



Exploration of high-resolution multispectral UAV imagery for non-destructive estimation of chlorophyll content in lemon leaves

By

Yonela Thungana (1975561)

Supervisor: Prof Elhadi Adam

A research report submitted in partial fulfilment of the requirements for the degree in Master of Science in GIS and Remote Sensing

Faculty of Science

University of the Witwatersrand

ABSTRACT

Unmanned Aerial Vehicle (UAV) technology also known as drones together with Remote Sensing (RS) have become evident as great potential for the agriculture industry in making informed decisions. The overall aim of using this technology is to improve productivity and support sustainable agricultural practices. South African farmers are faced with many challenges, including climate change, load shedding, and water shortages. Despite these challenges, they must sustain the environment and enhance food security. Chlorophyll plays a great role as a crop predictor of plant health status and can reveal nutrient deficiency. The estimation of chlorophyll requires selecting vegetation indices (VI) that are sensitive to chlorophyll or physiological properties of crops. Five indices were selected for this study: the Normalized Difference Vegetation Index (NDVI), Normalized Difference Red Edge (NDRE), Modified Soil Adjusted Vegetation Index (MSAVI), Leaf Chlorophyll Index (LCI), and Difference Vegetation Index (DVI). The study focused on estimating chlorophyll content in lemon leaves using VIs and validating these estimates with ground-measured chlorophyll data. The Partial Least Squares Regression (PLSR) method was employed to create a predictive model for chlorophyll content in the study area.

Through Pearson Correlation analysis, it was found that VIs that used the combination of the NIR and red band were more influential and sensitive indices to chlorophyll than the red edge indices. The DVI and MSAVI yielded the highest correlation, with values of 0.85 and 0.47 respectively. These were followed by the LCI, NDVI, and NDRE which had r values of 0.26, 0.23, and 0.22 respectively. Among the spectral bands, the NIR band exhibited the strongest correlation with a coefficient of $r = 0.89$, followed by the red edge, green, red, and blue bands with their respective r values of 0.57, 0.18, and 0.11. The blue band showed the lowest correlation with measured chlorophyll at $r = 0.04$. The PLSR estimation models were evaluated using the following metrics: coefficient of determination (R^2), Mean Absolute Error (MAE), and Root Mean Square Error (RMSE). The model based on UAV spectral bands achieved a slightly higher estimation accuracy with an $R^2 = 0.85$, and the lowest values of $RMSE = 2.11$ and $MAE = 1.79$. This model was subsequently used to create a chlorophyll concentration map for the study area. These findings demonstrate that high-resolution UAV data can be effectively utilized for field management in many ways including crop monitoring and field mapping. This data can significantly help in making more informed decisions.

Dedication

This research is dedicated to my family, especially my mother. She is the pillar of my strength. Thank you, Mommy, for holding my hand, giving me hope and instilling faith in this journey.

To my brother who gave me this opportunity to pursue my master's degree, I owe my life.

ACKNOWLEDGEMENTS

Firstly, I thank God for all the wisdom, courage, and perseverance that he gave me.

Secondly, to express my greatest gratitude to my supervisor Prof. Elhadi Adam for being there for me every step of the way and for nurturing me in the right direction.

My third gratitude goes to Dr Philemon Tsele from the University of Pretoria for assisting with drone data collection and processing.

To Ms Yingisani Chabalala from the University of South Africa thank you so much for availing yourself and showing interest in helping.

To Mr Mcebisi Qabaqaba from the University of Pretoria you have been so supportive and helpful throughout this project you made it all possible.

Lastly, to myself for hanging in there and showing up for my studies even when I felt discouraged, this was the most challenging time of my life mentally and financially, but I pulled through.

DECLARATION – PLAGIARISM

I, Yonela Innocentia Thungana (Student number: 1975561), hereby declare that.

- The research report titled “Exploration of high-resolution multispectral UAV imagery for non-destructive and estimation of chlorophyll content in lemon leaves" Is my work.
- All the sources quoted in this research have been re-written and acknowledged in the reference list.
- This research has never been submitted to any other University for examination or any degree.

Signature: YThungana

Date: 26-02-2024

Yonela Innocentia Thungana

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List of abbreviations

DVI - Difference Vegetation Index
GIS - Geographic Information Systems
GPS - Ground Positioning System
HS - Hyperspectral
LCI - Leaf Chlorophyll Index
MAE - Mean Absolute Error
MS - Multispectral
MSAVI - Modified Soil Adjusted Vegetation Index
NDRE - Normalized Difference Red Edge
NDVI - Normalized Difference Vegetation Index
NIR - Near Infrared
PA - Precision Agriculture
PLSR - Partial Least Squares Regression
r - Correlation Coefficient
 R^2 - Coefficient of Determination
RMSE - Root Mean Squares Error
RS - Remote Sensing
SPAD - Soil Plant Analysis Development
UAV - Unmanned Aerial Vehicle
UN - United Nation
VI - Vegetation Index

CHAPTER ONE

Introduction

1.1 General introduction

Agriculture plays a crucial role in Limpopo Province as a source of economic growth and sustenance for a large population (Oni et al., 2018). The province is the leading citrus grower in South Africa contributing significantly to foreign exchange earnings, according to DAFF (2017) it contributed 42% of total national citrus production, cultivated on over 26 960 hectares of land. Precision Agriculture (PA) is the use of innovative technology and analysis tools that provide a better understanding of the land under cultivation to enhance productivity, quality and reduce input costs (Sharma, 2023). It can be used to enhance food production while promoting sustainable agricultural practices (Demirbaş, 2018). Precision agriculture, also known as satellite agriculture, was once exclusively practiced by large commercial farmers who had access to high-technology resources or good infrastructure. However, in recent times, the agricultural sector has embraced technological advancement allowing small farmers to engage in satellite farming using tools like mobile apps, smart sensors, satellite images, and drones (Dhanaraju et al. 2022). Consequently, the ongoing developments in farming technology and their growing affordability helps small farmers to access the benefits of high-end farming methods which were previously only available to larger commercial farms (Gokool et al., 2023).

Chlorophyll functions as a key indicator of the health status of crops and can reveal their nutritional needs (Elsayed et al., 2023). The estimation of chlorophyll requires spectral indices that have strong response to the chlorophyll concentration, and modified indices that account for limitations associated with other indices such as background effects or soil interferences (Haboudane et al., 2002). High Nitrogen is associated with high chlorophyll content. According to Clevers et al. (2013), nitrogen can be used as an indicator of chlorophyll content to determine how much of the fertilizer is needed and to improve production across the field. The use of drones also known as Unmanned Aerial Vehicle (UAV) data in conjunction with machine learning and Vegetation Indices (VI) approach has shown great capabilities in monitoring chlorophyll content. Although traditional satellites and other conventional methods have successfully been used for decades, UAVs have emerged as potential alternatives for offering a cost-effective and highly spatial and temporal resolution to evaluate crop health status (Yang et al., 2022). Traditional satellites such as Landsat, Sentinel-2, RapidEye etc have been

optimized for agricultural applications, enhancing productivity and monitoring of crop health. However, Bacsa et al. (2019) indicated the benefits of integrating conventional methods with UAV as one of the best approaches to monitoring nutrient status. Substantially, all these different remote sensing platforms are significantly important, and each has its limitations and exceptional capabilities.

Hundreds of VIs have been developed and designed to assess a specific vegetation property and have the potential to differentiate healthy from diseased crops (Qiao et al., 2022). These VIs have been widely used in the agricultural industry to assess plant health and assist in nutrient management, the indices are mathematical combinations of spectral bands that highlight specific characteristics of vegetation (Vélez et al., 2023). The VIs that use the combination of red, red-edge, and near-infrared bands were selected for this study. Wong and He (2013) validated that numerous VIs have been developed focusing on both the Visible (VIS) and Near-Infrared (NIR) parts of the spectrum for estimating crop chlorophyll content. The main reason is that plants have high sensitivity in the visible and near-infrared regions. Shanmugapriya et al. (2022) suggested that chlorophyll-based indices are more sensitive than normalized indices. The results by Shanmugapriya et al. (2022) showed that chlorophyll-sensitive VIs performed better than the other indices that used green and red wavelengths.

Among the indices used in this study were the Normalized Difference Vegetation Index (NDVI), the most used index for the assessment of crop condition (Boiarskii, 2019), and Normalised Difference Red Edge (NDRE), which allows the assessment of chlorophyll activity in crops (Voitik et al., 2023). While these indices NDVI and NDRE have shown success, uncertainties and limitations persist in their application, particularly due to the reliance on spectral indices utilizing fewer bands (Croft et al., 2014). This poses challenges in remotely estimating chlorophyll content across various plant species or growth stages (Lu et al., 2019). The NDVI decreases in sensitivity in relatively dense vegetation and is affected by atmospheric contaminants (Yeom et al., 2019). The results by Qiao et al., (2022) also found that the Modified Soil Adjusted Vegetation Index (MSAVI) and NDVI saturated with the increase of chlorophyll content.

The calculated VIs were regressed and analysed to study the sensitivity of the different spectral bands and to examine their correlation with measured Soil Plant Analysis Development (SPAD) values. Marble Hall was selected to be the study area because of its agricultural significance with diverse crop types such as corn, wheat, cotton, rice, and other crops. The study will serve the purpose of validating the accuracy of VIs in measuring chlorophyll content.

Several studies have been conducted to estimate chlorophyll concentration on crops such as maize, potato, wheat, rice, cotton, and few studies focusing on citrus plantations. Qiao et al. (2022) estimated the chlorophyll content in maize and evaluated the responses of VIs under different crop coverages. Yin et al. (2023) conducted an analysis on UAV-based multispectral imaging for mapping chlorophyll content in potato crops, Wu et al. (2023) employed this approach to assess chlorophyll content in wheat crops. Such studies have proved the success of UAV-based approach in quantifying the chlorophyll content, it has been observed that the red-edge region correlates very well with the NIR region, and indices in these regions work better. The study focused on the use of high-resolution UAV imagery to estimate chlorophyll content in lemon leaves using vegetation indices and in-situ chlorophyll data for validation. Also compared the performances of different indices and regression models in chlorophyll measurement.

1.2 Problem Statement

Globally the population is increasing, and the majority of the population is facing a food crisis. There is more pressure on farmable land to produce, therefore crop management is essential crucial to providing and supporting sustainable food (Amitrano et al., 2023). One of the major contributing factors is climate change occurring at a faster rate than anticipated and Limpopo Province including the Sekhukhune District has been characterized by extreme climate events (Sikhwari et al., 2022). Marble Town is known as the agricultural hub of the region due to its extensive irrigation farming and faces the challenges of farmers having to manage both hot temperatures and electricity outages which could potentially affect fruit production (Fresh Plaza, 2022). These heatwave conditions greatly affect the quality and quantity of agricultural production (Joseph et al., 2021). There is also a lack of information about the benefits of geospatial technologies and a need for training, especially for small-scale farmers and poor communities. Many of these farmers may not be aware of the advantages that geospatial technology offers. Providing training is crucial to help them understand and effectively use these technologies, empowering them to make informed decisions and achieve better yields and sustainable agricultural practices (Smidt & Jokonya, 2022).

According to the Sustainable Development Goals (SDGs) in South Africa, Goal 2 emphasizes the necessity of promoting sustainable food production and implementing adaptable agricultural methods through utilization of advanced infrastructure and technology. The Corona pandemic globally has severely affected food security, especially for poor communities where millions were left without food and jobs (Mardones et al., 2020). To alleviate hunger

and help farmers for maximum production there is a need for transformation in food and agricultural systems (Smidt & Jokonya, 2022). The agricultural industry is integrating with remote sensing to utilize the advantages of field management and better yielding (Bacsa et al., 2019). This research can serve awareness, encourage proactive measures, and provide possible solutions.

Both small-scale and large-scale farms need to utilize the benefits of advanced technology in precision agriculture. The overuse of fertilizers, pesticides, herbicides, and other poorly managed intensive farming practices can cause environmental degradation (Rahman & Zhang, 2018), compromise food safety, and lead to increased unemployment. Measuring chlorophyll content provides valuable information for farmers to manage crop's chlorophyll levels for optimal fertilization. The use of drones is a transformative advancement for agriculture, offering many benefits such as improved data collection in a faster and more flexible way, ultimately leading to improved crop management and productivity (Istiak et al., 2023).

Farmers today are facing challenges such as climate change, price fluctuations, and high demand for agricultural land, and there is a need to farm in a more sustainable way than ever before. The future of smart farming involves the use of drones, Artificial Intelligence (AI), and sensors to collect data for monitoring crop health and yield. Integrating UAV data with other conventional methods can help to reduce risks and increase yield (Derraz et al., 2023). The town of Marble Hall in Limpopo was purposely chosen as the study area because it encompasses a large number of farms, it is the number one citrus producer in South Africa (DAFF 2017). The town is of obvious significance to the country's agricultural economy. Conventional satellite remote sensing has been extensively used for various agricultural applications, including monitoring crop health. However, UAVs offer a significant advantage by bridging the gap between high spatial and temporal resolution, allowing for faster and more flexible data collection at user-defined intervals. Therefore, there is a need to showcase the benefits of using drone-based data and advise the farmers and municipalities on the adoption of UAV data as a potential for long term investment (Bacsa et al., 2019).

1.3 Aims and objectives

1.3.1 The aim of the study

- i. The main purpose of the study is to explore the usage of high-resolution UAV imagery for non-destructive and real-time estimation of chlorophyll concentration in lemon leaves on a farm in the Marble Hall area of Limpopo Province.

1.3.2 Objectives

Specific objectives

- i. To examine spectral indices calculated from UAV imagery in predicting chlorophyll concentrations and correlate them with ground-measured chlorophyll data.
- ii. To use the Partial Least Squares Regression method to establish a predictive model for the estimation of chlorophyll content.
- iii. Generate a chlorophyll concentration map based on the best predictive model.

CHAPTER TWO

Literature review

2.1 Introduction

Farmers need to play a critical role in ensuring a balance between environmental management and food security to achieve sustainable living (Masipa, 2017). By adopting sustainable agricultural practices, they can increase their productivity while ensuring environmental sustainability (Brewer et al., 2022). Precision Agriculture (PA) jointly with Remote Sensing (RS) has pertinent answers to those requirements of balancing profitability with sustainability in the agricultural sector. Remote sensing has found numerous applications in agriculture most importantly to provide valuable information that helps farmers to make informed decisions (Liaghat, 2010). The convenience of Unmanned Aerial Vehicle (UAV) multispectral data with improved quality and high temporal resolution for chlorophyll estimation and other applications is a change maker for farmers (Tunca et al., 2024). Over the years both empirical and physical-based methods have continuously advanced improving the accuracy, reliability, and applicability of chlorophyll content estimation from UAV data (Simic Milas et al., 2018). The empirical spectral indices are perhaps the most popular and useful for quickly retrieving chlorophyll content.

Chlorophyll is a photosynthetic pigment that is found in crops plays a central role in plant productivity and is a key indicator of the health status of the crop (Qiao et al., 2022). Therefore, chlorophyll is directly proportional to nitrogen, and it determines the final yield of the crops Clevers et al. (2013). It is then very important that the farmers create a balance in supplying fertilizers in their crops while preventing harm to the environment. Climate change is a global issue with long-term negative impacts such as soil degradation, water scarcity and drought (Masipa, 2017). Among other challenges faced by farmers in Limpopo Province, they must manage both high temperatures and electricity outages, which could greatly affect the quality and yield production (Jansen, 2022). The poor communities are suffering from the adverse effects of climate change, especially small-scale farmers (Joseph et al., 2021). They lack information and training on smart agriculture for better yield and sustainability (Brewer et al., 2022). The Sustainable Development Goals (SDGs) 2 emphasizes the need to support farmers with advanced infrastructure and technologies to challenge climate change issues and promote sustainable agriculture.

Both multispectral (MS) and hyperspectral (HS) data have shown consistent results in various studies in accurately quantifying chlorophyll and other physiological variables. Many recent studies using spectral indices and UAV data have mainly focused on crops such as maize, wheat, grass, rice, and potato, and with few focusing on citrus plantations (Guendouz et al., 2012; Ban et al., 2022; Yang et al., 2022). However, a recent study by Naqvi et al. (2021) estimated the chlorophyll content in citrus groves, yielding moderate results with a DVI index of $R^2 = 0.59$. Over 150 vegetation indices have been developed to play a role in monitoring variations in vegetation (Huang et al., 2021). Normalized Difference Vegetation Index (NDV) is used to measure vegetation greenness (Boiarskii, 2019). Other indices that were selected for this study include NDRE, MSAVI, DVI, and LCI, all selected for their proven sensitivity to chlorophyll, allowing for accurate and reliable results. Several prediction models of chlorophyll estimation in agricultural fields were built based on the Partial Least Squares Regression method and have proven to be a valuable tool with effective results (Kiala et al., 2017).

It is prominent from the literature that the greatest capability of UAV data is providing high-quality real-time data that is more flexible and less affected by environmental variables (Caruso et al., 2023) conditional to the resolution of the sensor. Chlorophyll estimation can provide useful information about the crops guiding and improving crop yields (Elsayed et al., 2020). This study employed the integration of the conventional method of ground-based data collection and the implementation of UAV remote sensing in chlorophyll estimation. The usage of RS and smart technologies provides a more sustainable method that can assist in detecting early signs of food shortages (Brewer et al., 2022).

2.2 Precision agriculture and remote sensing

The world population is undeniably increasing, unemployment is rising and there is a high demand for the provision of affordable food to improve the livelihoods in rural areas where unemployment and poverty is severe (Oni et al., 2018). Rao (2023) explains that PA is an approach that utilizes specialized equipment and technology with a series of tools and strategies to maximize crop management and increase yields. This provides useful information to farmers as to what input is needed, where, and when. This approach cannot succeed without the integration of geospatial technologies, Artificial Intelligence (AI), Geographic Information Systems (GIS), and Remote sensing (RS) to make better-informed decisions (Sishodia et al., 2020). Precision agriculture involves the collection of massive data, analysis and responding on things like soil nutrients, chlorophyll estimation, presence of pesticides, weather focus, etc.

(Rao, 2023). Precision agriculture is getting more attention due to its ability to assess the health status of crops, promoting precision in planting, harvesting, or fertilizing results in a more economical, sustainable, and fast way (Sishodia et al., 2020).

This study aims to use UAV remote sensed data, along with other RS techniques to gather detailed information for assessing chlorophyll concentration in lemon leaves. This method is different from traditional farming essentially because it uses specialized technology to provide widespread information on the spatial changes in crops (Yuhao et al., 2020). Remote sensing uses multispectral/hyperspectral sensors that measure the reflected light from the crops where the health status of the crop can be evaluated based on its specific spectral characteristics in different growth stages (Mulla, 2013). Drones have transformed farming practices by providing real-time data at high spatial resolution, allowing farmers to track the location and health of the crops (Rejeb et al., 2022). These advanced tools help the farmers to improve productivity, sustainability and profitability. They play a vital role in ensuring food and nutrition security in agricultural systems by helping in the early identification of crop diseases, and nutrient requirements, enabling targeted actions (Bacsa et al., 2019).

There are still challenges faced by the agricultural sector, especially in poor regions with challenges such as climate change, scarcity of water, and land degradation which can result in less yield production. According to the report by Jansen, (2022), the farmers in Marble Hall are faced with heat waves throughout the province, plus the impact of load shedding could pose a large risk to fruit production. The mandate for most agricultural departments is to grow the economy and food security through the practice of sustainable agriculture (Oni et al., 2018.). Therefore, there is a need to intensify research and transform our poor communities with advanced technology for production, management, and cost control strategies. Farmers should be trained and educated about evolving technology, especially small-scale farmers in poor communities. Consequently, these solutions may be economically impractical for small-scale farms that lack the affordability of these advanced machines, lack the necessary skills, and resources for someone to operate (Istiak et al., 2023).

2.3 Chlorophyll estimation using UAV multispectral data

Remote sensing technology has rapidly advanced, and there is increasing interest in using UAVs because of their flexibility, affordability and superior quality which are essential for precision agriculture applications (Wang et al., 2023). The utilization of UAVs has enabled the collection of data with enhanced resolution, promoting informed decision making aimed at increasing production (Naqvi et al., 2021). Currently, there is a variety of RS sensors available that are used for chlorophyll estimation from multispectral and hyperspectral sensors operating at different spectrums with different spatial resolutions (Wang et al., 2022).

It is evident from the literature that multispectral Remote Sensing is efficient in agricultural applications. Multispectral-mounted drones collect and record light across different spectral bands in the visible spectrum and beyond. These multispectral cameras usually detect lights in three to five spectral bands (Tunca et al., 2024). The greenness produced by the healthy leaf is because of the absorbing the red and blue radiating lights and reflecting the green in the wavelength from 500 – 600nm. As the red light is absorbed between 600 – 700nm and beyond which the radiated light gets reflected from the red edge in the NIR region. The NIR provide valuable information about the physiological status of crops (Sishodia et al., 2020). Another important band is the red edge for the assessment of crop chlorophyll content. Literature has proven this region is useful in estimating chlorophyll and nitrogen content. Yuhao et al. (2020) indicated the importance of red edge-based indices to estimate chlorophyll and conclude that it has great potential for agricultural application.

The greatest benefit of using UAV multispectral imagery is that it provides high-quality data, uses non-destructive methods, and more flexible and convenient responses to crop-related problems (Wang et al., 2022). Drone data aims to exceed some of the limitations associated with previous multispectral sensors providing higher spatial and temporal resolution (Priyanka et al., 2023). Wang et al. (2022) used both MS and HS UAV-based data to estimate the sugar content of beetroot, and HS showed higher accuracy. Both these platforms allow an accurate exploration of various environmental characteristics and to understand the dynamics and processes occurring at ground level (Qi et al., 2021). This study used multispectral data at 3cm spatial resolution to evaluate and compare the performance of selected indices.

2.4 Vegetation indices in precision agriculture

The advancement of technologies like GIS, GPS, Remote Sensing, and other smart systems makes crop management a possibility (Sishodia et al., 2020). Remote sensing and vegetation indices (VIs) are considered as powerful tools in precision agriculture, enabling the assessment of plant health and photosynthetic activity in a non-destructive approach (Sun et al., 2021). The vegetation index is the transformation of multiple spectral bands into a single value that highlights different vegetation components such as biomass, photosynthetic activity, or health status of crops (Yang et al., 2022). Chlorophyll-based indices are sensitive in the red or visible region, while the near-infrared reflectance is sensitive to the photosynthetic tissues (Milas et al., 2018). The combination of traditional vegetation indices and chlorophyll-specific indices has been proposed to minimize the weaknesses associated with each while maximizing sensitivity to chlorophyll content providing a more accurate and reliable assessment of plant health (Naqvi et al., 2021). For chlorophyll estimation, farmers use VIs derived from satellite images or remote-sensing drones to analyse trends in crop health.

There has been incredible progress over the years in the use of remotely sensed data for retrieval of biophysical variables in vegetation, using vegetation indices (Huang et al., 2021). The NDVI is the most popular and widely used vegetative index developed by Rouse et al. (1973). The VIs are compiled using different mathematical formulas and designed to highlight different aspects of vegetation. The aim of this study is to select indices that can accurately reflect changes in chlorophyll levels through the use of UAV data. The NDRE Index is more responsive to the chlorophyll of the leaves and less to background effects (Boiarskii, 2019). The MSAVI index minimizes soil effects and emphasizes data from vegetation (Voitik et al., 2023). Leaf Chlorophyll Index (LCI) can monitor vegetation health through all seasons, sensitive to slight variations in reflectance caused by chlorophyll absorption (Qi et al., 2021). The Difference Vegetation Index (DVI) is one of the simplest indices developed by Richardson and Wiegand in 1977 to differentiate between water bodies, bare soils, and vegetated areas. It is clear from the listed VIs that NIR (700 – 1300nm) is an imperative region due to its sensitivity to vegetation health and biomass (Caruso et al., 2023).

There are limitations associated with VIs that can affect their reliability in estimation of chlorophyll content. These include sensitivity to environmental factors, saturation effects, temporal variability, and other factors (Qi et al., 2021). To overcome these limitations more revised and modified indices have been created over the years. Therefore, addressing these

drawbacks requires careful consideration and validation against ground truth data (Singhal et al., 2019). While hundreds of vegetation indices have been proposed, an appropriate index selection is very important to eliminate redundancy (Zhang et al., 2024). Numerous studies have been validated and proven successful in their estimation of biophysical attributes such as chlorophyll concentration, Leaf Area Index (LAI), and nitrogen concentration. These attributes are important to understand plant health, productivity, and environmental conditions (Bacsa et al., 2019).

2.5 Partial Least Squares Regression model for predicting chlorophyll content

Regression models are statistical tools powerful in remote sensing for non-destructively assessing relationships between spectral measurements and vegetation parameters (Quang et al., 2022). To generate precise diagnoses of the nutritional status of the crops, there is a need to employ a correlation-based evaluation method. The Partial least Squares Regression (PLSR) technique is a nonlinear and most popular technique that is commonly used to quantify spectral analyses (Jin et al., 2013). To test the goodness of fit of the models, we use the Root Mean Square Error (RMSE) and R-squared; generally, a good regression model should give high R-squared and lower RMSE values (Yin et al., 2023).

There is a collection of literature based on using regression models in predicting chlorophyll and quantifying relationships between applied techniques to evaluate its performance. The findings of several studies have confirmed PLSR to be a robust technique that has shown recurring accurate predictions for explaining vegetation features (Wang et al., 2017). Several studies compared parametric statistics, physical-based methods, vegetation indices, and PLSR and have shown outstanding performances in predicting Canopy Chlorophyll (Chen et al., 2023). Qiao et al., (2022) also compared two models' performances between PLSR and RF to estimate maize chlorophyll estimation, the findings show the PLSR performed better with NDRE as one of the optimal spectral variables. The PLSR technique is considered as a powerful modelling tool that specifies linear relationships between a set of dependent and independent variables (Lu & He, 2019). One of its advantages is that it does not need a huge sample size and can be used to suggest whether relationships exist or not (Du & Luo, 2024). This method is appropriate to deal with multicollinearity and to source important information regarding multiple variables, mostly spectral data (Silalahi et al., 2021). Regression models will serve the purpose of this study: to practice non-destructive estimation of chlorophyll content and to observe significant correlations between vegetation indices and in-situ measurements.

CHAPTER THREE

Materials and Methods

3.1 Study area

The study was conducted on a farm next to the Marble Hall area (Figure 3.1), which is a small town situated in the Sekhukhune District on the southern edge of the Limpopo Province in South Africa. The region is located 24.9651° S and 29.2815° E approximately 26 km from Groblersdal. The province is enriched with agricultural resources, serving as a key contributor to the production of vegetables and fruit (Sithole et al., 2023). This region is well known for its extensive irrigation farming, where high production of crops such as cotton, wheat, and citrus meets both national and international demand making the region a significant agricultural hub (LDARD, 2018).

The region is semi-arid, featured by low rainfall distribution, and sustained by the pivot farming irrigation system in the Loskop dam floodplains which allows all-season planting of crops (Taona, 2019). The Greater Sekhukhune District is largely rural, and the agricultural sector is the primary source of employment and livelihood (Oni et al., 2015). It is also rich in mineral deposits and takes its name the Marble Lime Mine in the area. The province is susceptible to various forms of degradation and climate change which may hinder agricultural production (Mpandeli et al., 2015).

Ephraim Mogale Local Municipality is situated near Loskop Dam it is the tourist hub of the district with several attractive natural areas offering great ecotourism and game farms (EPMLM, 2018). The municipality is characterized by immense geological structures that hold the world's biggest reserves of platinum group metals. Marble Hall's climate consists of summer average temperatures of between 15-31°C and winter months have average temperatures ranging from 4-23°C. The town normally receives an annual average precipitation of 634mm with most of the rainfall received in summer (COGTA: Sekhukhune, 2020). The rich soils and climate are devoted to the thriving agriculture in the province and the region boasts citrus farms. EL Sundew farm, the study area, is one of biggest citrus producers in the area getting water supply from the Loskop Dam enabling all-season planting of crops.

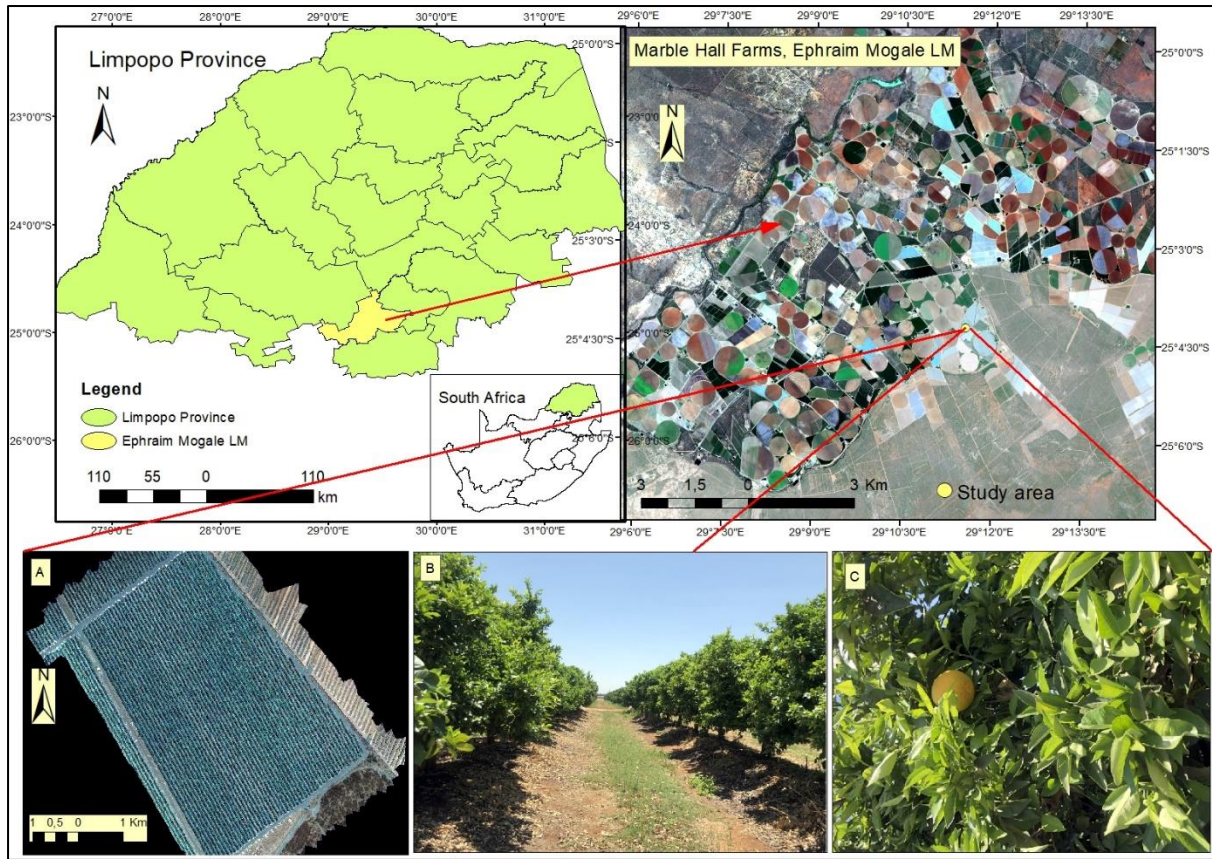


Figure 3.1: Shows the study area, EL Sundew Farm in Marble Hall, (A) UAV imagery of the study area (B), & (C) pictures of the lemon trees.

3.2 Data Collection

3.2.1 UAV image acquisition and processing

For image collection, a high-resolution multispectral Unmanned Aerial Vehicle (UAV) imagery was used. The data was collected in cloudless weather conditions with the sun at about 24-29°C on the 3rd of October in conjunction with in-situ chlorophyll data and GPS data collection. The target site imagery was acquired using DJI Matrice 300 RTK (M300) aircraft (Figure 3.1), known for its high performance and exceptional reliability. RGB and multispectral imagery were captured using M300 with two sensors simultaneously: DJI Zenmuse P1 RGB mapping camera and MicaSense RedEdge-MX/Dual multispectral sensor (Brewer et al., 2022). It weighs approximately 6kg, has up to 55 min flight time, and has a maximum flight speed of 23 m/s. It offers advanced AI capabilities, 6 Directional Sensing & Positioning (DJI enterprise). The flight time for the drone was between 11 am and 2 pm with a spatial resolution of

approximately 3 cm, and the landing spot was the same as the take-off spot. The multispectral cameras collected multispectral images in five bands.

3.2.2. Image pre-processing

Image processing was carried out using Agisoft Metashape software, which included geometric corrections and image stitching. An ortho-mosaic image was created by stitching the subsets of aerial photographs collected by the drone. The resulting image was a composite TIFF file with six spectral bands, displaying the study area as shown in Figure 3.1 (A).



Figure 3.2: A photo of the DJI Enterprise Matrice-300 series

Table 3.1: Multispectral sensor bands for the AUV image used for this study.

Sensor type	Sensor name	Band	Wavelength (nm)	Bandwidth (nm)	Spatial Resolution
Multispectral	MicaSense	Blue	450-590	32	3cm
		Green	530-570	27	3cm
		Red	640-680	14	3cm
		Red-Edge	730-740	12	3cm
		Near-Infrared	770-810	57	3cm

3.2.3 Ground Measurement of Chlorophyll Content

On the 3rd of October 2023, the study area was visited for field measurements at the harvesting stage of the lemon fruit. During the UAV data collection, ground chlorophyll data was also collected using a portable chlorophyll meter and a GPS (Global Positioning System). In situ measurement of leaf chlorophyll content in lemon leaves was conducted using a handheld device known as the SPAD chlorophyll meter. The SPAD-502 meter was used to calculate the relative amount of chlorophyll present in the crop by measuring the absorbance of the leaf in the red and near-infrared regions. This helps users to understand the condition of the crop and determine the optimal timing for fertilizer application (Raymond Hunt & Daughtry, 2014).

From each sample tree, five measurements were taken using a SPAD meter on different zones of the leaf from the middle to the upper level of the leaf. These five measurements were averaged to get precise results for the chlorophyll content of that sample tree. Abnormal records were excluded to ensure chlorophyll values between 40 and 60. The mean value was then calculated and recorded as the chlorophyll content of that sample tree. A total of 43 samples were collected and geolocated using a Garmin GPS meter each point corresponding to the location of the leaf samples. DNRGPS open software was used to download the GPS coordinates points and convert them into a shapefile. The shapefile was then imported into ArcMap 10.8.3 to view the location of the points. All the sample data was divided into two groups according to the ratio of 70:30, where 32 samples were used as training samples and 11 samples were used as validation samples of the model.

3.3 Spectral Vegetation Indices Calculated

To retrieve chlorophyll content and gain a better understanding of the vegetation status of the study area, five Vegetation Indices (VIs) known to have high correlation with crop growth status were selected and derived from AUV-based remotely sensed data (Milas et al., 2018). The selected VIs are closely related to crop growth status and provide valuable information about various aspects of plant health, including chlorophyll content, biomass, and overall health (Vélez et al., 2023). This study adopted five near-infrared (NIR) VIs to estimate chlorophyll content in lemon leaves. Yeom et al. (2019) compared the performance of NIR and RGB VIs both derived from UAV data and proved that NIR VIs are superior for agricultural analysis. It is therefore critical to select indices such as MSAVI, which compensate for and overcome various environmental interferences such as influence of soil background on VIs to achieve effective results (Boiarskii, 2019).

More improved indices with better estimation stability emerged to overcome the challenges faced such as soil background and other environmental disturbances (Naqvi et al., 2021). The VIs included the NDVI, NDRE, MSAVI, Difference Vegetation Index (DVI), and Leaf Chlorophyll Index (LCI). The NDVI developed by Rouse et al. (1973), it uses the red and near-infrared regions and has been used successfully to indicate crop health. As popular as it is, it is associated with the limitation of soil background interferences, atmospheric effects, and saturating effects (Huang et al., 2021). Even though it is a conventional index several studies have shown that NDVI is an efficient method for leaf chlorophyll content estimation especially in precision agriculture. In the study by Qi et al. (2021), eight indices were tested and NDVI showed the highest precision than other indices in chlorophyll estimation of peanut crops. It is also known that NDVI does saturate and may not always be the most effective index to determine the level of chlorophyll and that is why other indices such as red edge NDVI (RE-NDVI), modified red edge NDVI (mNDVI1705), and red edge position (REP) were developed.

The NDRE index uses the red-edge wavelength, which is more responsive to leaf chlorophyll and less influenced by background effects. This makes NDRE mainly useful for assessing plant health. Boiarskii (2019) emphasized that the NDRE can detect subtle changes in chlorophyll content, allowing easy identification of areas that might need fertilization. MSAVI is closely related to the estimation of crop chlorophyll and growth status. It offers a better approach to

addressing some of the NDVI's limitations related to soil reflectance by accounting for this factor and yielding more accurate results (Voitik et al., 2023).

The DVI index was developed to assess vegetation cover and biomass within a specific area, it is one of the simplest indices developed by Richardson and Wiegand in 1977 designed to differentiate between water bodies, bare soils, and vegetated areas. Naqvi et al., (2021) Showed that DVI can be effectively used to estimate chlorophyll content in citrus plants. The study concluded that DVI has less sensitivity in shrubs and grassland but is highly sensitive to vegetation cover and suitable for plant biomass. Narmilan et al. (2022) also revealed a significantly positive correlation between DVI and chlorophyll content of sugarcane crops.

Another chlorophyll indicator with significant benefits is the LCI; it can monitor vegetation health throughout all seasons and is sensitive to even slight variations in reflectance caused by chlorophyll absorption (Qi et al., 2021). Despite the challenges and variations in application among various VIs, they still produce robust and effective results in chlorophyll estimation (Hung et al., n.d.). The next step after image processing was to use the stacked multispectral image imported in ArcMap (10.8.2) software to generate corresponding vegetation index images. The values of reflectance in spectral bands were used to generate vegetation indices, and vegetative maps were produced. The vegetation index formulas utilized for the study are presented in Table 3.2 below.

Table 3.2: Vegetation indices and formulas derived from UAV imagery selected for this study.

Vegetation Index	abbreviation	Formula	Reference
Normalized Difference Vegetation Index	NDVI	$\frac{NIR - R}{NIR + R}$	Rouse et al. 1973
Normalized Difference Red Edge	NDRE	$\frac{NIR - REEDGE}{NIR + REEDGE}$	Fitzgerald et al. (2006)
Difference Vegetation Index	DVI	$NIR - R$	Richardson and Wiegand, 1977
Leaf Chlorophyll Index	LCI	$\frac{NIR - REEDGE}{NIR + R}$	Jordan, 1969
Modified Soil Adjusted Vegetation Index	MSAVI	$\frac{1.5 * (NIR - RED)}{(NIR + RED) + 0.5}$	Ren et al., 2011

3.4 Statistical Analysis

3.4.1 Pearson Correlation

The Pearson correlation coefficient (r) was used to measure the linear relationship between VIs, spectral bands, and ground-measured chlorophyll levels. The correlation coefficient was calculated to evaluate the strength of these relationships and to identify which VI or spectral band has the strongest correlation with measured chlorophyll. The mathematical formula for calculating r is shown in equation (1) where VIs are treated as the independent (x) and measured chlorophyll as dependent variables (y).

$$r = \frac{\sum_{i=1}^n (x_i - \bar{x})(y_i - \bar{y})}{\sqrt{\sum_{i=1}^n (x_i - \bar{x})^2 \sum_{i=1}^n (y_i - \bar{y})^2}} \quad (1)$$

3.4.2. Partial Least Square Regression

The PLSR method was selected as the model to estimate chlorophyll using R statistical programming language software package version 4.3 to build the model and produce statistical analysis. The PLSR is a widely used statistical technique for quantifying spectral analyses (Jin et al., 2013). Previous studies have highlighted the benefits of using the PLS model over other modelling techniques, as it integrates aspects of principal component regression, correlation analysis, and multiple linear regression analysis (Jin & Wang, 2019). The PLSR is very useful for multivariate data with correlations among independent variables, as it summarizes the data into small non-correlated components that can be used in the model. It builds a linear model as follows (Li et al., 2020):

$$Y = X\beta + \varepsilon \quad (2)$$

Where X and Y , refer to a mean-centered vector of independent and dependent variables, β refers to the regression coefficients, and ε refers to the regression residuals (Li et al., 2020).

3.4.3 K-Fold cross-validation

In this study, a repeated k-fold cross-validation approach was implemented to assess and test the model's performance by dividing the dataset into training and testing data. This approach is particularly suitable for small datasets, and it offers a much less biased estimate of model performance (Ryan & Ali, 2016). This method involves randomly splitting the training set into k smaller subsets. It is then trained using k-1 folds, with the remaining fold used as a test set to validate the model. This process is repeated up to a certain number of times. The primary metrics for evaluating prediction performance are R^2 , RMSE, and MAE

To assess the estimation precision of the PLS regression model, the coefficient of determination (R^2) and Root Mean Squared Error (RMSE) were selected as evaluation parameters. Lower RMSE values and high R-squared generally indicate a good regression model. The equations of R^2 and RMSE are represented by equations (3) and (4).

$$R^2 = \frac{\sum_{i=1}^n (y_i - \hat{y}_i)^2}{\sum_{i=1}^n (x_i - \bar{x})^2} \quad (3)$$

$$RMSE = \sqrt{\frac{\sum_{i=1}^n (y_i - \hat{y}_i)^2}{n}} \quad (4)$$

y_i indicates the actual values, $\hat{y}_i$ indicates predicted values, x_i is the observed values, and n is the number of variables.

To measure and assess the performance of a model on a dataset is by using the Mean Absolute Error (MAE). This metric was used to measure how close the predictions made by the model were to the actual data. A smaller MAE shows that the model's predictions are closer to the observations. The formula is calculated as follows.

$$MAE = \frac{\sum_{i=1}^n |y_i - \hat{y}_i|}{n} \quad (5)$$

Where n total number of observations, x_i and y_i are the predicted and measured values respectively.

3.5 Mapping chlorophyll spatial concentration

Two PLSR models were developed: one based on vegetation indices (NDVI, NDRE, MSAVI, LCI, and DVI) and another one based on UAV bands were created. The model based on UAV-based spectral bands achieved the highest estimation accuracy and was used further to create the chlorophyll concentration map for the study area. The best performed predictive model was chosen based on the following evaluation parameters R^2 , RMSE, and MAE. The selected model demonstrated the highest R^2 and the lowest values of RMSE and MAE. The resultant map shows the estimated values of chlorophyll representing chlorophyll concentration across the study area.

CHAPTER FOUR

Results

4.1 The variations in the spectral characteristics of leaf reflectance

The mean spectral reflectance curve for lemon leaves illustrates the normal spectral behaviour for green leaves. The graph provides an overview of how these leaves reflect different wavelengths and allows for the quick, and non-destructive assessment of chlorophyll content (Figure 4.1). As shown, chlorophyll absorbed blue and red light while reflected green light which corresponds to the green colour of the leaves. An increase in reflectance occurred in the red edge with the maximum reflectance found in the Near-Infrared region, indicating healthy leaves. This information is crucial for understanding the health and properties of vegetation.

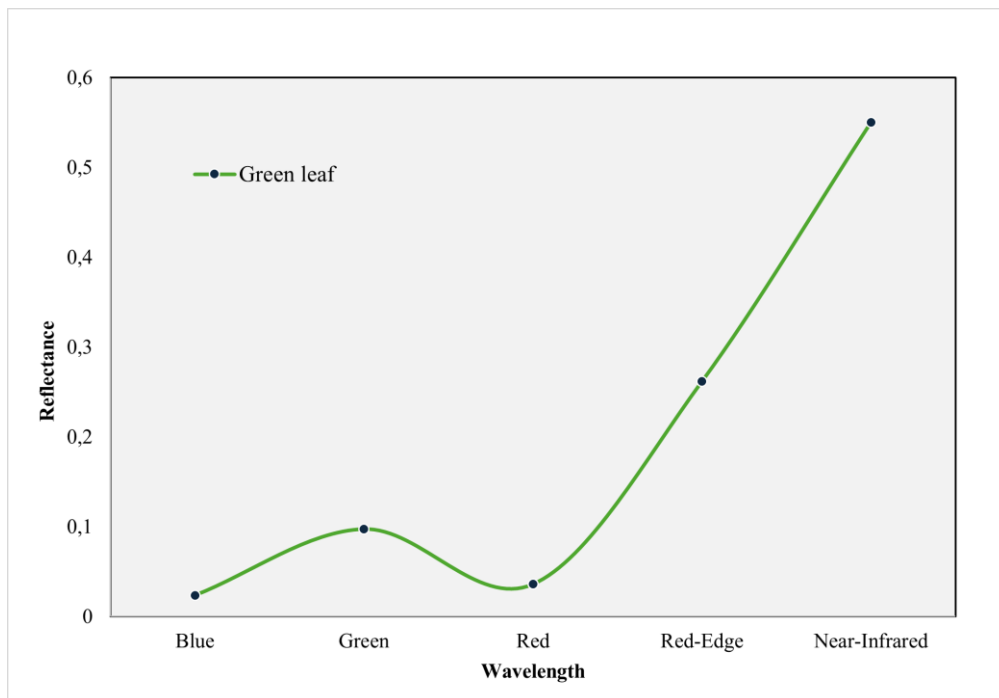


Figure 4.1: Assessment of chlorophyll content by reflectance spectra of lemon leaves.

4.2 Descriptive statistics of measured chlorophyll

The following analysis of 43 samples reveals a better understanding of the variability and distribution of chlorophyll content within the study area. Table 3 shows the statistical summary of the measured leaf chlorophyll content, with chlorophyll values ranging from 42.4 to 62.5. The minimum chlorophyll value recorded was 42.4, while the highest chlorophyll concentration was 62.5. The mean chlorophyll value was 50.3, with a standard deviation (SD) of 4.8 and a coefficient of variance (CV) of 9.5. The CV value indicates a low degree of variability in chlorophyll values across the study area.

Table 4.1: Statistical summary of the measured chlorophyll

	Samples	Minimum	Maximum	Mean	Standard Deviation	Coefficient of Variance (%)
SPAD						
	43	42.4	62.5	50.3	4.8	9.5

4.3 Correlation analysis between UAV spectral bands and measured chlorophyll

The Pearson correlation was used to determine the spectral bands that had the strongest correlation with measured chlorophyll. The scatter plots represent the correlation coefficients (r) between UAV spectral bands and measured chlorophyll (Figure 4.1). The analysis revealed that the visible spectrum bands (blue, green, and red) exhibited the lowest correlation with chlorophyll content. The findings are consistent with the known behaviour, where chlorophyll absorbs well in the blue and red region. Therefore, the lowest correlation was observed with the blue band with $r = 0.04$. The highest correlation appeared in the near-infrared and red edge at $r = 0.89$ and $r = 0.57$ respectively. In this transition zone, chlorophyll strongly reflects in the NIR hence showing the strongest sensitivity to chlorophyll levels.

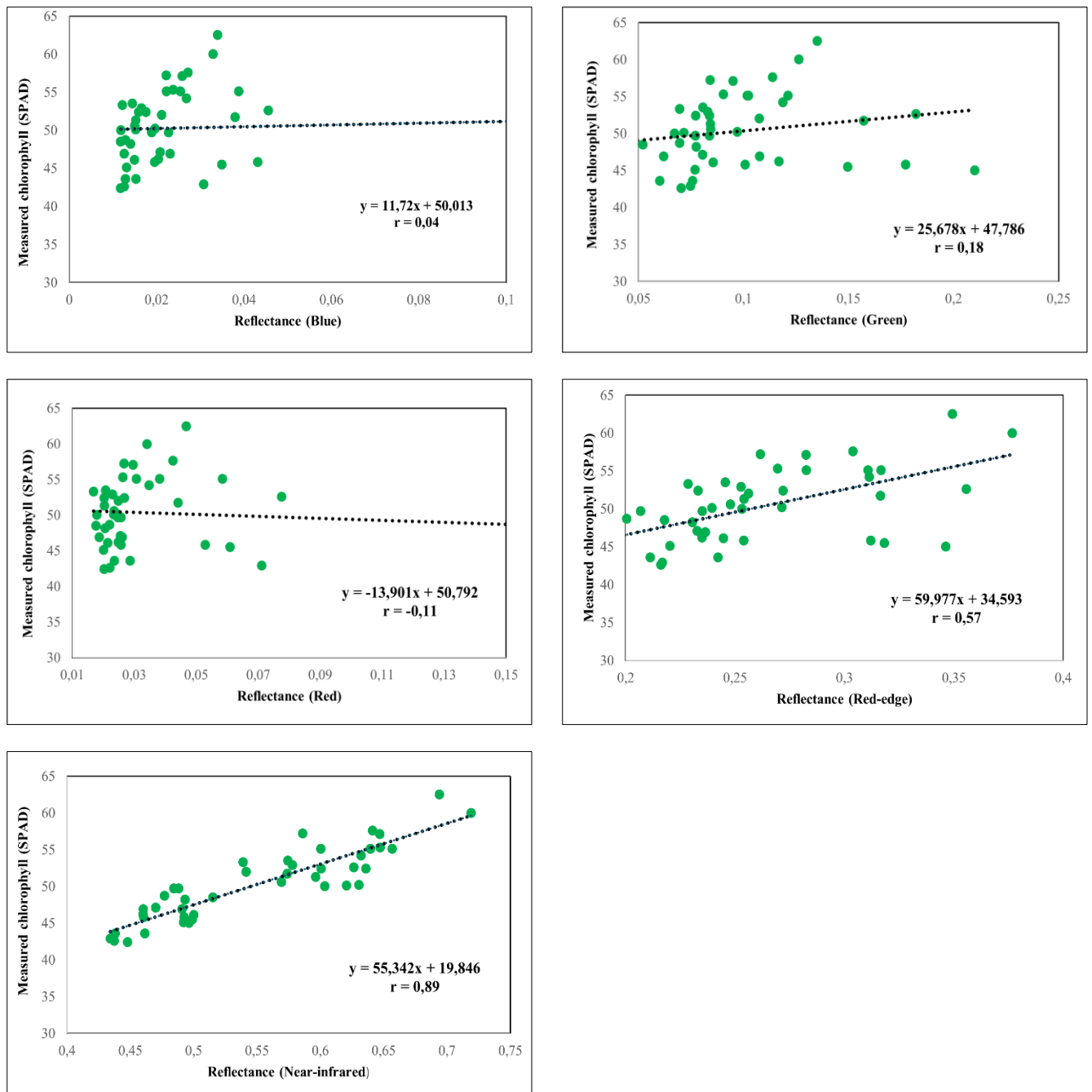


Figure 4.2: This figure displays the correlation coefficients (r) between the spectral reflectance values of different UAV bands and the measured chlorophyll content in lemon leaves.

4.4 Correlation analysis between vegetation indices and ground-measured chlorophyll

The correlation analysis was performed to evaluate the strength of the relationship between various VI and measured chlorophyll content. Five indices NDVI, NDRE, MSAVI, LCI, and DVI were tested to determine their sensitivity to crop chlorophyll as shown in scatter plots in Figure 4.2. The correlation coefficients (r) were used to assess the strength of the relationship between the two variables. All five VIs showed a positive correlation with measured chlorophyll, with r values ranging from 0.22 to 0.85, indicating that higher values of these indices are associated with higher chlorophyll values.

The results show that the combination of strong absorption in the visible region and strong reflectance in the near infrared yielded the strongest correlations with measured chlorophyll in this study. Specifically, the highest correlation coefficient was 0.89 between DVI and measured chlorophyll. This was followed by MSAVI at $r = 0.47$. On the other hand, VIs that used the combination of the red edge and NIR showed relatively weaker correlations. Among these, the NDRE obtained the lowest correlation.

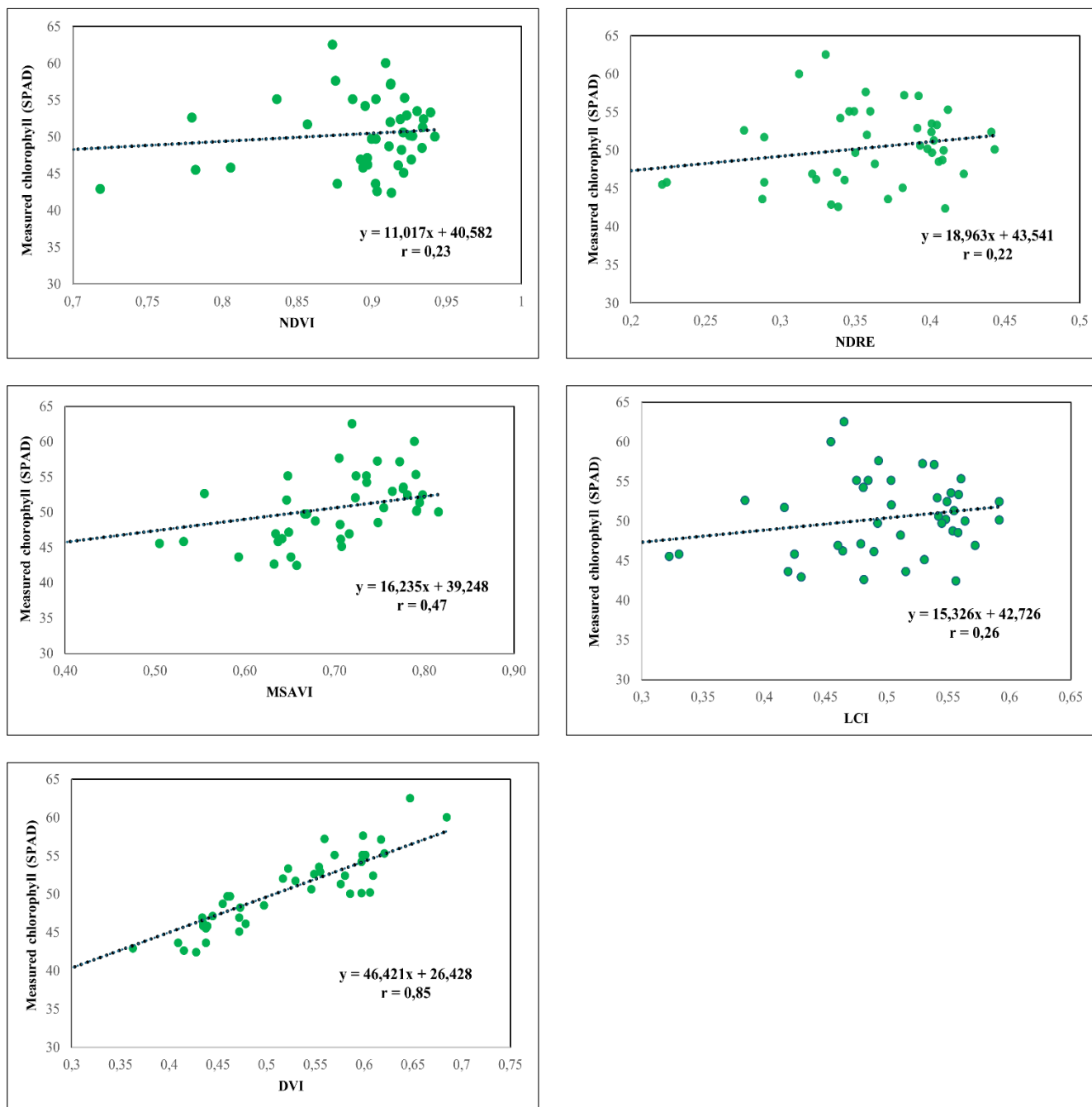


Figure 4.3: This figure displays the correlation coefficients (r) between vegetation indices and measured chlorophyll content in lemon leaves.

4.5 Model construction based on Partial Least Squares Regression

The PLSR technique was used to predict leaf chlorophyll, and a comparison was made between the models using the following performance metrics: R^2 , RMSE, and MAE. These metrics were employed to select the model that yielded the highest performance. Estimation models were created using all UAV spectral bands and the five vegetation indices (i.e., NDVI, NDRE, MSAVI, LCI, and DVI) utilized in this study.

The performance results for each model are presented below, both of which achieved high performances with R^2 values greater than 0.8. The models produced similar results with R^2 values of 0.85, RMSE ranging from 2.11 to 2.26 and MAE ranging 1.79 to 1.93. The model based on UAV-based multispectral bands performed slightly better, with smaller RMSE and MAE, further analysis was done using this model.

A 10-fold cross-validation repeated 5 times, was chosen to determine the optimal number of components for model development (Berrar, 2019).

Table 4.2: Performance evaluation between the two models of PLS Regression

Regression Model	Predictor variables	R^2	MAE	RMSE
PLSR	UAV Bands	0.85	1.79	2.11
	Vegetation Indices	0.85	1.93	2.26

4.6 Analysis and validation of the estimation models

Two PLSR models were developed to estimate chlorophyll content, considering two sets of input factors: one based on various VIs (including NDVI, NDRE, MSAVI, DVI, and LCI) and the other based on UAV spectral bands. The results are presented in scatter plots to assess how well the models' predictions align with the actual observed data.

As shown in Figure 4.4, a tight clustering of points around the 1:1 line is evident for both models, indicating high predictive accuracy. This tight clustering signifies that the models' predictions are highly aligned with the actual observed values.

In comparison, the model based on all VIs, although also achieved an R^2 of 0.85, had a slightly higher RMSE. These findings highlight the effectiveness of using UAV spectral bands and selected VIs in estimating chlorophyll content. The improved performance of the UAV bands suggests that these bands may provide more relevant or precise information for chlorophyll estimation than the VIs alone.

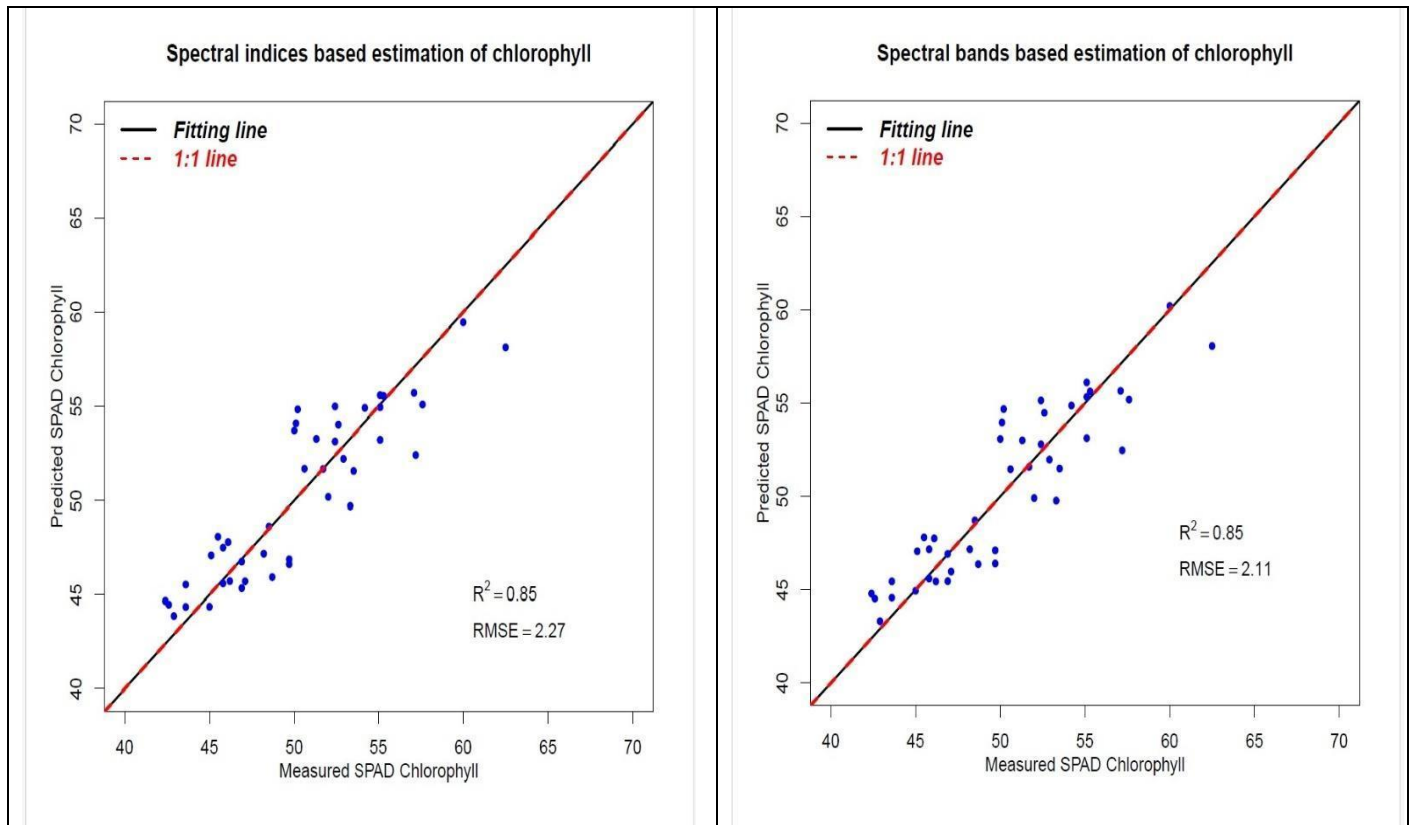


Figure 4.4: The relationship between measured versus predicted chlorophyll using Partial Least Square Regression

4.7 Variable Importance for the PLSR model

Below are the Variable Importance (VIP) score plots, showing which predictors had the most significant influence on estimating chlorophyll content. The most suitable predictor variables identified were NIR, DVI, NDVI, RE, and LCI; these variables are ranked from the most important to the least important as highlighted in Figure 4.4. The NIR band emerged as the most significant predictor, these results are consistent as it is an essential band for assessing crop health. DVI is another key indicator reflecting its ability in capturing differences in vegetation reflectance. These results show the influence of both spectral bands and vegetation indices in chlorophyll estimation.

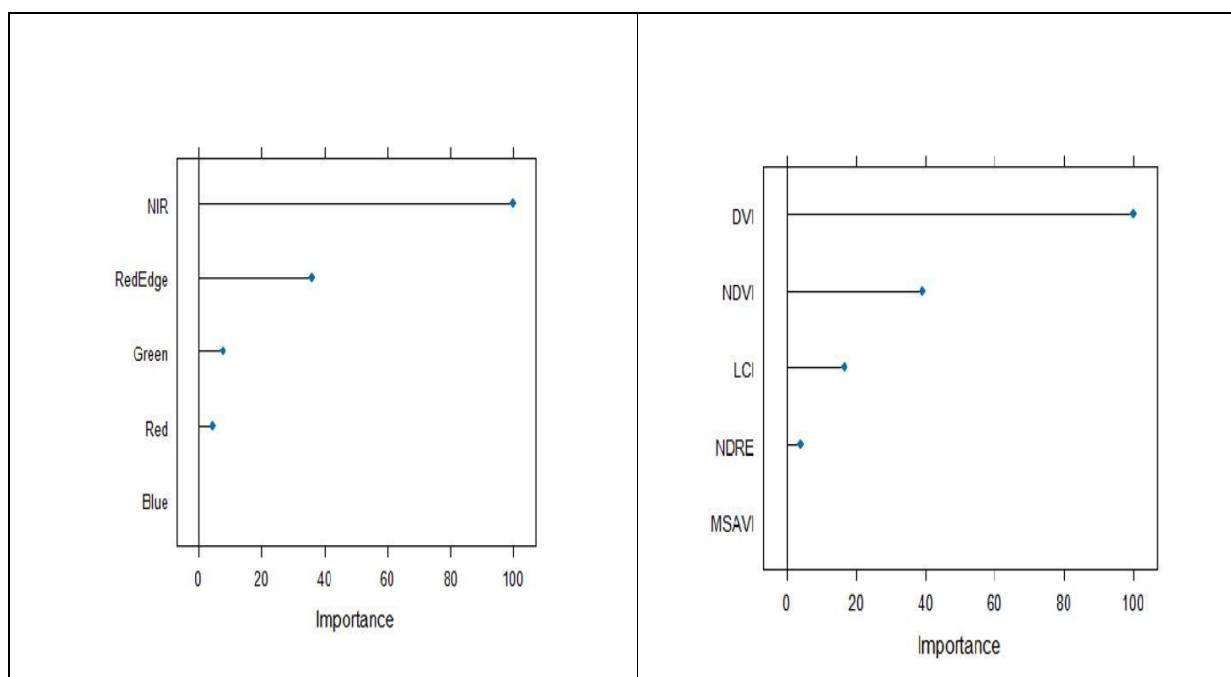


Figure 4.5: Variable Importance Plot for UAV raw bands and vegetation indices that were used in predicting chlorophyll.

4.8 Chlorophyll concentration map

The PLSR models both yielded similar accuracy results with an R^2 value of 0.85. However, the predictive model based on UAV spectral bands performed slightly better, as indicated by lower RMSE (2.11) and MAE (1.79) values. Consequently, the chlorophyll concentration map was created using this model. Figure 4.5 represents the spatial distribution of predicted chlorophyll levels across the study area, EL Sundew Farm generated by the PLSR model.

The resultant map shows a landscape of row cropland of lemon trees with estimated chlorophyll values ranging from 45 to 70. These estimated values are relative to chlorophyll values that were measured in the field but slightly higher with the highest chlorophyll value of 70. The chlorophyll values correspond to shades of blue, representing the row crops. Lower chlorophyll values are represented as small patches of green, observed between rows indicating the presence of weeds or small grass in the pathways. Non-vegetated areas and shadows are also represented on the map.

The map illustrates different chlorophyll levels across the study area. For instance, the eastern region of the field has lower chlorophyll concentration, while higher concentrations are observed towards the western part of the field. This information is of great importance as it can assist farmers in implementing precise, site-specific chlorophyll management practices.

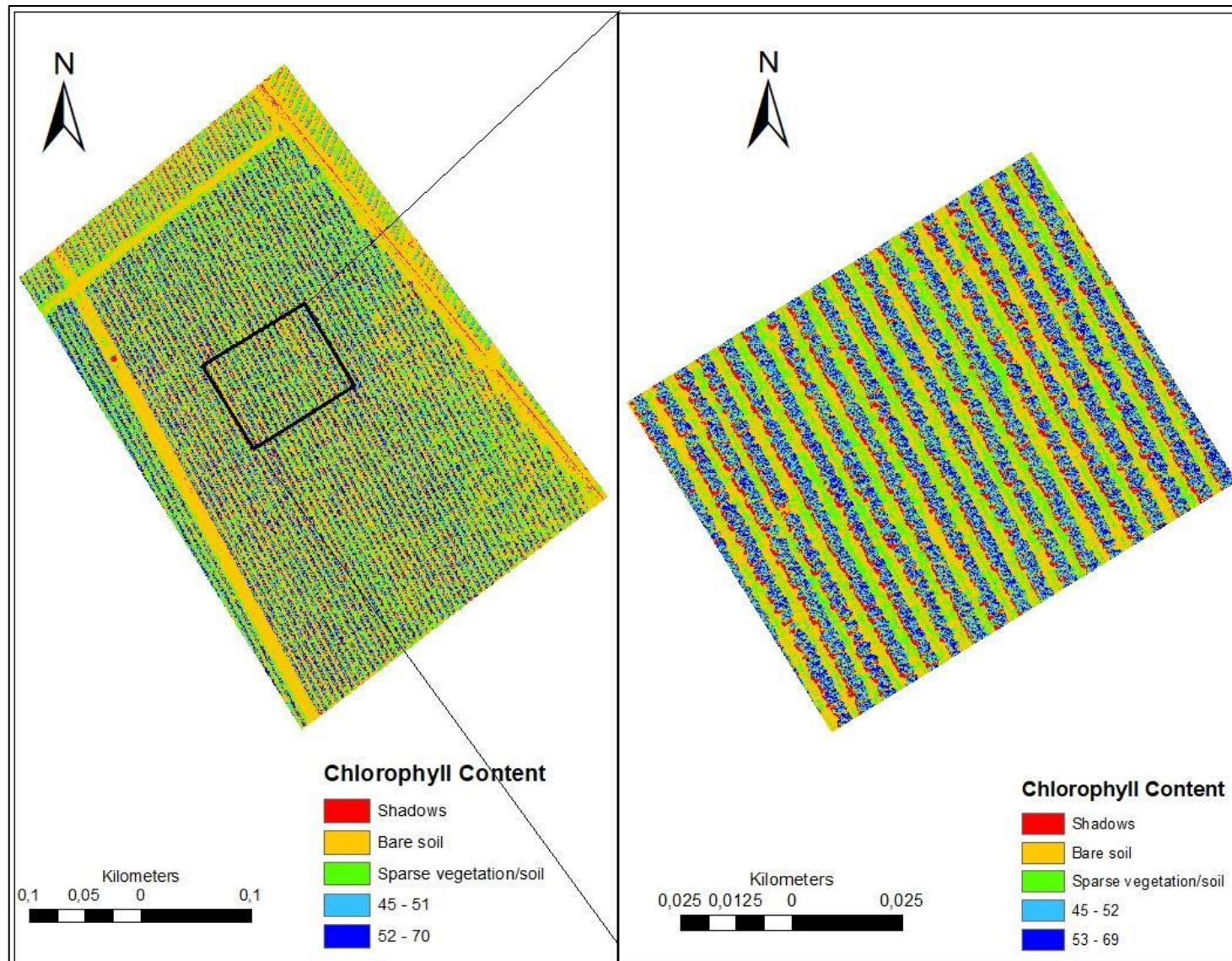


Figure 4.6: Chlorophyll Content distribution in the study area derived by applying the PLSR model.

CHAPTER FIVE

Discussion

5.1 Remote sensing and ground-based data collection approach

Ground-based data involves direct observations of the field and data collection using ground-based devices such as GPS, and SPAD meter. Field surveys serve the purpose of thorough investigation of the area of interest. Conventional methods for estimating chlorophyll content in crops such as field sampling or chlorophyll extraction are often destructive, can be expensive, time-consuming, and difficult to access in certain areas (Adam et al. 2014). In this study, a SPAD meter which calculates the relative amount of chlorophyll present in the crop was used in the field and this data was used to validate the data collected by UAV.

Remote Sensed data offers several advantages over field-based data such as retrieving the physiological properties of the crops on a non-destructive method and can cover large areas with a single image. UAV technology has emerged with the potential to conveniently allow farmers to monitor their crops and keep watch on their health status. It offers high spatial and temporal resolution, allows flexibility of data collection, and better accurate results. Gargiulo et al. (2023) verified various methods for improving accuracy in chlorophyll estimations. The results demonstrated that ground data can validate remotely sensed data, underscoring the importance of using various techniques to improve accuracy such as vegetation indices, radiative transfer models, machine learning, and ground truth data validation. This highlights the necessity of integrating different methods for precision results. The combined use of UAVs, remote sensed data, and ground-measured chlorophyll proved to be a reliable approach for monitoring nutrient status.

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5.2 The analysis of spectral characteristics of UAV bands, VIs, and chlorophyll content of lemon

The research findings show that among the five spectral bands, those in the visible spectrum showed the weakest correlations with measured chlorophyll, likely due to strong absorption in the blue and red bands (Yang et al., 2022). These results follow the typical spectral pattern of healthy vegetation, as confirmed by the study by Guendouz et al. (2012). A normal plant growth will decrease the reflection in the blue and red band and increase absorption with abrupt reflection in the NIR region (Wang et al., 2023). Reflection peaks were observed beyond the visible region, where the highest correlations ($r \geq 0.5$) were found (Figure 4.2). Previous studies have shown strong correlation in bands beyond the visible spectrum, collaborating the results by Xie et al. (2018), which identified the strongest correlation in the RE and NIR regions. Wu et al. (2023) demonstrated significant variations between different growth stages and the influence on spectral reflectance. In addition, the reflectance observed in the RE and NIR regions may also be influenced by the canopy and leaf structure (Qiao et al., 2022).

The VIs were also correlated with chlorophyll content to evaluate their relationship. The results revealed that VIs that used a combination of red band and NIR achieved the highest correlation in this study (Figure 4.3). The DVI obtained the highest correlation with an r value of 0.89, followed by MSAVI with an r value of 0.47; these were the only indices that achieved a correlation exceeding 0.5. A weak positive correlation was obtained by NDRE, NDVI, and LCI with r values of 0.22, 0.23, 0.26 respectively. The Red edge is a region where a sharp change in spectral reflectance of green vegetation is observed and often used for chlorophyll estimation. Many studies have shown VIs constructed with red edge generally perform better than those combining red and NIR; Brewer et al. (2022) showed that NIR and red edge VIs yielded better results in estimating chlorophyll in maize. Nevertheless, in this study the VIs that use the combination of red edge and NIR were found to produce weak correlation with measured chlorophyll. The results by Ban et al. (2022) found that most indices derived from the red and NIR such as NDVI and Ratio Vegetation Index (RVI), performed better than those incorporating the red edge when estimating rice leaf chlorophyll. Another study by Wang et al. (2023) in modelling vulgaris plant demonstrated the highest correlation and most reliable performance were obtained with indices that used the combination of red and NIR.

5.3 The assessment of VIs and PLSR model on chlorophyll estimation

The study investigated the capabilities of UAV imagery data using spectral indices and in-situ chlorophyll data to assess or measure the chlorophyll content of lemon leaves. Additionally, the study aimed to explore the use of the Partial Least Square Regression (PLSR) model in chlorophyll estimation. Five vegetation indices were selected for this study namely, NDVI, NDRE, MSAVI, LCI, and DVI. These indices were selected due to their high sensitivity to chlorophyll and based on their strong performance in previous or related literature.

The correlation between VIs and measured chlorophyll was assessed, revealing a linear and positive relationship for all VIs, with the strength of relationship ranging from weak to very strong. The strongest correlations were found for DVI and MSAVI (Figure 4.2), both of which use the combination of the red band and NIR. These results have strong matches with the study by Zhang et al. (2019) which demonstrated that the red band and NIR significantly contributed to winter wheat during the maturity stage. Another study by Wang et al. (2022) that successfully utilized VIs that combine visible bands and NIR to estimate rice aboveground biomass. However, several studies have highlighted the influence of red edge in chlorophyll estimation. For instance, Shanmugapriya et al. (2022) compared NDVI and NDRE and the results proved NDRE to be more robust with the highest $r = 0.97$, while NDVI had an r value of 0.48. Another study by Ali et al. (2020) demonstrated that the red edge region has the strongest sensitivity to chlorophyll among various spectral indices. Specifically, the modified simple ratio (mSR3) and the chlorophyll red edge (Clrededge) and had the highest correlation with canopy chlorophyll, with correlation coefficients of 0.75 and 0.74 respectively. Therefore, it is essential to consider key factors that may impact the ability of an index to estimate chlorophyll content.

In this study, two predictive models based on PLSR were constructed: one using five UAV spectral bands, and the other using five selected vegetation indices. The model based on UAV bands performed slightly better as shown in Table 4.2. Zhang et al. (2023) compared PLSR and Support Vector Regression (SVR) models for grassland chlorophyll estimation and found that PLSR achieved robust results with an R^2 of 0.80. PLSR has been successfully applied in several studies as a predictive model for chlorophyll estimation and other remote sensing

studies, including rice leaf chlorophyll estimation using UAV data (Ban et al., 2022) and UAV-based chlorophyll estimation of maize canopy (Qiao et al., 2022). The PLSR technique is popular due to its ability to model relationships between x and y variables and its strong performance with collinearity among dependent variables (Ryan & Ali, 2016). Jin et al. (2013) highlighted its fundamental capability to handle these aspects effectively.

The findings show that both UAV spectral bands and VIs were optimal predictor variables of chlorophyll content, with both models achieving significant results ($R^2 = 0.85$). However, the model based on UAV bands had a lower RMSE and MAE values, making it the preferred choice for mapping chlorophyll distribution in the study area. This study showed consistency with other studies demonstrating that UAV-based indices can serve as an effective tool for chlorophyll estimation. According to the variable importance projection plot NIR, DVI, NDVI, LCI were identified as the most optimal variables for chlorophyll estimation.

5.4 The distribution of chlorophyll content

Chlorophyll is a crucial pigment that helps us understand the health status of crops, enabling early predictions or interventions regarding final crop yield. A more robust regression model that is based on UAV spectral bands was selected to generate the chlorophyll distribution map (Figure 4.6). The map output displays the estimated chlorophyll content values reflecting the chlorophyll variations measured in the field. Higher chlorophyll values represent healthy row crops, while lower values represent bare soils. These findings are useful for farmers in monitoring crop health and identification of areas that need more attention in terms of nitrogen fertilization. Such maps can be used as references or guides for faster and non-destructive acquisition of chlorophyll content.

This research provides a meaningful analysis of chlorophyll distribution in the study area and aligns with previous reports that have demonstrated the success of UAV-derived VIs in chlorophyll estimation. The PLSR model based on UAV multispectral data successfully mapped significant chlorophyll variations. The integration of UAV technology with Remote Sensing and GIS permits real-time monitoring of chlorophyll status, allowing for adjustments in crop nutrient management (Liaghat, 2010). As robust as this method is there are certain challenges remaining in estimating chlorophyll based on VIs such as soil interferences or estimation stability under different changes in the environment.

5.5 Conclusion

The current study explored the use of UAV technology integrated with conventional methods to predict the chlorophyll concentration in lemon leaves. In-situ measurements were correlated with five vegetation indices adopted in this study. The study demonstrated a significant correlation between leaf chlorophyll and UAV-derived vegetation indices. The highest correlation was achieved by DVI ($r = 0.89$), followed by MSAVI with $r = 0.47$. Both DVI and MSAVI, which utilized visible bands and NIR yielded the best correlation results with measured chlorophyll. In contrast, VIs derived from the red edge did not show a clear advantage in this research. However, the role of the red edge band remains indisputable, as many research papers have demonstrated its high sensitivity to chlorophyll in this region. The bands beyond the visible spectrum yielded significant correlation results with measured chlorophyll having r values greater than 0.4 (Figure 4.2). These spectral characteristics could be attributed to the effects of saturation by vegetation density or the leaf structure. Overall, the study's results showed the efficiency and reliability of using UAV-based VIs to estimate chlorophyll. The PLS model provided significant prediction accuracies, demonstrating its effectiveness in RS predictive applications. The resulting chlorophyll map can be instrumental for large scale monitoring, revealing stressed or unhealthy crops that may require additional fertilizer.

5.6 Review of objectives

- The main purpose of the study was to explore the usage of high-resolution UAV imagery for non-destructive and real-time estimation of chlorophyll concentration

The study successfully explored the use of UAV imagery for non-destructive and accurate estimation of chlorophyll, which is very crucial especially in the context of precision agriculture. Achieving this objective the study demonstrated the capability to estimate chlorophyll variations within the study area. These results validate the efficiency of using UAV multispectral data offering the substantial advantage of high-quality data for efficient field assessment.

- To examine spectral indices calculated from UAV imagery in predicting chlorophyll concentrations and correlate them with ground-measured chlorophyll.

The study computed and validated five spectral indices (NDVI, NDRE, MSAVI, DVI, and LCI) derived from UAV using in-situ measured chlorophyll for accuracy and reliability. High correlation was achieved by two indices: DVI and MSAVI. However, with LCI, NDVI, and NDRE showed the lowest correlation, which could be tributed to various factors such as saturation effects, environmental conditions and other influencing variables.

- To use the PLSR method to create a predictive model for the estimation of chlorophyll content.

The objective of using the PLSR method to develop a predictive model for estimating chlorophyll content was successful. Two models were created one based on VIs and another based on spectral bands. The performance metrics of these models, including the R^2 and RMSE showed high accuracy and reliability.

- Generate a chlorophyll concentration map based on the best predictive model.

The last objective was successfully achieved by generating a chlorophyll concentration map using the best predictive model which was based on spectral bands. The resultant map accurately displays the variations in chlorophyll distribution across the study area. This map provides a valuable tool for visualizing and analysing chlorophyll distribution.

5.7 Recommendations

The main recommendations are as follows.

- The first recommendation would be to explore other band combinations such as green and Near-infrared for a comparative conclusion between the influence of these bands that is the green, red, and red edge. Also to have a clearer understanding of the impact of each spectral band and to be able to justify the limitations of some indices.
- A different sampling technique, more accurate sampling technology, and a much bigger sample size will better represent and provide more accurate results for future studies.
- Lastly, further studies may incorporate different growth stages to develop more robust and comparative model variables, leading to a better understanding of the influence of VIs in various growth stages.

5.8 References

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