applying more N to a field might increase yield and has an optimal N level, but over-fertilization can lead to unpredictable yield changes and lower amounts of N applied does not necessarily mean a lower yield will be achieved (Szuleta et al., 2023).

Conclusions

Based on the nitrogen rates we used a combination of spring and fall nitrogen is needed to maximize forage yield and quality. A minimum of 30 pounds of fall nitrogen is needed but when more fall nitrogen is applied lower spring nitrogen rates can be used to maximize winter cereal rye yield and quality. NDVI can be a tool further developed to assess the amount of fall nitrogen that was applied and make recommendations for spring nitrogen rates. Further research is needed to develop stronger NDVI reading recommendations for assessing fall nitrogen uptake of the cereal rye crop. This preliminary work however shows the value of using NDVI to assess the amount of fall nitrogen that may have been applied especially with nitrogen sources such as manure are used. In future work, a zero nitrogen spring application should also be used to better analyze the effects of spring nitrogen and fall nitrogen both alone and in combination.

Resources

Altom, W., Rogers, J. L., Raun, W. R., Johnson, G. V., & Taylor, S. L. (1996). Long-term rye-wheat-ryegrass forage yields as affected by rate and date of applied nitrogen. *Journal of Production Agriculture*, *9*(4), 510–516. https://doi.org/10.2134/jpa1996.0510

Chianucci, F., Lucibelli, A., & Dell'Abate, M. T. (2018). Estimation of ground canopy cover in agricultural crops using downward-looking photography. *Biosystems Engineering*, *169*, 209–216. https://doi.org/10.1016/j.biosystemseng.2018.02.012

De Mendiburu F (2023). _agricolae: Statistical Procedures for Agricultural Research_. R package version 1.3-7, <https://CRAN.R-project.org/package=agricolae>.(2) Lollato, R., Diaz, D. R., & Asebedo, R. (2017, February 3). *Precision Nitrogen Management for Wheat in Kansas Using Optical Sensor Technology*. Agronomy eUpdate. https://webapp.agron.ksu.edu/agr_social/m_eu_article.throck?article_id=1259

Lyons, S. E., Ketterings, Q. M., Ort, S., Godwin, G. S., Swink, S. N., Czymmek, K. J., Cherney, D. J., Cherney, J. H., Meisinger, J. J., & Kilcer, T. F. (2019). Nitrogen management for forage winter cereals in the northeastern USA. *Soil Science Society of America Journal*, *83*(4), 1111–1123. https://doi.org/10.2136/sssaj2019.01.0004

Patrignani, A., & Ochsner, T. E. (2015). Canopeo: A powerful new tool for measuring fractional green canopy cover. *Agronomy Journal*, *107*(6), 2312–2320. https://doi.org/10.2134/agronj15.0150

Prabhakara, K., Hively, W. D., & McCarty, G. W. (2015). Evaluating the relationship between biomass, percent groundcover and remote sensing indices across six winter cover crop fields in Maryland, United States. *International Journal of Applied Earth Observation and Geoinformation*, *39*, 1–15. https://doi.org/10.1016/j.jag.2015.03.002

RStudio Team (2020). RStudio: Integrated Development for R. RStudio, PBC, Boston, MA URL http://www.rstudio.com/

Sunoj, S., McRoberts, K. C., Benson, M., & Ketterings, Q. M. (2021). Digital image analysis estimates of biomass, carbon, and nitrogen uptake of winter cereal cover crops. *Computers and Electronics in Agriculture*, *184*, 1–11. https://doi.org/10.1016/j.compag.2021.106093

Szuleta, E., Shockley, J. M., Knott, C., Phillips, T., & Van Sanford, D. A. (2023). Influence of nitrogen rate on yield and profitability of Rye Grain Production. *Crop, Forage & Turfgrass Management*, *9*(2), 1–10. https://doi.org/10.1002/cft2.20243

White, C. (2023, May 28). *Using an NDVI sensor to estimate cover crop nitrogen content*. Penn State Extension. https://extension.psu.edu/using-an-ndvi-sensor-to-estimate-cover-crop-nitrogen-content