Assessment of Leaf Chlorophyll Content, Leaf Area Index and Yield of Corn (*Zea mays* L.) Using Low Altitude Remote Sensing

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ABSTRACT: The study focused on establishing relationships between remotely-sensed vegetation indices and selected agronomic parameters for corn, namely: leaf area index (LAI), leaf chlorophyll content (LCC), and yield. Four varieties of corn plants (UPLB Var 6 and 11, USM Var 10 and CSC Var 1) were planted following a split-plot arrangement in a randomized complete block design. These varieties were subjected to four different fertilizer treatments: F0 – no fertilizer, F1 – 100% commercial fertilizer, F2 - 100% organic fertilizer and F3 - 50% commercial, 50% organic fertilizer. Aerial images were acquired every 15 days starting at 30 days after sowing (DAS), until 75 DAS using a multispectral digital camera mounted in an unmanned aerial vehicle (UAV). Vegetation indices (VI), namely: GNDVI, BNDVI and ENDVI, were computed using the spectral bands (NIR, Green and Blue) from the multispectral images. Regression analysis revealed that GNDVI had the highest coefficient of multiple determination (R^2) for corn LAI, LCC, and yield at different growth stages. The relationship between GNDVI and LCC; and GNDVI and Yield both generated highest R^2 values of 0.66 and 0.47, respectively, at 45 DAS. Also, the relationship between GNDVI and LAI yielded highest R^2 of 0.44 at 75 DAS. Results of the study showed that remotely-sensed vegetation indices can be used for rapid and non-destructive monitoring corn growth parameters as early as vegetation stage for LCC and yield and reproductive stage for LAI.

INTRODUCTION

Chlorophyll is important in the photosynthesis of the plants; it is the green pigment found in plants. It absorbs blue light and portions of red light from the electromagnetic spectrum. It is essential as it traps light energy from the sun which is then combined with carbon dioxide and water to produce sugar and oxygen through photosynthesis. (ScienceDaily, 2019) Leaf Area Index is also an important indicator of the different processes that occur in the corn plant. It is a parameter for plant growth and yield as well as a measure for timely interventions such as in the application of fertilizers and proper irrigation. (Trimble, 2019)

Corn (*Zea mays L.*) is the second most important food crop in the Philippines, next to rice. It is the staple food of 20% of the Filipino population, especially in the regions of the Visayas and Mindanao. Also, the crop is important economically as a major ingredient of feeds for poultry and livestock and as raw material in other industrial products. (Greenpeace, n.d.)

Optical remote sensing uses sensors to detect the solar radiation reflected from the ground. Images are formed from the earth's surface with the use of visible, near infrared and short-wave infrared sensors. The different wavelengths absorb and reflect differently. Thus, remotely sensed images differentiate materials by their spectral reflectance signatures. (CRISP, 2001)

Drones or unmanned aerial vehicle (UAV) are now being used as a tool for low altitude remote sensing in agriculture. Drones over a field can also be used for data collection that may help farmers improve crop management. This study used the drone technology to monitor and assess chlorophyll content. A multispectral image sensor was mounted in a drone to capture image data at specific frequencies. The wavelengths were separated using filters or instruments that were sensitive to particular wavelengths.

Vegetation Indices (VIs) used reflectance properties of vegetation. These were combinations of surface reflectance at two or more wavelengths. Each of the VIs is designed to highlight a particular property of vegetation. (Harris Geospatial Solutions, Inc. 2019)

Monitoring of the different growth parameters is a tedious and time-consuming process. Also, some process may be destructive. This study showed that remotely-sensed vegetation indices using unmanned aerial vehicles can be used to assess and monitor chlorophyll content, leaf area index and yield non-destructively in lesser time. It can be used for data collection that may help farmers improve crop management, including determining the time and adequate application of nitrogen fertilizer.

MATERIALS AND METHODS

The study was conducted in a well-established corn field as part of National Corn Trials (NCT), located at the Central Experimental Station, University of the Philippines Los Baños, College, Laguna, Philippines. It is an experimental field next to the UPLB National Agrometeorological Station (NAS) with coordinates of 14˚11'N and 121˚15'E.

Four open-pollinated varieties of corn plants namely V1(CSC1); V2(IPB Var 6); V3(IPB VAR 11) and V4 (USM 10) were planted in the field following a split-plot arrangement in Randomized Complete Block Design (RCBD). The plants were subjected to four different fertilizer treatments: F0 – no fertilizer, F1 – 100% commercial fertilizer, F2 - 100% organic fertilizer and F3 - 50% commercial, 50% organic fertilizer, with three replications each.

Data Gathering

To represent the different growth stages of corn, measurements were obtained every 15 days starting at 30 days after sowing. The chlorophyll content of the corn leaves as well as the leaf area index were both measured from the ground, while aerial images were obtained from the drone. The yields of the different treatments were determined at harvest.

A hexacopter drone was used in the conduct of this study. A multispectral imaging camera was mounted to be able to acquire aerial images. A modified Canon SX260 HS commercial-grade digital camera was used in aerial imaging. This can acquire photos in blue, green and near infrared bands. The hexacopter was flown between 10:00 am to 1:00 pm maintaining an altitude of 25 m above ground to be able to have a clear and complete view of the experimental field.

The chlorophyll content of corn plants was measured using a Soil-Plant Analyses Development (SPAD) chlorophyll meter. Three (3) leaves were measured in each of the experimental plots at every growth stage. Ground measurements of LAI were then determined using a plant canopy analyser. There was at most a day difference between the aerial and ground measurements. At the end of the cropping season, the yield per hectare was determined.

Data Processing

The raw images acquired from the flight of the UAV were stitched. The orthomosaic images stitched were in high resolution to be able to extract the vegetation indices from the images properly. It was then further analyzed to determine the different vegetation indices, namely: the green, blue and enhanced NDVI in the Quantum Geographic Information System (QGIS). The vegetation indices of the corn plants subjected to the different fertilizer treatments were computed to assess the chlorophyll content, leaf area index and yield in the corn plants.

The raster calculator tool was used for computing the Green Normalized Difference Vegetation Index (GNDVI), Blue Normalized Difference Vegetation Index (BNDVI), and Enhanced Normalized Vegetation Index (ENDVI) using equations 1, 2 and 3, respectively.

From Zhou et. al, in 2017, the GNDVI that uses green and NIR reflectance is computed as:

$$\boldsymbol{GNDVI = \frac{(NIR - Green)}{(NIR + Green)}}$$

Where:

GNDVI = the Green Normalized Difference Vegetation Index
NIR = the digital number of the Red/NIR channel with the red-blocking filter
G = the digital number of the green channel

The BNDVI combines Blue and NIR reflectance to show the variation in the vegetation (Drone Deploy, n.d.). This index uses the formula:

$$\boldsymbol{BNDVI = \frac{(NIR - Blue)}{(NIR + Blue)}}$$

Where:

BNDVI = the Blue Normalized Difference Vegetation Index
NIR = the digital number of the Red/NIR channel with the red-blocking filter
Blue = the digital number of the blue channel

ENDVI shows a comparison of green and blue lights to the NIR and Red reflectance (Drone Deploy, n.d.). This index is computed using the formula:

$$\boldsymbol{ENDVI = \frac{((NIR + Green) - (2\ x\ Blue))}{((NIR + Green) + (2\ x\ Blue))}}$$

Where:

ENDVI = the Enhanced Normalized Difference Vegetation Index
NIR = the digital number of the Red/NIR channel with the red-blocking filter
Blue = the digital number of the blue channel
Green = the digital number of the Green channel

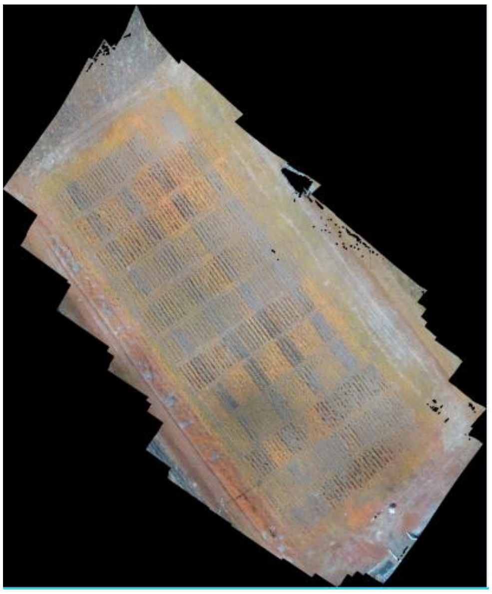
Significant differences in the experimental results were analyzed using Analysis-of-variance (ANOVA) of an experiment using split-plot arrangement in Randomized Complete Block Design with three replications. Treatment effects were considered significant when $p < 0.05$. (Gomez and Gomez, 1984)

Regression analysis was also done to be able to establish the relationship among the different variables measured in the experiment. The green, blue and enhanced NDVI values along with the chlorophyll content acquired from SPAD during ground measurements were correlated to establish the relationship of these values to the chlorophyll content in the leaves. The green, blue and enhanced NDVI values were also correlated with the LAI measurements from the plant canopy analyzer. The relationship of the green, blue and enhanced NDVI values with yield were also obtained. This was done to show at what growth stage the vegetation indices values have the greatest relationship to be able to predict the yield of the corn plants.

RESULTS AND DISCUSSION

Aerial Images

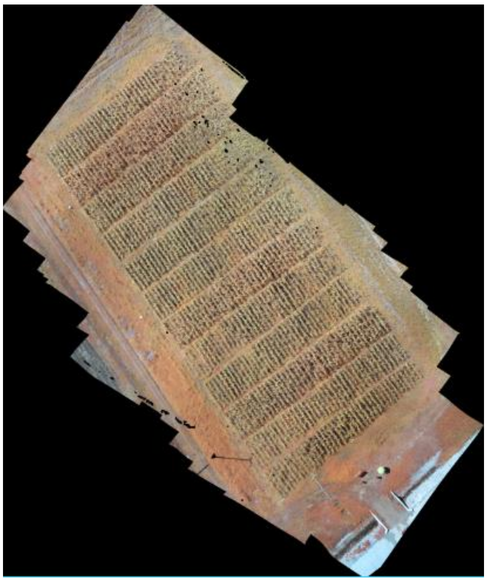
Aerial raw images were obtained using the multispectral camera mounted in the unmanned aerial vehicles. The raw images showed only parcels of the experimental plot, thus, these images were stitched. The stitched images of the different growth stages obtained from the unmanned aerial vehicle are shown below.



April 2 – 30 DAS

April 18 – 45 DAS

May 2 – 60 DAS



May 17 – 75 DAS

Figure 1. Stitched Images at the Different Growth Stages

Vegetation Indices are single numbers extracted from each pixel in a remote-sensed image. It is used to quantify vegetation biomass and/or plant vigor. Normalized difference vegetation index (NDVI) is the most common type of vegetation index used. (eXtension, 2014) NDVI uses near-infrared and red light to quantify the presence of vegetation in an area. NDVI is used to determine chlorophyll level, plant health and stress level, fertilizer use, nitrogen management; identify insects, pests, weed and plant diseases. It is also used for farm plant development, cultivation planning, and harvest planning. (Green Aero Technology Inc, 2016)

Blue Normalized Difference Vegetation Index (BNDVI), Enhanced Normalized Difference Vegetation Index (ENDVI) and Green Normalized Difference Vegetation Index (GNDVI) were used to assess the chlorophyll content, leaf area index and yield in the corn plants. Figure 2 below shows a sample (75 DAS) of the BNDVI, ENDVI and GNDVI maps generated.

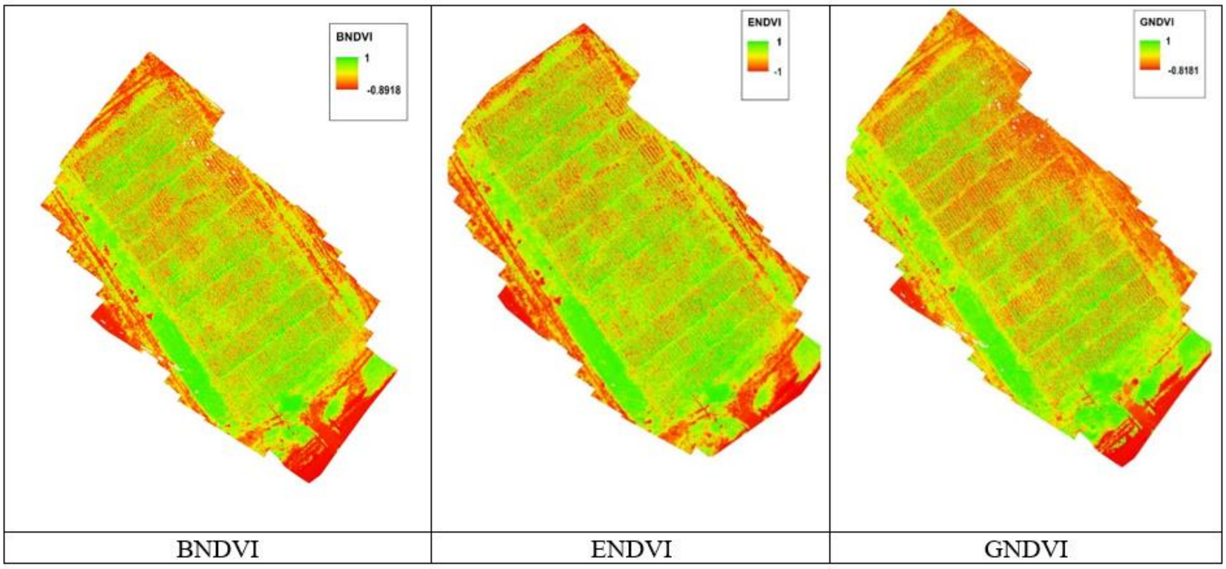


Figure 2. Samples images (75 DAS) of BNDVI, ENDVI and GNDVI maps

Figure 2 shows the sample vegetation images generated using QGIS. The red color shows no to low vegetation while the green color shows high and healthy vegetation in the area. NDVI always ranges from minus one (-1) to plus one (+1) for any given pixel. A value close to +1 (0.8 - 0.9) may indicate the highest possible density of green leaves. (Weier and Herring, 2000) However, there are no distinct boundary for each type of land cover. Negative values of NDVI may signify the presence of water while zero or close to zero value of NDVI may mean no presence of green leaves or an urbanized area. (GIS Geography, n.d.)

Analysis of variance (ANOVA) were performed for all the vegetation indices generated to show significant differences in the treatments. The ENDVI values generated for the different treatments were found to be only significantly different at 75 DAS, while BNDVI was also only significantly different at 45 DAS and 75 DAS. Only GNDVI values generated were significant for all stages of growth for both treatment and replicate.

It was observed that plants grown under the F1 and F3 treatments have higher vegetation indices generated values compared to plants grown under F0 and F2 treatments. Treatments containing recommended fertilizers generated significantly higher values for all vegetation indices. This is because recommended fertilizers have known nutrient content that is readily available for use by the plants. (AITC Canada 2019)

Chlorophyll Content and Vegetation Indices

Relationships between corn leaf chlorophyll content (LCC) and the different vegetation indices at the different growth stages are shown in Tables 1-4.

Table 1. Summary of line of best fit equation and coefficient of multiple determination for the different vegetation indices for corn LCC at 30 DAS.

VIs	LINE OF BEST FIT	R^2
GNDVI	y = 0.0041x - 0.0856	0.3971
ENDVI	y = 0.0007x + 0.1174	0.0116
BNDVI	y = 0.0024x + 0.0829	0.0794

At 30 DAS, the GNDVI exhibited the highest R^2 of 0.40 for its linear relationship between chlorophyll content and the different vegetation indices considered.

Table 2. Summary of line of best fit equation and coefficient of multiple determination for the different vegetation indices for corn LCC at 45 DAS.

VIs	LINE OF BEST FIT	R^2
GNDVI	y = 0.0046x - 0.0766	0.6569
ENDVI	y = 0.0009x + 0.1166	0.0385
BNDVI	y = 0.0029x + 0.083	0.2412

At the vegetative stage of the corn crop, the GNDVI also generated the highest R^2 among the different vegetation indices analyzed. The chlorophyll content of corn crops at 45 DAS had a strong linear relationship with GNDVI, with an R^2 of 0.66.

Table 3. Summary of line of best fit equation and coefficient of multiple determination for the different vegetation indices for corn LCC at 60 DAS.

VIs	LINE OF BEST FIT	R^2
GNDVI	y = 0.002x + 0.0211	0.491
ENDVI	y = 3E-06x + 0.1207	1E-06
BNDVI	y = 0.0009x + 0.1308	0.0633

The highest R^2 generated in analyzing the relationship of the chlorophyll content of corn crops at 60 DAS was also observed when GNDVI was used. The chlorophyll content and GNDVI of corn crops at 60 DAS has a moderate linear relationship with a 0.49 coefficient of determination.

Table 4. Summary of line of best fit equation and coefficient of multiple determination for the different vegetation indices for corn LCC at 75 DAS.

VIs	LINE OF BEST FIT	R^2
GNDVI	y = 0.0024x + 0.0008	0.4802
ENDVI	y = -1E-04x + 0.1664	0.0008
BNDVI	y = 0.001x + 0.1679	0.0556

At 75 DAS, it was also the GNDVI that generated the highest R^2 with chlorophyll content at 0.48. The GNDVI exhibited the highest coefficient of determination with chlorophyll content in all the growth stages. The R^2 values showed moderate to strong linear relationship between the chlorophyll content of corn and GNDVI. The GNDVI generated the highest R^2 for LCC in all the growth stages.

This result is in agreement with the findings of the experiment by Gitelson and Merzlyak in 2010 which showed that the sensitivity of NDVI to chlorophyll content increases by about five times when green and red channels are used.

Leaf Area Index and Vegetation Indices

The Leaf Area Index was also measured every 15 days starting 30 DAS using the Plant Canopy Analyzer. Several studies have shown a strong correlation between NDVI and LAI. One of which is by Wang et al in 2007 conducted a study entitled, "*New Vegetation Index and Its Application in Estimating Leaf Area Index of Rice.*" wherein the different bands were analyzed to show their sensitivity to LAI. It was found that red and blue bands were more sensitive when the LAI was less than 3, while the green band was more sensitive in a wider range of LAI values.

Relationships between corn leaf area index (LAI) and the different vegetation indices at the different growth stages are shown in Tables 5-8.

Table 5. Summary of line of best fit equation and coefficient of multiple determination for the different vegetation indices for corn LAI at 30 DAS.

VIs	LINE OF BEST FIT	R^2
GNDVI	y = 2.3606x + 0.9424	0.0644
ENDVI	y = 1.4191x + 0.8519	0.0213
BNDVI	y = 1.347x + 0.834	0.036

There is a very weak linear relationship observed with all the vegetation indices considered and LAI of corn at 30 DAS. The GNDVI, however, still generated the highest R^2 among the vegetation indices and LAI with R^2 of 0.06.

Table 6. Summary of line of best fit equation and coefficient of multiple determination for the different vegetation indices for corn LAI at 45 DAS.

VIs	LINE OF BEST FIT	R2
GNDVI	y = 5.8348x + 0.9648	0.28
ENDVI	y = 4.0676x + 0.7233	0.0888
BNDVI	y = 4.6824x + 0.5101	0.2035

The GNDVI still exhibited the highest R^2 among the different vegetation indices with LAI at 45 DAS. It was however a weak linear relationship with 0.28 coefficient of determination.

Table 7. Summary of line of best fit equation and coefficient of multiple determination for the different vegetation indices for corn LAI at 60 DAS.

VIs	LINE OF BEST FIT	R^2
GNDVI	y = 9.2952x + 1.0432	0.3277
ENDVI	y = 3.9966x + 1.1977	0.0636
BNDVI	y = 5.3477x + 0.8666	0.17

At 60 DAS, it was still the GNDVI that generated the highest R^2 of 0.33 with LAI of corn. GNDVI was followed by BNDVI then ENDVI which had the lowest R^2 for LAI of corn at 60 DAS.

Table 8. Summary of line of best fit equation and coefficient of multiple determination for the different vegetation indices for corn LAI at 75 DAS.

VIs	LINE OF BEST FIT	R^2
GNDVI	y = 10.09x + 1.3488	0.4422
ENDVI	y = 1.2076x + 1.793	0.0061
BNDVI	y = 3.9229x + 1.2318	0.0956

The Green NDVI generated the highest R^2 at 0.44, a moderate linear relationship, at 75 DAS.

The results are also in accord with the previous studies by Fumera in 2018 wherein the correlation coefficients obtained in his study between the LAI of corn in the vegetative stage and the BNDI, ENDVI and GNDVI are of 0.7189, 0.0526 and 0.8192, respectively. The study showed that GNDVI is a good indicator for the LAI of corn plants.

The knowledge of LAI during these different stages is important as it shows it is used as a parameter for plant growth. The knowledge of LAI can also be used as a tool for timely interventions such as the application of fertilizers and irrigation.

Yield and Vegetation Indices

The vegetation indices generated were also correlated with the yield data to show the best growth stage and vegetation index that can predict the yield of the corn plants. Several outliers were omitted to generate a better relationship with yield and vegetation indices.

Relationships between corn yield and the different vegetation indices at the different growth stages are shown in Tables 9-12.

Table 9. Summary of line of best fit equation and coefficient of multiple determination for the different vegetation indices for corn yield at 30 DAS.

VIs	LINE OF BEST FIT	R^2
GNDVI	y = 20.517x + 2.078	0.2557
ENDVI	y = 8.308x + 1.8354	0.0399
BNDVI	y = 9.4135x + 1.4917	0.0947

Considering the relationship between the vegetation indices generated at 30 DAS and the yield harvested, the GNDVI showed the highest R2 of 0.26, a weak linear relationship.

Table 10. Summary of line of best fit equation and coefficient of multiple determination for the different vegetation indices for corn yield at 45 DAS.

VIs	LINE OF BEST FIT	R^2
GNDVI	y = 25.913x + 1.5184	0.4684
ENDVI	y = 14.822x + 0.8626	0.1081
BNDVI	y = 18.356x – 0.1197	0.2829

The GNDVI of the plants at 45 DAS still generated the highest coefficient of determination. A moderate linear relationship was established with an R^2 of 0.47. BNDVI established a weak linear relation while ENDVI established a very weak linear relationship.

Table 11. Summary of line of best fit equation and coefficient of multiple determination for the different vegetation indices for corn yield at 60 DAS.

VIs	LINE OF BEST FIT	R^2
GNDVI	y = 36.486x + 0.5477	0.4151
ENDVI	y = 18.053x + 0.8148	0.124
BNDVI	y = 22.033x – 0.3379	0.2636

A moderate linear relationship was established between the GNDVI of 60 DAS corn plants with the yield data. This generated the highest R^2 of 0.42, followed by BNVI and ENDVI with R^2 of 0.26 and 0.12, respectively.

Table 12. Summary of line of best fit equation and coefficient of multiple determination for the different vegetation indices for corn yield at 75 DAS.

VIs	LINE OF BEST FIT	R^2
GNDVI	y = 30.156x + 1.0684	0.3868
ENDVI	y = 0.1337x + 2.9621	6E-06
BNDVI	y = 10.413x + 0.9545	0.0613

At 75 DAS, it is still the GNDVI that has the highest R^2 among the vegetation indices analyzed with an R^2 of 0.39. It was followed by BNDVI then ENDVI.

A moderate linear relationship for the GNDVI and yield were established, with 45 DAS having the highest R^2 of 0.47. It is followed by 60 DAS, 75 DAS and 30 DAS with R^2 of 0.42, 0.39 and 0.26, respectively. The stage that can best describe possible yield is at 45 DAS.

All the different growth stages showed that the GNDVI is the vegetation index that can best predict yield.

A summary of the best fit models for the agronomic parameters and the different vegetation indices is seen in Table 13 below.

Table 13. Summary of the best fit models for estimation of the different parameters

	GROWTH STAGE	VEGETATION INDEX	EQUATION	COEFFICIENT OF DETERMINATION
Chlorophyll Content	45 DAS	GNDVI	y = 144.1x + 21.184	0.66
Leaf Area Index	75 DAS	GNDVI	y = 10.09x + 1.3488	0.44
Yield	45 DAS	GNDVI	y = 25.913x + 1.5184	0.47

The summary table gives the best fit models that can be used to estimate leaf chlorophyll content, leaf area index of corn as well as the equation that can best predict yield. The GNDVI is the vegetation index that can be reliably used for estimation of the different parameters.

SUMMARY AND CONCLUSION

Plant leaves serve as the site for absorption and assimilation of carbon dioxide, light interception, evapotranspiration and other processes occurring necessary for plant growth. (Trimble, 2018) Assessment of chlorophyll content and leaf area index in the different growth stages is important as it serves as a guide for proper farm management practices that can be done in the field. However, it is often a labor intensive process.

It was observed that plants grown under the F1 and F3 treatments have higher measured values compared to plants grown under F0 and F2 treatments. Treatments containing recommended fertilizers generated significantly higher vegetation indices.

Relationships among the different variables measured in the experiment were established through regression analysis. The Green Normalized Difference Vegetation Index (GNDVI) generated the highest correlation coefficient (R^2) in all the growth stages for both SPAD and LAI measurements as well as in yield prediction.

Yield may be best predicted using GNDVI at 45 DAS. All the different growth stages showed that the GNDVI is the vegetation index that can best predict yield. A moderate linear relationship for the GNDVI and yield were established, with 45 DAS having the highest R2 of 0.47.

The models generated can be used as a tool to monitor the different growth parameters and be able to make the necessary interventions on a timely manner. Also, these can serve as guides to the proper amount of recommended fertilizer to give the plants to optimize their growth and lessen waste in giving excess fertilizers.

This study showed that the drone technology can be used to asses and monitor chlorophyll content and leaf area index non-destructively and in a much shorter period of time. UAV mounted with multispectral cameras serve as tools for low altitude remote sensing of agricultural fields and can be used for data collection. This can help farmers improve their crop management, including determining the time and adequate application of nitrogen fertilizer. Vegetation indices especially the GNDVI can be a reliable measure of leaf chlorophyll content of corn. Time in monitoring the leaf chlorophyll content, which can also indicate leaf nitrogen content, of corn can now be greatly lessened using remotely-sensed vegetation indices.

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